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# Relative distribution and biomass of invertebrates in fens and marshes in the boreal region of Northeastern Alberta

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**RELATIVE DISTRIBUTION AND BIOMASS OF INVERTEBRATES IN  
FENS AND MARSHES IN THE BOREAL REGION OF NORTHEASTERN  
ALBERTA**

By

**Kathryn L. Williams**

A Thesis  
Submitted to the Faculty of Graduate Studies  
through Biological Sciences  
in Partial Fulfillment of the Requirements for  
the Degree of Master of Science  
at the University of Windsor

Windsor, Ontario, Canada

2014

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FENS AND MARSHES IN THE BOREAL REGION OF NORTHEASTERN  
ALBERTA**

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13 December, 2013

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## ABSTRACT

Habitat selection determines the environment characteristics experienced by the individual. Arthropod assemblages are better predicted by plant community characteristics than by other environmental features. However, the role of local habitat characteristics (microhabitat structure, food) in regulating invertebrate distribution is less well known. The invertebrate fauna of northeastern Alberta's boreal peatlands and wetlands is especially poorly documented. I investigated invertebrate-vegetation associations of between and within fens and marshes, and variation across wetland hydrological zones. Family richness and biomass were greatest in wet meadow zones of marshes. Sampling instruments used to evaluate microhabitats collected complementary invertebrate types and different abundances. Vacuum sampling captured many phytophilous and soil associated fauna. Sticky traps caught mainly small-bodied, flying insects. Aerial sweep netting caught some large organisms but inadequately represented wetland biota. Overall, invertebrate composition was better predicted by vegetation zone than by hydrological regime or plant species richness within wetlands

*For my exceptional parents, Sharon and Dale Williams  
and for my big brother, Tom;  
for their enduring love and support.*

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## LIST OF ABBREVIATIONS

ANOVA= Analysis of Variance

EZ= Emergent vegetation Zone

NA= Naphthenic Acids

OSPW= Oil Sands Process Water

OSPM= Oil Sands Process Material

OW= Open Water zone

PCA= Principal Components Analysis

SE= Standard Error

SAV= Submergent Aquatic Vegetation

WM= Wet Meadow zone



## CHAPTER 1

### GENERAL INTRODUCTION

Wetlands represent a dominant feature of the boreal landscape of the northern hemisphere. Wetlands and peatlands comprise 60 – 80 percent of the area of the boreal plain region of northeastern Alberta (Rooney and Bayley 2011). The objectives of my research are to determine how invertebrate assemblages vary with respect to habitat complexity expressed as plant zonation across the hydrological gradient of boreal wetlands in northeastern Alberta. Wetland structural complexity arises from the diversity and distribution of hydrogeomorphic features that determine the distribution of a wetland's littoral and riparian vegetation (Remburg and Turner 2009). My goals are to evaluate how invertebrate (largely arthropod) community attributes (richness, biomass, abundance) vary with respect to the horizontal and vertical zonation of vegetation types.

#### ***Habitat Selection in Arthropods***

The ability of an organism to distinguish and choose among habitats is one of an organism's most important evolutionary adaptations as it determines the conditions under which it will survive to reproduce. This "habitat selection" is most commonly attributed to physical aspects of an organism's habitat. MacArthur and MacArthur (1961) demonstrated that as foliage complexity of trees increased, so did the species diversity of the tropical birds inhabiting them. This pattern has commonly been reported in a broad variety of small and motile animals (MacArthur and MacArthur 1961, Cody 1985,

McNett and Ryptra 2000). Vegetation structure refers to plants' three-dimensional arrangement and complexity that are able to support animal communities (McNett and Ryptra 2000, Warfe et al. 2008, Tokeshi and Arakaki 2012). Although ecologists often point to habitat complexity as being responsible for structuring the biotic community, often referred to as the "Habitat Heterogeneity hypothesis", there is still uncertainty about how habitat complexity mediates and regulates biological processes and brings about the pattern of species distributions seen in nature. The predictive capacity of the relationship between habitat characteristics and habitat selection is of intrinsic importance to ecologists. As such, it is necessary to determine what characteristics of vegetation are most important in mediating the patterns of diversity and abundance observed.

Studies of habitat selection have included many types of organisms including small tropical mammals (August 1982), mice (Wecker 1964), bats (Jung et al. 1999), arboreal lizards (Kiester et al. 1975) and arthropods (Southwood 1980). Vegetation is considered the most important determinant of arthropod species abundance (Schaffers et al. 2008). Arthropods are good model organisms in studies of habitat selection due to the diversity of habitats they occupy and their broad dispersal ability. Furthermore, in addition to responding to plants' physical structure, many herbivorous insect species exhibit host plant specificity (Andow 1991, Schaffers et al. 2008). The complexity of the relationship between insect diversity and their vegetative habitat led to an ecological controversy over whether insects respond to plant species diversity or to habitat complexity. The plant species diversity hypothesis predicts that arthropod abundance and diversity are positively correlated with plant species richness (Schaffers et al. 2008). Prevailing

explanations as to why plant species diversity may lead to increases in arthropod diversity are:

- 1) Diversity leads to patchiness, which in turn, uncouples the effects of overexploitation of herbivores on a lower-diversity patch (Andow 1991, Siemann et al. 1998, Schaeffers et al. 2008);
- 2) Stability of resulting herbivore populations could potentially cascade up the trophic food chain and increase in diversity of predators and parasitoids (Andow 1991, Hunter and Price 1992, Siemann et al. 1998);
- 3). Greater plant species or functional diversity can support greater productivity; this increase may also increase total arthropod abundance and allow rare species to persist locally (Tilman 1996, Siemann 1998, Wettstein and Schmid 2001).
- 4) The Taxonomic Diversity Hypothesis states that each additional plant species may have a specialized consumer (or group of consumers) (Siemann et al 1998, Brose 2003). Therefore, as plant species diversity increases, so does the potential insect taxa richness of a particular habitat.

Southwood (1980) discussed the relationship between insects and vegetation in relation to the successional sere of a forest habitat. While insect diversity was most highly correlated with plant species diversity at an early successional stage, at later vegetation stages insect diversity was strongly related to habitat complexity. This emphasizes that the question still remains as to which habitat attribute – taxonomic diversity or structural

diversity - is most important in determining patterns of insect abundance. Habitat Complexity is often described as the most important characteristic in maintaining insect diversity and abundance due to the presence of many trophic guilds (predator, herbivore, etc.) within insect taxa. Life history strategies play an important role in modulating the effects of habitat complexity (Ober and Hayes 2008). In mobile animals, for example, survival depends on habitat selection, which may depend on prey availability, host plant specificity, patch size, dispersal, an organism's trophic guild (predator, detritivore, parasitoid, etc.) or one of many other community structuring forces (Lewisohn 2005, Ober and Hayes 2008, Remsburg and Turner 2009). In order to assess the relationship between arthropod community attributes and characteristics of the vegetative landscape, it is important to choose a specific habitat that supports a taxa-rich community of insects that play an important role in habitat food webs.

Wetlands are a unique habitat that, together with riparian zones, contains elements of the ecosystem characteristics of both aquatic and terrestrial systems. Stream and wetland-derived aquatic insects provide an important subsidy to the terrestrial food web (Neiman et al. 1993). Paetzold et al. (2005) determined that predatory ground-dwelling arthropods are an important pathway for transformation of aquatic emergent insect production to the riparian ecotone bordering a stream. Subsidies from lentic and wetland systems can be equally important. For example, when Hoekman et al. (2011) placed the spent bodies of typical densities of adult midges (Chironomidae) in upland plots adjacent to an Icelandic lake, terrestrial arthropod biomass rose by 108% two years later. Hoekman et al. (2011) attributed this to a bottom-up response by detritivores and higher-order consumers to this

supplement. Wetland insects are extremely abundant during emergence events (Benke 1976) and provide food for many higher trophic order animals including vertebrates such as dabbling ducks (King and Wrubleski 1998), and aerial insectivores such as Tree Swallows (Gentes 2006) and bats (Barclay 1985).

Other studies have demonstrated a relationship between habitat complexity and secondary production in aquatic organisms (Benke et al. 1984). Secondary production is the accumulation of biomass by heterotrophs through time (Benke and Huryn 2007, Dolbeth et al. 2012). The important components of production are abundance and biomass. Secondary production in a wetland ecosystem can increase as a result of organism growth or through faunal recruitment of organisms from adjacent ecosystems. For this reason, it can be difficult to ascertain the true effect of habitat complexity on secondary production, i.e., whether increases in secondary productivity are due to plant species diversity or habitat complexity. An assessment of these relationships requires research of insect community structure measures such as diversity, abundance and biomass.

Studies of the wetland arthropod community have rarely assessed the effect of wetland zonation (presence of vegetation zones) on overall measures of production or diversity. More commonly, studies have focused on the habitat complexity of the aquatic ecotone of a wetland (reviewed by Kovalenko et al. (2012)). For example, Cremona et al. (2008) discussed the impacts of littoral plant zonation and plant architecture on the biomass of

benthic invertebrates. Though this work discussed non-emergent invertebrates, Cremona and colleagues discussed the effects of plant architecture on invertebrate biomass and abundance; the open water (submergent) zone of the studied wetland supported the greatest biomass of invertebrates due to submergent biomass of macrophytes that provide substrate for these invertebrates. Further, in a bed of emergent macrophytes, herbivores were most abundant when stem architecture was simplest (reeds) due to the increased penetration of light facilitating periphyton growth.

It is unclear what factors support riparian insect assemblages in a distinct wetland ecotone, whether this is variation in microhabitat structure, food resources, etc. In order to explain patterns of diversity and abundance that may result from vegetational characteristics, a major objective of my study is to determine how invertebrate assemblages vary with respect to habitat complexity that results from wetland zonation. Wetland structural complexity arises from the diversity of abiotic characteristics, littoral vegetation, and riparian vegetation (Remburg and Turner 2009). I evaluated how invertebrate community attributes (abundance, family richness, and biomass) varied within and among a series of northeastern Alberta boreal wetlands as a function of the horizontal and vertical zonation of vegetation types. Vertical zonation was defined as the vertical stratification resulting from plant structural diversity or complexity, primarily plant height. In contrast, horizontal zonation reflected the presence of distinct vegetation zones across a hydrological gradient.

### ***Wetland Types and Vegetation Zones***

The boreal forest of northern Alberta is part of a vast, temperate ecozone with worldwide distribution; this ecozone supports large areas of wetlands throughout its range. Boreal wetlands are estimated to cover approximately 3.8% of the world's ice-free area (500 billion ha; Vitt 1994). The extensive boreal wetlands in northern Alberta formed as a result of the actions of glaciation within the last 12000 years, through the formation of "pothole and kettle" landscape, and the restrictive permeability of glacial till soils (Batzer and Sharitz 2007).

For the purposes of this research boreal wetlands in the Athabasca Oil Sands region will be divided into 2 major groups: peatlands and marshes. The Canadian classification of wetlands divides wetlands into 5 types: shallow open water, marshes, fens, bogs, and swamps. Their characteristics depend on a number of factors including water sources (ground, surface, and/or precipitation), type of soil underlying the wetland, nutrient availability (oligotrophic to eutrophic), hydrodynamic regime (stagnant to very dynamic flow of surface or groundwater), and types and density of dominant vegetation (Zoltai and Vitt 1995, Smith et al. 2007; see Table 1.1). In northeastern, Alberta, approximately 90% of the landscape is wetlands, with fens comprising 65% of that area (Price et al. 2010, Rooney and Bayley 2011). Fens and marshes are receiving a major reclamation focus in the oil sands region.

**Table 1.1: Wetland classification based on nutrient regime, soil classification, moisture regime, dominant vegetation, and water source for dominant wetland classes in the boreal ecozone in northern Alberta. Adapted from Smith et al. 2007.**

	<b>Nutrient Status</b>	<b>Peat</b>	<b>Soil/Moisture Type</b>	<b>Dominant Vegetation</b>	<b>pH</b>	<b>Water Source</b>
Marsh	Mesotrophic -Eutrophic	No	Minerotrophic, Hydric	emergent herb-aceous vegetation	~7	Ombrogenous , Geogenous
Rich Fen	Mesotrophic	Yes	Minerotrophic, Hydric	brown mosses, sedges	7-8.5	Mainly Geogenous
Poor Fen	Oligotrophic	Yes	Minerotrophic, Hygic	<i>Sphagnum</i> sp., ericaceous shrubs	3.5-7	Mainly Geogenous
Bog	Oligotrophic	Yes	Ombrotrophic, Hygic	<i>Sphagnum</i> sp., ericaceous shrubs	3.5-5	Ombrogenous



Peatland and marsh formation occur under very different circumstances in the boreal region in northern Alberta. Marshes are a result of semi-permanent to permanent pooling of water above low permeability soils. The bathymetry of a shallow “pothole” combined with the creation of anoxic soils creates a shallow wetland surrounded by hydrophilic vegetation adapted to anoxia (Batzer and Sharitz 2007); this bathymetry creates distinct zonation across a marsh wetland (see General Marsh Characteristics, Methods for further information). Boreal marshes, therefore, are generally dominated by emergent vegetation including broadleaf cattail (*Typha latifolia*) and bulrushes (*Schoenoplectis* spp) and submergent vegetation including coontail (*Ceratophyllum demersum*) and northern water milfoil (*Myriophyllum spicatum*; Rooney and Bayley 2011). Further, the hydrological gradient from driest to most inundated results in distinct vegetation zones (as in McLaughlin and Harris 1990). The following definitions of hydrologic vegetation zones will include vegetation specific to boreal marshes in the study area: The driest zone is designated the wet meadow (WM) zone and is characterized by damp, periodically inundated soils. This zone is dominated by grasses and sedges (hence, sometimes referred to as “sedge meadows”) including *Carex aquatilis* Walenb. or *Carex utriculata* Boott. As this zone slopes towards the aquatic ecotone, transition into the emergent zone (EZ) will be characterized by flooding. *Typha* and bulrushes (*Schoenoplectus* spp.) dominate this zone as their root systems can survive in anoxia. Water depth in the emergent zone is typically between 1-100 cm (McLaughlin and Harris 1990). Water is more than 1 m deep, emergent plants are unable to grow, and this area becomes the open water zone (OW, often termed submergent aquatic vegetation zone (SAV)). Marshes in

Alberta commonly support submergent aquatic vegetation including Coontail (*Ceratophyllum demersum* L.), Common Pond weed (*Stuckenia pectinatus* (L). Boerner), and Water milfoil (*Myriophyllum sibiricum* Kom.).

Riparian areas are another region of wetlands that have received a great deal of focus in the recent past, especially in the context of stream health. Riparian areas are the interface between land and lotic systems. These areas are not discussed in the breadth of this research but are vitally important ecotones in their role in habitat biodiversity. The Athabasca River basin contains many thousands of km streams bordered by riparian vegetation, and these areas should be a reclamation focus in the future. Detailed review of riparian communities are provided by Pusey and Arthington (2003) and Naimen et al. (1993).

Fens occur in cool temperate areas in a depressional landscape where glacial mineral soils are situated above groundwater upwelling. This mineral rich water that bathes the plant-rooting zone supports a community made up of bryophytes, sedges, grasses, dicotyledonous herbs, and (stunted) coniferous trees (Vitt 1994, Batzer and Sharitz 2007; see General Fen Characteristics, Methods for more detail). Plant primary production exceeds decomposition leading to the accumulation of organic matter (carbon) and the creation of peat due to overall temperate environments and as a result of root-zone anoxia (Smith et al. 2007).

### ***Disturbance in the Athabasca Oil sands region - Surface Mining***

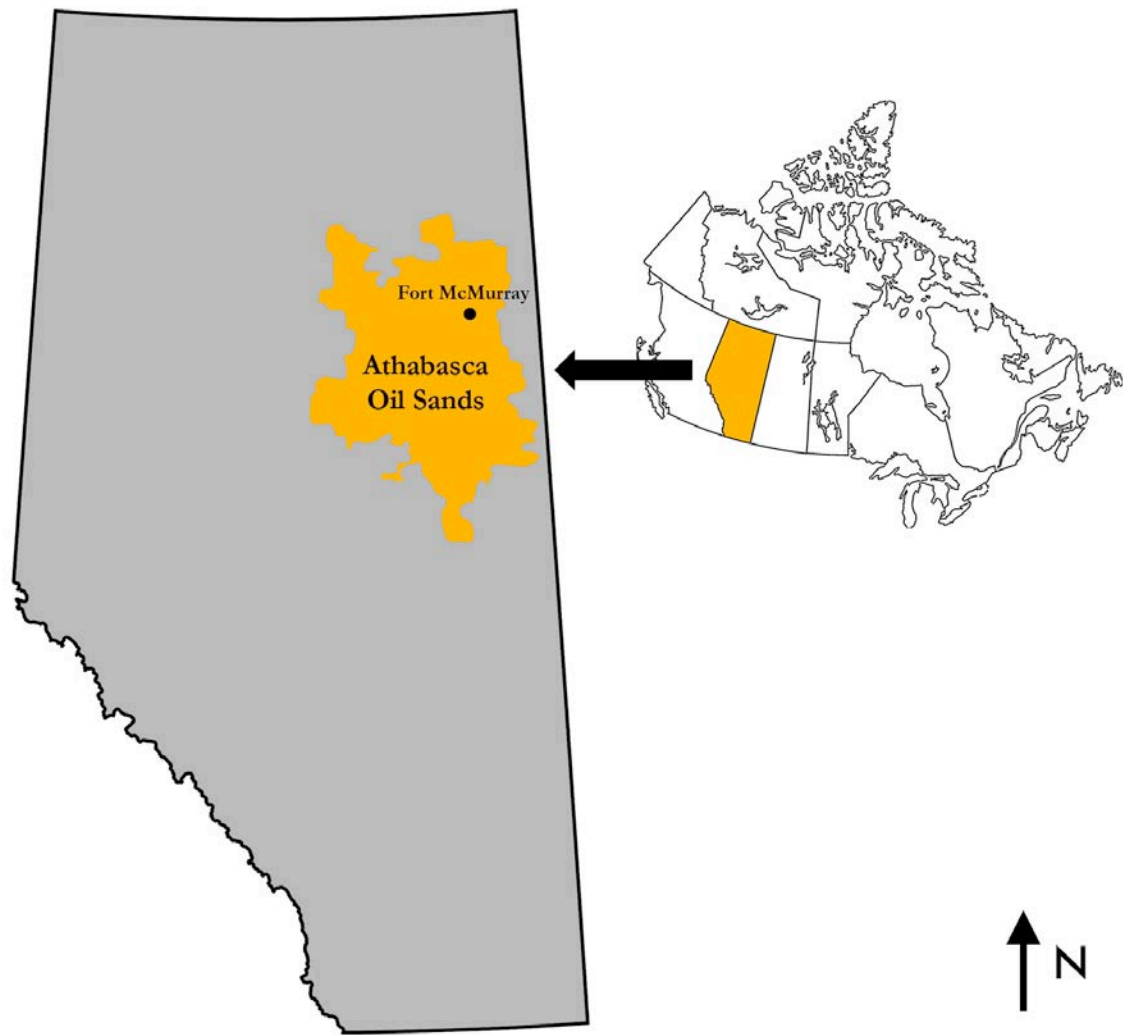
The extent to which habitat complexity can be used to predict other components of the food web has important implications with respect to efforts to restore and reclaim ecosystems that have been subject to extensive disturbance (mining, logging, etc.; Rooney and Bayley 2011). This is also true for the estimation of invertebrate biomass as a criterion for reclamation ‘success’ as for other food web components. If the goal of reclamation is to produce systems that can sustain pre-disturbance biota (Wray and Bayley 2006, Rooney and Bayley 2011), ecologists must determine if restoring natural vegetation will aid in this endeavor (Wray and Bayley 2006, Price et al. 2010).

Oil sands surface mining in the Athabasca region north of Fort McMurray, Alberta is a major source of environmental disturbance. The mining process initially involves stripping the landscape of standing vegetation and surface soils to expose the oil- rich bitumen beneath. This “overburden” material is stored in piles for later use in reclamation (Price et al. 2010).

Research into wetland reclamation in the Athabasca Oils sands region has focused on the creation of marshes (Price et al. 2010); marshes only constitute approximately 3% of the boreal region naturally, far less of a constituent than fens (Rooney and Bayley 2011). Previous reclamation efforts have emphasized the creation of “end-pit lakes” that would both facilitate storage of oil sands process water (OSPW), and fulfill the requirements of the Alberta Environmental Protection and Enhancement Act (AEPEA) to restore the land to “equivalent land capability” (OSWWG 2000).

Recently, however, the AEPEA adapted conceptual models for the creation of peatlands into its mandate and recommended that oil sands lessees reclaim these important wetlands to the best of their ability (Price et al. 2010, Vitt and Bhatti 2012).

Conventional knowledge of how fens form naturally suggests that the creation of a fen would take thousands of years (Vitt and Bhatti 2012). However, Price et al. (2010) proposed a fen model that, conceptually, could sustain a reclaimed fen by placing organic matter from overburden, in a contoured landform with hydrological properties that could “sustain the requisite wetness”, and planting techniques to re-establish the plant community (Price et al. 2010).



**Figure 1.1 Map of Alberta, Canada, showing the Athabasca oil sands deposits.**

The presence of fen landscape is an important element if the post-mining landscape is to retain regional biodiversity. If marshes and lakes comprised the only classes of aquatic ecosystem in a reclaimed landscape species of birds (Calme et al. 2002), amphibians (Mazerolle 2003), and large ungulates (moose and caribou, for example; Bradshaw et al. 1995) would be subject to habitat loss. Furthermore, peatlands are home to several species of provincially rare plants that occur only in these habitats including the purple pitcher plant (*Sarracenia purpurea*), and fen orchid (*Laparis loeselii*; Smith et al. 2007, Locky et al. 2005).

### ***Current Knowledge of Wetland Arthropods***

As previously stated, wetland arthropods and other invertebrates are a significant component of wetland productivity. Wetland arthropods subsidize energy from the aquatic to terrestrial ecotone through their position in the food web and as emergent biomass (Neiman et al. 1993). Marsh invertebrates are adapted to persist under a variety of adverse conditions that can periodically affect a marsh environment including low oxygen levels (Batzer et al. 1997), desiccation due to drought (Wiggins et al 1980), and stressful water quality parameters (Batzer et al. 1997). Beyond that, they occupy a wide range of trophic levels (Welborn et al. 1996, Batzer et al. 1999). In fact, many invertebrate orders typically occupy multiple positions in the foodweb of individual wetlands [e.g., midge larvae (Diptera: Chironomidae) can be both predaceous as in Tanypodinae species, suspension-feeding as in many Tanytarsini, “grazers” as in *Chironomus* sp., and detritivores (Orthocladinae); (Clifford 1994)].

Marsh habitats are generally considered transitional habitats between aquatic and terrestrial ecotones. As such, spatial heterogeneity within wetlands requires careful experimental design in order to interpret the results of bioassessment (Batzner et al. 1999, Benke et al. 1999, Gerth and Herlihy 2006). Due to the nature of a marsh – presence of multiple hydrological zones and microhabitats – sampling must account for the presence of these vegetation structures and their influence on the distribution of invertebrates (Innis et al. 2000). As previously stated, structural heterogeneity enhances invertebrate abundance and diversity (Southwood 1980, Frambs 1994). This relationship also holds true in wetland habitats (Benke et al 1999).

Research on the ecology of peatland arthropods is less well known (Danks and Footitt 1989), and is almost nonexistent in the context of habitat selection or reclamation. Although the boreal forest has worldwide distribution, habitat features that are unique to Canadian peatlands make comparison across its distribution difficult; these features include climate, geology, and soil type (Danks and Footitt 1989). Records of seminal work in Canadian peatlands include special issues of the *Memoirs of the Entomological Society of Canada* in 1987 and 1994; a 1989 issue of *The Canadian Entomologist* by Danks and Footitt entitled, “*Insects of the Boreal Zone of Canada*”; and, “*Insects of Boreal Peat Bogs*” by Spitzer and Danks (2006).

Danks and Rosenberg (1987) reviewed the literature and discussed the general trends in peatland invertebrate diversity; invertebrates are less abundant in peatlands than

elsewhere due to the nature of these extensive habitats. The combination of a limited nutrient regime and dissolved oxygen deficit in water and hygric soils of a peatland prevents the growth of bacteria and subsequently accounts for peat accumulation. This in turn limits the accumulation of invertebrate biomass (i.e. secondary production). Furthermore, Danks and Rosenberg (1987) related the unavailability of food, nutrient, and oxygen to unpredictable conditions and harsh environments such as the high arctic or temporary pools. They listed possible invertebrate adaptations to these unfavourable conditions. The lack of such adaptations would explain invertebrates' limited abundance and/or biomass in peatlands.

- 1) When nutrients are limited, some species may only grow to small size.
- 2) If conditions within the peatlands are heterogeneous, invertebrates may be patchily distributed, which would affect detection, enumeration, and biomass estimation;
- 3) Many invertebrates avoid adverse conditions via the timing of their development. Life history strategies can include periods of dormancy at times of food shortage, adverse climate, or to avoid desiccation; long life cycles that allow for slow growth during scarcity; and rapid development during the most suitable conditions. Any of these adaptations may make detection difficult during studies to estimate invertebrate abundance or biomass.

There are differing ideas in the literature as to how community composition in peatlands will reflect oligotrophy. Danks and Rosenberg (1987) suggested that in oligotrophic wetlands, including peatlands, a relative absence of herbivores will lead to significantly altered community composition (Mason and Standen, 1983, *in* Danks and Rosenberg



1987). This altered community composition would limit specialized competitors in favour of extreme generalists (Danks and Footitt 1989). In contrast, Spitzer and Danks (2006) summarized the community composition of bog-type peatlands and describe “Tyrphobiontic” species (those restricted to bog habitats) as being extreme specialists, namely, herbivores have become adapted to use the minimal nutrient value of ericaceous vegetation that dominates peatlands. These specialist herbivores include aphids (Hemiptera: Aphididae) and several species of butterfly and moth larvae (Lepidoptera). Specialist predators include species associated with (fen/bog) pools including dragonflies (Odonata: Aeshnidae, Corduliidae) and backswimmers (Hemiptera: Notonectidae). However, the majority of aquatic insects found in peatlands are not known to be specialists. Less frequently, terrestrial insect predators are tyrphobionts. Some ground beetles (Coleoptera: Carabidae) are specific to peatlands (Frambs 1994, Spitzer and Danks 2006). Frambs (1994) documented the role of the habitat heterogeneity hypothesis in driving ground beetle distribution within peatlands. These generalist predators are able to survive adverse conditions by relying on the “hummock-hollow” complexes, which concentrate prey species within these structurally complex microhabitats. Also, these ground beetles hibernate within the hummocks.

Censusing the distribution and abundance of invertebrates in wetlands can be challenging because of the numerous microhabitats available. Collection techniques for invertebrates in marshes have been review reviewed by several authors (e.g. Turner and Trexler 1997, Batzer et al. 2001, Anderson et al. 2013). Benke et al. (1999) noted that the presence of hydrological zones in marsh wetlands makes sampling complex and that often, more than

one technique is required to sample the entire community. Anderson et al. (2013) recently published a volume that reviews sampling methods for wetlands. These authors describe the benefits and limitations of each method's effectiveness within wetland types and between zones of marshes. However, fens lack the distinct zonation of marshes, and prior quantitative research on the study area is limited. Sampling techniques commonly used in the submerged aquatic vegetation and emergent areas of marshes often sample include D-frame nets, Ekman grabs, and benthic corers to sample the standing biomass (Benke et al. 1999). However, these techniques will not adequately address the terrestrial transition within wetlands, which is a major disadvantage.

Sampling in fens has generally been qualitative in nature, seeking to document the presence or absence of taxa (e.g. Aitchison-Benell 1994, Blades and Marshall 1994). Fens are often sampled with pan traps (Aitchison-Benell 1994, Blades and Marshall 1994, Finnamore 1994), and pitfall traps (Aitchison-Benell 1994). Research on the most suitable quantitative methods to assess invertebrates in peatlands is lacking.

When the ecological questions addressed within this research were framed, I considered the habitat, types of invertebrates likely to be present, and the commonly used techniques that habitat and faunal type suggested. Because peatlands are commonly compared to terrestrial habitat, and because the study sites were homogeneous, graminoid fens, I postulated that fen arthropods would be mainly terrestrial/ground dwelling. When Doxon et al. 2011 compared common collection techniques for terrestrial invertebrates (sweep

netting and vacuum sampling), they concluded that both sampling methods had merit depending on the size of organisms likely to be collected and well as structure and complexity of vegetation. My research endeavoured to evaluate invertebrate community measures including abundance, richness, and biomass. Consequently, I used both methods (vacuum samplers and sweep nets) to evaluate fen and marsh communities

### ***Research Objectives***

This thesis is organized into four chapters. The General Introduction identifies the broad hypotheses of habitat selection and explains the rationale behind the research project. Chapter 2 describes the distribution of arthropods collected in and above vegetation at various distances above the substrate in two classes of constructed wetland plots containing vegetation characteristic of fens and of marshes. The third chapter examines patterns of invertebrate abundance, family richness, and biomass as a function of vertical stratification, and horizontal zonation of natural fens and marshes. Chapter 4, provides general discussion, conclusions and identifies questions that should be addressed in future research. The studies were designed to address the following general research questions:

**1. How does invertebrate distribution vary with respect to vegetation type (Chapter 2) and with respect to vertical plant stratification (Chapter 2)?**

To address this question, we placed sticky traps in constructed wetland plots with fen or marsh vegetation as well as at varying heights with respect to plant vertical stratification.

**2. How does invertebrate distribution vary with respect to horizontal zonation (hydrological vegetation gradient) within wetlands? (Chapter 3)**

To address this question, we used sticky traps to sample arthropods in submergent aquatic vegetation, emergent vegetation and wet meadow zones of marshes.

**3. What zone of a marsh is the most productive (Chapter 3)?**

I used sticky traps to assess the abundance and relative biomass of flying insects within the submergent, emergent and wet meadow zones of marshes.

**4. Does fen arthropod community composition and biomass differ from that of equivalent habitat in marshes (Chapter 3)?**

I collected flying insects using sticky traps, plant-associated arthropods with an aerial sweep net, and phytophilic and soil-dwelling invertebrates using a vacuum sampler in 4 natural fens and 10 natural marshes. I analyzed these samples to determine how abundance and diversity of insects varied with respect to wetland type.

**4. What metrics of the vegetative environment best predict patterns of biomass and richness in invertebrates (Chapter 3)?**

To assess this relationship, plant community metrics were correlated with invertebrate biomass and richness.

In general, I expected to find marked differences in invertebrate community attributes (distribution, abundance, richness, biomass, etc.) between fens and marshes. Marshes are

productive habitats, with varied amounts of biomass present in each of the hydrological vegetation zones. Furthermore, I expected invertebrates to be distributed among zones in a manner that reflects their life history strategy or role within the wetland (e.g., herbivores were expected to be most abundant in the emergent zone, utilizing emergent vegetation as a food resource).

I undertook this investigation by studying natural fens and marshes as well as constructed wetland plots in the oil sands region of northeastern Alberta.

### ***Materials and Methods:***

#### ***Study Area and Wetlands***

My research was undertaken in a portion of northeastern, Alberta in the vicinity of the town of Fort McMurray. This region is part of the circumpolar boreal forest that covers much of northern North America and Eurasia. The landscape is composed of poplar, aspen, black spruce, and jack pine forest, transected by numerous streams and rivers that run into the Athabasca River, which flows northwards into Lake Athabasca (Smith et al. 2008).



**Figure 1.2: Satellite image of study area**

The study area includes regions of extensive anthropogenic disturbance as a result of surface mining. It is the province's largest and most accessible source of crude oil. Presently, surface mining leases north of Fort McMurray, Alberta, cover 663 km<sup>2</sup>, and are expected to eventually cover 4800 km<sup>2</sup> (FTFC 1995). The surface mining process involves removal of the forest, peat, and overburdens that overlie the oil sands deposits, as well as draining extensive wetland and peatland ecosystems that cover 65% of the region. In the post-mining landscape, oil sands lease holders are required to reclaim the landscape to where "the ability of the land to support various land-uses after conservation and reclamation is similar to the ability that existed prior to an activity being conducted on the land, but that the individual land uses will not necessarily be identical," (GOA, 1993). In light of this, there is an urgent need to identify science-based reclamation goals.

In the summers of 2011 and 2012 invertebrate sampling was undertaken to determine the relationship between influences of wetland type and wetland plant zonation on the community composition and biomass of invertebrates.

### ***General Fen Characteristics:***

Fens are a particular group of peatlands that constitute the predominant wetland type in the northern boreal forest in Alberta. Fens are one of the five recognizable wetland classes, (the other classes being marshes, bogs, swamps, and shallow open waters (Zoltai and Vitt 1995)). They are differentiated from these other wetland classes on the basis of

hydrologic regime, climate, mineral level, and water chemistry. These abiotic factors will influence the wetland biota to develop characteristic forms of vegetation cover, and in the case of fens, form peat. Peat is created over long periods of time in areas where primary productivity exceeds decomposition in cool temperate climates. More precisely, these characteristics are combined with stable, lentic waters whose limited flow will allow layers of bryophytes to develop over time in the presence of slow decomposition. Thus, over time, thick mats of peat will form (Zoltai and Vitt 1995). My study sites included both rich and poor fens. Rich fens are dominated by brown mosses, have a neutral pH, and are generally mesotrophic. The availability of nutrients may allow some vascular plants to grow in these wetlands. However, brown mosses and some grasses most often dominate rich fens. Poor fens are acidic and are dominated by acid tolerant mosses of the genus *Sphagnum* (sphagnum mosses). The acidic nature of these wetlands limits vascular plant growth. Although rich and poor fens differ in water chemistry, they share the common hydrologic characteristic of receiving minerotrophic geogenous water (and further reliance on upland waters) (Zoltai and Vitt 1995). From a habitat standpoint, both rich and poor fens tend to be spatially homogeneous across their expanse. They are characterized by large patches of moss and grass-dominated peat-hummocks, lack of horizontal relief, and relatively little vertical stratification (pers. obs.). Most notably, the vertical profile of the moss and grass layer is highly complex and allows the postulate that this profile would contain the majority of invertebrate biomass found in a fen-type wetland.

Fens will also be described in this study in terms of the dominant vegetation that is present. Graminoid fens are dominated by grassy and herbaceous plant species. A



patterned fen is a shrub and herbaceous plant dominated peatland characterized by a series of strings and flarks (peat ridges and hollows, respectively) (Vitt et al. 1975). Fens were characterized based on plant species present during data collection for *A Report on Fens of the Oil Sands Region* (Golder Associates 2011) or by personal observation.

In order to inventory invertebrates in each fen, as well as to estimate biomass within this rarely studied wetland type, I used 3 sampling methods during the summer of 2012 - sticky traps, aerial sweeps using a sweep net, and vacuum sampling (for detailed sampling methodology, refer to the general methods section)

### ***General Marsh Characteristics***

Marshes belong to another of the 5 most recognized classes of wetlands. These are high-nutrient, treeless wetlands whose seasonal water levels fluctuate greatly depending on the water table and precipitation events. Due to the presence of high levels of nutrients (nitrogen and phosphorus), primary productivity is relatively high. Because decomposition rates are also high they often do not form peat (Zoltai and Vitt 1995). In this study, marshes were divided into 3 vegetative zones based on the hydrological limitations (as in Voights 1976). From the driest to the most inundated, these zones are the wet meadow zone, the emergent vegetation zone, and the open (SAV) water zone (see above). All marshes sampled in this study were non-saline, alkaline, inland water wetlands. Also, with the exception of 3 wetlands (U-shaped Cell Test Plots, Wapisiw Marsh and Shallow Wetland), all wetlands used in this research formed naturally. The U-

shaped cell test plots are study plots that were constructed on Syncrude's Mildred Lake Lease, Alberta (Syncrude Canada, Ltd.; see further information in site description, below). Wapisiw Marsh is situated in the location of the first tailings pond (Suncor Pond 1) to be reclaimed on Suncor Energy's Lease adjacent to the Athabasca River (please see site description, below). Shallow Wetland is a constructed wetland on Syncrude's Mildred Lake Lease. This wetland was constructed without oil sands process materials (i.e. tailings, process water, etc.) and is therefore referred to as "reference" (Syncrude Canada, Ltd.; see further information in site description, below). Study sites were described in terms of their location, whether these were on the Syncrude Mildred Lake Lease, on the Suncor Lease on the Athabasca River; or offsite (i.e. not on land held by lease-holders).

### ***General Methods:***

#### ***Pilot Studies- May-August 2011***

In 2011, I conducted pilot studies on a series of boreal wetlands to become familiar with the study area and determine what sampling techniques would be most effective. I used several methods that would both permit me to make comparisons across zones and collect enough material to assess community characteristics within each zone. I sampled with benthic cores (Bologna 2006), emergent hoops (Swisterski et al. 2001), aerial sweep netting (Calver 1982), and sticky traps (King and Wrubleski 1998, Leonhardt 2003). In the 2012 field season, I sampled with aerial sweep nets, sticky traps, and a vacuum sampler (Hoekman et al. 2011; see below).

### ***May-August 2012 Field Season***

To determine which zone supports the greatest biomass of invertebrates, I sampled in each of the three marsh vegetation zones (wet meadow, emergent, and open water) using sticky traps, aerial sweeps, and vacuum sampling in 9 marshes and 4 graminoid fens.

## CHAPTER 2

### FLYING INSECT DISTRIBUTION RELATIVE TO FEN AND MARSH VEGETATION IN THE OIL SANDS REGION OF ALBERTA

#### *Introduction:*

Boreal wetlands are biologically and structurally diverse, reflecting the source of the water, nutrients, and topography that sustains them (Vitt and Bhatti 2012). The bathymetry of Alberta's "pothole and kettle" landscape, which sits on relatively impermeable glacial till creates anoxic soils that underlie semi-permanent pools of water that become surrounded by hydrophilic vegetation adapted to anoxia (Batzner and Sharitz 2006). This bathymetry creates distinct zonation that characterizes marsh wetlands (see General Marsh Characteristics in general introduction for further information). Fens occur in depressional landscapes where glacial mineral soils are situated in areas of groundwater upwelling. The upwelling provides a mineral-rich root zone for a community of plants adapted to low-nutrient conditions. Peat gradually forms in these wetlands (fens) when primary production exceeds decomposition due to anoxia (Smith et al. 2007). Naturally-forming wetlands of the boreal region can be categorized into 2 types: Marshes and peat-forming wetlands, including bogs and fens (Smith et al. 2007). Bogs are not common to the boreal region of northeastern, Alberta. However, the majority of peat-forming wetlands in Alberta's Boreal region are fens. Fens and marshes support different plant communities, reflecting differences in location of the water table in each. Comparative studies have found that the wet meadow zone of a marsh is hydrologically most similar to fens (Holmquist et al. 2011), resulting in similarities in

vegetation type (characteristic graminoid vegetation), and moist, minerotrophic soils (Garono and Kooser 2001). Holmquist et al. (2011) for example, compared the relative proportion of aquatic and terrestrial insect fauna between montane wet meadows and fens in the Sierra Nevadas and determined that these wetlands (zones) can support a similar assemblage of insects; terrestrial fauna were dominant in both of these habitats.

Similarities between fens and the wet meadow zone of marshes allow us to make predictions from one to the other. However, plant community composition can be markedly different, overall (See General Fen and General Marsh Characteristics, General Introduction). Natural fens are rich in plant species but lack the hydrological zonation that characterizes marshes, making the latter more structurally diverse. My objective in this chapter was to investigate how differences in vegetation types that characterize marshes vs. fens may affect attributes of the arthropod community. The distribution of flying insects among vegetation zones of marshes has been documented (see McLaughlin and Harris 1990, King and Wrubleski 1998). However, little data is available addressing the vertical distribution of arthropods within those zones.

Vertical stratification is herein defined as the availability of vertical space as well as the diversity of foliage height within a vegetation zone. The use of vertical structure has been widely researched in forest habitats for a wide range of taxa including birds (MacArthur and MacArthur 1961, Cody and Walter 1976, Wiens and Rotenberry 1981), bats (Hayes and Gruver 2000) and insects. A vertical component of habitat selection in

forests, referred to as “foliage height diversity”, often describes vegetation complexity through the comparison of the vertical distribution of canopy layers. Bird species diversity, for example, is positively correlated with the degree of plant structural complexity as a result of foliage height diversity (MacArthur 1964, Karr 1968). Although this relationship is well established for temperate forest ecosystems (MacArthur et al. 1962, Wiens et al. 1986, etc), wetland habitats have not received as much attention. Furthermore, the terminology that is used to describe “the availability of plant structures for animals” varies greatly from study to study, making comparisons of findings difficult (see McCoy and Bell 1991).

Habitat level patterns in vegetation are the most important determinant of insect abundance and diversity (Schaffers et al. 2008). Both habitat heterogeneity and plant species diversity have been identified as regulators of insect community composition, richness and biomass. However, the relative importance of each is a continuing topic of discussion. Schaffers et al. (2008) summarized the importance of the interaction between these two characteristics of plant communities: The key component of habitat complexity is the diversity of plant structures available to an arthropod. Consequently, the diversity of plant structures is expected to be positively correlated with plant species diversity (Frambs 1994). Schaffers et al. (2008) argued that that because few studies assess plant community composition in detail, insect diversity and abundance more often seem to correlate with plant complexity than with measures of diversity (e.g. richness.

Insects display patterns of diversity and abundance similar to the diversity of plant structures in forests (i.e. foliage height diversity). For example, vespid wasps (Hymenoptera: Vespidae) in temperate deciduous forests respond to difference in prey availability (caterpillars), which are controlled by vegetation complexity resulting from forest “layers” (Ulyshen et al. 2011).

Batzer and Wissinger (1996) reviewed the mechanisms by which wetland vegetation zones are maintained by a wetland’s hydrological gradient. They documented that the aquatic ecotone of a wetland (the boundary between the emergent zone and the open water zone) supports the greatest biomass of emergent insects. Less is known about the effects of habitat features influencing the insect assemblages of peatlands. Research on marsh and fen arthropod communities has focused on questions of biodiversity or distributional patterns of specific taxa. Specific groups studied in peatlands have included spiders and other arachnids (Aitchison-Benell 1994, Dondale and Redner 1994, Koponen 1994), odonates (Canning and Cannings, 1994), cicadellid homopterans (Hamilton 1994), sphaerocerid flies (Marshall 1994), and Hymenoptera (Finnamore 1994). Frambs (1987), however, studied ground beetle distribution (Coleoptera: Carabidae) to test the Habitat Heterogeneity hypothesis in peatlands of Sweden; New York state; and Maine. He concluded that vertically heterogeneous peatlands - those in which hummocks and hollows formed in *Sphagnum* beds- supported larger populations of carabid beetles than peatlands with little topography, likely due to beetles’ lower risk of detection by predators. McElligott and Lewis (1996) reported that the presence of hollows and hummocks in Labrador peatlands accounted for greater abundances of horse

and deer fly larvae (Diptera: Tabanidae). Mature larvae that moved into hummocks prior to pupating avoided drowning in flarks that became submerged by accumulating water.

Insects are ubiquitous, short-lived, and quickly populate a newly created wetland environment, making them excellent subjects for wetland assessment. Their wide array of life history traits, and breadth of ecological niches allows reclamation scientists to easily determine if specific ecosystem functions are being met by reclamation efforts (Garono and Kooser 2001).

This study was conducted to determine,

- a) whether and how flying insect assemblages vary according to the dominant vegetation that is characteristic of fens *versus* marshes and,
- b) whether differences in height and vertical structure of fen vegetation relative to wetland emergent vegetation affects the distribution of insects above and within vegetation.

#### *Predictions:*

Marsh hydrology supports a greater diversity of vegetation structures than fens. I expect that because marshes have more heterogeneous vegetation structure (short wet meadow plant species, tall emergent vegetation, and submergent aquatic vegetation) marsh wetlands will support greater arthropod richness and biomass than fens. The vegetation pattern of fens is structurally most similar to the terrestrial ecotone of a marsh (wet



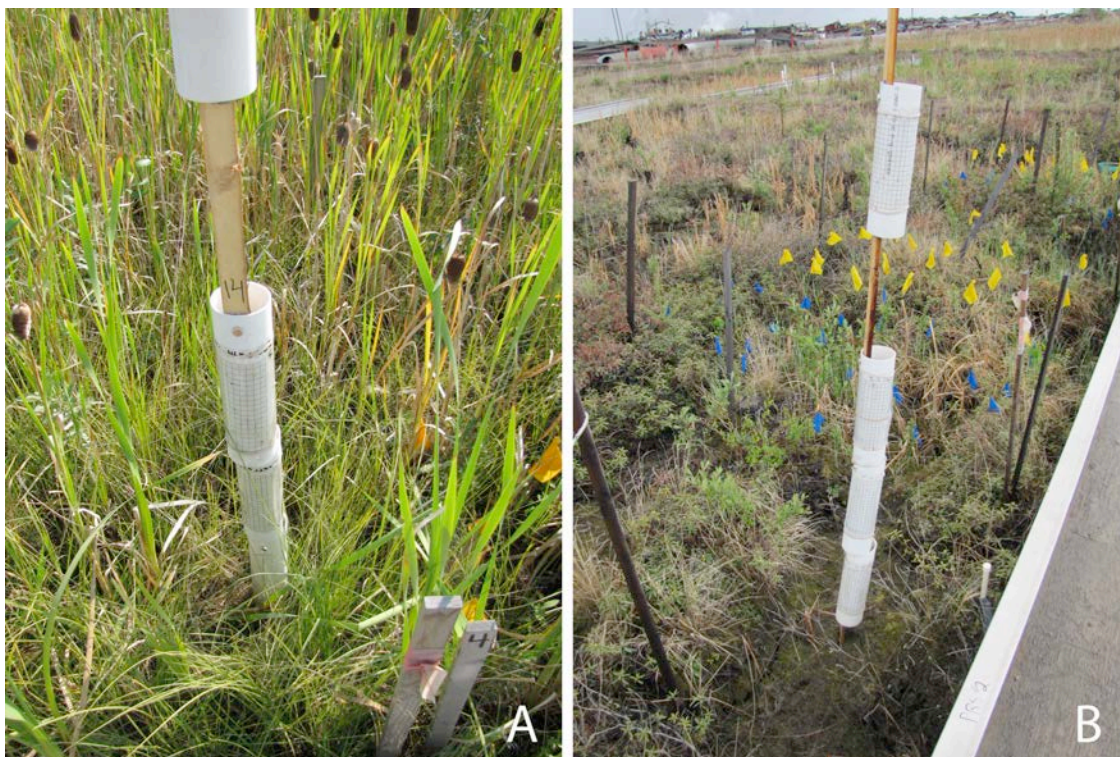
meadow zone). Both fens and marsh wet meadow zones are dominated by graminoid vegetation and exhibit similar spatial homogeneity (Holmquist et al. 2011). As such, I predict that fen wetlands will not support as rich a community of arthropods as would be found in marshes that support submergent aquatic vegetation, emergent vegetation, and wet meadow zones. Furthermore, I expect that the greater degree of vertical zonation provided by tall (emergent zone) plants characteristic of marshes will also lead to greater taxa richness and biomass in marsh vegetation than in fens. I also expect to find greater abundance of aquatic organisms in marshes than in fens.

### ***Materials and Methods:***

#### *Sampling Site- “U-Shaped Cell”, Syncrude Canada Ltd., Mildred Lake Lease*

To examine flying insects’ vertical distribution and use of contrasting wetland vegetation types, I placed passive “sticky trap” samplers (King and Wrubleski 1998) in twenty-eight 10 x 20 m experimental wetland cells created in 2008 (ref? Vitt?). At the time of sampling, cells were 4 years old. Experimental cells were created using substrate from one of 2 sources: living or stockpiled peat. Mats of living fen vegetation and the soil (peat) upon which there were growing was excavated and transported intact from a nearby fen. ‘Stockpiled peat’ consisted of surface soil that had been removed from peat-forming wetlands 10-15 y prior to the wetlands’ excavation for surface mining (BCG Engineering Company, Inc. 2009).

Plot types differed in plant composition. The vegetation of the live peat plots survived the transplant process and was representative of the composition of a peat-forming wetland (natural rich fen, see General Fen Characteristics, Chapter 1). In contrast, stockpiled peat plots lacked a viable seed bank. Consequently, they were colonized by prevalent “weedy” wetland plants (see General Marsh Characteristics, Chapter 1), characteristic of the emergent and wet meadow zones of marshes (see figure 2.1). Of the 28 cells, 12 cells were “fen” cells and 14 cells were “marsh” cells. The remaining 2 cells were “reference” as they did not contain peat amendments.



**Figure 2.1: In situ experiment design. A) photograph of sticky trap installed in marsh-type cell (Stockpiled peat) in May 2012. B) sticky trap installed in fen-type cell (live peat) showing differences in vegetational composition.**

The trap design was based on the units developed by Leonhardt (2003). Sticky traps were constructed using 7.6cm (3-inch) diameter polyvinyl chloride (PVC) piping, cut into 30-cm long sections. Sections were attached to a 125 cm tall x 5 cm wide wooden stake by a pair of threaded bolts at four heights above the substrate: 5-35 cm, 35-65 cm, 65-95 cm, and 125-155 cm. The trapping surface consisted of a clear, acetate overhead transparency with a 1 cm x 1 cm grid photocopied onto it, painted with Tanglefoot (Tanglefoot Company, Grand Rapids, MI), a natural plant resin. A transparency was wrapped around each PVC tube and secured with rubber bands (Fig. 2.1). I placed one stake (with 4 traps on it) in each cell for 3 fair weather days, following the recommendations of Leonhardt (2003). The transparencies were subsequently removed from the PVC tubing, covered in plastic film and stored frozen until processing in the laboratory.

Placement of a sticky trap within the cell was constrained by the distribution of boardwalks across experimental plots. Non-destructive sampling of plots required that sticky traps be within reach of these boardwalks to avoid the necessity of stepping into the cells. Furthermore, other sampling techniques (e.g., aerial sweeping) were not permitted, to minimize disturbance that might influence other experimental studies being conducted in the plots. Readings of meteorological information including air temperature, wind speed, and precipitation were available from a meteorological station set up in the center of the experimental plot field (Figure 2.2). Plots were watered daily to maintain peat saturation. However, the cells did not contain standing water capable of supporting truly aquatic fauna.



**Figure 2.2: Aerial photo of experimental field. Circle indicates position of meteorological station. Rectangle indicates a cell boardwalk (2 boardwalks in each 10 x 20 m cell). Note the white pickup truck in the lower left corner for scale.**

The sticky-trap tube heights were chosen to assess flying insect activity over and within the vegetation of both marsh and fen plants. The 5-35 cm tube sampled “within fen vegetation” insects, the 35-65 cm tube was “above fen vegetation”, the 65-95 cm tube was “within marsh vegetation”, and the 125-155 cm tube was situated “above (marsh) vegetation”. Marsh-cell vegetation consisted largely of tall, emergent cattails (*Typha latifolia*). At the time of this study, no cattails were taller than 125 cm. In contrast, the tallest vegetation in the live-peat cells was shorter than 35 cm at the time of sampling. Although these cells originally had contained some black spruce shrubs (*Picea mariana*) the trees did not survive the transplantation process.

#### *Sample Handling and Processing*

Sticky trap samples were transported frozen and upright, to avoid compression, in filing boxes from the field to the University of Windsor. In the laboratory, insects were removed from sticky traps by submersing the acetate sheets in B-X Safety Solvent (Bird-X Inc., Chicago, IL), which dissolved the Tanglefoot. The insects were rinsed in hexane, which is miscible with both polar and non-polar solvents, and transferred to 95% ethanol, which serves as a preservative.

Samples were processed in stratified-random order to minimize possible identification bias based on wetland plot type. Trapped insects’ exposure to the elements over the deployment period of 72 h resulted in a significant number of insects being damaged.

Consequently, I limited identifications to the family level, using the keys of Marshall (2007).

Life history (or membership in wetland insect categories) was also determined using knowledge of insect family life history. Categories were descriptive of an arthropod's relationship to a wetland. The categories: aquatic, transient, resident and soil inhabitant describe how an arthropod uses a wetland habitat.

1) Aquatic, those taxa that are obligately associated with a wetland habitat due to spending at least one phase of their life (most often larval) in a wetland habitat (e.g. chironomid midges (Diptera: Chironomidae)).

2) Transient taxa are those taxa that are not tied to a wetland environment but are “passing through” in search of food resources, or further away mating/oviposition sites (e.g. black flies (Diptera: Simuliidae) do not develop in wetlands.. Their larvae develop only in running water).

3) Herbivorous insects do not necessarily require a wetland habitat, but utilize the dense vegetation that is characteristic of wetland habitats (e.g. aphids (Homoptera: Aphididae)).

4) Soil/Peat resident taxa are those that live on or within the soil stratum, these may or may not be tied to a wetland habitat. Those dependent on wetland habitat may be Diptera or Coleoptera larva. Taxa that may not be tied to a wetland may be certain taxa of mites (Acari: Oribatidae, Prostigmatidae, etc.).

Because the larval and adult stages of many insects differ in their habitats or feeding behavior, I assigned membership on the basis of the life history of larval stages (generally the longest life stage). Saprophagous insects were placed in “soil” category (Table 2.1). These arthropod categorizations will aid analysis of data for patterns within wetland types and among wetland vegetation.

Biomass of identified arthropods was estimated by image analysis. Specimens were placed under a dissection microscope equipped with a digital camera. Magnification was adjusted so that an insect took up 20-50 percent of the diameter of the field of view beside a 10-mm scale bar, which was subsequently used as a frame of reference for calibration. Photos were digitized using SPOT advanced 5.1 software (SPOT Imaging Solutions, Sterling Heights, MI). Damaged specimens were not photographed for accurate future estimation of biomass using body size metrics.

ImageJ 1.47 for Mac OS X software (U. S. National Institutes of Health, Bethesda, Maryland, USA) was used to digitally determine body length measurements from images. Images were measured from the most anterior portion of the head to the anus. Appendages (including antennae, mouthparts, cerci, genitals, ovipositors, etc.) were not included in the measurements.



### *Statistical Analysis*

Statistical analyses were performed using STATISTICA 7 software (Statsoft Inc., Tulsa, OK). Family richness, abundance, and biomass data were input into statistical software files and were expressed as means to eliminate error due to loss of traps due to cell flooding. Richness data were represented as the mean number of families (Mean Family Richness) per cell or per trap, depending on analysis. Abundance data were expressed as mean numbers of individuals per cell or per trap (height). Data were  $\text{Log}_{10}$  transformed to meet assumptions of parametric tests (equal variances). Differences in family richness or total abundance between wetland plot types or among median trap heights were compared using randomized block ANOVA. To determine if there was a statistically significant difference between wetland plot types, a type 1 error (alpha) value of 0.05 was chosen.

To interpret patterns of community composition, the relative abundance of each family within each wetland type was expressed as a percentage and transformed into Octaves ( $\text{Log}_2+1$ ) to reduce the dominance effects of common taxa (Gauch 1972). Families that occurred in less than 15% of samples were excluded from further analysis. These transformed relative abundance data were further analyzed using Principal Component Analysis (PCA) in order to reduce the number of variables/families to a few independent principal components (as in Leonhardt 2003, Kennedy 2012). Components were rotated using varimax rotation.

**Table 2.1: Taxa captured on sticky traps in 12 fen (live peat) and 14 constructed marsh (stockpiled peat) vegetation plots and classified according to 4 “life history” categories.**

<b>Order</b>	<b>Aquatic</b>	<b>Resident</b>	<b>Transient</b>	<b>Soil</b>
<b>Diptera</b>	Ceratopogonidae Chironomidae Culicidae Dixidae Ephydriidae Psychodidae Tipulidae Sciomyzidae	Chloropidae Ulidiidae	Simuliidae	Anthomyiidae Bibionidae Dolichopodidae Empididae Muscidae Phoridae Rhagionidae Sepsidae Scatopsidae
<b>Coleoptera</b>	Dytiscidae Haliplidae Scirtidae	Chrysomelidae Coccinellidae Curculionidae Mordellidae	Anobiidae	Elateridae Latridiidae Ptilidae Staphylinidae
<b>Hemiptera/ Homoptera</b>	Corixidae	Alydidae Aphididae Cicadellidae Delphacidae Miridae Pentatomidae Pseudococcidae Tingidae		
<b>Hymenoptera</b>		Bethylidae Braconidae Ceraphronidae Eulophidae Ichneumonidae Mymaridae Scelionidae		Formicidae
<b>Thysanoptera</b>		Thripidae		
<b>Trichoptera</b>	Hydroptilidae			
<b>Collembola</b>				Sminthuridae

## ***Results:***

### *Family Richness*

Mean family richness in plots containing marsh vegetation (i.e. stockpiled peat plots) was  $19.18 \pm 1.10$  (n=14; 61 families in total), having on average 2 more families than fen plots (mean  $\pm$ SE was  $17.30 \pm 1.07$ , (n=12; 40 families in total)). As predicted, fewer families were present in fen-vegetation (live peat) plots than in marsh-vegetation plots. However, the difference was not significant ( $p=0.27$ , one-way ANOVA; Figure 2.3).

Mean family richness did not vary significantly among the 4 trap heights ( $p=0.96$ ) when all plots were treated as a single group. Family richness in the 80 and 140 cm median trap height in live peat plots was significantly less than at equivalent heights in stockpiled vegetation plots. (n=12 live peat plots and n=14 stockpiled peat plots; Two-way ANOVA; Figure 3.4/ Table 2.2).

**Table 2.2: Analysis of variance (ANOVA) of numbers of families of emergent insects on sticky traps in 12 fen plots and 14 marsh plots. Analysis is based on randomized block two-way ANOVA**

<b>Source</b>	<b>Df</b>	<b>SS</b>	<b>MS</b>	<b>F</b>	<b>p</b>
<b>Height</b>	3	14.570	4.857	0.200	0.895
<b>Vegetation Type</b>	1	81.791	81.791	3.630	0.0826
<b>Interaction</b>	3	12.381	4.127	1.70	0.9152
<b>Error (within)</b>	18	435.78	24.210		
<b>Total</b>	25	544.521			

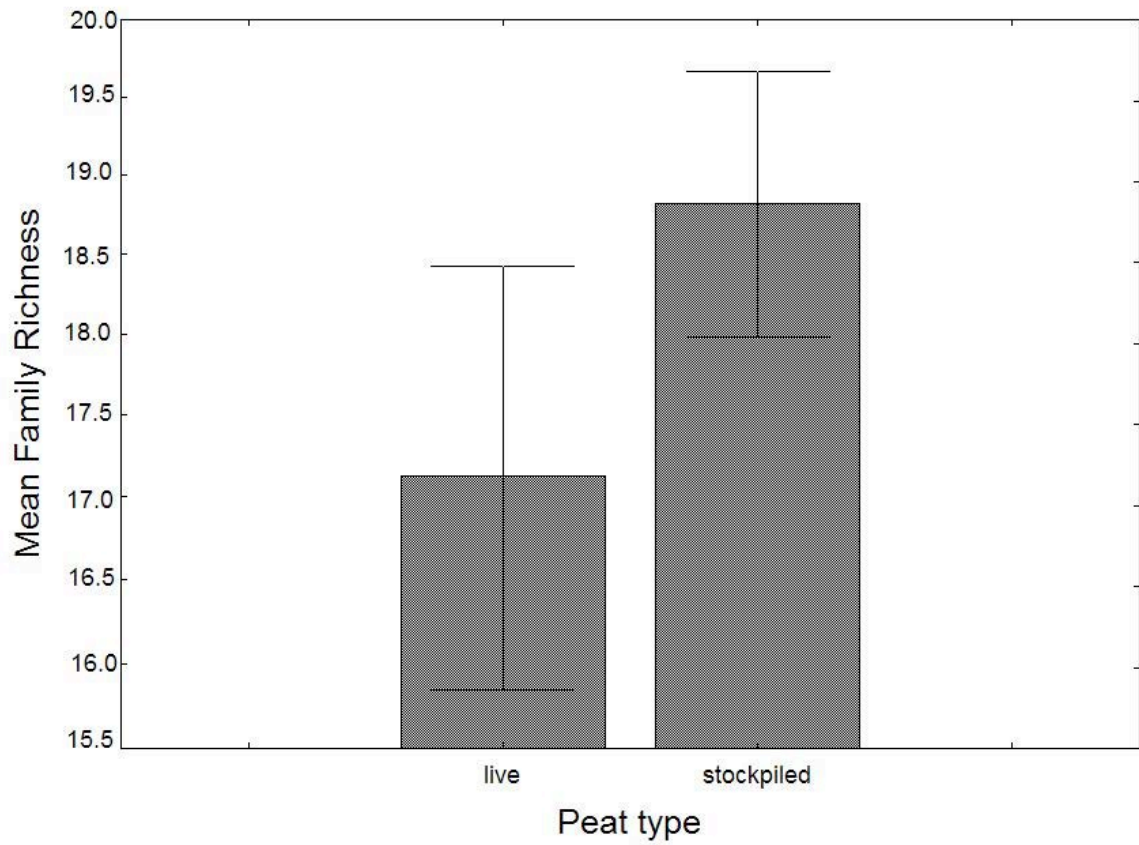


Figure 2.3: Mean ( $\pm$  1SE) family richness in live peat (n=12) and stockpiled peat (n=14) plots (p=0.27).

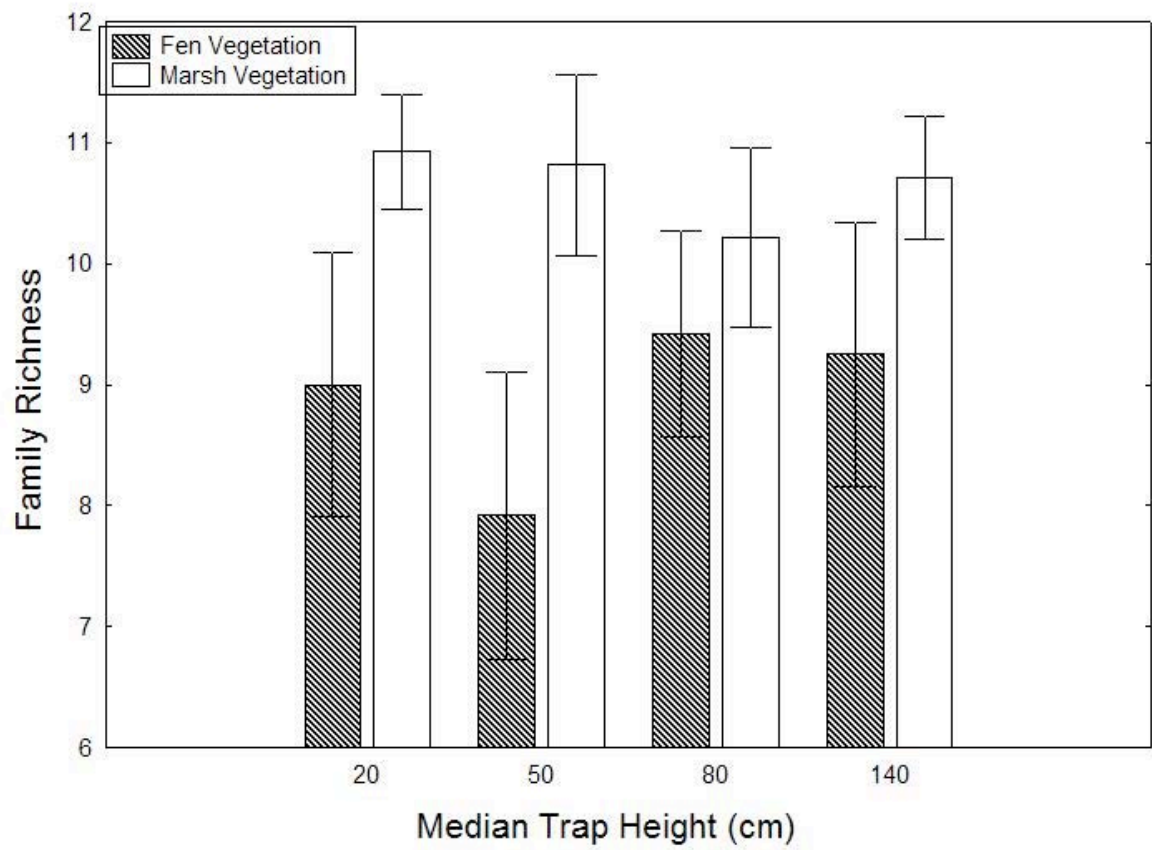


Figure 2.4: Mean ( $\pm$  SE) family richness at 4 median trap heights in 12 live peat and 14 stockpiled peat plots ( $p=0.88$ ).

### *Total Abundance*

There was also no significant difference in the total number of insects caught per cell between fen vegetation (live peat) and marsh vegetation (stockpiled peat) plots ( $p=0.90$ ;  $n=12$  live and  $n=14$  stockpiled peat plots; one- way ANOVA; Figure 2.5). Mean ( $\pm$ SE) total abundance was  $69.23 \pm 4.68$  and  $73.00 \pm 7.49$  individuals per cell, for live and stockpiled peat, respectively. On average, fen-plots collected more insects than marsh plots, contrary to our predictions; the difference was not statistically significant.

There was no significant difference in abundance among the trap sheets mounted at different heights in either peat type ( $p=0.8958$ ;  $n=12$  live and  $n=14$  stockpiled peat plots; factorial ANOVA; Figure 2.6). In contrast to the trends for of family richness, in which most taxa tended to be found at the highest elevation, the fewest insects were captured at the 140 cm trap in both fen and marsh wetland plots.

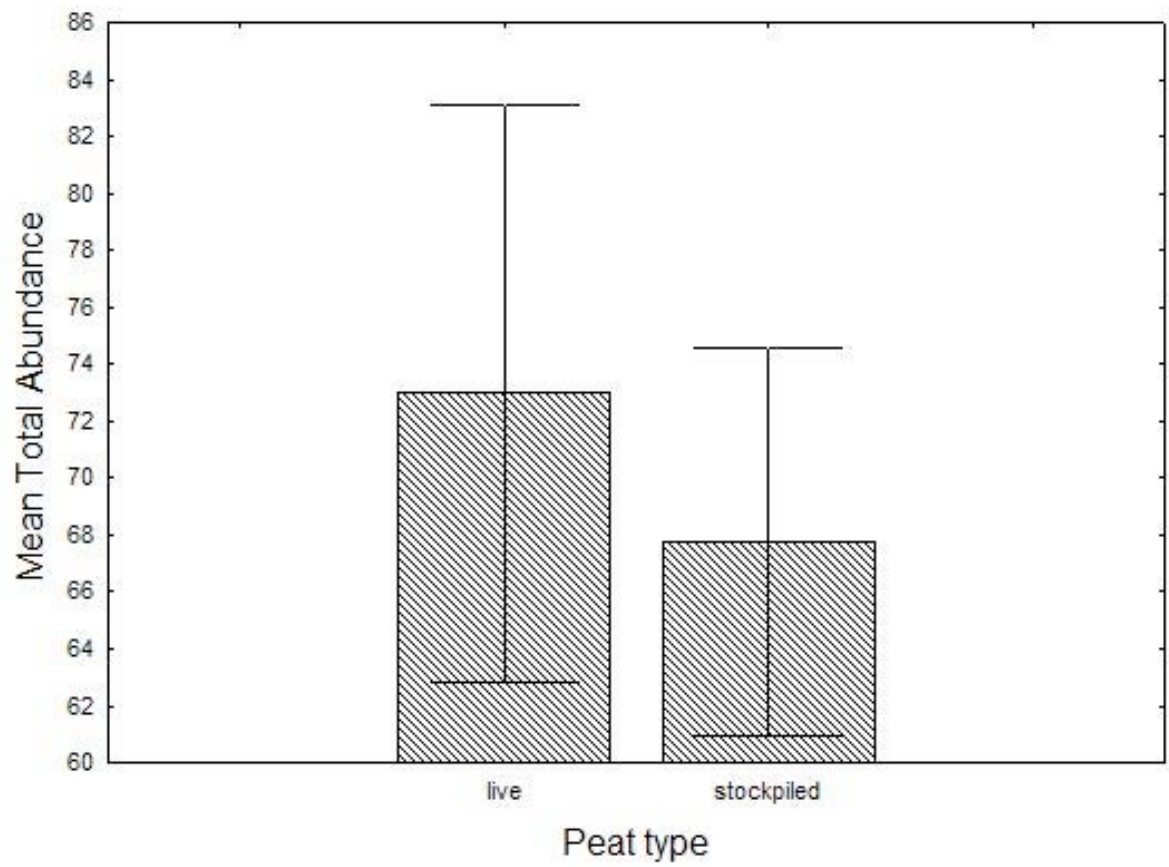


Figure 2.5: Mean ( $\pm 1$ SE) abundance for 12 live peat and 14 stockpiled peat cells ( $p=0.9$ ).



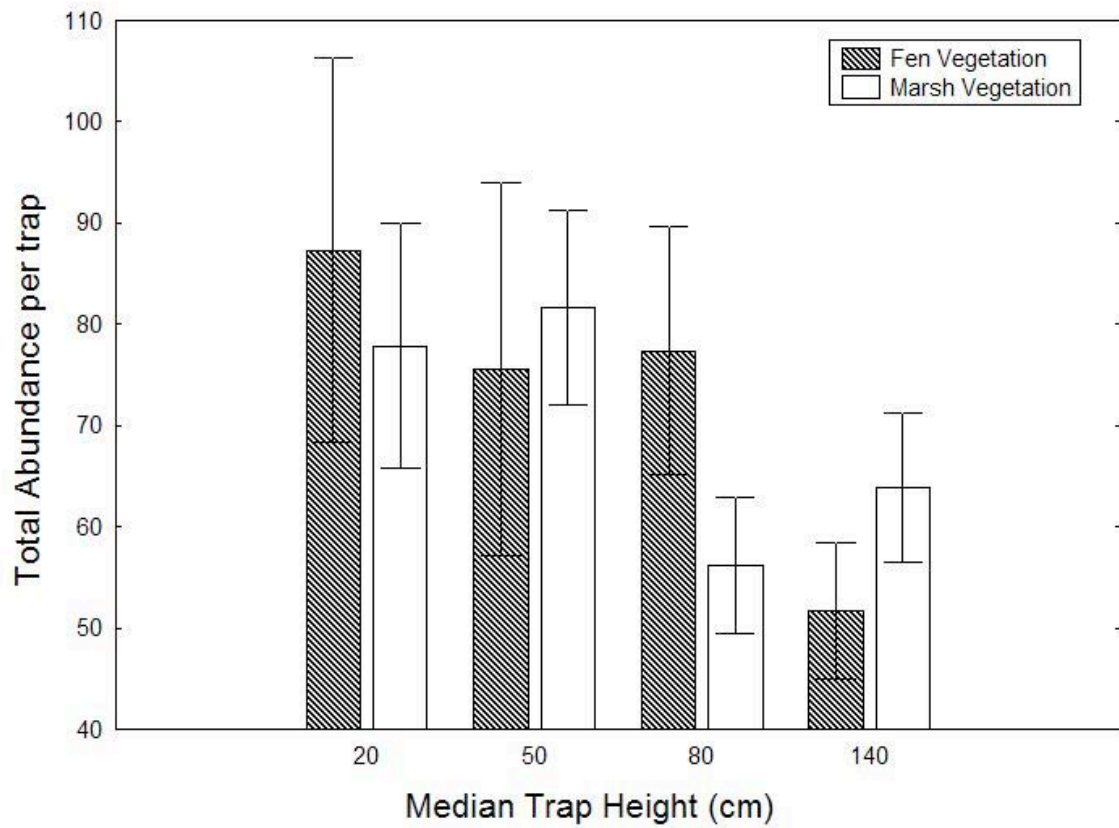


Figure 2.6: Total abundance of insects (mean number $\pm$  SE) in traps set at 4 median trap heights in 12 live peat and 14 stockpiled peat plots ( $p=0.16$ ).

### *Biomass in Live and Stockpiled Peat Plots*

There was no significant difference in total biomass emergent between fen and marsh plots ( $p=0.75$ ,  $n=12$  live peat plots and  $n=14$  stockpiled peat plots; one-way ANOVA; Figure 2.7). Mean biomass in fen plots was  $15.88 \pm 2.39$  mg DM per cell. Mean biomass in Marsh plots was  $14.72 \pm 2.61$  mg DM per cell.

In relation to biomass at median trap heights, there was also no significant difference between mean biomass per cell among the 4 median heights ( $p=0.15$ ,  $n=12$  live peat plots and  $n=14$  stockpiled peat plots; factorial ANOVA; Figure 2.8). There was a general trend, however, that biomass was greatest at the 2 middle heights (50 and 80 cm).

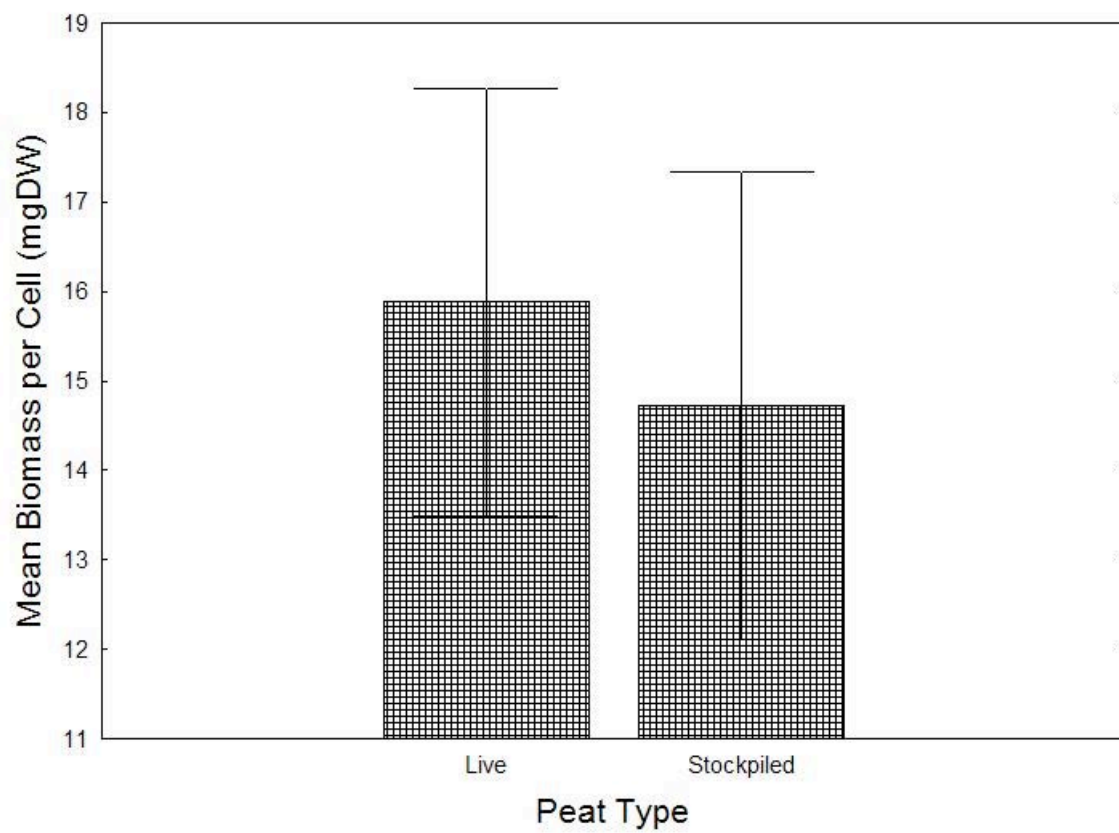


Figure 2.7: Mean cell biomass ( $\pm$  SE) between 12 live peat and 14 stockpiled peat plots ( $p=0.75$ ).

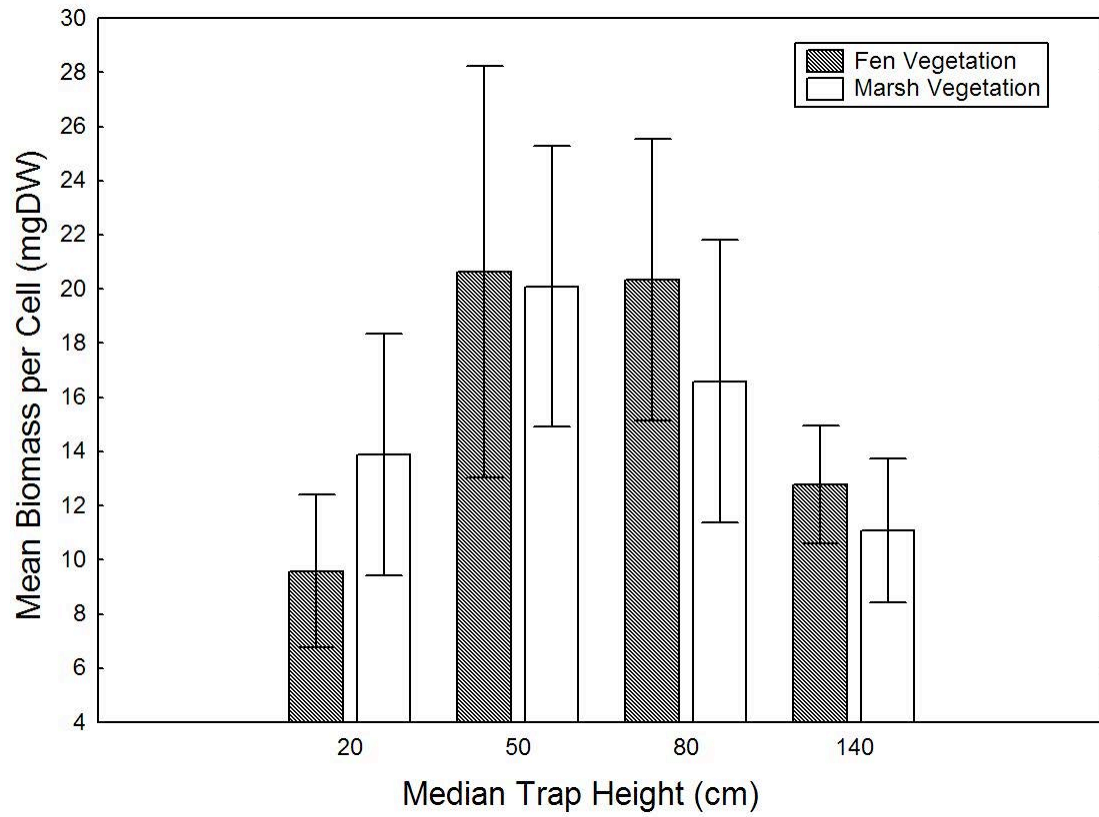


Figure 2.8: Mean cell biomass ( $\pm$  SE) between 12 live peat and 14 stockpiled peat plots at 4 median sticky trap heights ( $p=0.16$ ).

### *Composition of Arthropods- Principal Component Analysis (PCA)*

To further characterize the communities in fen and marsh plots, I compared the relative abundances families captured in experimental plots to those in control plots. The two control plots (plots without peat amendment) did not differ from experimental plots very much in terms of family richness and abundance, suggesting that there was little influence of the vegetation itself. However, three dipteran families (Ceratopogonidae, Chironomidae, and Dolichopodidae) were consistently more abundant in traps in experimental plots than in control plots.

Fifty-four families were identified among the over 7000 individuals collected from the experimental plots. Rare families (those appearing in less than 15% of samples) were omitted from a Principal Components Analysis (PCA). Covariation in 13 of the most common families was summarized using a PCA. Six principal components with eigenvalues  $>1.0$  were derived. These components explained 70% of the variation in the data (Table 2.3). Of note, the relative abundances of Ceratopogonidae, Dolichopodidae, and Chironomidae each loaded on one of the first three factors. The relative abundances of Ceratopogonidae were negatively correlated with PC1. PC1 scores positively correlated with Staphylinidae and Aphididae. Dolichopodidae was positively and negatively correlated to PC1 and PC2 scores, respectively. The relative abundances of Chironomidae were negatively correlated with scores of PC3, which was also positively correlated with Thripidae. PC loadings of four families that are significantly different from control plot abundance are detailed in table 2.4.

**Table 2.3: Eigenvalues of six principal components derived from relative abundance (Octaves) of thirteen dominant wetland families among 28 samples.**

Factor	Eigenvalue	Variance Explained (%)	Cumulative Variance explained (%)
PC1	2.21	17.0	17.0
PC2	1.40	11.0	28.0
PC3	1.40	11.0	39.0
PC4	1.38	11.0	49.0
PC5	1.29	10.0	59.0
PC6	1.45	11.0	70.0

**Table 2.4: Correlations (factor loadings) between relative abundance (octaves) of each of the 13 dominant families and principal component scores. Loadings that are greater than |0.5| are boldfaced.**

Family	PC1	PC2	PC3
Aphididae	<b>0.821</b>	0.003	0.005
Staphylinidae	<b>0.556</b>	0.213	0.388
Lathridiidae	0.458	-0.442	0.149
<b>Ceratopogonidae</b>	<b>-0.883</b>	-0.001	0.099
<b>Ephydriidae</b>	0.084	<b>0.700</b>	0.207
<b>Dolichopodidae</b>	-0.042	<b>-0.703</b>	0.072
Thripidae	0.183	-0.310	<b>0.545</b>
<b>Chironomidae</b>	0.127	-0.112	<b>-0.786</b>
Explained Variance	2.21	1.40	1.40
% of total variance explained	17.0	11.0	11.0

*Patterns in most abundant families- Ceratopogonidae, Chironomidae, Dolichopodidae*

Results of the above PCA analysis determined the next set of statistical tests. The goal was to determine how abundances of 3 dipteran families (Ceratopogonidae, Chironomidae, Dolichopodidae) varied among vegetation heights, indicating that their distribution was dependent on the vegetation rather than by random chance. The three families, as previously mentioned, were the only ones whose abundance was significantly higher in experimental plots than in control plots (plots that did not contain peat). Randomized block two-way ANOVA was performed using abundances of each of the three dipteran families as the dependent variable. Independent variables were median trap height and vegetation type (fen or marsh)

**Table 2.5 Randomized block ANOVA comparing abundance of Chironomidae among cells and Heights**

Factor	DF	SS	MS	F	p
Cells	25	2346.340	93.85359	3.114311	0.000079
Heights	3	210.181	70.06018	2.324782	0.081639
Remainder	75	2260.217	30.13623		
Total	103	4816.738			

**Table 2.6 Randomized block ANOVA comparing abundance of Ceratopogonidae among cells and Heights**

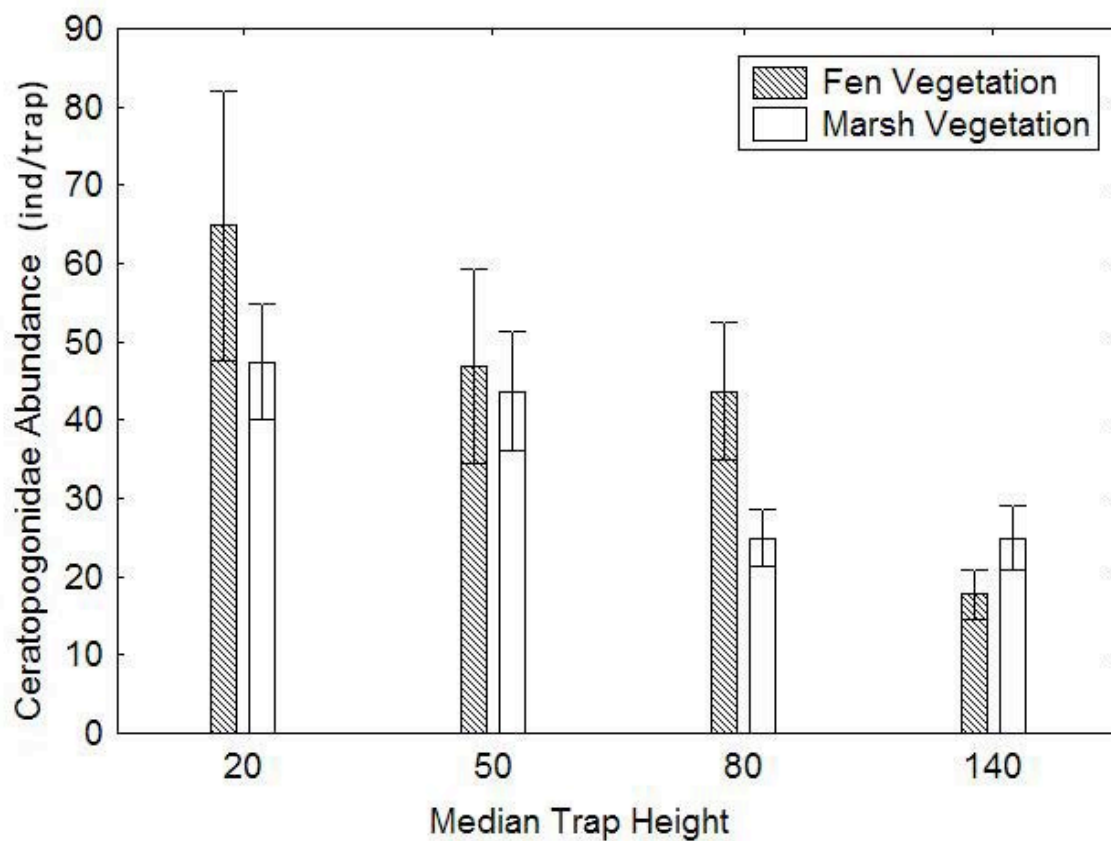
Factor	DF	SS	MS	F	p
Cells	25	37637.26	1505.491	2.264788	0.003550
Heights	3	16657.13	5552.377	8.352732	<b>0.000073</b>
Remainder	75	49855.34	664.738		
Total	103	104149.7			

**Table 2.7 Randomized block ANOVA comparing abundance of Dolichopodidae among cells and Heights**

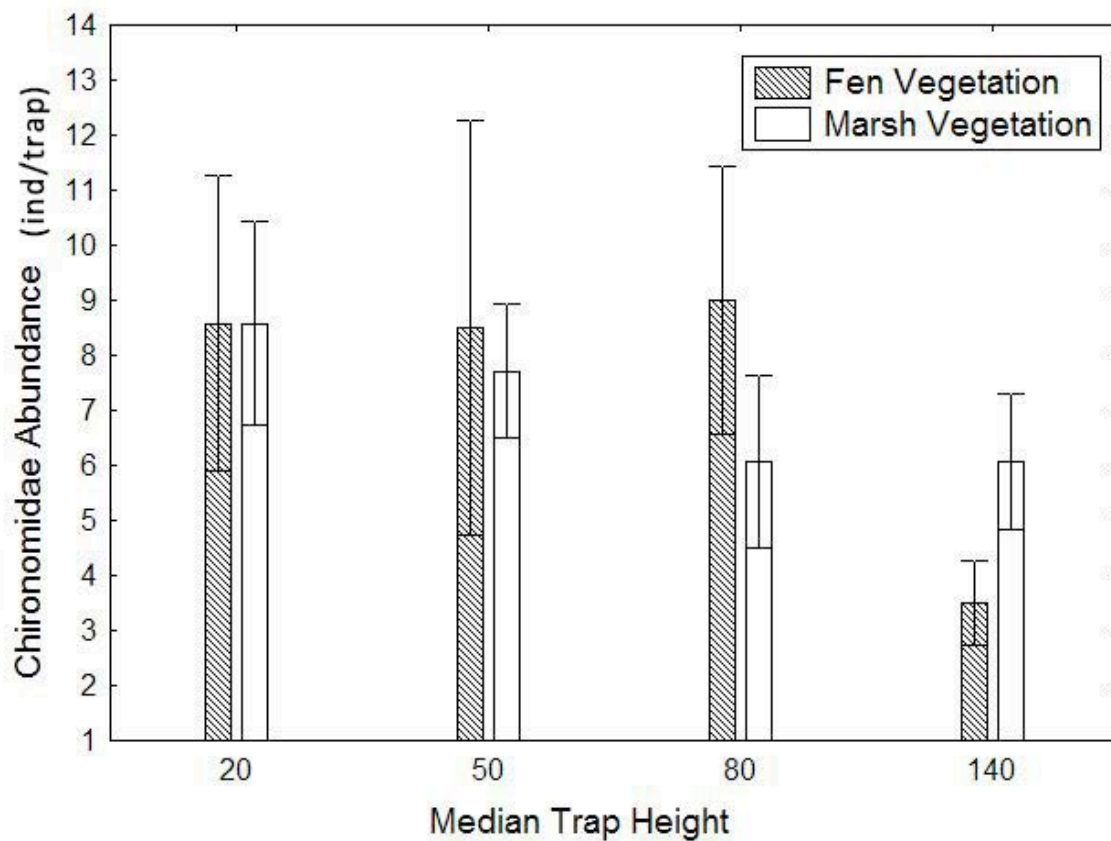
Factor	DF	SS	MS	F	p
Cells	25	449.505	17.98020	1.159521	0.304792
Heights	3	88.330	29.44327	1.898761	0.137062
Remainder	75	1162.993	15.50657		
Total	103	1700.828			



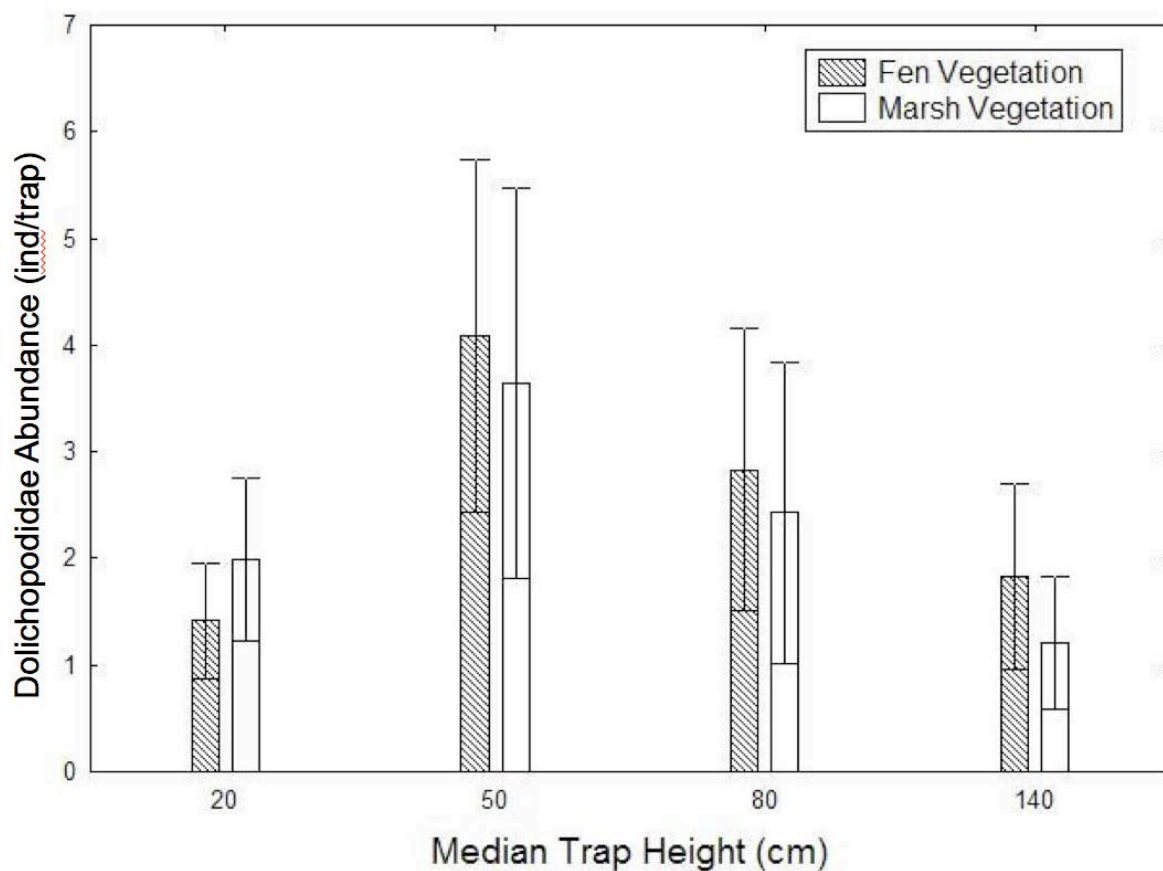
The abundance of 3 aquatic families of Diptera (Chironomidae, Ceratopogonidae, Dolichopodidae) was lowest in the sticky traps places at the greatest height, and tended to be greatest nearest the substrates. The trends were reflected in randomized block ANOVA of abundances of all three aquatic families. However, only the differences in ceratopogonid abundances among heights were statistically significant ( $F=8.35$ ,  $p=0.000073$ , Fig. 2.9, table 2.6). Those for Chironomidae ( $F = 2.43$ ,  $p = 0.08$ , Fig. 2.10, table 2.5) and Dolichopodidae ( $F=1.90$ ,  $p=0.14$ ; Fig. 2.11, table 2.7) were not statistically significant.



**Figure 2.9: Ceratopogonidae abundance (individuals per trap  $\pm$  SE) between 12 live peat and 14 stockpiled peat plots ( $p=0.56$ ) at 4 median sticky trap heights ( $p=0.32$ ).**



**Figure 2.10: Chironomidae abundance (individuals per trap  $\pm$  SE) between 12 live peat and 14 stockpiled peat plots ( $p=0.77$ ) at 4 median sticky trap heights ( $p=0.83$ ).**



**Figure 2.11: Dolichopodidae abundance (individuals per trap  $\pm$  SE) between 12 live peat and 14 stockpiled peat plots ( $p=0.71$ ) at 4 median sticky trap heights ( $p=0.77$ ).**

### ***Discussion:***

Abundance, family richness, and biomass were similar between constructed wetland plot types, indicating that functional attributes of flying insect assemblages didn't distinguish between the two vegetation types at the scale evaluated. The fact that relatively few taxa were more abundant over or in experimental cells relative to control cells suggests that most of the individuals captured do indeed represent the regional insect community rather than being locally representative. Nevertheless, two aquatic families of Diptera that typically dominate the fauna of zoobenthos (Chironomidae and Ceratopogonidae) and one semiaquatic family (Dolichopodidae) were consistently more abundant within than outside of experimental cells. This suggests that the sticky traps do sample specimens that likely represent the local habitat. Furthermore, abundances of these taxa were vertically stratified, and more abundant at heights nearest the substrate than at the greatest height. It is difficult to ascertain whether the individuals collected at low heights are those that may have recently emerged, or if these small-bodied individuals are merely more common at heights among plant stems that are sheltered from the wind.

Cryptic biodiversity may play a role in the lack of significant differences. Finer taxonomic resolution may be able to distinguish response of insects to structural and vegetative characteristics in these plots.

The lack of more pronounced differences between any of the experimental factors, especially with respect to vegetation type may be a function of the construction design of the plots. Because cells were only 10 x 20 m and separated from other plots by only a

few meters, the units may have been too small and closely associated to be perceived as independent patches by the insects. Flying insects can easily disperse much greater distances than this (Smith et al. 2007). Also, emergent insect populations in natural wetlands may experience several emergent events throughout the summer season. Should this experiment have captured an emergence event, it is likely that marsh associated aquatic insects would have increased total abundance captured and influenced the significance of these data.

Age of the plant communities at the time of sampling likely influenced non-significance of data. At the time of sampling, these plots were only 4 years old. Leonhardt (2003) compared benthic invertebrate community successional trajectories in oil sands constructed wetlands and suggested that aquatic invertebrate family richness and composition stabilized at around 7 years of age. The plots used in my study would be considered “young” by this definition. Only limited research is available regarding the successional trajectory of fen vegetation or arthropod biomass. The trends are likely similar to those of marshes, although perhaps on a far longer timescale. Natural boreal fens are long living successional seres, with some area wetlands being upwards of 10,000 years old (Batzer and Sharitz 2007). Fens are also relatively plant species rich wetlands, which presents one of the greatest challenges to reclamation efforts. Although most of the natural fen vegetation of live peat plots survived the transplanting process, tree species (Black Spruce, *Picea mariana*) did not persist, likely due to severed taproots. Much research has demonstrated that plant species richness accounts for greater arthropod diversity as, for example, each new plant species could provide a host plant relationship

for a particular taxon of arthropod (Siemann et al 1998, Brose 2003). Loss of plant species during transplantation may account for lack of significant data trends.

To describe why trends in data are non-significant and explain the relationship between taxa and biomass, we must further characterize these experimental plots. These cells most closely resemble wet-meadow and emergent zones of a marsh, and the overall homogeneous environment of a fen. These cells did not naturally contain standing water, but were watered to maintain plant communities. Many studies have demonstrated that emergent and open water zones can support the greatest biomass of emergent arthropods. For example, Voight's (1976) study of Iowa marshes demonstrated that overall aquatic arthropod abundance was greatest in open water zones that supported submergent vegetation. Though Voight's study focused on aquatic taxa, he also concluded that chironomid midges dominated complex submergent vegetation (which would be available as emergent biomass during emergence events). McLaughlin and Harris' (1990) study of Great Lakes marshes characterized these wetlands using 4 vegetation zones (wet meadow, dense emergent, sparse emergent, and open water). They concluded that the sparse emergent zone of diked marshes supported the greatest biomass of emergent adult insects. However, their study wetlands didn't support submergent vegetation (McLaughlin and Harris 1990). The need for standing water to support insect biomass has been well documented. The lack of these habitats in these constructed cells likely accounts for the non-significance of biomass, abundance, and family level richness data.

The abundance patterns of the three Diptera families that have aquatic or semiaquatic larvae revealed trends, but only one was statistically significant. Most individuals of all taxa tended to be most abundant at the 20-cm median trap height.

### ***Conclusions***

Sticky traps placed in the two vegetation types that dominated the constructed wetland plots collected similar family richness, abundance and biomass of flying insects.

However, the great majority of specimens captured were terrestrial or transient aquatic species. Only 3 families, all with aquatic or semiaquatic larval stages were consistently more abundant in the test cell catches than in control cells. The lack of differences may reflect the nearness of cells to one another, or the coarse level of taxonomic resolution used in this study. However, the differences in vertical stratification of aquatic taxa suggests that sticky traps do sample the local emergent fauna and that these insects are most common within 30 cm of ground level. Continued research would benefit from identification of taxa to the genus level.



## CHAPTER 3

### INVERTEBRATE RICHNESS AND BIOMASS IN BOREAL WETLANDS OF NORTHEASTERN ALBERTA, CANADA

#### *Introduction:*

As was previously discussed in the general introduction, vegetation is one of the most important habitat characteristics influencing arthropod community attributes. In wetlands, vegetation composition and distribution reflects local environmental conditions including shoreline morphometry, hydrology, nutrient regime, and hydrodynamic regime (Zoltai and Vitt 1995, Smith et al. 2007, Remburg and Turner 2009). Arthropod assemblages in turn relate to the local vegetative characteristics necessary for completion of their life cycles. Marshes are characterized by the presence of distinct, hydrologically-determined vegetation zones, each supporting invertebrate communities defined by particular taxa richness, abundance, and biomass. For example, submerged aquatic vegetation (SAV) influences biotic processes by driving nutrient dynamics (Asaeda et al. 2000) and providing the physical and chemical environment for aquatic invertebrates; macrophytes also alter the physical environment by increasing habitat complexity and serving as prey refugia (Mittelbach 1988, Cyr and Downing 1988), increasing food availability (Campeau et al. 1994, Taniguchi et al. 2003), and providing attachment sites and “building materials” for case-building invertebrates (Dudley 1988). deSzalay and Resh (2000) determined that wetlands with greater structural complexity (provided by macrophytes) supported higher invertebrate biomass than similar open water wetlands. Benke et al. (1999) compared taxon richness and abundance across zones of marshes and

found that richness was related to habitat heterogeneity. Richness was lowest in open water zones that did not support submergent vegetation. Furthermore, semiaquatic structures provided by *Nymphaea* water lilies supported the greatest emergent abundance of chironomid midges, demonstrating the importance of these macrophyte microhabitats. These findings are similar to those of McLaughlin and Harris (1990) for Great Lakes wetlands. Emergent insect biomass was greater from the sparse emergent zone than from open water zones that lacked significant submergent macrophyte growth. Several studies have demonstrated the importance of SAV in the open water zone as habitat for aquatic invertebrates, resulting in large emergences that are transported into the terrestrial ecotone of a marsh (Paetzold et al. 2005, Hoekman et al. 2012).

Vegetation structures invertebrate assemblages both in the SAV zone of a wetland, and in the emergent and riparian plant zones. These transitional habitats also support a wide albeit different array of invertebrates (McLaughlin and Harris 1990, Benke et al. 1999). However, the community attributes of arthropods in these transitional habitats not as well known. Questions arise as to whether invertebrates play the same role in peatlands as they do in the semi-terrestrial zone of wetlands (Holmquist et al. 2008), (i.e., sedge meadows and wet meadows).

Sedge meadows are described here as an aquatic-terrestrial transition zone. These habitats comprise a large portion of wetland area and are dominated by graminoid vegetation. The sedge genus *Carex* (Cyperaceae) is dominant in these habitats (Beaulieu

and Wheeler 2005). Structurally, wet meadows are relatively homogeneous habitats due to the dominance of a single plant type (Beaulieu and Wheeler 2005). Wet meadows have received increasing recent attention due to encroachment of development in many parts of North America (Benke et al. 1999). Wet meadows both receive a carbon subsidy from the aquatic ecotone (wetlands) to which they are adjacent (Beaulieu et al 2005, Hoekman et al. 2012), and support their own unique community of arthropods (Hury and Gibbs 1999). Notably, Beaulieu and Wheeler (2001) surveyed higher flies (Diptera: Brachycera) of sedge meadows in southern Quebec and identified 362 species in 35 families including many species whose larvae develop in semi-aquatic soils. However, despite the apparent diversity of brachycerans, only a small proportion was restricted to these habitats. Most species were generalists. This is the case for many other ecological studies of sedge meadow Diptera. Other brachyceran families that have been studied in sedge meadows include Chloropidae (Todd and Foote 1987a), Ephydriidae (Todd and Foote 1987b), and Scathophagidae (Wallace and Neff 1971).

The semi-terrestrial nature of wet meadows provides habitats suitable for aquatic, semi-aquatic, and terrestrial taxa. Also, because soils are only intermittently inundated (unlike the permanently saturated sediments of the zones of emergent and submergent aquatic vegetation) they provide an environment that supports communities of soil invertebrates (Davis et al. 2006, Riggins et al. 2009). Davis et al. (2006) described the diversity of soil invertebrate communities, identifying 73 taxa (39 of which were considered soil inhabitants). Most of the invertebrates captured (using soil cores) were earthworms, isopods, scarab beetles, and click beetles. They noted the potential impacts of altering

water table depth on the natural biota of these habitats; anthropogenic disturbance in these areas and surrounding water bodies would alter the community composition of wet meadows towards fewer, more moisture-tolerant species. The relatively short generation time of soil invertebrates allows them to respond to adverse conditions. However the limited dispersal ability of these invertebrates makes post-disturbance recolonization difficult (Riggins et al. 2009). These characteristics make soil invertebrates excellent indicators of environmental condition; Soil arthropods have previously been used to assess terrestrial reclamation success (Finnamore 1994).

As previously mentioned, peatlands are often compared to wet meadows due to their similar hydrology, homogeneity, and the dominance of graminoid vegetation. Characteristic peatland fauna were briefly discussed in the General Introduction to this thesis. The major limitations of peatland arthropod studies have been a lack of expertise in identifying invertebrates to a sufficient level of taxonomic resolution (Marshall et al. 1999, Beaulieu and Wheeler 2001). and the use of different trapping methods (Rosenburg and Danks 1994.), as well as, perhaps most importantly, the lack of taxonomic resolution within the arthropods. Marshall et al. (1999) reviewed the arthropod faunal diversity of peatlands and noted that approximately 3600 species of arthropod have been identified from Canadian peatlands of which about 10% are obligate inhabitants. In contrast to the characteristics of sedge meadows, permanent inundation of peat does limit the abundance soil arthropods. However, the structure of vegetation typical of Canada's boreal peatlands can support a variety of typical soil arthropods including spiders (Araneae), mites (Acari), and springtails (Collembola). Behan-Pelletier and Bissett (1994) described the

structure of waterlogged *Sphagnum* mosses as habitat for many oribatid mites (Acari: Oribatidae).

The most visually striking difference between fens and marshes exists between the type and structure of plant communities that each of these wetland types support. This study was designed to assess how the marked differences in hydrologic and vegetation structure affect invertebrate community composition. I sampled various microhabitats to determine where most invertebrates reside within a fen: within the profile of peat, associated with grasses common to a rich fen, or associated with patchy, marsh-like vegetation. In comparing of the fauna of these two wetland types I assessed the differences in fauna among the vegetation zones established in natural wetlands by the systems' hydrological structure. The objectives were to determine:

- a) how fens and marshes differ in terms of invertebrate family richness and biomass;
- b) how invertebrate richness, biomass and abundance vary with respect to horizontal zonation within a marsh; and
- c) which vegetation characteristics (plant species richness or aspects of vegetation structure) best correlate to increases in arthropod family richness and/or biomass

### *Predictions*

Generally, I postulated that invertebrate abundance, family richness and biomass would vary significantly among hydrologically regulated vegetation zones in wetlands; highest invertebrate community attributes (biomass, abundance, richness) will be produced in the SAV zone of marshes. Furthermore, I anticipated that marshes overall would support

more invertebrate biomass than fens due to their greater diversity of habitats. Because marshes have a longer hydrological gradient, they exhibit greater habitat heterogeneity than fens, and therefore support greater abundance and biomass of invertebrates.

Because wet meadows and fens are often argued to have similar vegetation structure and type (Holmquist et al. 2011), I expected that they would support similar invertebrate biomass and abundance; I expected there to be no significant difference between biomass and abundance of invertebrates from the wet meadow zones of natural fens and marshes. Alternatively, postulates similar to those of Schaffers et al. (2008) anticipate that the greater plant species richness of fens would support greater invertebrate taxa richness, abundance, and biomass. I observed that though both marsh wet meadows and fens have similar homogeneous expanses of graminoid plant species, graminoid fens are more species rich than the wet meadow zone of marshes (pers. obs.), which could support greater invertebrate richness in fens than is found in marsh wet meadows.

### ***Materials and Methods:***

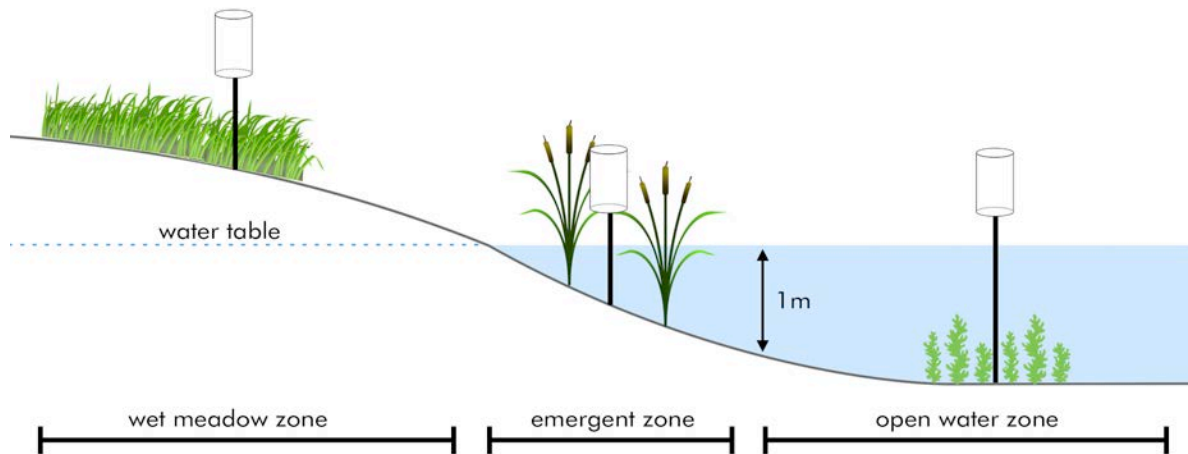
#### *Study Locations*

Study wetlands were located near the town of Fort McMurray in the Athabasca Oil Sands region of northeastern, Alberta, Canada. All wetlands occurred in relatively undisturbed landscapes. Wetland locations and their detailed characteristics are described in Appendix 1. Wetlands were sampled between May and August 2012 using three sampling methods.

### *Sticky Trap Sampling (all zones)*

Detailed sampling and processing methods were described in chapter 2. Sticky traps consisted of sheets of acetate (21.6 x 27.9 cm overhead transparencies) that were painted with Tanglefoot™, a natural plant resin wrapped around a 30-cm long x 10 cm diameter plastic pipe mounted on a stake at a height of 60-80 cm above the substrate (Fig 3.1). Organisms alighting on the sheet became stuck to the resin. Traps were deployed for periods of 3 days. This height was selected based on results of a pilot study undertaken in 2011, which indicated this height would capture the greatest abundance of insects.

To determine how insects were distributed among plant zones, 3 traps each were placed in each of the submerged aquatic vegetation (SAV), emergent vegetation (EZ) and wet meadow (WM) zones. Traps were arranged in three radii across the hydrological zones of the wetland. Traps placed in the SAV zone were placed as close to the center of the open water as was possible, given the limitations of wading depth (<100 cm).



**Figure 3.1 Schematic diagram of a wetland showing vegetation zones and locations of sticky traps (not to scale).**

Although fens often lack the 3 vegetation zones that typify marshes, traps were placed similarly in wetlands for consistency. Open pools of water were limited in fens sampled, so samplers were more randomly distributed. Traps were allotted to “zones” based on dominant plant species. Three of the 4 fens sampled (Beaver Lodge, Maqua Lake Fen, and Gravel Pit Fen) had areas of emergent and submergent vegetation, and so traps were placed within these zones in those wetlands.

Sticky trap samples were stored, transported and processed in the same manner as described in detail in Chapter 1. Insects were identified to the family level using keys in Marshall (2007).



The biomass of each insect was estimated from its length using previously published length-biomass regression equations derived for insect families (Sample et al. 1993, Ganihar 1997, Stagliano et al. 1998, Sabo et al. 2001). Specimens were photographed beneath a camera-equipped dissection microscope, and the images were measured to determine the length of an individual. A 10-mm scale bar in each photo was used to calibrate images. Images were analyzed using ImageJ 1.47 for Mac OS X software (U. S. National Institutes of Health, Bethesda, Maryland, USA). Insects were measured from most anterior part of head to the level of the anus (Rogers et al. 1977). Appendages (antennae, cerci, ovipositor, etc.) were not included in the length measurement. Eighty percent of the insects collected per family per sample were randomly selected and measured. These length measurements were used to estimate the biomass of each individual from a family-specific regression equation, and the resulting biomasses were averaged to calculate a “mean weight per insect” for that family (mg dry mass per insect).

#### *Aerial Sweep Net Sampling (Wet Meadow Zone)*

Aerial sweep net sampling is perhaps the most commonly used method to collect terrestrial invertebrates from vegetation. I collected semi-quantitative sweep net samples from the wet meadow zones of all fens and marshes on relatively calm days (wind speed <5 km/h). Samples were collected using a heavy-duty sweep net with a 38.1-cm (15”) diameter opening (Catalog #7635HS, Bioquip Products Inc. Rancho Dominguez, CA). Three samples per wetland were collected sequentially on single occasions at each wetland. A sample consisted of a timed, one-minute, walk while sweeping continuously

through the tops of wet meadow vegetation. A single individual collected all of the samples to maximize consistency. Material in the net bag after each sweep was transferred into a jar containing 95% ethanol. In the laboratory, specimens were sorted from debris beneath a dissecting microscope and identified to family. The lengths of 80% of the specimens were measured and used to estimate sample biomass as was described for sticky trap catches.

#### *Vacuum Sampling (Wet Meadow Zone)*

Sampling of a habitat's entire complement of invertebrates is seldom achieved through one method alone (Anderson et al. 2013). Sticky traps will not sample nonflying insects or invertebrates that reside within the structure of vegetation. Aerial sweeps capture primarily the fauna associated with the upper portions of vegetation. I employed vacuum sampling to actively sample the invertebrates associated with both the vegetation and surface substrate of wet meadows (see Hoekmann 2012).

The sampler used was modified from the design described by Hoekman et al. (2011). A Stihl7 model SH87c Leaf blower/vacuum (Stihl Incorporated Canada, London, ON) was used as a vacuum to collect wet meadow invertebrates. An insect sweep net bag (Bioquip Products, Rancho Dominguez, CA) was fitted into the mouth of the intake tube of the suction sampler and secured with self-gripping Velcro tape to retain the materials drawn into the sampler. To ensure that the sweep net bag and arthropod samples were not destroyed by being pulled into the mulching blades, the suction tube was also fitted with a 6.3-mm mesh steel "net stopper" device 45 cm from the suction tube opening. During

sampling, a net bag was placed into the intake tube of the leaf blower, and the intake tube was held perpendicular to the ground while the barrel opening was repeatedly tapped on the soil surface to draw invertebrates into the net bag.

Sampling plots were randomly selected from the wet meadow zone of a wetland. A 30 x 43 cm (11 x 18 inch) box sampler was used to delineate a portion of the substrate.

Sampling proceeded in 3 stages to collect material from vegetation proper, from the surface litter, and from the soil, respectively:

1) Vegetation sampling - The sampler was used to collect invertebrates from the leaves and stems of vegetation by lowering the intake tube over the leaves and stems of vegetation until it touched the substrate surface. The tube was then raised. This procedure was done 12 times at various locations within the 30 x 43 cm delineated area. The sampler was switched off, and net bag was then removed from the vacuum sampler, and turned inside out into a small plastic bag. Bags were placed into a cooler containing several ice packs until the sample bags could be frozen.

2) Vegetation Clippings – The height of the tallest plant within the box sampler was measured and recorded. All vegetation within the area delineated by the box sampler was then clipped to at the level of the soil's surface, placed in another labeled plastic bag and stored on ice in the cooler.

3) Soil Fauna - After clippings had been removed, the now-exposed soil surface was again vacuumed (using the tapping method, 12 times). The contents of the net bag were placed in a third labeled plastic bag and stored on ice in the cooler.

On the day of sampling, soil fauna samples were placed in a Berlese funnel for 24 h to extract soil invertebrates. Light and heat from a 40-W incandescent bulb secured 10 cm above the sample induced invertebrates to move downward in the funnel. They then fell down the funnel's stem and into preservative. These specimens were preserved and stored in 95% ethanol. The Vegetation Suction and "Vegetation Clippings" samples were kept in a freezer until processing.

In the laboratory, invertebrates were separated from vegetation by immersing the sample in warm, soapy water in a 20-L plastic pail for 30 min and then pouring the bucket contents through a nested series of 7 brass sieves (mesh apertures 8.00, 4.00, 1.00, 0.50 , 0.25, 0.18, and 0.09 mm). Material in the sieves was further separated by placing the stack under a tap and gently rinsing with cold running water until it was clear that the material remaining in the top sieve would not pass through that sieve. The top sieve in the stack was removed and its material was emptied into a Petri dish. The material in the next coarsest sieve was similarly rinsed, and retained material placed in another Petri plate. This process was repeated until 7 Petri plates had been prepared, each with a relatively uniform size fraction of material. The invertebrates in each dish were separated from debris under a dissecting microscope at 40X magnification and preserved in 70% ethanol

for later identification. Specimens were subsequently identified to family. The remaining organic debris was placed in a drying oven at 65 degrees C until constant weight was achieved and then weighed to the nearest 0.01 g.

Biomass of vacuum sample invertebrates was calculated using a regression equation derived by Reger et al. (1982) as modified by Ciborowski (1984). Reger et al. (1982) provided an equation relating zoobenthic ash-free dry mass (AFDM, mg) to sieve aperture size that would retain invertebrates rinsed through a series of 13 brass sieves arranged in  $2^{-0.5}$  decrements (i.e. 8.0, 5.7, 4.0, 2.8, 2.0, . . . . 0.63 mm). Ciborowski (1984) modified this equation to pertain to a series arranged in  $2^{-1}$  decrements. I used this regression equation to estimate the AFDM of invertebrates collected using vacuum sampling. The following sieve (mm) -AFDM per invertebrate (mg) estimates were used: 8.00 – 10.893, 4.00 – 2.819, 1.00 – 0.189, 0.50 – 0.049, 0.25 – 0.013, 0.18 – 0.007, 0.09 – 0.002. Total biomass per sample was estimated by multiplying the number of invertebrates per sieve size fraction by the estimated AFDM per individual.

### *Statistical Analyses*

Statistical analyses were performed using STATISTICA 7.0 software (Statsoft Inc., Tulsa, OK). For zone-specific and among-wetland comparisons, data from replicate samples were pooled and expressed as total abundance, biomass, or richness per zone or per wetland, as appropriate for each sampling. Sample totals were then  $\text{Log}_{10}$  transformed

to meet the assumptions of parametric testing; assumption of equal variances. Univariate tests were employed to determine if arthropod family richness, total abundance, and biomass differed significantly between wetland types or among zones using an alpha value of 0.05, corrected for multiple tests.

Variation in sticky trap family richness and total biomass between wetland types and among vegetation zones were assessed using two-way main-effects ANOVA. Null hypotheses for all data sets were:

- 1) Wetland type has no effect on insect family richness/biomass, and
- 2) Family richness/biomass did not vary among wetland zones.

A randomized block design was used to determine the statistical significance of difference among vegetation zones for each wetland type. In this analysis, factor A (wetland zone) was a fixed factor whose means were compared among wetlands, treated as blocks. Because we are performing multiple analyses on the same data set (5 factorial ANOVAs), we used the Holm modification of Bonferroni correction (Holm 1979). To reduce the probability of committing type 1 error, we used a p-value of 0.01 (Bonferroni correction =  $K/\alpha$ , where K is the number of statistical analyses) for analysis of sticky trap data.

Estimates of family richness, abundance, and biomass based on vacuum sampler and aerial sweep net catches pertained only to the wet meadow zone. Samples for individual

wetlands were pooled, and variation between wetland types was assessed using one-way ANOVA. Estimates of family richness, biomass, and abundance calculated for vacuum samples were pool between “vegetation sampling”, “vegetation clipping” and “soil fauna” per vacuum box plot sample.

Multiple regression analysis was used to determine which aspects of wetland vegetation - plant species richness or vegetation structure - most significantly influenced arthropod family richness and biomass. These analyses were conducted using vegetation and arthropod data from the wet meadow portion of wetlands. Independent variables for this analysis included: plant species richness determined by Marie-Claude Roy (University of Alberta, personal communication), mean plant biomass (g dry mass per vacuum sample box plot), and maximum plant height (cm) within vacuum sample box plot. Data for these analyses were  $\text{Log}_{10}(Y+1)$  transformed meet the assumption of equal variances.

## ***Results:***

### *Sticky Trap Samples*

Sticky traps placed in marshes collected significantly more families of flying insects than traps in fens ( $18.41 \pm 1.04$  families (n=9) vs.  $11.74 \pm 1.12$  n=3); ANOVA,  $p < 0.001$ ; Table 3.1). There was no significant difference among vegetation zones ( $p = 0.639$ ) when all wetlands were considered as replicates..

**Table 3.1: Summary of analysis of variance (ANOVA) of numbers of families of emergent insects on sticky traps (n=9 marshes, n=4 fens). Analysis is based on two-way main-effects ANOVA. Error degrees of freedom are adjusted to reflect lack of independence of zones within wetlands**

Source	df	SS	MS	F	p
Whole	1	45.297	45.297	384.854	<b>&lt;0.0001</b>
Wetland Type	1	0.317	0.317	18.621	<b>0.0035</b>
Wetland Zone	2	0.008	0.004	0.241	0.639
Type x Zone	2	0.010	0.005	0.305	0.598
Error (Within)	<u>7</u>	<u>0.118</u>	<u>0.017</u>		
Total	12	0.453			



**Table 3.2: Summary of analysis of variance (ANOVA) of numbers of families of emergent insects on sticky traps in a field experiment within n=9 marshes, n=4 fens. Analysis is based on three separate one-way ANOVA**

Source	df	SS	MS	F	p
<b>Wet Meadow</b>	1	0.157	0.157	14.651	<b>0.003</b>
WM Error	<u>11</u>	<u>0.118</u>	<u>0.011</u>		
WM Total	12	0.276			
<b>Emergent Zone</b>	1	0.105	0.105	8.309	<b>0.015</b>
EZ Error	<u>11</u>	<u>0.139</u>	<u>0.013</u>		
EZ Total	12	0.245			
<b>Open Water</b>	1	0.064	0.064	2.327	0.155
OW Error	<u>11</u>	<u>0.304</u>	<u>0.028</u>		
OW Total	12	0.368			

Because wetland zones were nested within wetlands, no independent interaction term was available to determine whether between-wetland differences depended on the wetland zone considered. Consequently, simple one-way ANOVAs were performed on each wetland zone to determine the source of the significant variation in family richness. These analyses (Table 3.2) indicated that although family richness in marshes consistently exceeded that of fens (Fig. 3.2) the differences were statistically significant only for the wet meadow (WM) and emergent zone (EZ). The differences in richness for the SAV zone was not significant ( $p > 0.05$ ) largely because there was large variation among the replicate fen samples.

Randomized block ANOVA analysis indicated that there were no differences in family richness among zones within either marshes alone ( $p = 0.087$ ; Table 3.3) or fens alone ( $p = 0.783$ ).

**Table 3.3: Summary of randomized block two-way analysis of variance (ANOVA) for family richness of emergent insects on sticky traps. Table summarizes the results of two separate randomized block analyses for fens and marshes.**

	<b>df</b>	<b>SS</b>	<b>MS</b>	<b>F</b>	<b>p</b>
<b>Marsh</b>	2	0.020	0.010	2.481	0.087
Remainder	16	0.057	0.004		
<b>Fen</b>	2	0.004	0.002	0.254	0.783
Remainder	6	0.051	0.008		

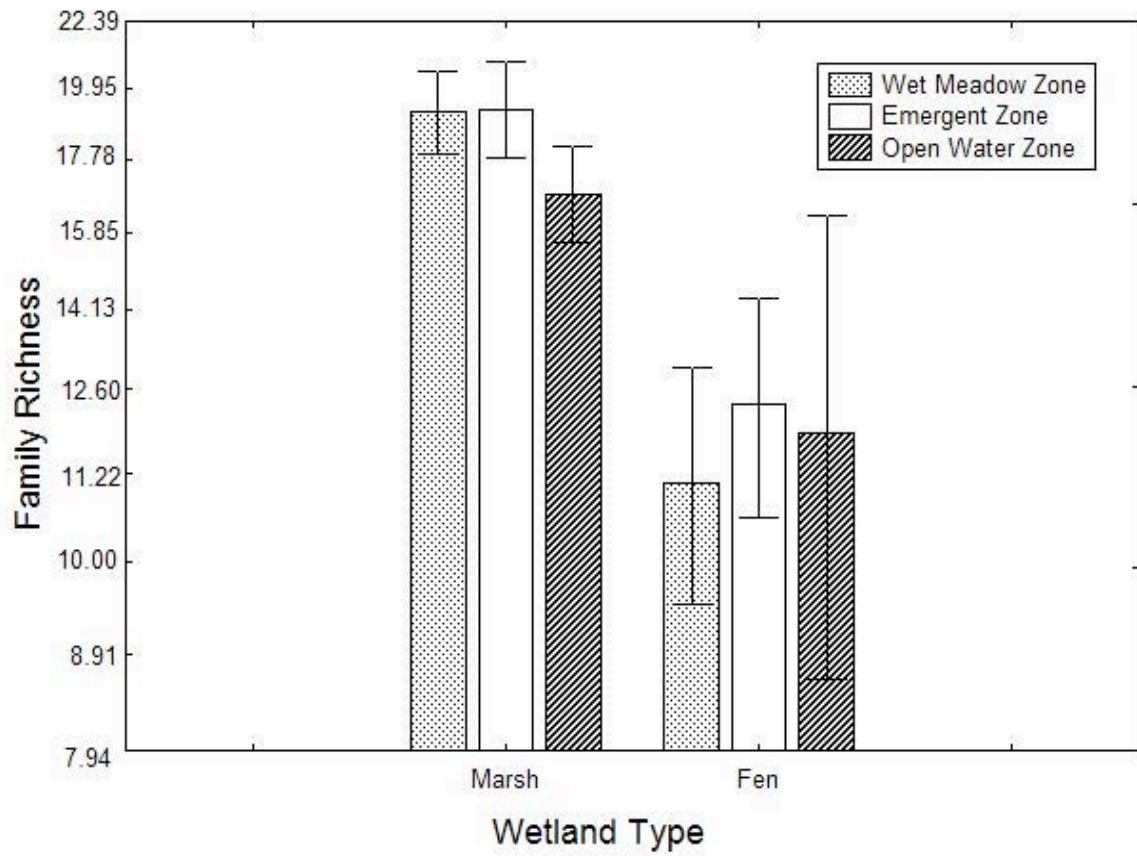


Figure 3.2: Mean ( $\pm$  1SE) family richness of insects collected on sticky traps in 11 marshes and 4 fens.

### *Sticky Trap Biomass*

There were no significant differences in flying insect biomass between wetland types (117.49±1.13 mg DM for marshes vs. 110.14±1.39 for fens;  $p=0.139$ ; Two-way ANOVA Table 3.4; Fig 3.3). Although mean biomass increased from wet meadow emergent vegetation to the submergent vegetation zone, the differences among zones were not statistically significant ( $p=0.855$ ; Table 3.4). The differences among vegetation zones were deemed to be marginally significant ( $p<0.011$ ) among replicate marshes, but not among replicate fens (Table 3.6).

**Table 3.4: Summary of analysis of variance (ANOVA) of total biomass of emergent insects on sticky traps in a field experiment within n=9 marshes, n=4 fens. Analysis is based on randomized block ANOVA. Error degrees of freedom are adjusted to reflect lack of independence of zones within wetlands**

Source	df	SS	MS	F	p
Whole	1	141.036	141.036	467.160	<b>&lt;0.0001</b>
Wetland Type	1	0.011	0.011	0.036	0.855
Wetland Zone	2	1.604	0.802	2.656	0.139
Type x Zone	2	0.194	0.097	0.322	0.735
Error (Within)	<u>7</u>	<u>2.113</u>	<u>0.302</u>		
Total	12	3.922			

**Table 3.5: Summary of analysis of variance (ANOVA) of biomass of emergent insects on sticky traps in a field experiment within n=9 marshes, n=4 fens. Analysis is based one-way ANOVA**

Source	df	SS	MS	F	p
<b>Wet Meadow</b>	1	0.064	0.064	0.709	0.418
WM Error	<u>11</u>	<u>0.994</u>	<u>0.090</u>		
WM Total	12	1.058			
<b>Emergent Zone</b>	1	0.004	0.004	0.076	0.788
EZ Error	<u>11</u>	<u>0.601</u>	<u>0.055</u>		
EZ total	12	0.605			
<b>Open Water</b>	1	0.137	0.137	0.914	0.360
OW Error	<u>11</u>	<u>1.651</u>	<u>0.150</u>		
OW Total	12	1.788			

**Table 3.6: Summary of randomized block two-way analysis of variance (ANOVA) for biomass of emergent insect on sticky traps. Table summarizes the results of two separate randomized block analyses for fens and marshes.**

	df	SS	MS	F	p
<b>Marsh</b>	2	0.555	0.278	5.969	<b>0.011</b>
Remainder	16	0.744	0.047		
<b>Fen</b>	2	1.052	0.526	2.687	0.147
Remainder	6	1.174	0.195		

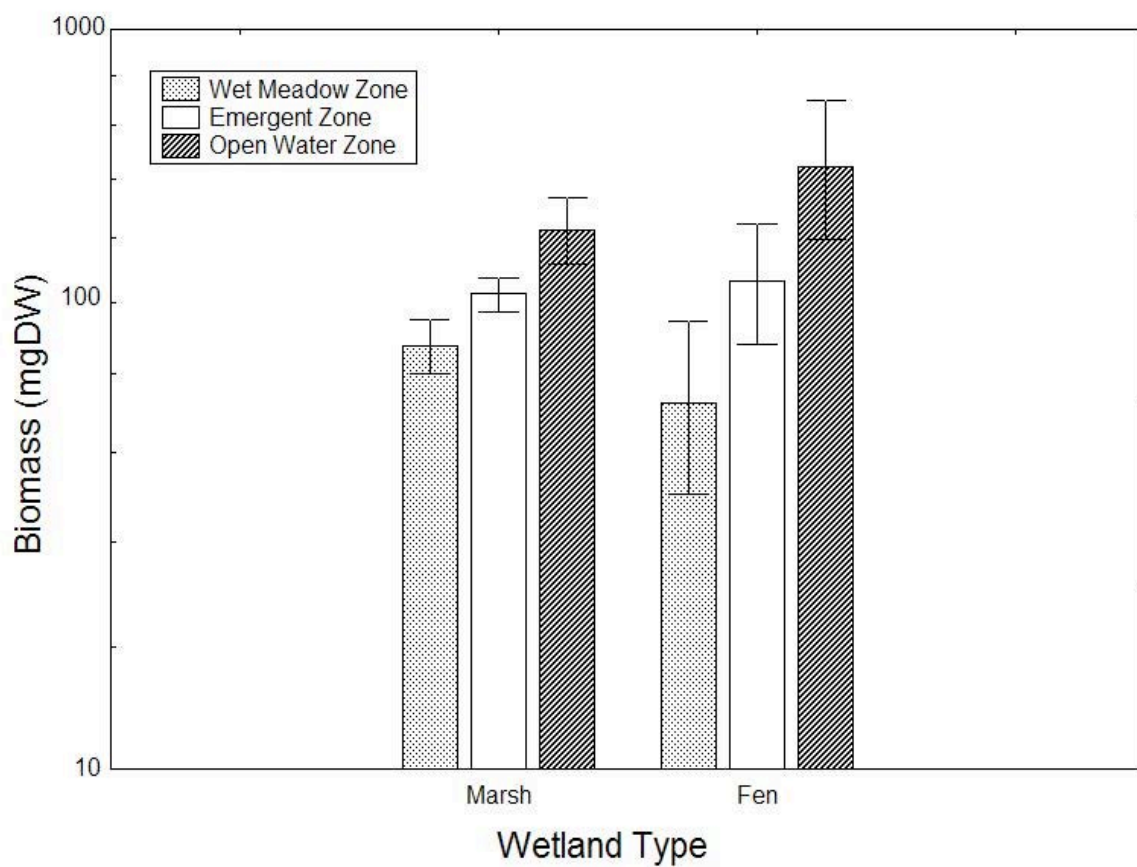


Figure 3.3: Mean ( $\pm$  1SE) biomass of insects collected on sticky traps in 11 marshes and 4 fens.



### *Vacuum Samples*

Vacuum samples contained greater richness and biomass as well as a different composition of species than sticky traps. Mean  $\pm$  1SE family richness from marshes was on average twice the richness in fens ( $40.18 \pm 1.07$  families per marsh vs.  $17.19 \pm 1.15$  families per fen Fig. 3.4); the difference was highly significant (one-way ANOVA  $p=0.000182$ ). Invertebrate biomass was also much greater in marshes than in fens ( $39.39 \pm 1.30$  mg AFDM in marshes vs.  $17.98 \pm 1.37$  mg AFDM in fens). However, the difference was not statistically significant (one-way ANOVA,  $p=0.094$ , Fig. 3.5).

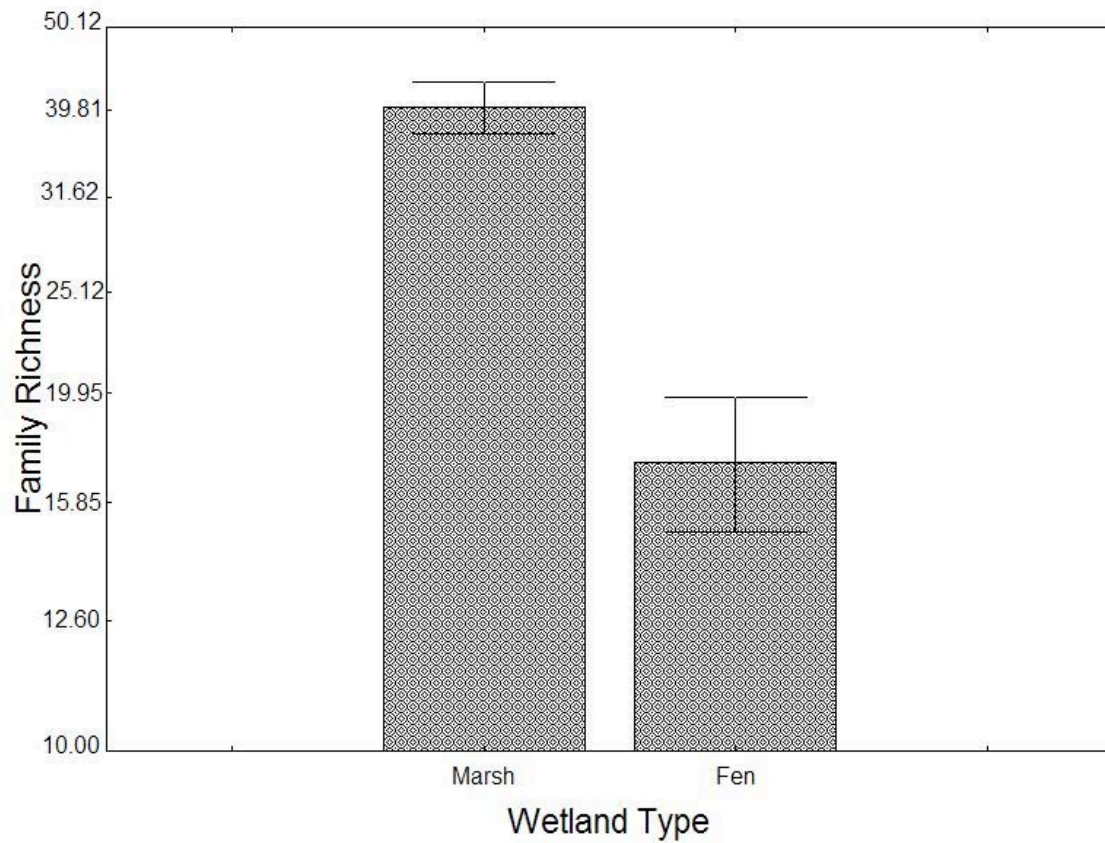


Figure 3.4: Mean ( $\pm$  1SE) family richness of invertebrates collected in vacuum samples at 11 marshes and 4 fens.

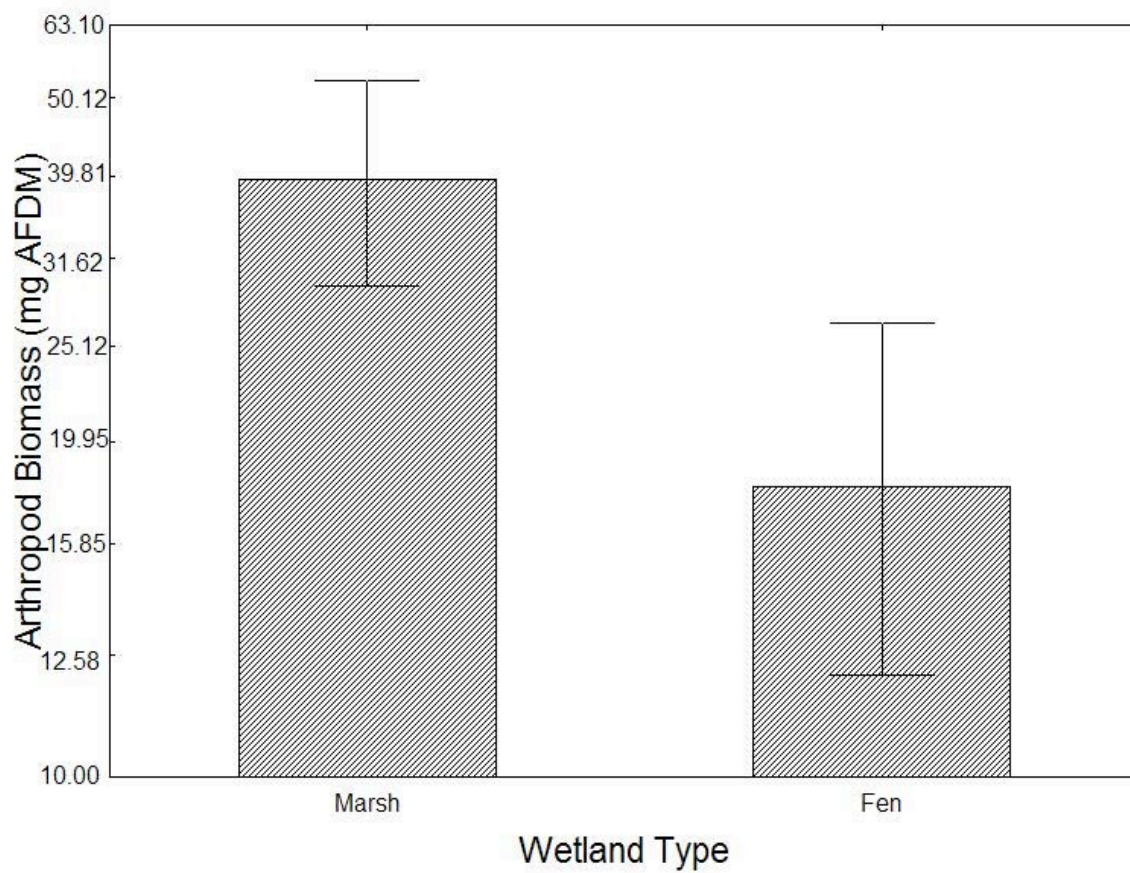


Figure 3.5: Mean ( $\pm$  1SE) biomass (mg AFDM) of invertebrates collected in vacuum samples at 11 marshes and 4 fens.

### *Aerial Sweep Samples*

Aerial sweep samples collected fauna that had elements of both sticky trap and vacuum sample assemblages. In addition, they contained some motile, large-bodied invertebrates that were not captured by either of the other methods. Yet, family richness of sweep samples was intermediate between that of sticky traps and vacuum samples. Mean  $\pm$  1SE family richness from fens was  $17.78 \pm 1.58$  whereas richness for marsh samples was  $26.30 \pm 1.05$ . There was marked among-fen variability in family richness (Fig. 3.6) relative to among-marsh variation. Consequently, the difference in richness between fens and marshes was not statistically significant (one way ANOVA,  $p=0.153$ ). In contrast there was a significant difference in biomass between biomass between fens and marshes ( $p=0.004$ ; one-way ANOVA; see figure 3.7). Mean  $\pm$  SE biomass collected from fens was  $19.67 \pm 10.82$  mg DM. Mean biomass of invertebrates from marshes was, on average 4 times greater averaging  $81.66 \pm 13.28$  mg DM. The presence of large bodied odonates and Lepidoptera in marshes but not fens accounted for most of the difference in biomass.

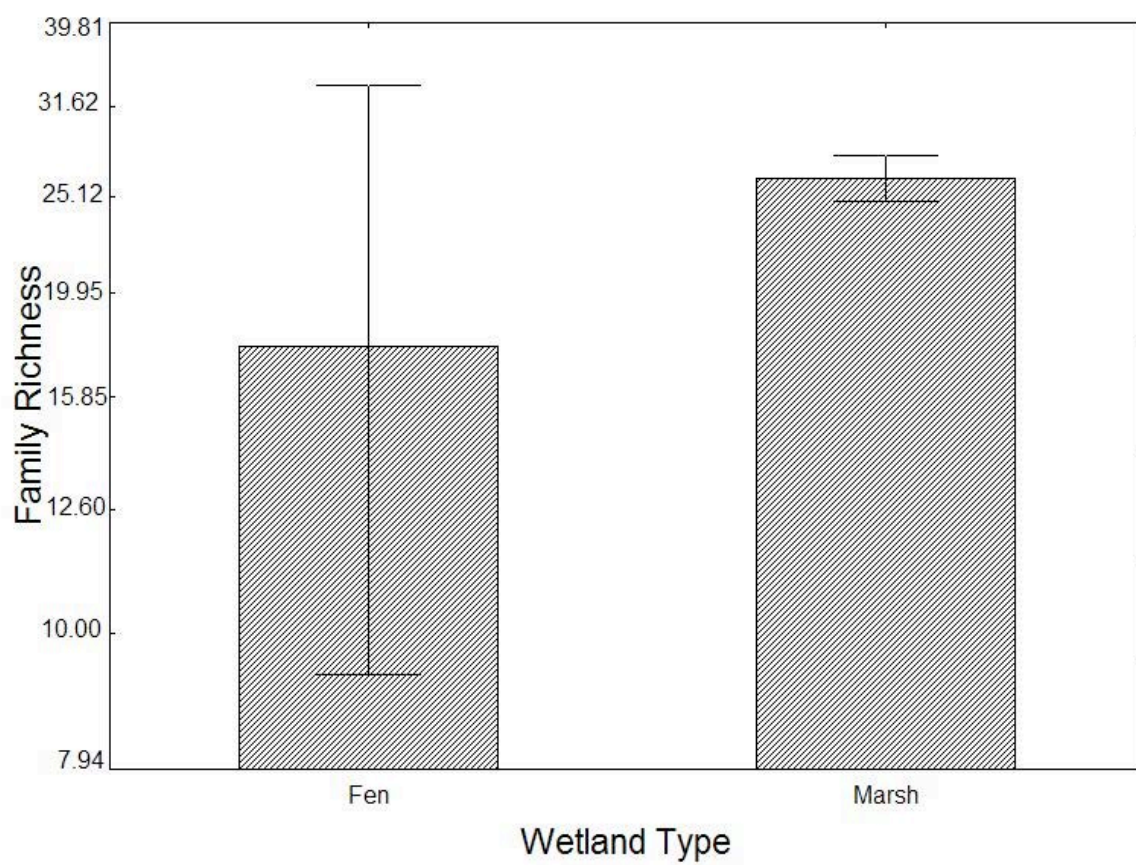


Figure 3.6: Mean ( $\pm$  1SE) family richness of invertebrates collected in aerial sweep samples at 11 marshes and 4 fens.

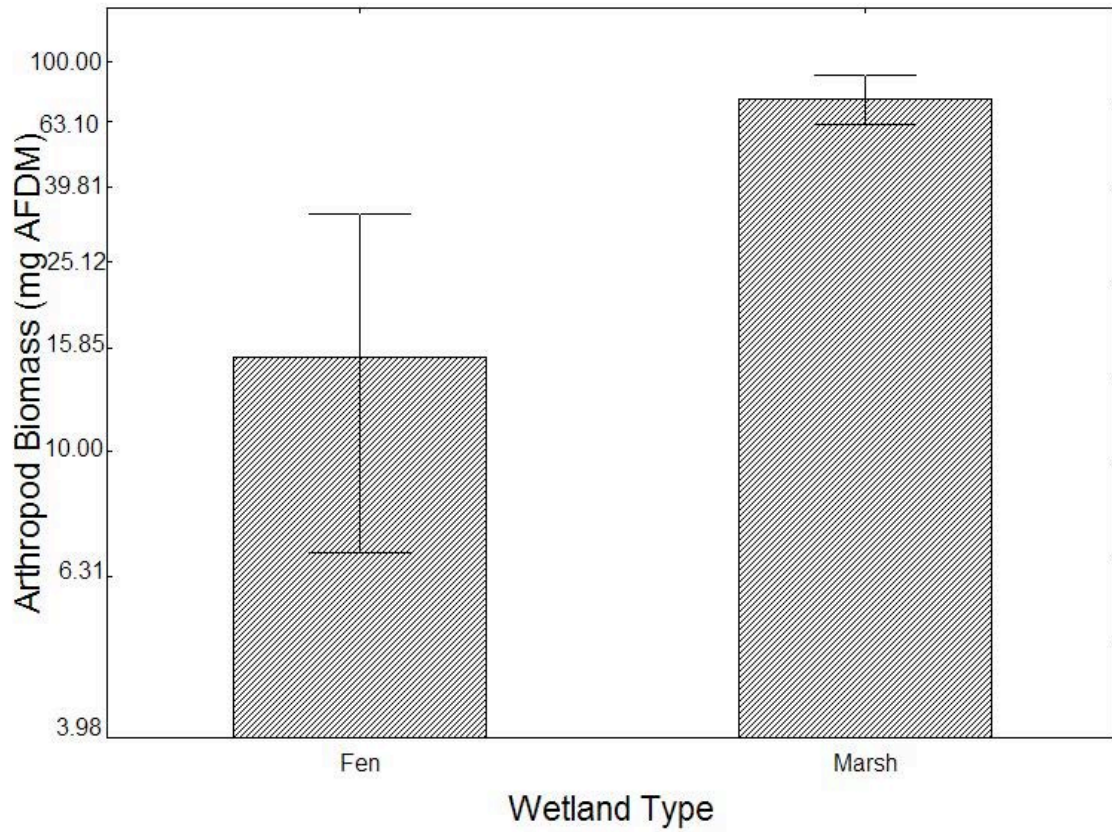


Figure 3.7: Mean ( $\pm$  1SE) biomass (mg AFDM) of invertebrates collected in aerial sweep samples at 11 marshes and 4 fens.

### *Relationship between Vegetation Attributes and Invertebrate Community Characteristics*

To determine which vegetation aspects could best account for invertebrate family richness, abundance, or biomass in vacuum samples, two sets of analyses were performed. Although multiple regression was the preferred method the small sample size available precluded its use. Instead, I summarized the simple correlations between the invertebrate community measures and the vegetation structure summaries (Table 3.7 and Fig 3.8).

Although sample sizes are too small to permit more than subjective evaluations to be made, an assessment of the correlation matrix suggests that all associations were positive. Family richness was most highly associated with plant height, abundance was positively associated with plant biomass, and invertebrate biomass was most strongly positively associated with plant species richness.

**Table 3.7. Correlation matrix summarizing associations between invertebrate community attributes (rows) and wetland vegetation characteristics (columns) based on measurements from 7 marshes and 1 fen. Significant Correlations are bold-faced**

	Wetland Type	Plant Biomass	Plant Species richness (WM zone)	Total Wetland Plant species richness	Maximum plant height (cm)
Invert. Fam. Rich. (WM zone)	<b>-0.87</b>	0.57	0.63	0.67	<b>0.84</b>
Invert. Fam. Rich. (all zones)	<b>-0.86</b>	0.59	0.53	0.57	<b>0.87</b>
Invert. Abundance (WM zone)	-0.51	<b>0.76</b>	0.61	0.68	0.62
Invert. Abundance (all zones)	-0.64	<b>0.81</b>	0.68	0.68	<b>0.69</b>
Mean Biomass/ sample (WM zone)	<b>-0.95</b>	0.42	<b>0.82</b>	0.79	0.61
Mean Biomass/sample (all zones)	<b>-0.81</b>	0.56	<b>0.69</b>	0.51	0.44



**Table 3.8. Non-parametric (Spearman's) correlation matrix summarizing associations between invertebrate community attributes (rows) and wetland vegetation characteristics (columns) based on measurements from 7 marshes and 1 fen. Significant Correlations are bold-faced**

	Plant Biomass	Plant Species richness (WM zone)	Total Wetland Plant species richness	Maximum plant height (cm)
Invert. Fam. Rich. (WM zone)	0.60	0.39	0.52	<b>0.64</b>
Invert. Fam. Rich. (all zones)	0.62	0.23	0.38	<b>0.74</b>
Invert. Abundance (WM zone)	0.62	<b>0.75</b>	<b>0.81</b>	0.36
Invert. Abundance (all zones)	<b>0.69</b>	0.64	<b>0.70</b>	0.43
Mean Biomass/ sample (WM zone)	0.60	<b>0.75</b>	<b>0.81</b>	0.19
Mean Biomass/sample (all zones)	<b>0.69</b>	0.61	0.52	-0.05

### ***Discussion:***

Trends in invertebrate attributes between and within wetlands varied according to the sampling method used. Sticky trap sampling indicated that family richness was greater in marshes than in fens (especially in wet meadow and emergent vegetation zones) but that biomass did not differ. Similarly, vacuum samples showed that marsh wet meadow zones supported the greatest richness of family taxa, whereas biomass showed no significant difference. Aerial sweeps indicated that biomass was greatest in marsh wet meadow zones. Family richness sampled using aerial sweeps was extremely variable, indicating that there may be a trend; marsh wet meadow biomass tended to be higher. Although multiple forms of sampling can be necessary to assess a spatially variable and complex system such as a wetland (Benke 1999), their use makes analysis and interpretation of trends difficult.

Results of statistical analyses of sticky trap data were consistent with predictions in some aspects but not others. Although biomass was not significantly different between wetland types or within wetland zones, differences in family richness were consistently evident. The wet meadow and emergent vegetation zones of marshes were more family-rich than their counterparts in fens. This result is contrary to my predictions for fens. I had expected that the comparable structural homogeneity, dominant graminoid vegetation and hydrology would result in no difference in flying insect community attributes between marshes and fens. I had also expected that fen family richness would be greater in fens due to fens' greater diversity of plant species. My findings for the vacuum sampler data

were not consistent with this prediction. Family richness was significantly greater in marshes. These patterns suggest that the structure of the vegetation in marsh wet meadow zones may better explain the greater taxa richness. The greater plant species richness of fen meadows did not support a concomitantly greater number of invertebrate families.

Also contrary to my predictions, the family richness and biomass of flying insects captured on sticky traps in the SAV zone did not differ significantly between wetland types. These sampling methods cannot directly measure the richness and biomass of invertebrate fauna within the water, but they reflect patterns of insect assemblages associated with areas of open water. This indicates that flying insects seem to be distributed similarly over water regardless of wetland type. Flying adult insects associated with water in wetlands will use it in oviposition. Whelly (1999) studied oviposition patterns of chironomids in oil sands affected wetlands and found that gravid females were equally likely to lay eggs in oilsands process affected water as in reference wetland water. He concluded that the presence of chironomids in SAV zones of wetlands was determined by the tolerance of larvae rather than by adult oviposition behavior. This suggests that aquatic samples are likely to be more representative of wetland water quality than estimates of adult insect biomass and taxa richness collected from sticky traps over open water pools in fens.

Analysis of vacuum sampler data revealed a similar pattern of relative richness and biomass between fens and marshes. There was no significant difference in invertebrate biomass of wet meadow fens vs. marshes. However, marsh family richness was significantly greater than that of fens. These results are complementary to those calculated for sticky traps. As previously stated, fens and marsh wet meadows are considered to be similar in terms of hydrology, spatial homogeneity, and dominant graminoid vegetation. However, wet meadows of marshes differ slightly by experiencing some dry phases (Batzer and Wissinger 1999), possibly because of differences in soil characteristics. Peat has higher water-holding capacity than the inorganic soils that may underlie the wet meadows of marshes (Smith 2007). The continual saturation of peat with anoxic water within plant rooting zones is what reduces rates of bacterial decomposition and allows peat to form (Vitt 1994). In contrast, dry phases experienced by wet meadows of marshes allow detritus to decompose under the aerobic conditions (releasing nutrients), which would in turn, allow these habitats to support higher taxa richness (Batzer and Wissinger 1999). Therefore, it follows that our findings suggest higher taxa richness in wet meadows.

Aerial sweep net samples showed different patterns of biomass and richness than were observed for sticky traps and vacuum samples. Biomass differed significantly between wetland types whereas family richness did not differ. This is largely due to the capture of large-bodied Odonata, Lepidoptera, and Hymenoptera in marshes but not in fens.

Addressing the ecological question: does plant species diversity or vegetation structure best explain arthropod community attributes using correlation yielded some spurious significant relationships. Significant results were: family richness of arthropod was positively correlated with maximum plant height within the box plot samples and arthropod abundance and biomass from the wet meadow zones were positively correlated with total plant species richness. These results are in agreement with the findings of Schaeffers (2008) who stated that this vegetation metric's importance for arthropod richness is only underrepresented or under supported in the literature due to the necessary time and skill it takes to measure plant species richness. He further expanded upon this relationship by saying that not only is plant species richness a better predictor of arthropod community metrics, but these two vegetation variables are intrinsically tied; as plant species richness increases, so would vegetation structure as a result of each species unique architecture.

### ***Conclusions:***

Data from sticky traps and vacuum samples indicate that the wet meadow and emergent vegetation zones marshes support greater family richness than equivalent zones in fens.

Biomass estimates collected from sticky traps demonstrate a trend of increasing biomass across hydrological zones; SAV zones tended to support greater biomass. Although there was not a significant difference, biomass tended to be higher in the SAV zones of

marshes. Aerial sweep net samples, on the other hand indicated that biomass was higher in marshes, though this comparison was limited to the wet meadow zone.

An important consideration when sampling wetland arthropods is the type and quantity of samples needed to be representative of the variation in microhabitat and resulting taxa, and more practically, efficient (defined as the relationship between sampling effort and taxa collected). Three sampling types were utilized here in order to meet these criteria. However, each sampling type has its own specific biases. Briefly, sticky traps are versatile in their use across hydrological gradients, but are only able to capture flying insects. Sticky traps used alone would underrepresent the taxa inhabiting any wetland especially fens, as they are often considered to be terrestrial transition habitats. Arthropod metrics calculated using aerial sweep nets were highly variable in this study. Although aerial sweeping is often the most convenient method of arthropod sampling, insect catches are extremely dependent on humidity, temperature, and sampling intensity. I do not recommend the use of aerial sweep nets for calculation of taxa richness or biomass. With regards to sampling fen habitats, aerial sweep nets do not sample, with any consistency, terrestrial, ground-dwelling taxa. This is especially important as the majority of fen taxa live within the profile of low-lying vegetation. Finally, vacuum sampling is a novel method for sampling taxa that live within the profile of grassy, shrubby vegetation. It was effective at sampling flying taxa as well as ground-dwelling taxa. However, it became less effective as wetland substrates became heavily inundated with water, which is a characteristic of fen substrates. I recommend the use of vacuum sampling in marsh wet meadows.

Time constraints limited the level of taxonomic resolution that could be used to assess trends in invertebrate richness among zones and between wetlands. Identification of taxa to the genus or species level, especially of aquatic taxa would improve the understanding of patterns of invertebrate community attributes. However, the nature of a sticky trap limits the taxonomic resolution of identification in this study. Previous research has observed that sticky traps left in place for 3 days collected more insects than traps left in place for shorter periods of time. However, the proportion of damaged specimens increased as a function of time in a trap. Other researchers have left sticky traps in place for only 24-h periods to minimize the effects of weather damage. They were able to catch large number of insects by timing their sampling to correspond with periods when large emergences of insects occurred, in mid- June. I collected samples from traps left in place for 3 consecutive days in “ideal” weather, but many of the insects lost limbs, wings or antennae over this interval. A more suitable approach would be to sample over 3-day intervals but to collect and replace the sticky sheets every 24 h.

Future research in wetlands that seeks to document relationships between vegetation characteristics (i.e. structure, species richness) and invertebrate biomass and taxa richness will require comprehensive sampling for both invertebrates and vegetation. The understanding of this relationship in wetlands will become extremely important in the future, as large reclamation projects will seek to certify reclaimed lands. Without knowledge of plant-arthropod interactions in natural habitats, science-based reclamation targets will be lacking a key component.

## CHAPTER 4

### GENERAL CONCLUSIONS

#### *Project Objectives*

The objectives of this project were to determine the influence of wetland zonation, both vertical and horizontal, on invertebrate community attributes. Because vegetation characteristics are most commonly correlated with arthropod biomass, abundance, and taxa richness, I investigated how the presence of hydrological vegetation zones would influence the distribution of invertebrates along this gradient. Furthermore, in my study area, wetland habitats are primarily peat-forming fens. As such, I investigated the role of vegetation characteristics of these unique habitats on invertebrate community attributes of family richness, abundance and biomass.

#### *Major Findings*

Sticky traps deployed in constructed wetland plots that supported marsh and fen vegetation collected similar family richness, abundance, and biomass of flying insects. When experimental cells (those containing peat) were compared to control plots (without peat) only 3 families were significantly more abundant in test cells than control cells. These families (Ceratopogonidae, Chironomidae, and Dolichopodidae) all utilize an aquatic or semi aquatic habitat as larvae. The lack of significant differences in this (Chapter 2) study may reflect the nearness of cells to one another, or the coarse level of taxonomic resolution used in this study. However, the differences in vertical stratification of aquatic taxa suggest that sticky traps do sample the local emergent fauna. Furthermore,



one can be confident that the assemblages of insects within study plots are present based on the relatively large distance between the experimental field and adjacent wetlands. The closest known body of freshwater is Mildred Lake, which is approximately 3.5 km away. This indicates that insects are responding to vegetation and wetland conditions.

In natural wetlands, patterns became more apparent. Natural marshes exhibited greater family richness and biomass, overall than fens. Furthermore, when family richness was compared across zones, wet meadow and emergent vegetation zones of marshes supported significantly higher family richness than the same zones of fens. Wet meadow zones of marshes are similar to fens due to dominance of graminoid vegetation as well as their structural homogeneity. In contrast, marsh wet meadows experience dry phases, which would cause detrital decomposition and in turn supports greater taxa richness (Batzer and Wissinger 1999). Vacuum sampler data also indicated that marsh wet meadows produced the greatest family richness between wetland types.

SAV zones of marshes and those adjacent to fens did not appear to support significantly different arthropod communities with respect to biomass and family richness. This indicates that arthropods are responding to open water, independent of wetland type. Indeed, Whelley's (1999) investigation of chironomid oviposition in oil sands affected wetlands demonstrated that abundance is determined by larval survivorship rather than female choice as females oviposited in open water regardless of water quality.

Sampling using an aerial sweep net captured the greatest biomass of arthropods in marshes, but this was likely an artifact of sampling bias associated with sweep nets as well as the presence of a few large-bodied taxa.

Correlation matrices were used to analyze the ecological controversy that asks: does plant species diversity or vegetation structure best explain arthropod community attributes? Family richness of invertebrates was positively correlated with maximum plant height within the box plot samples, and invertebrate abundance and biomass from the wet meadow zones were positively correlated with total plant species richness. These results are in agreement with the findings of Schaeffers (2008) who stated that plant species diversity is the best predictor of arthropod richness.

### *Limitations of Research*

Limitations of the sampling procedure used in this thesis research meant that although invertebrates were captured in all available microhabitats and wetland zones, the open water and emergent zones were only sampled using one sampling method, sticky traps, due to their versatility. However, knowing the bias of this trapping type towards small, emergent aquatic insects, their trapping is not representative of the entire pool of invertebrates in wetlands. Furthermore, their bias towards small-bodied invertebrates means that larger inhabitants of the emergent and open water zones are not being captured, identified, and their additional biomass is not counted. Taxa excluded by sticky traps could include many families of Lepidoptera, Odonata, Trichoptera, Ephemeroptera,

and Coleoptera as well as aquatic invertebrates that do not have flying stages (Annelida, Crustacea, Mollusca). Techniques to sample the inundated zones of wetlands that are not biased towards small bodies aquatic insects should be utilized in future. For example, sampling of odonates could include direct observation of flight activity, or elevated pan traps as well as using dipnets, cores or other techniques suitable for aquatic collecting.

This research was conducted using one set of data from each wetland. As such, calculation of “energy” could only be measured in biomass, rather than productivity that would require measures over time.

When plant community attributes were correlated with invertebrate attributes in boreal marshes and a single fen, significant correlations were observed. Multiple regression would have been the preferred statistical method. However, I was limited by the type and quantity of data available for this procedure. The plant community attributes that were used in the correlation analysis were plant species richness, maximum plant height within box plot samplers, and plant biomass (mg DW). In future, further measures of plant structural complexity and density are required to better address the ecological controversy in question (see Chapter 3).

#### *Recommendations for Future Research*

Three sampling methods were used to sample the variety of wetland microhabitats present in fens and marshes. However, due to the bias associated with each sampling type, a comparison of methods for sampling efficiency (the relationship between number

of taxa collected and sampling effort) and detection biases would be useful to inform future research in boreal fens.

Fen wetlands are diverse in nutrient regime and vary from poor fens (nutrient poor) to rich fens (nutrient rich). Therefore, studies of the effects of nutrient level on arthropod biomass and taxa richness would provide an interesting contrast to knowledge of wetland invertebrates. Nutrient regime is only one aspect of the diversity of peatland forms.

Peatlands in boreal Alberta also differ in terms of dominant vegetation. Graminoid fens were chosen for this research to contrast with marsh wet meadow zones. However, many fens in this region are wooded fens. The invertebrate community attributes of wooded fens should be addressed, and perhaps contrasted with that of graminoid fens.

Fen reclamation has recently become government mandated and oil sands leasees have already begun the process of testing conceptual reclamation models. In light of these new conceptual models, research involving faunal recruitment is sure to follow. Reclamation of peatlands in the boreal region of Alberta is vital for maintaining regional biodiversity.

In designing future sampling protocols, oil sands leasees must first establish “what is the purpose of sampling?” Measuring biological performance is generally the goal of invertebrate sampling. Oil sands leasees can determine the biodiversity, functional measures (biomass, productivity), or community composition of habitats based on the goals of sampling. After determining goals, a protocol can be designed that addresses these.

Invertebrates are a vitally important component of ecosystem function, and the description of their role in future reclamation of wetlands can give clues as to its success.

Invertebrate biomass or productivity in marshes, describes nutrient availability for primary consumers, food resources available to secondary consumers, and allows researchers to make assertions about wetland biological condition, as marshes are, generally, productive habitats. In contrast, fens are by nature, non-productive habitats. This lack of productivity (due to anoxia) fosters the creation of peat and makes fens an important carbon sink. Invertebrate productivity measured from reclaimed fen (peat-forming) wetlands would give clues about the future sustainability of the wetland. High invertebrate productivity in reclaimed fens would indicate oxic conditions that would result in peat decomposition, food resources for primary consumers, and perhaps eventually a failure of the reclamation goal. However, future research needs to determine which invertebrate species are “resident” in reclaimed fens, indicating non-sustainable peat reclamation, versus which species are transient, or are utilizing fen resources in a capacity that is non-destructive to peat accumulation. This future research could culminate in the identification of “ fen reclamation indicator species” that would aid oil sands leasees in establishing guidelines for assessment of successful reclamation.

Specific recommendations from this thesis research will be useful for future invertebrate sampling in fens and marshes. Firstly, regarding sticky traps: this sampling technique is useful for collection of emergent adult insects in a variety of habitats. However, they are biased towards smaller-bodied insects and therefore should be used in conjunction with another sampling technique. If the sampling protocol requires higher-level taxonomic identification, they are not recommended as deployed in this study because insects became damaged during collection. To minimize damage to collected specimens, I

recommend collection after 24 h over three consecutive days, rather than recovering traps after three continuous days.

The modified Stihl leaf blower/vac that was used in this research was very effective at collecting invertebrates from grassy plants and the plant-soil interface. Invertebrates collected were not damaged and would be suitable for higher-level taxonomic identification. This sampling procedure used alone would likely underrepresent flying insect taxa, so it should be used in conjunction with another sampling type if the goal is to sample all invertebrate types/microhabitats within a habitat. I observed few disadvantages to this sampling technique, although one major disadvantage is the inability to sample in inundated habitats. This sampling technique is ineffective at sampling habitats with >1cm water above soil surface. I sampled invertebrates within vegetation by dividing sampling into three strata (see Chapter 3). This technique was extremely time consuming. In future, I recommend not processing the clipped vegetation for invertebrates.

Aerial sweep net sampling is not effective in fen or wet meadow habitats. The net cannot accurately collect insects from within dense vegetation and therefore, underrepresents invertebrate community attributes. Aerial sweep netting would be appropriate to subsidize other sampling techniques for measurement of biodiversity.

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## APPENDIX 1: SAMPLING SITE DESCRIPTIONS

### ***MARSH SITE DESCRIPTIONS***

Wetland names, below, correspond to wetland inventories compiled by Golder Associates (2003) and CEMA (2014) where additional information can be found. Wetlands that have not been previously surveyed are denoted in descriptions.

#### ***Syncrude Sites***

##### *U-Shaped Cell Test Plots (USC, Constructed; UTM: 12 V, 0460221E, 6323155N)*

Twenty-eight wetland plots were created in 2008 to provide pilot data on plant development potential as a precursor to the creation a 50 ha fen on Syncrude's Mildred Lake Lease. The 20 x 10 m plots were created by placing either live peat (peat dug from a nearby fen and transported and placed intact onto the substrate) or stockpiled peat (peat that had been collected during the process of overburden removal prior to the opening of a new surface mining site and stored for a period of 10 y) on a substrate of mine tailings sand. The natural fen vegetation of plots that were created using live peat survived the transplanting process, and these plots resemble a fen wetland. The long storage period of the stockpiled peat resulted in death of propagules in the seedbank. Consequently, these plots were colonized by airborne seeds and became dominated by species that are typical of marshes (primarily *Typha latifolia*). When stockpiled peat is placed in wetland plots, these plots resemble marshes. These wetland plots were well suited to assessing the flying insect fauna associated with marsh vs. fen wetland vegetation.

*Shallow Wetland (SW, Constructed, reference; UTM: 12V, 0457759E, 6326653N)*

This 2.75 ha wetland was constructed in 1993 on the Syncrude Canada Ltd. Mildred Lake lease on a substrate of tailings sand. It was filled with fresh water from a diverted stream at the time of construction. This wetland has been extensively sampled for diversity, productivity and water chemistry; Shallow Wetland has is 0.5-0.75 m deep and has the characteristic wetland vegetation zones. The open water zone supports many species of macrophyte including Floating Pond weed (*Potamogeton pectinatus*), Coontail (*Ceratophyllum demersum*) and Water milfoil (*Myriophyllum angustifolium*), etc. The emergent zone contains large stands of Cattail and Greater Bulrush. The wet meadow zone supports a wide variety of plants with the majority being sedges (*Carex aquatilis* and *Carex utriculata*).

*Katie's Sedge Meadow (KSM, Natural; UTM: 12V, 0458283E, 6317709N)*

This wetland is located just to the west of Syncrude's South Tailings pond. It is a naturally-formed wetland of indeterminate age, although the presence of mature white spruce trees at the periphery of the riparian zone indicates that this is an established, natural wetland. This wetland is part of a long string of natural wetlands that most likely would have drained into Mildred Lake prior to Oil sands development. KSM has an extensive sedge meadow zone that surrounds the small, deep (>2 m in the centre) wetland. This zone is dominated by sedges of the genus *Carex* and many different shrubs including Willow (*Salix* spp.) and Prickly Rose (*Rosa acicularis*). The emergent zone of KSM is dominated by Cattails (*Typha latifolia*) with intermittent stands of Horsetail

(*Equisetum* spp.). The open water zone at the time of study (20-23 June 2012) was covered in Common Duck Weed (*Lemna* spp.), further submergent macrophytes were not visible.

*Southwest Sands Beaver Wetland (SWSS, Natural; UTM: 12V, 0456571E, 6315836N)*

SWSS Beaver Wetland is located in the Southwest Sands Storage area of Syncrude's Mildred Lake Lease. This wetland is a natural wetland of indeterminate age, but just as is the case for KSM, the presence of old growth riparian vegetation indicated that this is an established, natural wetland. To access the emergent and open water zones of this wetland, one must walk through an extensive wet meadow zone dominated by sedges and small shrubs. The open water zone is partitioned by islands created by the past activity of a beaver.

***Off-Site Wetlands***

*Ruth Lake Marsh (RLM, Natural; UTM: 12V, 0465627E, 6316229N)*

This marsh is an opportunistic marsh that was formed as a result of increasing water levels in the adjacent Ruth Lake. Ruth Lake (and eventually Ruth Lake Marsh) was formed after 1975 when surface water was diverted from the Beaver Creek reservoir in service of surface mine creation on the Syncrude Mildred Lake Lease. Although this marsh is located in close proximity to oil sands activity, it is free of oil sands effluent. Ruth Lake marsh is a relatively deep wetland (<2 m in the center) formed beside a very

steep embankment. The majority of this marsh is an extensive emergent zone (and inaccessible without a boat). A very limited wet meadow zone is accessible by a small path to the north of the marsh, and leads into the small, deep open water zone. The wet meadow zone is dominated by Willow (*Salix* sp.) shrubs and *Carex* sedges. The emergent zone is an extensive zone of Cattails (*Typha latifolia*). Submergent vegetation was not observed in the open water zone, due to the steep slopes and deep water.

*Tower Road Spruce Pond (HSB, Natural; UTM: 12V, 0463684E, 6290569N)*

This reference wetland is located within Fort McMurray and is one of the "Tower Road Wetlands" (a string of reference wetlands on Tower Road, located in the Timberlea subdivision of Northern Fort McMurray). Wetlands on Tower Road are natural wetland of indeterminate age (Golder Associates 2003). These wetlands are intermittently disturbed by heavy equipment due to their proximity to Tower Road. During 2012, these wetlands were undisturbed. This large wetland (7.8 ha) has extensive sedge meadow on the western and northern shores. The wet meadow is composed *Carex* sedges and Willow (*Salix*) shrubs. The emergent zone is composed primarily of a thick stand of cattails that extends into the open water zone. The open water zone is >1 m deep. In June and July 2012, this wet meadow region of this wetland was flooded to a depth of up to 20 cm.

*Moose Wetland (MW, Natural; UTM: 12V, 0469520E, 6289123N)*

Moose wetland is also located on Tower Road. This natural wetland is very similar to Tower Road Spruce Pond with an extensive *Carex* wet meadow zone, emergent zone dominated by cattails, and a deep open water zone ( $> 1$  m deep). Moose wetland is (3.39 ha).

*Tower Road 1 (NI, Natural; UTM: 12V, 0469718E, 6289158N)*

Tower Road 1 is located on Tower Road within 500 m of a housing subdivision in Fort McMurray, Alberta. This small (2.11 ha) wetland is a typical marsh; open water is greater than 1 m in depth, the emergent zone is dominated by Cattails, and wet meadow vegetation is composed primarily of *Carex* sedges. The wet meadow extends into stands of Willow shrubs and Aspen.

*Rhyno's Watering Hole (RWH, Natural; UTM: 12 V, 0479419E, 6274282N)*

Rhyno's Watering Hole (RWH) is a newly monitored wetland for 2012, no previous research on this waterbody is known. In 2012, however, this wetland was monitored for Avian species richness and Plant community characteristics by Sheeva Nakhaie and Marie-Claude Roy, respectively. This marsh is of indeterminate age. It lies in a depression between large stands of white spruce at the edges of the wet meadow, indicating that it is a mature, natural wetland. RWH is located in the east side of HWY 63 approximately 5 km from the airport road turnoff south of Fort McMurray.. This large (5.21 ha) wetland has an extensive wet meadow zone that transitions directly from white

spruce, and vegetation often resembles that of a fen. Wet meadow vegetation is primarily *Carex* grasses. However, patches of brown mosses and other typical fen plants indicate that the surrounding forest is a wooded fen. The emergent zone is comprised of thick stands of cattails that grow >1 m tall. The open water zone is >1 m deep.

*Sam's Rodeo (SRW, Natural; UTM: 12V, 0481490E, 6278822N)*

This marsh-type wetland is located approximately 2.5 km down HWY 69 from the HWY 63 turn off. This wetland is a relatively small in area (1.91 ha). This wetland is also a new wetland monitored for the 2012 season, no previous research on this wetland is known prior. In 2012, however, this wetland was monitored for Avian species richness and Plant community characteristics by Sheeva Nakhaie and Marie-Claude Roy, respectively. During 2012, this wetland was within 100 m of road construction; Sam's rodeo is within 20 m of HWY 69. The wet meadow zone of this wetland is small (a maximum of 20 m wide on the accessible northern side) and is comprised of *Carex* sedges and willow (*Salix*) shrubs. The emergent zone is narrow (<10 m wide) and is composed mainly of *Equisitum* interspersed with *Typha*. The open water zone is approximately 50 m wide and >1 m deep. There were many species of submergent macrophyte present including *Potamogeton* sp., and *Ceratophyllum demersum*.

## ***BOREAL FEN SITE DESCRIPTIONS***

*Maqua Lake Fen Complex (MLF) (UTM: 12V, 482923E, 624666N)*



Maqua Lake Fen serves as a natural Fen- type wetland in this study. This fen is approximately 75 ha in area, which includes a patterned fen, a graminoid fen, and at the most eastern portion, a lake and the attached Maqua Lake Provincial Recreation Area. This wetland area was used as a natural fen. However, unlike the other fens that were sampled in this study, this fen was adjacent to a large expanse of open water (Maqua Lake). The peat mat of the patterned and graminoid fen ends abruptly on the western side of Maqua Lake. Samples were taken in this open water body in order to contrast the biomass of fen habitat to the more typically “marsh” habitat of open water. Because Maqua Fen complex is also part of a greater Maqua Lake Provincial Recreation area, this fen is disturbed annually by recreational vehicle traffic (Golder Assoc. 2011). For this reason, samples were taken in obviously undisturbed areas of peat, remote from tire ruts in the moss.

*Sphagnum* mosses, herbaceous shrubs, and grasses that exist in a homogeneous mat across the fen dominate the vegetation of Maqua Fen Complex. This is a graminoid fen, exhibiting little vertical stratification (presence of tall, emergent vegetation) or vegetational zonation (presence of distinct wetland zones). Consequently, I postulated that most invertebrate biomass in this fen would exist within the moss strata within the patterned fen portion. However, I also expected that there would be a great deal of biomass contributed by the open water “lake” portion of the fen complex.

*Pauciflora Fen (UTM: 12V 485378E, 6248068N)*

This fen is characterized as a poor fen, meaning that it is fed by groundwater. However, it is oligotrophic and acidic, and as such, it supports many acid-tolerant plant species. This fen is approximately 7 ha in area and is situated in an expanse of narrow, flat valley that sits at the base of two hills, covered in typical boreal mixed coniferous forest. The main expanse of moss-dominated open fen is surrounded in stunted Black Spruce (*Picea mariana*) Measurement of water quality revealed the typical acidity of a poor fen (pH 4.2). The dominant vegetation of this wetland was also indicative of a poor fen. The main expanse is covered in a thick mat of acid tolerant Sphagnum moss, as well as many sedge species including *Carex pauciflora* (for which this wetland is named), and Cottongrass (*Eriophorum* spp.); small cover of woody shrubs including Leatherleaf (*Chamaedaphne calyculata*) as well as other herbaceous vegetation including species of Bog Cranberry (*Oxycoccus microcarpus*), Bog Laurel (*Kalmia polifolia*).

The majority of this fen is an undisturbed expanse of *Sphagnum* peat mat. Samples taken in this fen sampled the gradient of vegetation present. Samples extended from the ring of stunted Black Spruce surrounding, into the homogeneous peat mat dominated by *Sphagnum*.

#### *Gravel Pit Patterned Fen (GPF) (UTM: 12V, 467790E, 6311975N)*

Gravel Pit fen is the largest of the fens sampled in this study. It is approximately 200 ha in area, although the borders are not clearly defined. This fen also sits in the valley of two mixed conifer covered hills, which gives the margin of this wetland wooded fen borders that transition into shrubbery. The main expanse of moss-dominated peat is

characterized as a patterned and/or graminoid fen. Unique to this fen was the expanse of marsh plants adjacent to the access road at the Southern edge. Cattails (*Typha latifolia*) and Greater Bulrush (*Scirpus hudsonianus*) grew in discrete clumps on top of peat hummocks. Further into the fen, there are pools of stagnant water that seem to denote breaks in the peat layer (pers. observ.). Vegetation in this fen is typical of rich fens: brown mosses, *Carex* sedges, Leatherleaf, and Pitcher Plant (*Sarracenia purpurea*), for example. Water chemistry of Gravel Pit fen was also indicative of a rich fen. It was slightly alkaline (pH 7.8) with specific conductivity of 383 uS/cm. Samples taken in this fen sampled along a gradient from South to North, from the shrubby fen margins into the expanse of graminoid fen that makes up the majority of this wetland.

*Beaver Lodge Fen Wetland (BLW) (UTM: 12V, 0483271E, 6263298N)*

This wetland is a smallest fen-like wetland that was sampled for this study. It was also an especially suitable location at which to assess variation in arthropod distribution with respect to vegetation zonation and vertical stratification zonation, as it contains an opportunistic marsh habitat within it. Although, the age of the marshy area is unknown, it likely formed as the result of beaver activity (pers. observ.). Access the fen portion of the wetland, required either wading through the shallow marsh, or walking across an old, grown-over beaver dam. The fen portion of this wetland is best described as a graminoid fen transitioning from a wooded fen (situated at the southern border of the wetland). Typical fen plants characterize the fen portion of this wetland: brown mosses, leatherleaf (*Chamaedaphne calyculata*), cottongrass (*Eriophorum angustifolium*), etc. The marsh

portion of this wetland transitions directly from fen vegetation into the emergent zone, dominated by cattails (*Typha latifolia*). The open water portion of this wetland is shallow (<1 m deep) and is dominated by submergent macrophytes including floating pondweeds (*Potamogeton pectinatus*), Coontail (*Ceratophyllum demersum*) and Water milfoil (*Myriophyllum angustifolium*). To sample the gradient of insect activity, and to contrast the influence of fen and marsh vegetation on insect biomass, sticky traps were placed in the fen vegetation, the emergent zone of the contained marsh, and the open water portion of the marsh.

## APPENDIX 2

### A COMPARISON OF STICKY TRAP, VACUUM, AND AERIAL SWEEP NETTING METHODS FOR SAMPLING TERRESTRIAL INVERTEBRATES FROM NATURAL BOREAL WETLANDS

#### *Introduction:*

Wetlands are widespread and important habitats whose landmass is likely to decrease in the future due to anthropogenic habitat destruction and climate change (Batzler and Sharitz 2007). Wetland conservation not only promotes the preservation of regional biodiversity, but wetlands provide a wide variety of ecosystem services. These services include those of ecological and anthropogenic importance: ecological services include nutrient cycling, flood regulation, waste treatment, and water supply; Anthropogenic services include recreation, food production and cultural uses (Costanza et al. 1997).

In recent history, one disturbance of particular interest is that of surface mining in the Athabasca oil sands region of Northeastern, Alberta. This region lies within the greater biome of the boreal forest. This biome is distributed across subarctic regions worldwide including regions of Russia, Fennoscandia, and Canada; the boreal forest makes up approximately 35% of Canada's total land area (Vitt and Bhatti 2012). Alberta's boreal forest includes large areas of wetlands, including natural marshes and peat-forming fens that make up approximately 60% of the natural landscape in the boreal region of Alberta (Rooney and Bayley 2011). Actions of oil sands related surface mining are decreasing

the constituent of natural wetlands. This decline in wetland area is of great concern for the preservation of regional biodiversity.

### *Oil Sands Disturbance and Reclamation Requirements*

Due to the shallow nature of oil sands deposits, Oil sands extraction requires surface mining. During this process, shallow mine pits (~100m in depth) are dug to reveal bitumen deposits. Prior to digging a new mine, overburden is removed from the area, disturbing entire landscapes. Overburden removal generally includes draining of wetlands, removal of peat, and clear-cutting forest. Of note is the practice of storing overburden materials in discrete piles for use in later reclamation (Price et al. 2010). By law, under the Alberta Environmental Protection and Enhancement Act (AEPEA), oil sands companies are required to reclaim the land to “equivalent land capacity” (GOA 1993). As such, the pre-mining proportion of wetland landscape must be reclaimed.

As was previously discussed in the general introduction, wetland reclamation requires knowledge of the pre-disturbance landscape. Research surrounding wetland reclamation has, until the recent past, focused on the reclamation of marsh-type wetlands due to their relatively simple hydrology (Daly et al. 2013). Marshes are also commonly opportunistic and may develop in poorly drained soils, even in disturbed habitats (Harris 2007).

Marshes amount to a relatively small proportion of the natural landscape of the Athabasca Oil Sands region (about 3%), as such, and to resolve concerns regarding loss of regional biodiversity, the AEPEA has included conceptual fen reclamation models into

their mandate (Price et al. 2010). The necessity of this results in one of the largest reclamation projects in Canadian history, as such there is an urgent need for science-based reclamation goals.

### *Arthropod Sampling in Wetlands*

Wetland arthropods constitute a significant component of wetland productivity, function, and diversity and therefore, the assessment of reclamation success will require accurate estimation of arthropod taxa richness, abundance, and biomass within wetlands; this will be achieved through accurate and representative collection methods. Representative sampling of the distribution and abundance of wetland arthropods is difficult due to the variety of microhabitats within a wetland; sampling often requires more than one collection technique (Benke 1999). Wetlands are aquatic-terrestrial transitions zones, yet it is rare to find research that simultaneously investigates aquatic and terrestrial arthropod fauna (Holmquist et al. 2011). This is due in part to the lack of method development for simultaneous sampling of aquatic and terrestrial taxa (Benke 1999).

Prior to considering the type of invertebrate sampling or the appropriate sampling effort, it is crucial to consider “what is the purpose of sampling?” When summarizing biological data, it is crucial to understand the significant questions that any sampling protocol is to address; this is especially true of post-disturbance assessments. There are three general biological measures of performance (Ciborowski et al. in prep):

- 1) **Biodiversity of Invertebrates** is the most high profile community attribute, as the concept is most easily communicated. Reclamation or restoration often emphasizes the recreation or recruitment of pre-disturbance diversity (Brown and Batzer 2001). However, this measure emphasizes rare species and requires intensive sampling of all available microhabitats within the area of question (Benke et al. 1999), as larger samples are more likely to collect rare species.
- 2) **Functional measures of biological data** (including productivity and biomass) measure biological “currency” (carbon) in amounts of biomass, for example, per unit area. These approaches emphasize common species but allow for the creation of reclamation targets, and prediction of trajectories that speak to habitat sustainability.
- 3) **Compositional measures** (Community composition, functional composition) include general biological traits of organisms (e.g. dispersal ability, size, feeding guild, reproductive habits) and indicate ecological functions that are comparable across habitats and taxa. Patterns of functional composition are often related to disturbance and can be used to compare similarity to reference conditions (Statzner et al. 2001).

As the questions within this research were framed, I considered the characteristics of wetland habitats and their resident arthropods, and decided upon three commonly used sampling techniques that would address the research questions. The goal of this chapter is to explore sampling efficiency and biases associated with three sampling types used to



collect invertebrates from boreal fens and marshes in Alberta, in hopes of accelerating research techniques for quantitative sampling of peatlands.

Research on boreal wetland invertebrates, fens especially, has mainly been focused on the biodiversity of these habitats. Though the determination of biodiversity was outside the scope of this project, it remains a “hot topic” for research, as wetlands are considered important for biodiversity. Biodiversity, however, requires a degree of taxonomic precision that is beyond the scope of much research. Relatively few species lists exist for Alberta’s wetland invertebrates. However, Wrubleski and Ross (\*\*\*\*) reviewed the available species lists of aquatic invertebrates in prairie pothole wetlands in Alberta. The list of reported species reached 401 from a reported 68 families of aquatic invertebrate, this is an average of 6 species per family. However, the number of reported species per family can be as high as 68 for chironomid midges and 70 for predaceous diving beetles (Coleoptera: Dytiscidae).

Research on fens in general was rare in the past, perhaps due to the misidentification of these unique wetlands (Danks and Rosenberg 1987). Arthropod biomass in fens is not commonly studied. However, research that seeks to elucidate patterns in food web structure use biomass as biological currency, to understand energy flow through an ecosystem. Policy makers, oil sands leasees, and reclamation scientists would find information on invertebrate biomass especially important in advising future reclamation efforts.

The purpose of this research was to determine:

- a) which method(s) are the most efficient for estimating biomass and composition of arthropods from different wetland types;
- b) which invertebrate taxa/ microhabitats are collected using each sampling type, this will also elucidate the biases associated with each sampling type, and
- c) which methods are recommended for future sampling of microhabitats of fen and marshes?

#### *Sampling Bias of Sticky Traps, Vacuum Samplers, and Sweep Nets*

Sticky traps are used to sample the abundance and activity of flying and/or emerging insects. They have been used to assess food availability for wetland ducklings (King and Wrubleski 1998), insectivorous birds (Csada et al. 1992, Whitaker et al. 2000), insectivorous bats (Kuntz 1988) and lizards (Sabo and Power 2002). Sticky traps are most appropriate for sampling smaller bodied (>6.4 mm) insects (Taylor 1962), but have the advantage of being versatile in their placement with a habitat (Doxon et al. 2010). King and Wrubleski (1998) used sticky traps to sample insects to determine food availability for ducklings at the surface of the water amongst different zones within wetland vegetation. Their sticky traps sampled a large variety of families, with Diptera being the most abundant order. However, they note that a major limitation of sticky trap sampling,

is the tendency for insects to become damaged as a result of being coated in adhesive, or during removal from trapping surface. Because of this limitation, many researchers choose to amend their biomass estimates using voucher specimens collected using aerial sweep netting (Table 2.1).

Aerial sweep netting is a very common method for sampling terrestrial invertebrates. However, it is limited in its sampling versatility. Sampling is limited by foliage density and intensity, and is biased towards larger-bodied arthropods. Furthermore, large variation in catch efficiency exists as a result of human error (Doxon et al. 2010). Aerial sweep netting is also affected by the weather and wind conditions. Sampling is best performed on a warm, low-wind day (Romney 1945). Finally, aerial sweep netting may not be appropriate for accurately estimating insect community attributes (e.g. biomass) due to overestimation of biomass due to body size bias, for example (Doxon et al. 2010; see table A.1).

Finally, I employed a novel method of vacuum sampling using a leaf blower/vac.

Vacuum sampling is not uncommon in entomological studies. However, most utilize a specially designed Dietrick vacuum (D-vac; Dietrick 1961). Previous research using the vacuum sampling has elucidated the biases of this sampling technique to be its inadequacy to capture large-bodied arthropods and its expense. My specific method was similar to that described in Hoekman et al. (2012). Hoekman and colleagues used a leaf blower/vac to sample the upland riparian area of Icelandic lakes for ground dwelling invertebrates to measure changes in food web dynamic post amendment of experimental

plots with the spent bodies of midges (Diptera: Chironomidae). Our research utilized this specific vacuum type due its usefulness for sampling at the ground level. Limitations that we made note of include the inability to perform when soil becomes inundated, an important characteristic when sampling in wetlands (pers.obs.; Table 2.1)

### *Predictions*

- 1) I predicted that vacuum sampling would be the most efficient for estimating taxa richness due to its ability to sample multiple strata of a vegetation profile. Also, unlike sticky trap sampling this active sampling technique is able to sample ground dwelling, soil, flying, and perching insects.

**Table A.1: Brief summary of the advantages and disadvantages of sticky trap, vacuum, and sweep net sampling.**

	<b>Advantages</b>	<b>Disadvantages</b>
<b>Sticky Traps</b>	-cost effective	-bias to insects >6.4 mm
	-versatility of placement within habitat	-difficult to transport trapping surface to lab
	-ease of use	-susceptible to wind/weather
	-effective at range of wind speeds	-insects susceptible to weather damage
		-insects difficult to remove from trapping surface
		-rely on insect activity, collection affected by weather
		-messy
<b>Vacuum Sampling</b>	-effective at collecting foliar and near-ground arthropods	-expensive
	-effective in dense vegetation	-cumbersome, heavy
		-not effective in inundated areas
		-requires additional processing to remove arthropods from plant material
<b>Sweep Netting</b>	-lightweight	-bias towards foliar insects
	-maneuverable	-bias towards heavier, more active arthropods
	-inexpensive	-results can be highly variable based on sampling intensity, weather, wind
		-net susceptible to damage during collection
		-ineffective in dense vegetation

### ***Methods:***

Sampling methods are as in Chapter 4.

### ***Data Analysis***

#### ***Sampling efficiency***

For the purposes of this study, sampling efficiency will be referred to as sampling effort and the resultant abundance of taxa collected. I used rarefaction to determine this relationship. Rarefaction (see Gotelli and Colwell, 2001) randomly re-samples a dataset numerous times and provides an “expected” value for each taxon in the previous dataset. The statistical program “R” (R core team 2013) was used to calculate rarefaction curves using the “vegan” community ecology package (Oksanen et al. 2013). Differences between rarefaction curves will be compared graphically. Rarefaction curves will be plotted for each sampling type for comparisons across wetlands. Rarefaction used raw abundance data for vacuum samplers and sticky traps. Due to the small number of taxa collected per sample using aerial traps, raw data was multiplied by 10 to increase the range on the x-axis on rarefaction curves.

Sampling effort was compared across 3 collection methods (vacuum sampler, sticky trap, and aerial sweep netting). Sampling effort was defined as the sum of time required for travel (set up/take down), processing time, and identification time. Travel time for sticky traps was calculated as double that of aerial sweep netting and vacuum sampling because

two “trips” are required in order to set-up and retrieve samples. Processing time was calculated as the time in minutes required to prepare one sample for identification. Aerial sweep netting does not require any sample processing as invertebrates taken from net are killed and preserved in one step. Identification time was calculated as the average time required to identify one full sample for each method. This time did not include time needed to identify taxa that were not previously encountered.

#### *How different are samplers?*

Knowing that wetlands contain many microhabitats, it is necessary to utilize more than one sampling type. Consequently, it is important to know that sampling types that are used collect different taxa from different microhabitats.

Statistical analysis was performed using STATISTICA 7.0. The relative abundance of each family within each wetland type was expressed as a percentage and transformed into Octaves ( $\log_2 + 1$ ) to reduce the dominance effects of common taxa (Gauch 1972).

Families that occurred in less than 15% of samples were excluded from further analysis. These transformed relative abundance data were further analyzed using Cluster analysis to group samples (wetlands, sampler type; e.g. Shallow Wetland- Vacuum Sample) according to taxa that are most commonly collected. Clusters were formed using Ward’s Method for linkage rules and Euclidian distances for distance measures. Clusters were plotted on a horizontal hierarchical tree plot to visualize cluster groups.

Samples belonging to each cluster were then assigned to groups; these groups were used in planned comparison analyses to determine which invertebrate taxa were representative of each sampling type.

***Results:***

A total of 124 families and 18,180 individuals were sampled from wetlands using aerial sweep nets, a vacuum sampler, and sticky traps. Sticky traps captured an average ( $\pm$  S.E.) of  $416.8 \pm 84.98$  individuals. Aerial sweeps captured  $86.4 \pm 24.27$  individuals. Vacuum Sampling captured  $372.2 \pm 95.18$  individuals. The most abundant families captured using sticky traps were Chironomidae, Ceratopogonidae, Thripidae, Aphididae, and Simuliidae. Using vacuum samplers, we captured small, soil dwelling invertebrate families, most abundance being Oribatidae, Vertinigdae, Cicadellidae, Nematoda, and Isotomidae. Aerial sweep nets captured small phytohilous invertebrates, most abundant being Cicadellidae, Prostigmatidae, Aphididae, Chloropidae, and Isotomidae. These most abundant taxa, in all sampling types, represented more than 50% of the total abundance collected with each method (see table A-3 and A-4).

In terms of family richness collected, on average, vacuum sampling was able to collect the greatest number of families per sample ( $20.61 \pm 2.56$ ), sticky traps captured  $15.6 \pm 1.46$ , aerial sweep netting captured on average,  $23.75 \pm 1.91$  families per sample.



*Addressing measures biological performance:*

*Biological* – Biodiversity was not addressed in the framing of biological questions for this project. However, I will use abundance and family richness as proxy for this measure as they are a component of diversity calculations. To illustrate the effectiveness of sampling types, the “top 5” most abundant taxa were compared across wetlands and total and cumulative abundance were reported (Table A-3). Vacuum Samples consistently collected the greatest invertebrate abundance per wetland. Common taxa collected using this method included families of soil mites, snails, and springtails. Cumulative abundance of these taxa commonly exceeded 70%, mean cumulative abundance was 73.8%.

Sticky traps collected large numbers of flying insects within each wetland. Cumulative percentage of the 5 most abundance families commonly exceeded 90%, mean cumulative abundance was 89.8%. Most abundant families consistently included Chironomidae, Ceratopogonidae, Thripidae, Simuliidae, Aphididae.

Aerial sweep nets collected the lowest abundance of invertebrates. Cumulative abundance as well as dominant taxa sampled were quite variable. Dominant taxa included flying insects, soil taxa, as well as phytophagous grass-associated invertebrates. Mean cumulative abundance was 64.10% but ranged from 46% - 91%.

*Functional Measures (Biomass)* – total and cumulative biomass were reported across wetlands for all sampling types. As opposed to abundance measures, sticky traps consistently collect the greatest amount of biomass per wetland, over aerial sweep net samples and vacuum samples. Mean cumulative percentage of biomass of top 5 most

abundant taxa per wetland collected using sticky traps was also the highest among the three sampling types (54.86%). Aerial sweep net samples and vacuum samples had similar cumulative biomass at 41.76% and 43.88%, respectively (see table A-4).

Compositional measures were not addressed within the scope of this research.

**Table A-2: Sampling effort for comparison of three sampling types based on typical travel time (trips), typical processing time (minutes per sample), and typical identification time (minutes per sample).**

	Travel	Processing	Identification
<b>Aerial Sweep Netting</b>	1	0	20
<b>Vacuum Sampling</b>	1	90	60
<b>Sticky Trap Sampling</b>	2	30	20

Table A-3: Percentage and cumulative percentage of abundance of “top 5” taxa per wetland for 3 sampling types

	Sticky Traps				Vacuum Sampler				Aerial Sweep Nets			
		Sum	%	Cumul. %		Sum	%	Cumul. %		Sum	%	Cumul. %
<b>Shallow</b>	Ceratopog.	118	19.63%	19.63%	Prostigmat.	161	25.97%	25.97%	Chironomidae	8	12.90%	12.90%
	Chironomidae	102	16.97%	33.11%	Vertingidae	73	11.77%	35.97%	Cicadellidae	6	9.68%	22.58%
	Thripidae	81	13.48%	46.59%	Vallionidae	62	10.00%	45.97%	Philodrom.	6	9.68%	32.26%
	Aphididae	66	10.98%	57.57%	Isotomidae	56	9.03%	55.00%	Coenagrion.	5	8.06%	40.32%
	Simuliidae	50	8.32%	65.89%	Euconulidae	38	6.13%	61.13%	Pseudococcid.	4	6.45%	46.77%
	<b>Total</b>											
	<b>Abundance</b>	601				620				62		
<b>Katie's Sedge Meadow</b>	Thripidae	896	56.96%	56.96%	Nematoda	505	44.03%	44.03%	Cantharidae	16	14.81%	14.81%
	Chironimidae	354	22.50%	79.47%	Oribatidae	120	10.46%	54.49%	Syrphidae	15	13.89%	28.70%
	Simuliidae	111	7.06%	86.52%	Sminthuridae	88	7.67%	62.16%	Coccinellidae	13	12.04%	40.74%
	Aphididae	65	4.13%	90.65%	Tardigrada	82	7.15%	69.31%	Chloropidae	10	9.26%	50.00%
	Ceratopog.	34	2.16%	92.82%	Cicadellidae	46	4.01%	73.32%	Cicadellidae	8	7.41%	57.41%
	<b>Total</b>											
	<b>Abundance</b>	1573				1147				108		
<b>Southwest Sands Beaver Pond</b>	Thripidae	947	57.92%	57.92%	Nematoda	2534	63.29%	63.29%	Chloropidae	9	11.54%	11.54%
	Chironomidae	264	16.15%	74.07%	Oribatidae	309	7.72%	71.00%	Coccinellidae	9	11.54%	23.08%
	Simuliidae	150	9.17%	83.24%	Chironomidae	247	6.17%	77.17%	Cicadellidae	7	8.97%	32.05%
	Ceratopog.	74	4.53%	87.77%	Prostigmat.	148	3.70%	80.87%	Sciomyzidae	7	8.97%	41.03%
	Aphidae	68	4.16%	91.93%	Isotomidae	114	2.85%	83.72%	Muscidae	5	6.41%	47.44%
	<b>Total</b>											
	<b>Abundance</b>	1635				4004				78		

		Sticky Traps			Vacuum Sampler				Aerial Sweeps			
		Sum	%	Cumul. %	Sum	%	Cumul. %		Sum	%	Cumul. %	
Ruth Lake Marsh	Chironimidae	1327	56.49%	56.49%				Sciomyzidae	4	13.79%	13.79%	
	Thripidae	512	21.80%	78.29%				Muscidae	3.5	12.07%	25.86%	
	Ceratopog.	171	7.28%	85.57%		n/a		Ulidiidae	3	10.34%	36.21%	
	Hydroptilidae	69	2.94%	88.51%				Ichneumonid.	3	10.34%	46.55%	
	Aphididae	47	2.00%	90.51%				Formicidae	2.5	8.62%	55.17%	
	Total											
	Abundance	2349							29			
Tower Road Spruce Pond	Chironomidae	1006	49.80%	49.80%				Thripidae	35	41.18%	41.18%	
	Thripidae	510	25.25%	75.05%		n/a		Chironomidae	8	9.41%	50.59%	
	Aphididae	144	7.13%	82.18%				Miridae	4	4.71%	55.29%	
	Simuliidae	127	6.29%	88.47%				Pseudococcid.	4	4.71%	60.00%	
	Ephydriidae	43	2.13%	90.59%				Cicadellidae	3	3.53%	63.53%	
	Total											
	Abundance	2020							85			
Tower Road Moose Wetland	Chironomidae	515	60.66%	60.66%	Vertingidae	69	14.81%	14.81%	Cicadellidae	28	28.28%	28.28%
	Aphididae	146	17.20%	77.86%	isotomidae	60	12.88%	27.68%	Araenidae	8	8.08%	36.36%
	Simuliidae	81	9.54%	87.40%	Nematoda	56	12.02%	39.70%	Chloropidae	7	7.07%	43.43%
	Thripidae	46	5.42%	92.82%	Cicadellidae	48	10.30%	50.00%	Cercopidae	5	5.05%	48.48%
	Ceratopog.	21	2.47%	95.29%	Prostigmat.	29	6.22%	56.22%	Aphididae	4	4.04%	52.53%
	Total											
	Abundance	849				466			99			

		Sticky Traps				Vacuum Sampler				Aerial Sweep		
		Sum	%	Cumul. %		Sum	%	Cumul. %		Sum	%	Cumul. %
<b>Tower Road 1</b>	Chironomidae	970	63.90%	63.90%	Vertingidae	63	15.29%	15.29%	Prostigmat.	18	20.45%	20.45%
	Thripidae	168	11.07%	74.97%	Isotomidae	48	11.65%	26.94%	Chloropidae	17	19.32%	39.77%
	Simuliidae	111	7.31%	82.28%	Sminthuridae	43	10.44%	37.38%	Cicadellidae	11	12.50%	52.27%
	Aphididae	108	7.11%	89.39%	Nematoda	42	10.19%	47.57%	Sminthuridae	8	9.09%	61.36%
	Chloropidae	37	2.44%	91.83%	Cicadellidae	41	9.95%	57.52%	Simuliidae	8	9.09%	70.45%
	<b>Total</b>											
	<b>Abundance</b>	1518				412				88		
<b>Rhyno's Watering Hole</b>	Chironomidae	193	41.68%	41.68%	Cicadellidae	141	48.12%	48.12%	Coccinellidae	14	24.14%	24.14%
	Thripidae	94	20.30%	61.99%	Vertingidae	30	10.24%	58.36%	Chironomidae	8	13.79%	37.93%
	Certatopog.	61	13.17%	75.16%	Prostigmat.	15	5.12%	63.48%	Muscidae	5	8.62%	46.55%
	Aphididae	27	5.83%	80.99%	Sminthuridae	11	3.75%	67.24%	Syrphidae	5	8.62%	55.17%
	Simuliidae	18	3.89%	84.88%	Thomosidae	11	3.75%	70.99%	Cicadellidae	4	6.90%	62.07%
	<b>Total</b>											
	<b>Abundance</b>	463				293				58		
<b>Sam's Rodeo Marsh</b>	Chironomidae	282	38.52%	38.52%	Oribatidae	1981	78.42%	78.42%	Sminthuridae	148	44.44%	44.44%
	Thripidae	230	31.42%	69.95%	Nematoda	339	13.42%	91.84%	Aphididae	51	15.32%	59.76%
	Simuliidae	81	11.07%	81.01%	Prostigmat.	44	1.74%	93.59%	Thripidae	24	7.21%	66.97%
	Sminthuridae	25	3.42%	84.43%	Sminthuridae	31	1.23%	94.81%	Chironomidae	21	6.31%	73.27%
	Chloropidae	17	2.32%	86.75%	Isotomidae	26	1.03%	95.84%	Cicadellidae	15	4.50%	77.78%
	<b>Total</b>											
	<b>Abundance</b>	732				2526				333		

		Sticky Traps				Vacuum Sampler				Aerial Sweeps		
		Sum	%	Cumul. %		Sum	%	Cumul. %		Sum	%	Cumul. %
<b>Maqua Lake Fen</b>	Hydroptilidae	587	31.54%	31.54%	Oribatidae	146	44.65%	44.65%	Oribatidae	218	73.90%	73.90%
	Chironomidae	387	20.80%	52.34%	Prostigmat.	38	11.62%	56.27%	Isotomidae	20	6.78%	80.68%
	Simuliidae	299	16.07%	68.40%	Chironomidae	20	6.12%	62.39%	Sminthuridae	16	5.42%	86.10%
	Thripidae	249	13.38%	81.78%	Nematoda	20	6.12%	68.50%	Prostigmat.	7	2.37%	88.47%
	Ceratopog.	174	9.35%	91.13%	Mesostigmat.	16	4.89%	73.39%	Hypogastur.	6	2.03%	90.51%
	<b>Total Abundance</b>	1861				327				295		
<b>Gravel Pit Fen</b>	Simuliidae	851	50.38%	50.38%	Nematoda	105	17.68%	17.68%	Cicadellidae	7.5	23.44%	23.44%
	Thripidae	489	28.95%	79.34%	Oribatidae	94	15.82%	33.50%	Sciomyzidae	3.5	10.94%	34.38%
	Ceratopogon.	149	8.82%	88.16%	Hypogasturid.	82	13.80%	47.31%	Muscidae	2.5	7.81%	42.19%
	Chironomidae	99	5.86%	94.02%	Isotomidae	58	9.76%	57.07%	Coccinellidae	2.5	7.81%	50.00%
	Hydroptilidae	13	0.77%	94.79%	Prostigmat.	58	9.76%	66.84%	Chironomidae	1.5	4.69%	54.69%
	<b>Total Abundance</b>	1689				594				32		
<b>Pauciflora Fen</b>	Simuliidae	6042	97.66%	97.66%	Oribatidae	1118	67.88%	67.88%	Cicadellidae	3	27.27%	27.27%
	Chironimidae	57	0.92%	98.58%	Nematoda	293	17.79%	85.67%	Muscidae	2	18.18%	45.45%
	Aphididae	45	0.73%	99.30%	Mesostigmat.	53	3.22%	88.89%	Ichneumonid.	2	18.18%	63.64%
	Ceratopog.	27	0.44%	99.74%	Sminthuridae	29	1.76%	90.65%	Philodromid.	2	18.18%	81.82%
	Thripidae	9	0.15%	99.89%	Isotomidae	26	1.58%	92.23%	Mymaridae	1	9.09%	90.91%
	<b>Total Abundance</b>	6187				1647				11		

		Sticky Trap				Vacuum Sampler		
		Sum	%	Cumul. %		Sum	%	Cumul. %
<b>Beaver Lodge Wetland</b>	Chironomidae	151	44.94%	44.94%	Oribatidae	117	45.00%	45.00%
	Ephydriidae	57	16.96%	61.90%	Vertingidae	46	17.69%	62.69%
	Hydroptilidae	43	12.80%	74.70%	Coccidae	21	8.08%	70.77%
	Ceratopog.	32	9.52%	84.23%	Euconulidae	13	5.00%	75.77%
	Simuliidae	25	7.44%	91.67%	Succineidae	10	3.85%	79.62%
	<b>Total</b>							
	<b>Abundance</b>	336				260		n/a



**Table A-4: Percentage and cumulative percentage of biomass of “top 5 most abundant families within each wetland collected with 3 sampling types.**

		Sticky Traps				Vacuum Samples				Aerial Sweep Nets		
		Sum	%	Cumul. %		Sum	%	Cumul. %		Sum	%	Cumul. %
<b>Shallow Wetland</b>	Ceratopog.	7.906	4.47%	4.47%	Prostigmat.	1.07	1.74%	1.74%	Chironomid.	7.91	4.47%	4.47%
	Chironomid.	14.17	8.02%	12.49%	Vertingidae	13.80	22.37%	24.11%	Cicadellidae	14.18	8.02%	12.49%
	Thripidae	2.754	1.56%	14.05%	Vallionidae	11.72	19.00%	43.11%	Philodrom.	0.40	0.23%	12.72%
	Aphididae	12.40	7.02%	21.07%	Isotomidae	0.73	1.18%	44.29%	Coenagrion.	0.14	0.08%	12.79%
	Simuliidae	5.15	2.91%	23.98%	Euconulidae	7.18	11.64%	55.93%	Pseudococc.	5.15	2.91%	15.71%
	<b>Total</b>											
	<b>Biomass</b>	176.8				61.68				176.79		
<b>Katie's Sedge Meadow</b>	Thripidae	30.46	9.76%	9.76%	Nematoda	0.87	1.64%	1.64%	Cantharidae	0.03	0.01%	0.01%
	Chironimid.	49.21	15.77%	25.53%	Oribatidae	0.80	1.50%	3.14%	Syrphidae	0.11	0.04%	0.05%
	Simuliidae	11.43	3.66%	29.20%	Sminthur.	4.31	8.10%	11.24%	Coccinellid.	0.44	0.14%	0.19%
	Aphididae	12.22	3.92%	33.11%	Tardigrada	0.14	0.27%	11.51%	Chloropidae	0.03	0.01%	0.20%
	Ceratopog.	2.278	0.73%	33.84%	Cicadellidae	8.69	16.33%	27.84%	Cicadellidae	0.58	0.19%	0.38%
	<b>Total</b>											
	<b>Biomass</b>	312.1				53.23				312.04		
<b>Southwest Sands Beaver Pond</b>	Thripidae	32.19	12.23%	12.23%	Nematoda	4.38	4.64%	4.64%	Chloropidae	0.15	0.06%	0.06%
	Chironomid.	36.69	13.94%	26.16%	Oribatidae	2.06	2.18%	6.82%	Coccinellid.	0.29	0.11%	0.17%
	Simuliidae	15.45	5.87%	32.03%	Chironomid	46.68	49.44%	56.26%	Cicadellidae	4.81	1.83%	1.99%
	Ceratopog.	4.958	1.88%	33.91%	Prostigmat.	0.99	1.04%	57.31%	Sciomyzidae	0.07	0.03%	2.02%
	Aphidae	12.78	4.86%	38.77%	Isotomidae	1.48	1.57%	58.88%	Muscidae	0.28	0.11%	2.12%
	<b>Total</b>											
	<b>Biomass</b>	263.3				94.42				263.31		

		Sum	%	Cum. %			Sum	%	Cum. %			Sum	%	Cum. %
<b>Ruth Lake Marsh</b>	Chironimid.	184.5	29.83%	29.83%			n/a			Sciomyzidae		26.83	16.34%	16.34%
	Thripidae	30.46	4.93%	34.76%						Muscidae		3.20	1.95%	18.29%
	Ceratopog.	171.1	27.67%	62.43%						Ulidiidae		4.09	2.49%	20.78%
	Hydroptilid.	151.8	24.55%	86.98%						Ichneumon.		5.08	3.09%	23.87%
	Aphididae	8.836	1.43%	88.41%						Formicidae		1.85	1.13%	25.00%
	<b>Total</b>													
	<b>Biomass</b>	618.3										164.13		
<b>Tower Road Spruce Pond</b>	Chironomid.	139.8	20.18%	20.18%			n/a			Thripidae		87.65	30.44%	30.44%
	Thripidae	17.34	2.50%	22.68%						Chironomid.		16.63	5.77%	36.21%
	Aphididae	27.07	3.91%	26.59%						Miridae		9.98	3.47%	39.68%
	Simuliidae	13.08	1.89%	28.48%						Pseudococc.		13.76	4.78%	44.46%
	Ephydriidae	122.7	17.71%	46.19%						Cicadellidae		28.60	9.93%	54.39%
	<b>Total</b>													
	<b>Biomass</b>	692.9										287.96		
<b>Tower Road Moose Wetland</b>	Chironomid.	71.59	44.43%	44.43%	Vertingidae	13.04	26.12%	26.12%	Chironomid.	71.59	44.43%	44.43%		
	Aphididae	27.45	17.03%	61.46%	isotomidae	0.78	1.56%	27.68%	Aphididae	27.45	17.03%	61.46%		
	Simuliidae	8.343	5.18%	66.64%	Nematoda	0.097	0.19%	27.87%	Simuliidae	8.34	5.18%	66.64%		
	Thripidae	1.564	0.97%	67.61%	Cicadellidae	9.072	18.17%	46.04%	Thripidae	1.56	0.97%	67.61%		
	Ceratopog.	1.407	0.87%	68.48%	Prostigmat.	0.191	0.38%	46.43%	Ceratopog.	1.41	0.87%	68.48%		
	<b>Total</b>													
	<b>Biomass</b>	161.1					49.93					161.13		

		Sum	%	Cum. %		Sum	%	Cum. %		Sum	%	Cum. %
<b>Tower Road 1</b>	Chironomid.	134.8	29.68%	29.68%	Vertingidae	11.91	24.42%	24.42%	Prostigmat.	134.83	29.68%	29.68%
	Thripidae	5.712	1.26%	30.94%	Isotomidae	0.624	1.28%	25.70%	Chloropidae	5.71	1.26%	30.94%
	Simuliidae	11.43	2.52%	33.45%	Sminthur.	2.107	4.32%	30.02%	Cicadellidae	11.43	2.52%	33.45%
	Aphididae	20.30	4.47%	37.92%	Nematoda	0.073	0.15%	30.17%	Sminthur.	20.30	4.47%	37.92%
	Chloropidae	53.28	11.73%	49.65%	Cicadellidae	7.749	15.89%	46.06%	Simuliidae	53.28	11.73%	49.65%
	<b>Total</b>											
	<b>Biomass</b>	454.3				48.76				454.28		
<b>Rhyno's Watering Hole</b>	Chironomid.	26.83	16.34%	16.34%	Cicadellidae	26.65	52.58%	52.58%	Coccinellid.	87.65	30.44%	30.44%
	Thripidae	3.196	1.95%	18.29%	Vertingidae	5.67	11.19%	63.77%	Chironomid	16.63	5.77%	36.21%
	Certatopog.	4.087	2.49%	20.78%	Prostigmat.	0.099	0.20%	63.97%	Muscidae	9.98	3.47%	39.68%
	Aphididae	5.076	3.09%	23.87%	Sminthur.	0.539	1.06%	65.03%	Syrphidae	13.76	4.78%	44.46%
	Simuliidae	1.854	1.13%	25.00%	Thomosidae	2.079	4.10%	69.13%	Cicadellidae	28.60	9.93%	54.39%
	<b>Total</b>											
	<b>Biomass</b>	164.1				50.68				287.96		
<b>Sam's Rodeo Marsh</b>	Chironomid	39.19	18.34%	18.34%	Oribatidae	13.19	42.75%	42.75%	Sminthur.	39.20	18.34%	18.34%
	Thripidae	7.82	3.66%	22.00%	Nematoda	0.586	1.90%	44.65%	Aphididae	7.82	3.66%	22.00%
	Simuliidae	8.343	3.90%	25.91%	Prostigmat.	0.293	0.95%	45.60%	Thripidae	8.34	3.90%	25.91%
	Sminthur.				Sminthur.	1.519	4.92%	50.52%	Chironomid	0.21	0.10%	26.00%
	Chloropidae	24.48	11.46%		Isotomidae	0.338	1.10%	51.62%	Cicadellidae	24.48	11.46%	37.46%
	<b>Total</b>											
	<b>Biomass</b>	213.7				30.86				213.70		

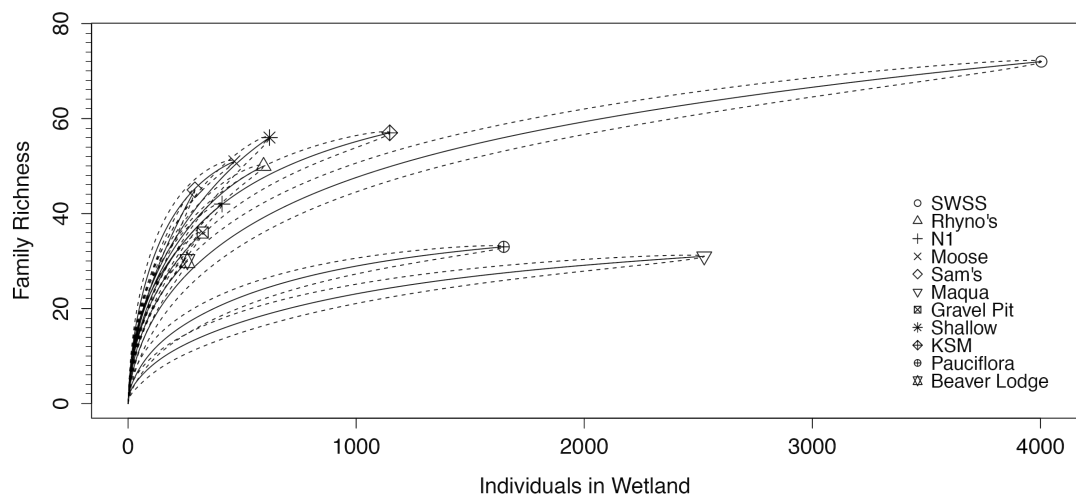
		Sum	%	Cum. %			Sum	%	Cum. %			Sum	%	Cum. %
<b>Maqua Lake Fen</b>		1291.4					1291.4					1291.4		
	Hydroptilid.	1291	78.37%	78.37%	Oribatidae	0.972	6.14%	6.14%	Oribatidae	0	78.37%	78.37%		
	Chironomid.	53.79	3.26%	81.64%	Prostigmat.	0.006	0.04%	6.18%	Isotomidae	53.79	3.26%	81.64%		
	Simuliidae	30.79	1.87%	83.51%	Chironomid.	0.189	1.19%	7.37%	Sminthur.	30.80	1.87%	83.51%		
	Thripidae	8.466	0.51%	84.02%	Nematoda	0.035	0.22%	7.59%	Prostigmat.	8.47	0.51%	84.02%		
	Ceratopog.	11.66	0.71%	84.73%	Mesostigma	0.107	0.67%	8.27%	Hypogastur.	11.66	0.71%	84.73%		
	<b>Total Biomass</b>	1647				15.84				1647.8				
<b>Gravel Pit</b>	Simuliidae	87.65	30.44%	30.44%	Nematoda	0.181	0.41%	0.41%	Cicadellidae	87.65	30.44%	30.44%		
	Thripidae	16.62	5.77%	36.21%	Oribatidae	0.626	1.42%	1.83%	Sciomyzidae	16.63	5.77%	36.21%		
	Ceratopog.	9.983	3.47%	39.68%	Hypogastur.	1.066	2.41%	4.24%	Muscidae	9.98	3.47%	39.68%		
	Chironomid	13.76	4.78%	44.46%	Isotomidae	0.754	1.70%	5.94%	Coccinellid.	13.76	4.78%	44.46%		
	Hydroptilid.	28.6	9.93%	54.39%	Prostigmat.	0.386	0.87%	6.81%	Chironomid.	28.60	9.93%	54.39%		
	<b>Total Biomass</b>	287.9				44.24				287.96				
<b>Pauciflora Fen</b>	Simuliidae	622.3	76.56%	76.56%	Oribatidae	7.446	32.53%	32.53%	Cicadellidae	87.65	30.44%	30.44%		
	Chironimid.	7.923	0.97%	77.54%	Nematoda	0.507	2.21%	34.74%	Muscidae	16.63	5.77%	36.21%		
	Aphididae	8.46	1.04%	78.58%	Mesostigma	0.353	1.54%	36.29%	Ichneumon.	9.98	3.47%	39.68%		
	Ceratopo.	1.809	0.22%	78.80%	Sminthur.	1.421	6.21%	42.49%	Philodrom.	13.76	4.78%	44.46%		
	Thripidae	0.306	0.04%	78.84%	Isotomidae	0.338	1.48%	43.97%	Mymaridae	28.60	9.93%	54.39%		
	<b>Total Biomass</b>	812.9				22.89				287.96				

		Beaver Lodge Wetland			Cum. %				
		Sum	%	Cum. %	Sum	%	Cum. %		
Beaver Lodge Wetland	Chironomid.	20.99	7.05%	7.05%	Oribatidae	0.779	3.28%	3.28%	n/a
	Ephydriidae	162.6	54.60%	61.64%	Vertingidae	8.694	36.59%	39.87%	
	Hydroptilid.	94.6	31.76%	93.41%	Coccidae	3.969	16.70%	56.58%	
	Ceratopog.	2.144	0.72%	94.13%	Euconulidae	2.457	10.34%	66.92%	
	Simuliidae	2.575	0.86%	94.99%	Succineidae	0.189	0.80%	67.71%	
	<b>Total</b>								
	<b>Biomass</b>	297.9				23.76			

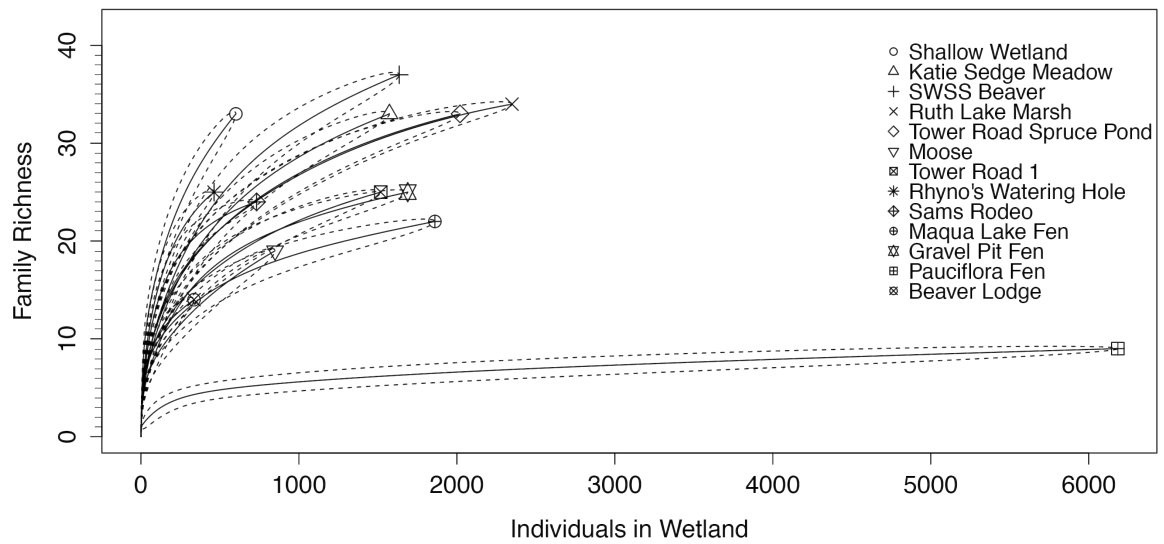
### *Sample Efficiency*

Previously, I determined that marsh wet meadow zones contained significantly more families than fens. To confirm that this was not an artifact of sample size, rarefaction curves were used to interpolate family richness captured at smallest sample size collected using each sampling method. Interpolated sample sizes were analyzed using ANOVA and results indicate that marsh wet meadow family richness is significantly higher than fen family richness when sampled using vacuum sampling ( $p=0.01$ ) and sticky traps ( $p=0.02$ ). Similar to non-rarified results, there was no significant difference in family richness in wet meadows and fens sampled using aerial sweep nets ( $p=0.57$ ).

Rarefaction curves indicated that vacuum samples estimate family richness more adequately than sticky traps. The shape of vacuum sample curves indicates that sampling was adequate in all but 3 wetlands, where the curve does not plateau (Rhyno's, Maqua Lake Fen, Shallow Wetland, see figure A-1). The number of families sampled from wetlands using sticky traps increased with additional individuals sampled in all wetlands except Pauciflora fen where family richness was small, but abundance was composed almost entirely of black flies (Diptera: Simuliidae; Figure A-2). Rarefaction curves generated for aerial sweep nets indicated that sampling was adequate to collect family richness present; all curves plateau indicating that sampling was sufficient (Figure A-3).

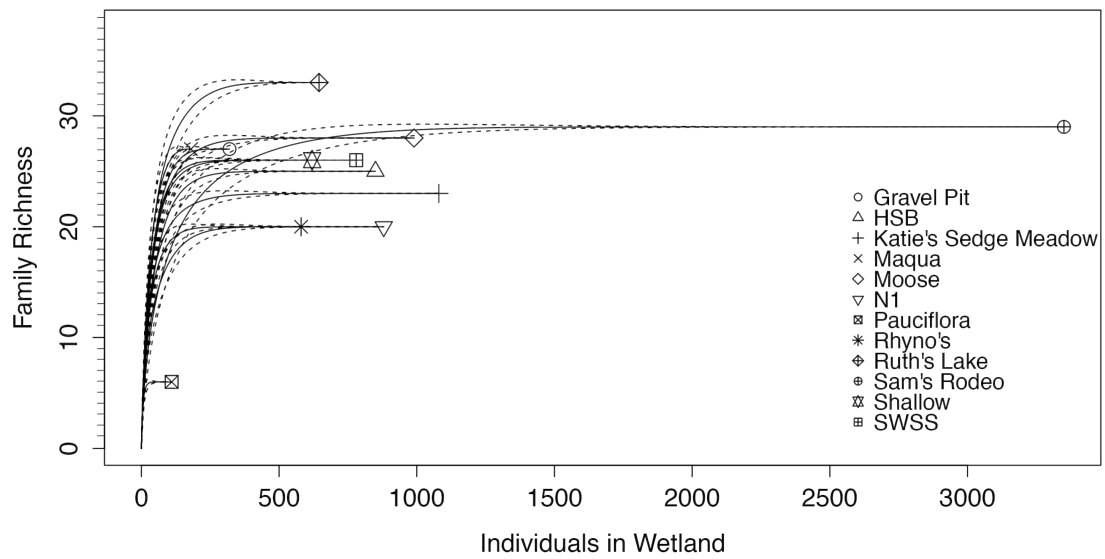


**Figure A-1: Rarefaction curves for vacuum sampling in 11 wetlands.**



**Figure A-2: Rarefaction curves for sticky trap samples in 13 wetlands.**





**Figure A-3: Rarefaction curves for aerial sweep netting in 12 wetlands.**

### *Sampling Effort*

Sampling types did not differ much in terms of taxa occurrence. However there was a large difference in terms of sampling effort (Table A-2). Vacuum sampling took, on average, triple the amount of time to prepare a sample for identification due to sieve stack methods that were used (see Chapter 4, Methods). Identification of insects from sticky traps and aerial samples required on average the same amount of time (60-min), which was roughly half the time required to identify all the invertebrates in vacuum samples. Therefore, for a similar number of taxa per sample, vacuum sampling takes roughly triple the effort. The primary cause of greater time required for identification was due to the large number of small, bodied invertebrates caught within the filtrate vegetation. Although vacuum sampling required the greatest amount of sampling effort, sticky trap samples require the greatest amount of travel/ field time due to retrieval.

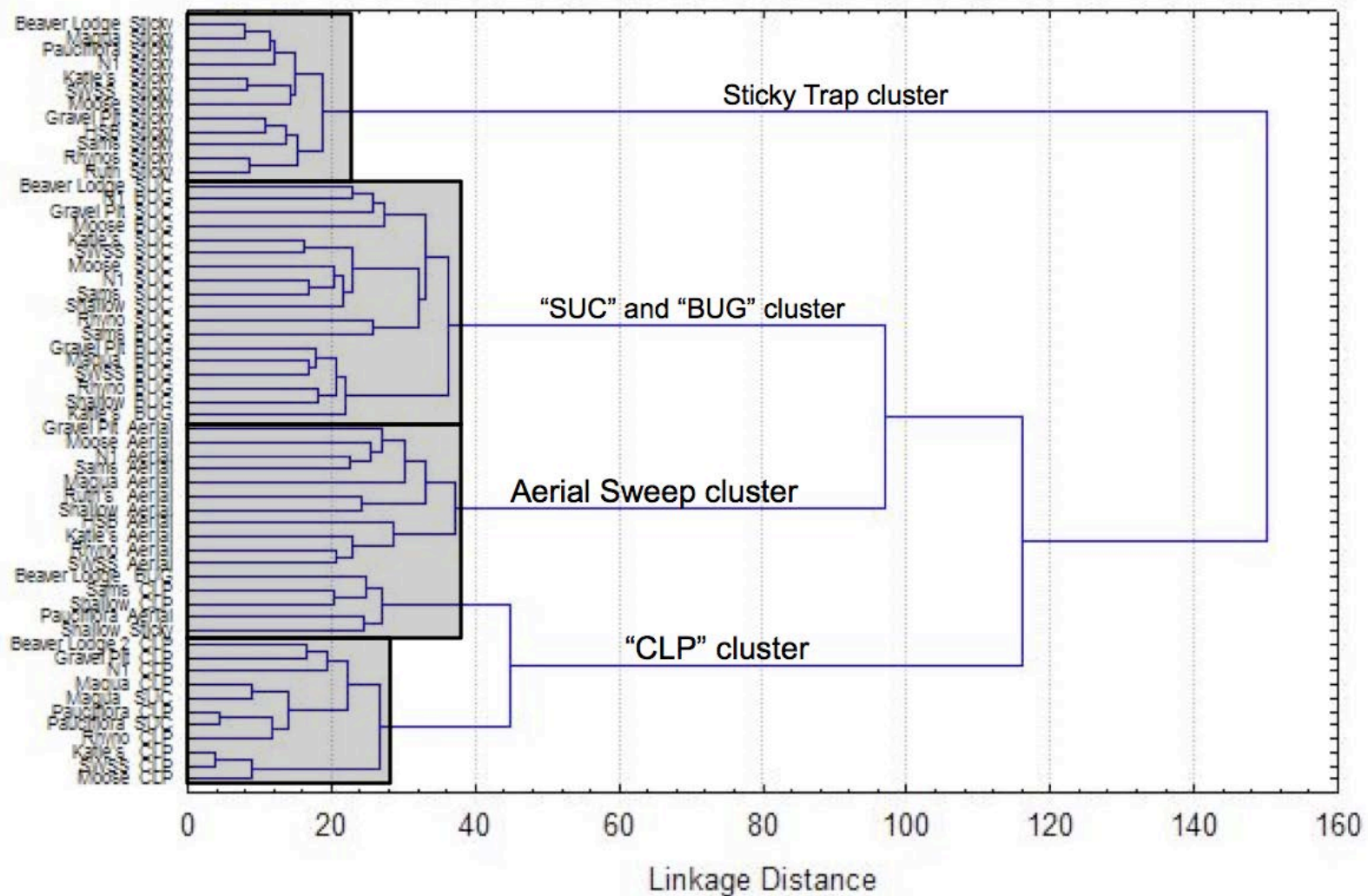
### *Cluster Analysis*

Cluster analysis of the samples taken in wetlands classified samples into 4 distinct groups based on dominant taxa collected using each sampling method. Vacuum samples were divided into 3 strata (as per methods in Chapter 3), based on the microhabitat that each stratum of vacuum samples sampled (“BUG”, “SUC”, and “CLP”). “BUG” being the materials collected from the soil’s surface after vegetation had been clipped, clustered with “SUC” which was the material initially collected from the vegetation within the box plot. “CLP”, representing the invertebrates associated with the vegetation that was clipped from the box plot, clustered separately from other

vacuum sample strata. Aerial sweep net samples clustered together, as did sticky trap samples (Figure A-4). Clusters are named according to the sampler type that dominated the cluster. Generally, traps clustered extremely clearly.

#### *Planned Comparison of Clusters*

Samples in clusters were assigned a group name and used in planned comparison analyses. Results of planned comparison of clusters are summarized in table A-4. Taxa that are associated with each sampling type are listed within the cluster with which they clustered. Samples assigned to the sticky trap clusters were composed mainly of flying, aquatic (wetland) insects. However, there were a few families of phytophilous insects (Aphididae, Tingidae), one spider (Araneidae), and a hymenopteran family (Tiphidiidae). The “CLP” cluster was very small and was associated with a spider family (Anapidae), and nematodes. The aerial sweep cluster contained 22 families of invertebrates including, phytophilous insects (e.g. Alydidae, Aphididae, Cercopidae, etc.), emergent adults of aquatic species (Chironomidae, Coenagrionidae, Hydroptilidae, etc.), predatory invertebrates (e.g. Salticidae, Coccinellidae), and two families of forest-dwelling beetles (Cerambycidae and Buprestidae). The “SUC” and “BUG” cluster contained 25 families of phytophilous and soil dwelling invertebrate.



**Figure A-4: Cluster Analysis of invertebrate samples taken from 11 marshes and 4 fens using 3 sampling methods. Clusters based on taxa associated with sampling type. Clustered using Ward's method. Linkage distances are Euclidean distances.**

**Table A-5: Results of planned comparison of clusters: taxa associated with 4 clusters resulting from trap type. Relative abundance of a taxon listed under a trap type is significantly greater than its mean relative abundance in the other three trap types.**

	Sticky Trap Cluster	“CLP” Cluster	Aerial Sweep Cluster	“SUC” and “BUG” cluster
<b>Oligochaeta</b>				Lumbricidae
<b>Gastropoda</b>				Euconulidae Succineidae Vallionidae Vertingidae
<b>Nematoda</b>	Nematoda	Nematoda		Nematoda
<b>Acari</b>				Mesostigmatidae Oribatidae Prostigmatidae
<b>Aranea</b>	Araneidae	Anapidae	Salticidae	Araneidae Corrinidae Lycosidae Philodromidae

	Sticky Trap Cluster	“CLP” Cluster	Aerial Sweep Cluster	“SUC” and “BUG” cluster
<b>Collembola</b>			Sminthuridae	Entomobryidae Isotomidae Hypogasturidae
<b>Ephemeroptera</b>	Baetidae			
<b>Odonata</b>			Coenagrionidae Lestidae	
<b>Trichoptera</b>	Hydroptilidae		Hydroptilidae	
<b>Thysanoptera</b>	Thripidae		Aphididae	
<b>Hemiptera</b>	Aphididae Tingidae		Alydidae Cercopidae Miridae	Delphadidae Reduviidae
<b>Coleoptera</b>			Buprestidae Cerambycidae Chrysomelidae Coccinellidae	Cantharidae Carabidae Hydrophilidae Staphylinidae

	Sticky Trap Cluster	“CLP” Cluster	Aerial Sweep Cluster	“SUC” and “BUG” cluster
<b>Diptera</b>	Ceratopogonidae		Chironomidae	Bibionidae
	Chironomidae		Chloropidae	Diptera pupa
	Simuliidae		Dolichopodidae	
	Tabanidae		Tipulidae	
	Ephydriidae		Muscidae	
	Phoridae		Sciomyzidae	
			Utilidae	
<b>Hymenoptera</b>	Tiphidiidae		Syrphidae	
			Tenthredinidae	

### ***Discussion:***

When addressing “What is the purpose of sampling?” prior to designing a sampling regime, the goal is, generally, to evaluate one or more of the three measures of biological performance. When addressing biodiversity, taxonomic resolution is the first consideration. Studies seeking to evaluate a wetland’s species pool must utilize a sampling procedure that can sample non-destructively, and must address the multiple habitats within a wetland. Though identification to the species level was beyond the scope of this research, a comparison of aquatic families from Wrubleski and Ross (2011), to terrestrially captured invertebrates from this research revealed an average family: species ratio of 1:12 species per family.

We have previously illustrated how effective these trapping methods are for evaluating functional measures (e.g. biomass). Tables A-2 and A-3 illustrate that each sampling type captured unique “dominant taxa” and that these taxa tend to make up a large proportion of total biomass.

### ***Method Comparisons***

The results of this methods comparison indicate that vacuum sampling and sticky trap sampling each produces a higher estimate of taxonomic richness than aerial sweep netting in the natural fens and marshes that were sampled for this research. Rarefaction analysis indicates that vacuum sampling and sticky trapping more adequately estimated family richness in natural fens and marshes than aerial sweep netting. Family richness was also significantly higher in marsh wet meadows than fens, and this relationship



remained significant when data was rarefied. Rarefaction curves created using sticky trap data were steeper than those created using vacuum sampling data, indicating that when using the vacuum sampler, few additional taxa would result from more intensive sampling. In contrast, sticky trap rarefaction indicates more families could potentially be collected with more intensive sampling.

Family richness collected using aerial sweep net sampling did not differ significantly between fens and marshes using raw or rarefied data. Furthermore, rarefaction of aerial sweep netting data produced curves that plateaued at the maximum family richness reported per wetland, indicating that the low abundance per family did not allow for accurate rarefaction estimates. The wetland that produced the most appropriately shaped curve was that generated from the data from “Sams Rodeo” wetland. Invertebrate abundance in Sam’s Rodeo wetland was 335 individuals. These results indicate that aerial sampling is an inappropriate sampling technique for estimating invertebrate community attributes in wetland vegetation. As previously discussed, prior research regarding the biases of this sampling technique and these findings are consistent with our own. Aerial sweep netting is inappropriate for estimating invertebrate community attributes associated within vegetation as the lightweight net is unable to penetrate high density foliage (Doxon et al. 2010). Foliage reported from Sam’s rodeo wetland is less dense in the wet meadow zone, being mainly comprised of sparse *Carex* patches, and horsetail (*Equisitum fluviatile*), which allowed for more effective sweeps (pers. obs.).

### *Sampling Effort*

Total family richness was estimated to be very similar among all 3 sampling types. However, processing and identification time are important considerations in any sampling regime. Vacuum sampling takes a great deal of time to process due to the nature of the microhabitat that they are suited to sample. The dense, wet meadow or fen vegetation presents challenges when extricating invertebrates for enumeration and identification. Nevertheless, this method collected a large number of taxa that were not caught by the other samplers.

### *Cluster Analysis*

Cluster analysis of samples based on invertebrate taxa revealed 4 distinct clusters, each containing a particular sampling type. This indicates that the taxa captured by each sampling type are very different. This further demonstrates that utilizing multiple sampling techniques in wetland microhabitats will increase estimates of wetland taxa richness. Planned comparison of clusters further explains the taxonomic basis for the individual groups. For example, the sticky trap cluster of samples was composed mainly of emergent aquatic insects as well as some small-bodied vegetation-associated insects.

### ***Conclusions:***

The goal of any sampling regime is accurate, representative data that addresses a sampling goal, likely to estimate one or more of the previously mentioned measures of biological performance. In wetlands, the numerous microhabitats require more than one sampling technique. Because wetlands are composed of both aquatic and terrestrial ecotones, sampling should represent and seek to sample invertebrates from these habitats. Truly aquatic microhabitats can be sampled using standard, well-established techniques. Vacuum sampling is an excellent sampling technique to capture representative invertebrate richness and biomass from the terrestrial ecotone. It efficiently samples a diverse community of invertebrates from the vegetation and from the soil-vegetation interface. although processing is time consuming, the process can be expedited. I collected vacuum samples in 3 strata (vegetation sampling, vegetation clipping, and soil suction; see Chapter 3 methods). However, only the “vegetation sampling” and the “soil fauna” strata contained abundant amounts of material and unique taxa. Invertebrates in the “CLP”(vegetation clippings) cluster were primarily Nematoda and the spider family Anapidae. As a result, I recommend not processing this stratum.

Vacuum sampling requires a complementary sampler for the multiple habitats within a wetland. I recommend sticky trapping to collect flying insects from the aquatic ecotone. Sticky traps are easily set up, and insects are quickly processed for identification. Sticky traps provide a good estimate of family richness of aquatic insects, but are biased towards smaller-bodied insects. I do not recommend using sticky traps for biodiversity

studies as the nature of the adhesive plant resin results in damage to diagnostic features. Furthermore, to reduce damage, we recommend placing traps for 24 h over 3 consecutive days rather than leaving traps in place for continuous days of sample collection.

Aerial sweep netting is not recommended for research seeking to address functional measures of biological performance, as in this research. Aerial sweep netting may be appropriate for biodiversity studies. Abundance measures estimated using this technique were extremely variable. This sampling type, above the others, is extremely affected by phenology, foliage density, and human sampling bias. Sampling protocols that call for aerial sweep netting must minimize this error, or only use aerial sweep netting as supplementary to less biased protocols.

Other considerations for sampling invertebrates in wetlands of the Athabasca oil sands region include estimations of biological performance in riparian habitats. The Athabasca River drainage basin includes over 130,000 km<sup>2</sup> of boreal Alberta. Riparian and floodplain wetlands are likely an important habitat for regional biodiversity.

Phenology and the spatial arrangement of samples should also be considered in designing a sampling program. Sampling for this study was limited in terms of phenology by time constraints. However, previous research in wetlands has been able to utilize knowledge of local phenology to sample during emergence events or “peak production” to generate a more accurate estimate of biomass as well determining the identity of dominant taxa. Furthermore, researchers could sample multiple times throughout “peak season” to capture invertebrates that emerge at different times of the year, or that have multiple cohorts.. For example, “Early”, “Mid-season” and “Late

season” sampling could be used to evaluate taxa that overwinter as larvae, taxa associated with peak plant growth and flowering, and slow growing/taxa that are associated with peak plant biomass, respectively. Spatial arrangement is an especially important consideration in wetlands where the habitat is heterogeneous. Future research should carefully sample the various microhabitats using multiple sampling techniques. In fens, habitats may appear to be homogeneous, but many fens contain hummock and hollow or string and flark patterns of peat ridges and hollow spaces (which may result from slow-moving surface water). This spatial heterogeneity can create very local concentrations of invertebrates within fens. Consequently, trap placement may be even more important in these patterned fens than in low-relief wetlands.

Analysis of the invertebrate inhabitants of wooded fens was beyond the scope of this project. However, such fen forms are quite dominant in the Athabasca oil sands region, and their assessment and comparison with other fen types is a significant research need

**APPENDIX 3: SUMMARY OF INVERTEBRATE AND ENVIRONMENTAL SAMPLING DATA****A3-1: U SHAPED CELL SAMPLING DATA AND WATER CHEMISTRY**

Cell	Peat Type	Vegetation Type	Peat Depth (cm)	Water Type	Salinity (ppt)	pH	Conductivity (uS)	Number of Traps Recovered
2	Live	Fen	50	OSPW				4
5	Live	Fen	100	OSPW				4
6	Live	Fen	15	Fresh	0.1	7.6	301.8	4
11	Live	Fen	15	Fresh	0.2	7.7	3844	4
12	Live	Fen	100	OSPW				4
15	Live	Fen	15	OSPW				4
18	Live	Fen	50	OSPW	0.8	7.1	1641	4
19	Live	Fen	100	Fresh	0.2	6.9	334.1	4
22	Live	Fen	50	Fresh	0.2	6.7	386.4	4
24	Live	Fen	15	OSPW				4
26	Live	Fen	50	Fresh	0.2	7	352.9	4
27	Live	Fen	100	Fresh				4
1	Stockpiled	Marsh	100	OSPW				4
3	Stockpiled	Marsh	100	OSPW				4
4	Stockpiled	Marsh	100	Fresh	0.1	7.3	354.1	4
7	Stockpiled	Marsh	15	OSPW				4
8	Stockpiled	Marsh	50	OSPW	1.9	7.3	3580	4
9	Stockpiled	Marsh	50	OPSW	0.3	7.2	571	3
10	Stockpiled	Marsh	100	OSPW	1.7	7	3206	4
13	Stockpiled	Marsh	100	Fresh				4
14	Stockpiled	Marsh	50	OSPW	1.8	7	3370	4

# APPENDICES

Cell	Peat Type	Vegetation Type	Peat Depth (cm)	Water Type	Salinity (ppt)	pH	Conductivity (uS)	Number of Traps Recovered
17	Stockpiled	Marsh	50	Fresh	0.2	7.1	489.1	3
20	Stockpiled	Marsh	15	OSPW	2.2	8	4484	3
21	Stockpiled	Marsh	15	Fresh	0.2	7	364.1	4
25	Stockpiled	Marsh	15	Fresh				4
28	Stockpiled	Marsh	15	OSPW				4
16	Mineral Soil	CONTROL	20	n/a				4
23	Mineral Soil	CONTROL	20	n/a	0.2	6.9	1466	4

\*only cells that were flooded at time of collection had sufficient water for water quality testing (Conductivity, pH, etc)

<b>A3-2: U-CELL BIOMASS SUMMARY</b>			
<b>Cell</b>	<b>Cell Type</b>	<b>Average biomass/trap (mgDW)</b>	<b>Total Biomass/cell (mgDW)</b>
1	Marsh	19.21	76.86
2	Fen	13.50	54.01
3	Marsh	15.61	62.44
4	Marsh	23.49	93.94
5	Fen	29.57	118.26
6	Fen	16.87	67.47
7	Marsh	11.04	44.15
8	Marsh	7.77	31.10
9	Marsh	4.07	12.20
10	Marsh	14.60	58.42
11	Fen	22.29	89.16
12	Fen	15.95	63.82
13	Marsh	12.22	48.87
14	Marsh	12.51	50.06
15	Fen	7.66	30.64
17	Marsh	3.47	10.40
18	Fen	15.67	62.68
19	Fen	7.99	31.97
20	Marsh	1.56	4.68
21	Marsh	17.67	70.67
22	Fen	12.64	50.56
24	Fen	26.09	104.35
25	Marsh	38.07	152.29
26	Fen	0.51	2.04
27	Fen	21.87	87.46
28	Marsh	24.84	99.37



## A3-3: WETLAND SAMPLING SUMMARY DATA

Wetland Name	Wetland Code	Wetland Type	UTM coordinates 12V,		Sampling Dates		
			Easting	Northing	Sticky Trap	Vacuum Sampler	Aerial Sweeps
Southwest Sands Beaver Pond	SWSS	Marsh	456519	6315878	20 June 2012	11 June 2012	11 June 2012
Shallow Wetland	SSWL	Marsh	458078	6326544	20 June 2012	14 July 2012	14 July 2012
Katie's Sedge Meadow	KSM	Marsh	458278	6317693	20 June 2012	15 July 2012	15 July 2012
Ruth Lake Marsh	RLM	Marsh	463354	6316229	21 June 2012	n/a	21 June 2012
Tower Road Spruce Pond	HSB	Marsh	463700	6290570	26 June 2012	n/a	26 June 2012
Moose	TRM	Marsh	469529	6289121	26 June 2012	9 July 2012	9 July 2012
Tower Road 1	N1	Marsh	469720	6289153	26 June 2012	8 July 2012	8 July 2012
Rhyno's Watering Hole	RWH	Marsh	479425	6274201	6 July 2012	7 July 2012	7 July 2012
Sam's Rodeo	SRW	Marsh	481462	6278833	6 July 2012	10 July 2012	10 July 2012
Beaver Lodge	BLF	Fen	483271	6263298	6 August 2012	9 August 2012	9 August 2012
Maqua Lake Fen	MLF	Fen	482895	6246673	8 July 2012	11 July 2012	11 July 2012
Gravel Pit Fen	GPF	Fen	467641	6312068	13 July 2012	13 July 2012	13 July 2012
Pauciflora Fen	PFF	Fen	485437	6248091	1 August 2012	1 August 2012	1 August 2012
Wapisiw Marsh	WAP	Cons. Marsh	471782	6315495	3 August 2012	n/a	n/a
U-Shaped Cell	UCCELL	Cons. Plots	460214	6323167	2 June 2012	n/a	n/a

## A3-4: METEOROLOGICAL AND WATER QUALITY DATA FOR WETLAND SAMPLING

Wetland	Wetland Type	Date Range Reported	Weather		Cond. (uS)	Water Quality		D.O.
			Air Temp (°C)	Avg. Wind Speed (km/h)		Salinity (ppt)	pH	
<b>Southwest Sands Beaver Pond</b>	Marsh	20-23 June 2012	20.3	7.2	1278	0.6	7.7	5.75
<b>Shallow Wetland</b>	Marsh	20-23 June 2012	20.3	7.2	421.6	0.2	7.8	3.10
<b>Katie's Sedge Meadow</b>	Marsh	20-23 June 2012	20.3	7.2	1246	0.6	7.9	1.65
<b>Ruth Lake Marsh</b>	Marsh	21-24 June 2012	24.6	7.2	437.6	0.2	8.9	5.35
<b>Tower Road Spruce Pond</b>	Marsh	26-29 June 2012	27.3	10.8	360.2	0.2	8.2	3.65
<b>Moose</b>	Marsh	26-29 June 2012	27.3	10.8	428.5	0.2	8.0	5.82
<b>Tower Road 1</b>	Marsh	26-29 June 2012	27.3	10.8	346.5	0.2	9.3	7.15
<b>Rhyno's Watering Hole</b>	Marsh	6-9 July 2012	23.0	3.6	270.3	0.1	13.2	6.09
<b>Sam's Rodeo</b>	Marsh	6-9 July 2012	23.0	3.6	193.6	0.1	8.3	6.63
<b>Beaver Lodge</b>	Fen	6-9 August 2012	24.4	7.2	49.0	0.1	7.0	5.37
<b>Maqua Lake Fen</b>	Fen	8-11 July 2012	30.6	5.4	44.7	0.0	7.1	5.05
<b>Gravel Pit Fen</b>	Fen	13-16 July 2012	25.5	9	52.5	0.0	7.8	5.34
<b>Pauciflora Fen</b>	Fen	1-4 August 2012	25.3	7.2	38.0	0	4.2	5.37
<b>Wapisiw Marsh</b>	Cons. Marsh	3-6 August 2012	24.2	7.2	1823	0.9	8.9	5.70
<b>U-Shaped Cell</b>	Cons. Plots	2-5 June 2012	25.7	9	n/a	n/a	n/a	n/a

## A3-5: STICKY TRAP HEIGHTS AND WATER DEPTHS FOR NATURAL WETLANDS

Wetland Name	Wetland Type	Wetland Zone	Water Depth (cm)	Trap Height (cm)	UTM	
					Easting	Northing
SWSS	Marsh	WM1	0	70	456571	6315836
		EZ1	23.5	71	451590	6315827
		OW1	44	72	456589	6315823
		WM2	0	72	456589	6315797
		EZ2	8	70	456612	6315805
		OW2	48	70	456615	6315809
		WM3	0	75	456624	6315822
		EZ3	18	75	456621	6315822
KSM	Marsh	OW3	59	78	456623	6315809
		WM1	0	75	458283	6317709
		EZ1	33	70	458284	6317727
		OW1	65	70	458286	6317727
		WM2	0	70	458282	6317754
		EZ2	24	70	458283	6317743
		OW2	68	68	458287	6317740
		WM3	0	77	458403	6317724
Shallow	Marsh	EZ3	22	69	458297	6317740
		OW3	74	74	458294	6317735
		WM1	0	84	458073	6326555
		EZ1	43	70	458074	6326561
		OW1	60	80	458881	6326571
		WM2	3	73	458057	6326565
		EZ2	24	73	458067	6326595
		OW2	51	80	458078	6326596
RLM	Marsh	WM3	0	77	458063	6326622
		EZ3	25	66	458083	6326638
		OW3	53	72	458091	6326645
		WM1	0	63	464650	6317011
		EZ1	0	67	464472	6316952
		OW1	61	87	464470	6316948
		WM2	0	77	464657	6317018
		EZ2	22	55	464459	6316945
		OW2	70	75	464450	6316944
		WM3	0	54	464643	6317008
		EZ3	48	42	464456	6316442
		OW3	61	83	464443	6316940

# APPENDICES

Wetland Name	Wetland Type	Wetland Zone	Water Depth (cm)	Trap Height (cm)	Easting	Northing
N1	Marsh	WM1	0	66	469718	6289158
		EZ1	15	50	469724	6289170
		OW1	33	53	469727	6289177
		WM2	0	69	469715	6289202
		EZ2	16	56	469713	6289202
		OW2	42	55	469722	6289201
		WM3	0	60	469695	6289213
		EZ3	12	47	469700	6289233
		OW3	26	51	469706	6289226
Moose	Marsh	WM1	0	80	469520	6289123
		EZ1	30	76	469516	6289119
		OW1	45	69	469519	6289116
		WM2	0	70	469498	6289133
		EZ2	44	70	469492	6289119
		OW2	58	65	469497	6286112
		WM3	30	82	469488	6289134
		EZ3	25	56	469476	6289101
		OW3	57	60	469487	6286104
HSB	Marsh	WM1	24	66	463684	6290569
		EZ1	45	67	463689	6290520
		OW1	87	87	463690	6290548
		WM2	35	77	463697	6290564
		EZ2	43	55	463700	6290558
		OW2	66	75	463695	6290549
		WM3	39	54	463693	6290561
		EZ3	42	42	463677	6296550
		OW3	83	83	463679	6296549

# APPENDICES

Wetland Name	Wetland Type	Wetland Zone	Water Depth (cm)	Trap Height (cm)	Easting	Northing
RWH	Marsh	WM1	4	63	479419	6274282
		EZ1	28	60	479421	6274286
		OW1	70	70	479436	6274278
		WM2	10	65	479425	6274259
		EZ2	15	60	479434	6274255
		OW2	63	65	479439	6274261
		WM3	0	68	479434	6274213
		EZ3	22	60	479445	6274213
		OW3	48	62	479447	6274252
SRM	Marsh	WM1	5	70	481490	6278822
		EZ1	36	73	481489	6278810
		OW1	69	73	481479	6278806
		WM2	0	68	481454	6278828
		EZ2	45	60	481454	6278825
		OW2	74	74	481453	6278815
		WM3	7	66	481442	6278831
		EZ3	28	63	481444	6278823
		OW3	76	76	481444	6278817
MLF	Fen	WM1	0	65	482873	6246669
		EZ1	0	60	482920	6246666
		OW1	70	62	482933	6246628
		WM2	0	65	482589	6246852
		EZ2	0	62	482606	6246828
		OW2	70	69	482586	6246816
		WM3	0	74	482554	6246836
		EZ3	0	68	482555	6246843
		OW3	73	74	482525	6246859

# APPENDICES

Wetland Name	Wetland Type	Wetland Zone	Water Depth (cm)	Trap Height (cm)	Easting	Northing
GPF	Fen	WM1	12	67	467677	6312097
		EZ1	5	67	467664	6312111
		OW1	27	60	467616	6312013
		WM2	0	58	467635	6312079
		EZ2	9	60	467628	6312091
		OW2	22	65	467607	6312100
		WM3	0	68	467621	6312067
		EZ3	6	60	467615	6312082
		OW3	27	62	467598	6312100
PFF	Fen	WM1	1	60	485439	6248062
		EZ1	4	67	485437	6248049
		OW1	2	66	485435	6248028
		WM2	1	55	485425	6248074
		EZ2	3	56	485419	6248056
		OW2	4	55	485417	6248043
		WM3	0	50	485400	6248014
		EZ3	3	54	485409	6248017
		OW3	4	55	485426	6248007
BLF	Fen/Marsh	WM1	0	60	483271	6263298
		EZ1	30	65	483285	6263342
		OW1	75	61	483285	6263355
		WM2	0	59	483308	6263300
		EZ2	30	68	483312	6263341
		OW2	75	75	483292	6263355
		WM3	0	54	483290	6263301
		EZ3	30	55	483301	6263340
		OW3	75	60	483305	6243361

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