

Chapter 1

The Theory and Application of Biological Indicators: A Literature Review

1.1 Biological Indicator Theory

1.1.1 *What is a biological indicator?*

The fundamental principle behind biological indicator theory is that organisms provide information about their habitats. A biological indicator (or bioindicator) is a taxon/taxa selected based on its sensitivity to a particular attribute, and then assessed to make inferences about that attribute. In other words, they are a surrogate for directly measuring abiotic features or other biota. Bioindicators are evaluated through presence/absence, condition, relative abundance, reproductive success, community structure (i.e. composition and diversity), community function (i.e. trophic structure), or any combination thereof (Hellowell 1986, Landres et al. 1988). Communities (i.e. organisms living and interacting with one another in a specific habitat) are generally regarded as the most appropriate indicators for conservation biology since inferences can be made at the ecosystem level, as opposed to being limited to an individual species or population (Kovacs et al. 1992).

Much of the discussion surrounding this theory concerns what exactly it is that biological indicators ‘indicate’, and whether or not inferences – beyond the condition of that particular taxon – are legitimate. Irrespective of validity, these inferences fall into three categories: 1) the condition of their habitat, 2) population trends of other taxa, or 3) the diversity of other taxa in that habitat, defining three types of biological indicators herein referred to as habitat, population, and biodiversity indicators respectively (adapted

from Caro & O’Doherty 1999). One bioindicator may simultaneously fulfill more than one role.

Flagship and umbrella species are the other two types of surrogate species (Meffe & Carroll 1994, Caro & O’Doherty 1999). Flagships are chosen based solely on their ability to provoke public and political compassion, and serve science by garnering public support for conservation efforts (Caro & O’Doherty 1999). A classic example is the World Wide Fund for Nature’s (WWF) adoption of the Giant Panda (*Ailuropoda melanoleuca*) as a symbol of international conservation due its massive public appeal. Problems arise when limitations are ignored and flagships are deemed suitable bioindicators based on popular opinion rather than scientific merit (Andelman & Fagan 2000, Simberloff 1998). This improper application of a flagship species is a misuse of limited conservation resources that serves to discredit single-species management in general. Taxa that require vast areas of habitat (e.g. Grizzly Bear, *Ursus arctos horribilis*; Caribou, *Rangifer tarandus*; or Green Sea Turtles, *Chelonia mydas*) are often selected as umbrella species and used to delineate protected area boundaries with the supposition that providing habitat for the taxa that demands the most will automatically provide for the rest. This approach is criticized for relying more on blind-faith than science since it overlooks both stenotopic species and species at risk (Simberloff 1998, Schwartz 1999, Andelman & Fagan 2000).

1.1.2 Why indicator species?

Bioindicators make a broad and intangible concept such as ‘biodiversity’ or ‘ecosystem monitoring’ manageable by breaking it down into a specific set of

quantifiable indicators (Noss 1990). Inference through biological indicators replaces direct measurement when such measurements are not possible, too expensive/difficult, or too direct (Landres et al. 1988, Caro & O'Doherty 1999). Some historical events are impossible to observe directly, but can be inferred via bioindicators, such as reconstruction of lake pH using diatom communities in lake sediments from known dates (Renburg & Hellburg 1982), or examination of museum bird skins as bioindicators of past mercury concentrations (Thompson et al. 1992). Infinite natural complexity and finite management resources dictate that certain parts must be chosen as surrogates for studying the whole; it is impossible to measure every single abiotic and biotic attribute. Biological indicator theory serves to select surrogates that offer managers the largest universe of extrapolation.

Environmental conditions are often highly variable, making it too difficult or too expensive to get accurate measurements. Some substances are altered very quickly after they enter the environment and may be easily missed by infrequent measurements. It may not be cost effective to monitor as frequently as is needed to obtain an accurate reading, especially for brief (e.g. industrial releases) or unpredictable intermittent events (e.g. sewer overflows, storm run-off). Measuring water quality is a good example since water chemistry is inherently so variable; there are temporal and spatial challenges with data collection; measurements only really provide information on conditions at that moment in time (Spellerburg 1991, Resh et al. 1996). By monitoring organisms in addition to physical/chemical attributes a temporal aspect is inherently introduced since organisms incorporate past, as well as present, conditions (Rosenburg & Resh 1996).

Direct measurements of abiotic (i.e. pollution) or biotic (i.e. introduced species) variables are important but also too direct to provide insight into ecological effects, especially when considering the synergistic effects of multiple factors; we lack complete understanding of synergistic interactions and often of the appropriate choice of substances to measure (Karr 1981, Croonquist & Brooks 1991, Karr 1991, Spellerburg 1991, Kremen 1992, Lambeck 1997, Paoletti 1999, Hilty & Merenlender 2000, US EPA 2002a). For example, mercury concentrations in arctic ice are too low to get an accurate (uncontaminated) reading, yet unhealthy concentrations are found in porpoises, birds, humans and bears due to bioaccumulation (Thompson et al. 1992). Measuring abiotic parameters exclusive of the impact of these conditions on biotic indicators may provide incomplete or inaccurate information on the state of an ecosystem.

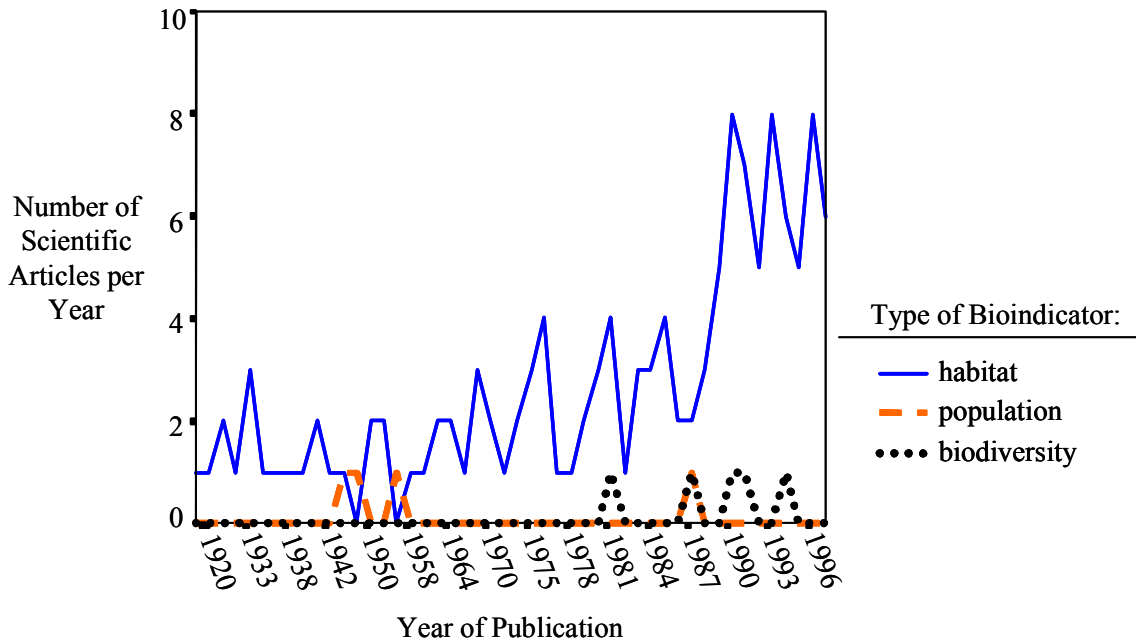
1.1.3 Evolution of biological indicator theory

The term “bioindicator species” was coined by Kolkwitz and Marsson in 1908 and 1909 regarding the impact of organic pollution (i.e. sewage) on aquatic organisms (Rosenburg & Resh 1996). Bioindicator literature has since developed to include the concepts of population and biodiversity indicators (i.e. originating around 1942 and 1980, respectively), though the bulk of articles remain on the topic of habitat indicators (Figure 1–1; Appendix-A).

The idea of habitat indicators is definitely the most accepted and studied type of bioindicator in the scientific literature. This concept arose in the field of botany, therefore the bulk of the early literature deals with plants as indicators of soil chemistry or habitat quality (i.e. pollution), but other commonly studied taxa include vertebrates and

aquatic invertebrates (Figure 1–2; Appendix-A). Scientific merit, not frequency of use, determines the suitability of a bioindicator and the most recommended taxa for bioindicator use are aquatic macroinvertebrates and algae (i.e. recommended by 27% and 25% of the reviewed articles; other taxa included protozoa, bacteria, fish, macrophytes, fungi, yeasts, and viruses) (Hellowell 1986).

Figure1–1: Timeline of the types of biological indicators discussed in the scientific literature during the 20th century (earliest publication to 1998). Results of a JSTOR© database search (Appendix-A for details).

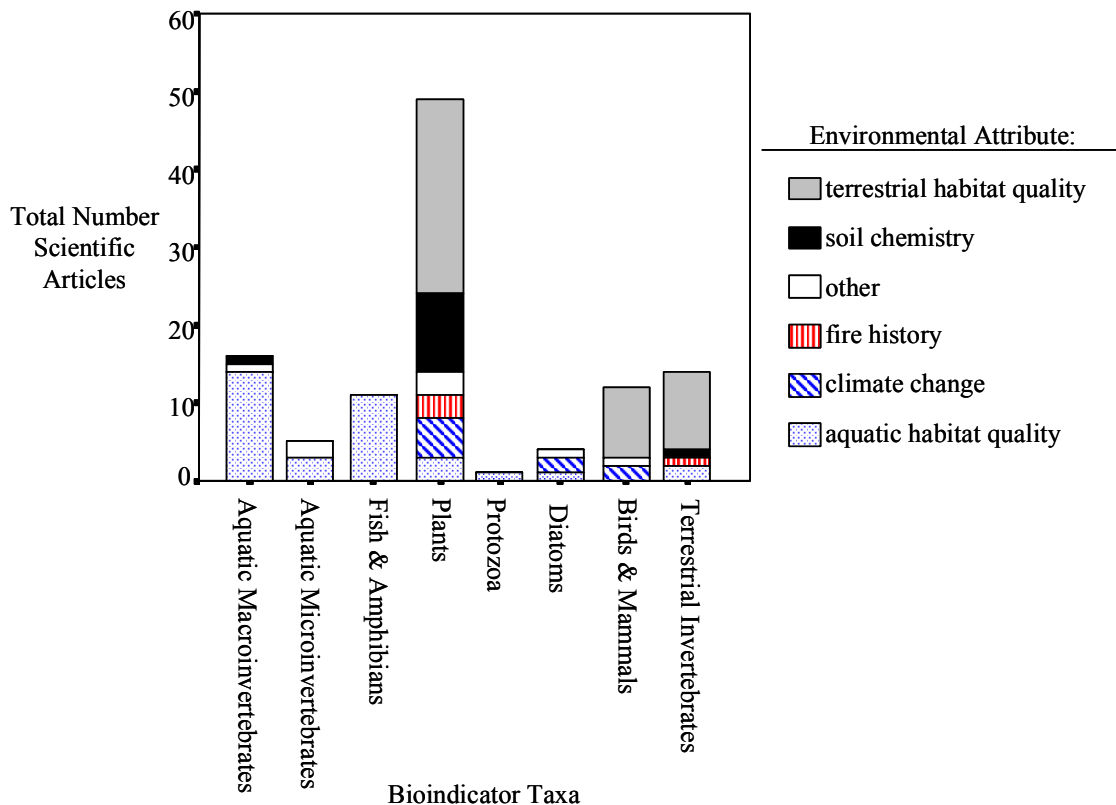


Indices of Biotic Integrity (IBI) and other similar multi-metric concepts have developed since the 1970’s as methods to quantitatively assess environmental condition through habitat indicators (Rosenburg & Resh 1996). IBI’s are an alternative to statistical analyses that aim to condense and clarify complicated ecosystem interactions into a numerical index based on scores for “high” or “low” environmental quality from

various metrics (i.e. habitat indicators; usually 8-12 per site) (US EPA 2002a). Karr (1981) coined the methodology for quantitative assessments that has since been widely applied and developed as a tool to evaluate riparian health (almost exclusively in lotic systems) by many state and federal agencies in the U.S.A., most prominently the U.S. Environmental Protection Agency (US EPA 2002a).

Figure 1–2: Distribution of scientific articles (earliest – 1998) between different bioindicator taxa.

Results of a JSTOR© database search (Appendix-A for details).



Biological indicators are currently used and promoted by numerous conservation agencies as a means to tackle biological monitoring and assess human impacts, including the World Conservation Union (IUCN), World Conservation Monitoring Centre (UNEP),

U.S. Environmental Protection Agency (US EPA), as well as the Nature Conservancy, World Wide Fund for Nature (WWF), Friends of the Earth (FOE), and Greenpeace (IUCN 1989, US EPA 2002a, UNEP 2002).

1.1.4 Critique of biological indicator theory

Biological indicators have high intuitive appeal and have been eagerly embraced by many conservation managers, frequently without regard for selection criteria or empirical evidence (McGeoch & Chown 1998). The most common criticism of indicator species is that scientifically invalid criteria have been used for their selection, whether this is socio-economic pressure to study charismatic macro-fauna (Landres et al. 1988, Niemi et al. 1997, Simberloff 1998, Hilty & Merenlender 2000), or the desire to study a convenient or favourite taxa (Williams & Gaston 1994, Stork & Samways 1995, McGeoch 1998). A lack of standardized techniques for bioindicator-based research has been implicated as a reason for high discrepancy with respect to its validity (Reyers et al. 2000). These criticisms can be avoided by basing indicator taxa selection on research objectives, rather than vice versa, and by following the guidelines and selection criteria offered in the literature (Phillips 1980, Hellowell 1986, Pearson 1994, McGeoch 1998, Caro & O'Doherty 1999, New 1999, US EPA 2002b). Odonates were chosen as bioindicators for this thesis based on published research on their sensitivity to environmental disturbances in lotic systems (Watson et al. 1982, Takamura et al. 1991, Clark & Samways 1996, Samways & Stetler 1996, Stewart & Samways 1998), and the results of preliminary research within the study area in 1999 (Hornung & Rice 2003 In press).

Even when adequately selected, controversy remains over what exactly it is that indicators ‘indicate’ (i.e. what is the legitimate universe of extrapolation?). Some argue that biological indicators only provide information about that particular taxon and it is conjecture, not science, to extrapolate findings to other taxa (Simberloff 1998). Others argue that it is perfectly reasonable to infer how other taxa are faring once a relationship has been established between indicator and indicatee (McGeoch 1998, Caro & O’Doherty 1999). The ramification is that this relationship must be established for each region (i.e. at least each continent – see Pearson & Carroll 1997) and system (e.g. lotic vs. lentic aquatic habitats), and this need for empirical confirmation diminishes the practical appeal of bioindicators as management short-cuts, at least in the short-term.

Each of the three types of biological indicators is critiqued further below.

1.1.5 Habitat indicator species

Habitat indicators provide information about the quality of their habitat through their physical condition or presence/absence, thereby effectively functioning as “biological litmus paper” (Hellawell 1986). There is little controversy concerning their validity since they involve very little inference and have a long history of study in the fields of botany and environmental toxicology (Landres et al. 1998, Paoletti 1999). The scientific community appears to accept the need to assess how changes in the physical environment materialize in the biotic community (Karr 1981, Landres et al. 1988, Karr 1991, Paoletti 1999, US EPA 2002a), as well as the concept that managing for the most sensitive taxa accounts for other less sensitive taxa (Lambeck 1997, but see Simberloff 1998).

1.1.6 Population indicator species

Population indicator species are also selected based on their sensitivity to particular environmental attributes. An important distinction is that their population trends are extrapolated to reflect those of similar species, rather than the condition of their environment (Landres et al. 1988, McGeoch 1998, Caro & O’Doherty 1999). This “guild-indicator approach” is charged with oversimplifying interspecific relationships since species are different in their response to habitat changes and mechanisms of population control (e.g. sensitivity to disease resistance), and the removal or decline of one species may actually benefit a similar species by freeing up limited resources (Landres et al. 1988, Caro & O’Doherty 1999). To ensure the validity of population indicators the relationship between the indicator species and its guild members must be empirically established. Little correlation has been shown to exist between indicator taxa and guild members (e.g. forest bird communities; Mannan et al. 1984, Szaro et al. 1986), except concerning predator – prey relationships (e.g. fewer Cape Gannets (*Morus capensis*) when anchovies were scarce (*Engraulis capensis*), or correlations between abundance of White-backed Woodpeckers (*Dendrocopos leucotos*) and saproxylic beetles (Order Coleoptera); Oatley et al. 1992 and Martikainen et al. 1998, respectively).

1.1.7 Biodiversity indicator species

The most recent application of bioindicator theory developed from the concept of “biological diversity”, which was made commonplace by E.O. Wilson in the late 1980’s (Wilson 1988) and the Rio Earth Summit in 1992 (UNEP 1992) (Figure 1–2). There is much discussion in the scientific literature as to the validity of biodiversity indicators,

with studies that both support (Pearson 1992, Kremen 1994, Pearson 1994, Carroll and Pearson 1998, and Reyers et al. 2000) and oppose this theory (Landres et al. 1988, Kremen 1992, Prendergast 1993, Faith & Walker 1996, and van Jaarsveld et al. 1998). Biodiversity indicators have been used to infer lower taxon (i.e. species) richness by surveying higher taxonomic levels (i.e. family) (supported by Oliver & Beattie 1993, Gaston 2000; disputed by Goldstein 1997, Prendergast and Eversham 1997), however they are more commonly used to identify hubs of biodiversity by inferring overall diversity from that of an indicator taxa (McGeoch 1998, Caro & O'Doherty 1999).

The ability to infer overall biodiversity from a single index is disputed (Faith & Walker 1996, Landres et al. 1988, Simberloff 1998, van Jaarsveld et al. 1998) and has been shown to be invalid in some cases (Goldstein 1997). For example, Oliver & Beattie (1992) found invertebrate diversity inaccurately represents vertebrate diversity, Kremen (1992) found Malagasy butterflies were inappropriate indicators of plant diversity, while Prendergast et al. (1993) found little correlation between the species richness of birds, butterflies, dragonflies, and aquatic plants in Britain. In addition, the presence of threatened or endangered species does not necessarily coincide with areas of high biodiversity, consequently making species at risk inadequate biodiversity indicators (Bonn et al. 2002), and biodiversity indicators poor indicators of rare or endemic species (Reyers et al. 2000). However, correlations between the diversity of some related taxa do exist though these may be valid only within a particular geographic area, such as a continent or eco-region (Pearson & Cassola 1992, Flather et al. 1997, Pearson & Carroll 1998, Anderson & Vondracek 1999). Research in support of biodiversity indicator

theory usually has a very limited realm of extrapolation, such as using one butterfly genus to indicate overall butterfly diversity (Kremen 1994).

Relationships between taxa must be validated rather than assumed in order to accurately identify biodiversity indicator taxon (Prendergast 1993, McGeoch 1998, Sahlen & Ekestubbe 2001), and more than one taxon should be used to infer overall biodiversity and avoid great oversimplification (Wilson 1988, Croonquist & Brooks 1991, Kremen 1992 and 1994, Kremen et al. 1993, Saetersdal & Birks 1993, Launer & Murphy 1994, Lambeck 1997, Caro & O'Doherty 1999). The need for multiple taxa and validation within each region erases the initial intuitive appeal of bioindicators as a quick and easy answer for monitoring biodiversity. Indeed, the initial stages of identifying, testing, and selecting habitat, population, or biodiversity indicators will be (temporarily) resource intensive, but this is a necessary first step in order to ensure the validity of the indicator taxon and the accuracy of subsequent inferences.

1.2 Introduction to the Study

1.2.1 Cattle grazing and the prairie landscape

Wetland management on the prairies has increased in intensity in recent years due to the enormity of historical wetland loss, and the increased prevalence of drought. Conservation efforts have focused almost exclusively on water birds, with invertebrates largely ignored except as food sources for waterfowl and shorebirds (Chura 1961, Ashley et al. 2000). This is changing since the ecological and intrinsic value of invertebrates, (especially large charismatic taxa like odonates and butterflies), is gaining appreciation among the public and, concurrently, wetland managers. For example, odonates are now

included on species inventory and species at risk lists internationally (Moore 1997), nationally (NCC 2001), and provincially (Rice 2000, ANHIC 2002).

Cattle (*Bos taurus*) are given free access to prairie wetlands in the study area as a source of drinking water and supplementary forage. Cattle were commonly observed targeting their grazing efforts on wetland vegetation and effectively removing most of the emergent vegetation (e.g. *Typha latifolia* and *Scirpus* sp.). Unrestricted cattle access to wetlands is a well-established management strategy that has arisen out of convenience and economy rather than ecological study. It has been demonstrated that keeping cattle out of wetlands and providing on-site watering troughs can improve cattle health and weight gain, and in turn increase rancher's profits (Willms et al. 1994).

1.2.2 Aquatic invertebrates as bioindicators

Invertebrates are generally regarded as better bioindicator candidates than vertebrates due to their diversity, relatively quick response to environmental change, and inability to adapt outside of particular physiological constraints (Pearson & Cassola 1992, Kremen et al. 1993, Weaver 1995, but see Mensing et al. 1998). Sedentary invertebrates can be used to identify the location of a particular environmental stress, long-lived (i.e. years) species facilitate studying the effects of sporadic disturbances, and collecting invertebrate samples is simple, inexpensive (e.g. dip nets), and publicly acceptable (Hellowell 1986, Resh et al. 1996). The disadvantages to studying invertebrates are mainly logistical (i.e. laboratory work is time consuming, laborious, and requires a certain type of expertise; Hellowell 1986), and ecological; the inherently patchy distribution of some invertebrates may pose sampling problems, and invertebrate

presence or abundance may be confounded by factors other than environmental disturbance (e.g. seasonal variation, substrate availability, dispersal capabilities) (Hellowell 1986, Resh et al. 1996).

Aquatic macroinvertebrates are the most frequently used taxa for monitoring habitat quality (Hellowell 1986), with a well-developed body of information for lotic systems (Hilsenhoff 1987, Kiffney & Clements 1993, Day & Reynoldson 1996, Rosenberg & Resh 1996, Clausen & Biggs 1997, Brooks et al. 1998, Scrimgeour et al. 1998, Barton 2000, Birge et al. 2000, Giggelman & Bocanegra 2000, Barbosa et al. 2001, Braukmann 2001, Lotufu 2001). Most research involves IBI's or similar multi-metric 'rapid assessments' that compare quantitative indices based on numbers and types of species for disturbed versus reference sites, and monitoring programs employing these techniques are underway in the USA and United Kingdom (Resh et al. 1995, Reynoldson et al. 1997, Karr & Chu 1997, US EPA 2002a & b). IBI's have mainly been developed for specific habitats (i.e. small streams) and pollutants; more research is needed regarding other aquatic habitats (i.e. wetlands) and non-point source pollutants (Rosenburg & Resh 1996).

Research on macroinvertebrates as indicators in lentic systems has only recently begun (i.e. mid to late 1990's) and consequently is far less prevalent in the literature (Resh et al. 1995, Rosenberg & Resh 1996, US EPA 2002a). Procedures for lentic habitat indicators have mainly been adapted from the work of stream biologists (for wetlands see US EPA 2002a; for lakes see Somers et al. 1998 and O'Conner et al. 2000). The US EPA has arguably become the authority on wetland rapid assessments, and has compiled comprehensive on-line information and guidelines for all types of aquatic

ecosystems (e.g. wetlands, estuaries, small streams and rivers, lakes and reservoirs, and coral reefs) (US EPA 2002a & c). Otherwise, few studies specifically investigate wetland-tailored IBI's (van Dam et al. 1998, Burton et al. 1999, Kashian & Burton 2000), or macroinvertebrates as wetland habitat indicators (Hicks & Larson 1997, Zimmer et al. 2000, Spieles & Mitsch 2000, and Cohen et al. 2001).

There is a lack of research on aquatic invertebrates as population or biodiversity indicators in both lotic or lentic systems.

1.2.3 Odonates as bioindicators

Odonates are characterized as excellent habitat indicators of present and past (long-term) environmental conditions in aquatic habitats (Watson et al. 1982, Clark & Samways 1996, Samways & Stetler 1996, Stewart & Samways 1998). Concerning their scientific merit as appropriate bioindicator taxa, odonates satisfy most published selection criteria, rank among the top 20% for all candidate taxa, and are one of the best when considering aquatic taxa alone (Table 1–1; Brown 1991, Clark & Samways 1996).

Odonates inhabit both terrestrial and aquatic habitats during their life cycle and therefore may better reflect disturbance to the riparian buffer than other strict wetland obligates.

Regardless of their suitability, odonates have been employed as habitat indicators relatively infrequently in lotic systems (Carle 1979, Watson et al. 1982, Ferreras Romero 1984, Carchini & Rota 1985, Takamura et al. 1991, Clark & Samways 1996, Samways & Stetler 1996, Stewart & Samways 1998), and even less frequently in lentic systems (Chovanec & Raab 1997, Rith-Najarian 1997a & b).

Table 1-1: Suitability of Order Odonata as bioindicator taxa.

(H=habitat indicator, P=population indicator, B=biodiversity indicator;
 *=satisfies criteria, ?=unknown. Adapted from Pearson 1994, McGeoch
 1998, and Caro & O'Doherty 1999)

	Selection Criteria	Bioindicator			Order Odonata
		H	P	B	
Measurement Attributes	Represents other species		*	*	?
	Taxonomically well-known	*	*	*	*
	Easy/cheap to sample	*	*	*	*
	Accessible breeding site	*	*		*
	Single species	*		*	*
	Species assemblage		*		*
	Baseline data available	*			not often
Life-history Attributes	Small body size	*			*
	Short generation time	*	*		variable
	High metabolic rate	*			*
Ecological Attributes	Medium home range size	*			*
	Resident (not migratory)	*	*		almost all
	Particular trophic level	*	*		*
Attributes of Commonness	Abundant	*	*		most species
	Ubiquitous	*	*	*	most species
	Habitat specialist			*	*
Environmental Sensitivity	Sensitive to human disturbance	*	*		*
	Low variability in response	*	*		?
Social Attributes	Intrinsic/economic value recognized	*	*	*	*

To provide insight into reasons for their response,(e.g. natural or anthropogenic disturbance? organic or inorganic pollution?), bioindicators should be monitored in concert with relevant environmental data (Faith & Walker 1996). Adult odonate species richness has been shown to be correlated with macrophyte richness (Rith-Najarian 1997a & b, Stewart & Samways 1998, Painter 1999, Sahlen & Ekestubbe 2001, Hornung & Rice 2003 In press). Declines in odonate richness have been linked to activities that trample and remove vegetation from the littoral zone including intensive sport fishing (Muller et al. 2003), and wild buffalo trampling (Stewart & Samways 1998).

1.3 Need for this study

This thesis undertakes a novel approach to researching cattle grazing at prairie wetlands since environmental data (i.e. water quality and wetland vegetation) are investigated together with long-lived predaceous aquatic invertebrates (i.e. odonates) to provide insight into how the physical impacts of cattle grazing affect wetland biota. Data collected here will help develop bioindicator theory by addressing the gap in research concerning lentic habitats, population or biodiversity indicators, and non-point source agricultural pollution.

1.4 Thesis outline

This study examines the suitability of odonate community structure as an accurate bioindicator of cattle grazing at prairie wetlands. Chapter 1 provides a literature review of bioindicator theory including its applications and limitations, focussing on macro-invertebrates and aquatic habitats, to provide context and rationale for this thesis.

Chapter 2 investigates odonates as habitat indicators by exploring relationships between larval odonates and cattle impacts on wetland water quality, addressing the following hypotheses:

Hypothesis #1: Cattle affect the water quality of a wetland by urinating and defecating directing into the water.

***Prediction:** Wetlands with greater exposure to cattle grazing (i.e. more continuous grazing regimes) will have poorer water quality than those with truncated or no cattle grazing (as measured by higher concentrations of fecal*

coliforms, ammonium, nitrates + nitrites, total Kjiedahl nitrogen, total phosphorus, chlorophyll-a, and total dissolved solids).

Hypothesis #2: The odonate community at a wetland is sensitive to changes in water quality due to cattle grazing.

***Prediction:** If cattle grazing decreases wetland water quality, then larval odonate genus composition will be altered and genus richness, abundance, and diversity will decreased.*

Chapter 3 addresses the impact of cattle grazing on wetland vegetation, and any subsequent impact on the adult odonate fauna of that wetland, with the following hypotheses:

Hypothesis #3: Cattle grazing directly affects the wetland vegetation community.

***Prediction:** The vegetation community at wetlands with greater exposure to cattle grazing (i.e. more continuous grazing regimes) will have lower species richness, abundance (i.e. % cover), vertical structure (i.e. average height/species), and diversity.*

Hypothesis #4: Wetland vegetation (providing emergence substrates, perches, oviposition sites, and cover) directly affects the adult odonate community.

***Prediction:** If cattle grazing decreases wetland vegetation richness, abundance (i.e. % cover), vertical structure, and diversity, then adult odonate richness, abundance, and diversity will also decrease, and species composition will be altered.*

Odonates as biodiversity indicators are further investigated in Chapter 4 by examining their accuracy in predicting aquatic macro-invertebrate and potential prey

diversity at wetlands with different grazing regimes. The suitability of odonates as bioindicators of the wetland community is assessed by synthesising results from the previous two chapters. The hypotheses tested are:

Hypothesis #5: Larval odonate community structure is an accurate population and biodiversity indicator of the aquatic macroinvertebrate community at a wetland.

Prediction: If odonate prey and overall aquatic macroinvertebrate richness, diversity, or abundance is low, then larval odonate richness, diversity, or abundance will also be low.

Hypothesis #6: Odonates are an accurate biological indicator of cattle grazing impacts on the water quality, vegetation structure and diversity, and aquatic macroinvertebrate community of prairie wetlands.

Prediction: If cattle grazing decreases wetland water quality, then overall larval odonate composition will be altered and taxa richness, abundance, and diversity will decrease.

Prediction: If cattle grazing decreases wetland vegetation richness, abundance (i.e. % cover), vertical structure, and diversity, then overall adult odonate composition will be altered and richness, abundance, and diversity will decrease.

Prediction: If cattle grazing negatively impacts the aquatic macro-invertebrate community this impact will be mirrored by the larval odonate community.

Chapter 5 provides a synopsis of odonates as biological indicators at prairie wetlands, and discusses the implications of this study to biological indicator theory and the practical utilization of odonates to prairie bio-monitoring.

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Chapter 2

Larval odonates as bioindicators of cattle grazing and water quality at prairie wetlands

2.1 Introduction

Prairie potholes were formed during the last glaciation on the Great Plains of south-central Canada and north-central United States, and are characteristically small (<50ha) and collectively numerous. Greater than 50% of these wetlands have been lost, primarily due to agriculture (Mitsch & Gosselink 1993). Ephemeral natural wetlands undergo a 5-20 year drought cycle (van der Valk & Davis 1978), however many remaining potholes are intensively managed for anthropocentric purposes (i.e. drinking reservoirs for cattle, irrigation basins, waterfowl breeding/hunting grounds), effectively truncating natural draw-down periods and decreasing hydrologic variation.

Prairie marshes are typically basic to alkaline, somewhat saline, and highly productive (Mitsch and Gosselink 1993). Nitrogen and phosphorus are the two most important nutrients in freshwater systems, nitrogen being the most limiting, and excessive amounts of either can cause eutrophication (Mitsch and Gosselink 1993). These nutrients enter wetlands via decomposing organic mater, animal wastes (i.e. human sewage, livestock manure), agricultural or industrial run-off, nitrogen fixation, and erosion or re-suspension of phosphorus in parent geological material. Wetlands are generally regarded as nitrogen and phosphorus sinks, and as such, are frequently constructed to amend nutrient loading from organic pollution (i.e. livestock feed lots, human sewage ponds) (Neely & Baker 1989, Mitsch and Gosselink 1993, Peterson 1998).

Grazing is one of the main sources of disturbance on the prairies, and wetlands are utilized for the water and vegetation they provide livestock. Prairie wetlands are inherently variable systems that have evolved with wild fire and bison grazing (van der Valk & Davis 1978, Mitsch and Gosselink 1993), and may in fact flourish with some level of disturbance. This study is well suited for testing the Intermediate Disturbance Hypothesis (IDH) (i.e. moderate disturbance increases diversity, Connell 1978) concerning prairie wetlands and cattle grazing. Cattle remove emergent and submergent vegetation, trample shorelines, and deposit feces and urine in and around the wetland. Water quality can be degraded by the addition of fecal coliforms via manure, or increased sedimentation via shoreline trampling and grazing (Meehan & Platts 1978, Mosley et al. 1999). Grazing impacts can be visually significant (Figure 2–1), however, the ecological consequences on wetland nutrients is less apparent with conflicting research that both confirms (Schepers et al. 1982, Jansen & Robertson 2001, Scrimgeour & Kendall 2002) and refutes (Buckhouse & Gifford 1976, Bohn & Buckhouse 1985, Clark 1998, Nader et al. 1998) cattle’s negative impact on water quality.

Figure 2-1: Differences in appearance between intensively grazed and ungrazed prairie wetlands.



Ambiguity regarding the ecological impact of grazing on wetlands may be explained by differences in grazing practices (i.e. intensity, timing, frequency) and landscapes (i.e. vegetation communities, topography) (Clark 1998, Mosley et al. 1999). Soil-water interactions, diurnal or seasonal fluctuations, and storm events further affect the buffering capacity of a wetland (Mitsch & Gosselink 1993, Harker et al. 1998). Furthermore, water chemistry is so inherently variable that any detectable decrease in water quality due to grazing only really provides insight into what conditions are at the time of data collection rather than the significance of these conditions to the aquatic flora and fauna (Spellerburg 1991, CAST 1992, Resh et al. 1996).

Biological indicators are valuable for interpreting the ecological consequences of environmental conditions to biota, especially when considering intermittent disturbances (Rosenburg & Resh 1996). Larval odonates have been recognized as sensitive water quality indicators in lotic (Carle 1979, Watson et al. 1982, Ferreras Romero 1984, Carchini & Rota 1985, Takamura et al. 1991, Clark & Samways 1996, Samways & Stetler 1996, Stewart & Samways 1998) and lentic systems (Chovanec & Raab 1997, Rith-Najarian 1997a & b), and are prime candidates for investigating the effects of non-point source agricultural pollution (e.g. cattle grazing) at prairie wetlands.

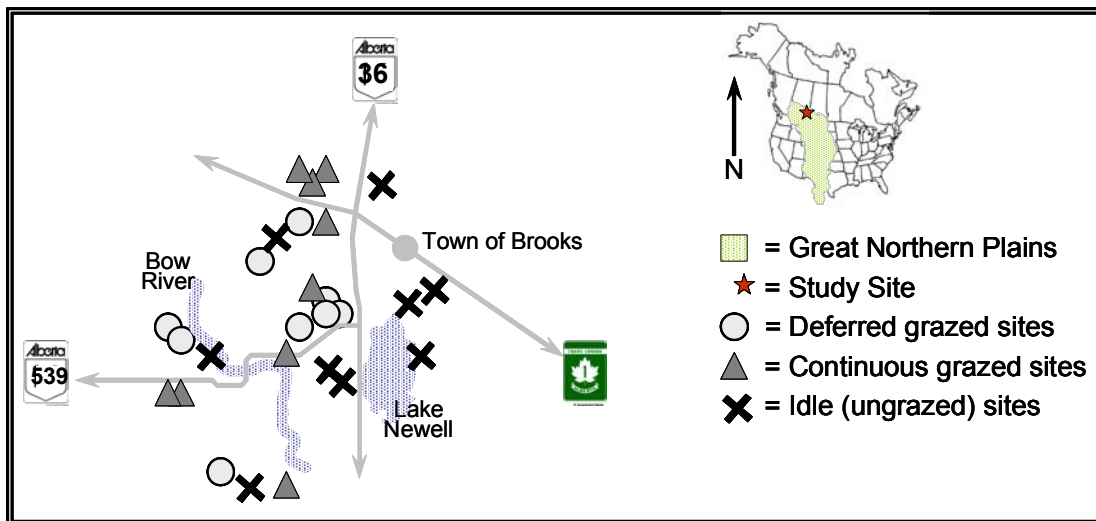
2.2 Objective

This chapter investigates the impact of three cattle grazing regimes on the water quality of prairie wetlands, and the subsequent ecological impacts on the larval odonate (Order Odonata) fauna.

2.3 Study Area

Study sites are located within an 1800 km² area of the dry mixed-grass prairie of south-eastern Alberta, Canada (50° 30' N, 111° 55' W) (Figure 2-2). Agriculture (i.e. cropland, irrigation infrastructure, beef cattle grazing) and oil and gas extraction are the main contemporary sources of disturbance within the study area. Free-roaming bison herds and wildfires were past sources of landscape disturbance, but these have been eliminated as 94% of Alberta's native grasslands have been converted to cropland (Environment Canada 2002). The dry mixed-grass prairie still supports substantial tracts of grazing land because precipitation is insufficient for dependable cropping regimes.

Figure 2–2: Study area location.



Moisture deficits are typical in this dry and windy eco-region, particularly at the end of summer (i.e. 250mm mean annual precipitation; 19 km/h mean annual wind speed) (Environment Canada 2002). Study site ponds are not typical of naturally occurring ephemeral prairie wetlands, however, because water levels are artificially controlled by

the Eastern Irrigation District (EID) and Ducks Unlimited Canada (DUC) via a network of irrigation canals. These managed wetlands remain wet into late summer and have been created to serve as irrigation basins and sources of drinking water for beef cattle and to a lesser degree, to provide wildlife habitat.

2.4 Methods

2.4.1 Experimental Design

Wetlands were selected based on five criteria: grazing regime during the previous five years, basin characteristics (i.e. size, source, and degree of isolation represented at least once in each treatment), surrounding landscape (i.e. grasslands only, no croplands), wetland class (i.e. Stewart and Kantrud (1971) Class IV or V), and access permission.

Manipulating pasture sizes and stocking rates for the sake of this project was logistically infeasible. Continuous and rotational grazing are the two predominant grazing strategies employed within the study area. Continuous grazing regimes keep cattle in one pasture all season (i.e. May to August) and consequently involve small herds of cattle on small sections of private land. More commonly, rotational grazing merges many herds to form a single ‘super herd’ managed by a local grazing association. To ensure pasture re-growth and a constant supply of forage, cattle are directed through a series of pastures beginning with early-germinating tame pastures (i.e. exotic species) and ending on native grasslands. Deferred grazing regimes are a derivation of rotational grazing systems and are promoted by DUC in an attempt to decrease waterfowl nest exposure and mortality due to foliage removal and cattle trampling. DUC negotiates

contracts with grazing managers to delay cattle access to specific wetlands until after July 15th, at which time most waterfowl broods have fledged (NAWMP 1999).

The experimental unit for this study were the individual wetlands. Wetland characteristics were estimated from a continuous 250m stretch of shoreline that was selected to best represent the variation in wetland vegetation and shoreline structure. All data collection took place along these 250m segments (delineated into six 50m sections selected as random sub-samples) to standardize sampling wetlands of various sizes. The grazing regime in the pasture surrounding each wetland supported one of three treatments: deferred grazing (i.e. mid-July to mid-August), continuous grazing (i.e. approximately May to August), and idle (ungrazed) pastures (i.e. control treatment). Larval odonates were sampled using random stratified design (Krebs 1989); strata were delineated based on distance from interface of vegetation and open water. Pseudoreplication was avoided since experimental units (i.e. wetlands) were replicated nine times in each of the three treatments, and each wetland is far enough apart (i.e.>1km) to be considered statistically independent (Hurlbert 1984). Although wetlands are connected via irrigation canals they are distinct habitats (i.e. flowing vs. standing water, sparsely vegetated banks vs. heavily vegetated shorelines), and canals are dry and unsuitable for aquatic organisms for a large portion of the year (i.e. late summer through spring); therefore wetlands were considered biologically unconnected and statistically independent. Grazing was ubiquitous throughout the study area and as a result the limitation on sample size was the number of ungrazed (idled) pastures in local protected areas. A total of twenty-seven wetlands were selected for a balanced design of nine replicates in each of the three treatments (Figure 2-2). Response variables included

wetland water quality and larval odonate community metrics. Wetland vegetation structure, adult odonate community, and other aquatic invertebrates are addressed in following chapters.

2.4.2 Measuring Wetland Water Chemistry

Basic water chemistry data were recorded with a handheld multi-meter (YSI Model 85 Dissolved Oxygen and Conductivity Meter) at each wetland for all rounds beginning with round three in 2000. Three of the six survey points were randomly selected for each sampling period and measurements were averaged to obtain a composite measurement for each site. Temperature (°C), pH, salinity (g/L, temperature corrected), and dissolved oxygen (%) were recorded at each survey point at 1m into the open water zone and at a depth of 15-25cm immediately upon arrival to avoid confounding effects from research activities that disturb wetland sediments.

2.4.3 Measuring Water Quality

Budget constraints dictated that the minimum number of water samples be submitted for analysis. All samples were collected three or more days after a storm event to minimize effects from run-off. Three of the six survey points were randomly selected, and 1L of water was collected from each point at 1m into the open water zone at a depth of 30cm. The three samples were combined and one 0.5L composite sample was packed in ice and shipped to the lab that day.

Two water samples were collected from all wetlands: 1) before grazing at deferred sites (i.e. mid-July 2000), herein called pre-cattle sample, and 2) during cattle

grazing at deferred sites (i.e. late-August 2000), herein called post-cattle sample. These samples were analysed for ammonium (NH_4^+), nitrates and nitrites (NO_2+NO_3), total Kjeldahl nitrogen (TKN), total phosphorus (TP), total dissolved phosphorus (TDP), total dissolved solids (TDS), and Chlorophyll-a concentrations (i.e. algae productivity) at the Limnology Lab at the University of Alberta. An additional sample was collected from all ponds in late August 2000, and analyzed locally at Lakeside Packers (Brooks, AB) for fecal coliform concentrations.

2.4.4 Collecting Larval Odonates and Detecting Fish

During each sampling round, odonate larvae were collected along with other aquatic macro-invertebrates using a D-frame sweep net (one of the best devices for sampling aquatic invertebrates in heavily vegetated wetlands; Turner & Trexler 1997). Three of the six survey points were randomly selected, and three distinct micro-habitats were sampled at each point including: emergent vegetation (i.e. 1m outward from open water), emergent vegetation /open water interface, and deeper open water (i.e. 1m inward from vegetation/water interface). Five 1m sweeps were collected from each microhabitat for a total of forty-five sweeps per wetland. Larvae were preserved in 70% ethanol solution until later identified to genus (i.e. lowest identifiable taxonomic level). Primary references for identification were Clifford (1991) and Merritt and Cummins (1996).

Fish were detected by randomly placing baited minnow traps at three of the six delineated survey points in the open water zone of each site for a two to three day period (checked daily) during the 2001 field season.

2.4.5 Statistical Analysis

The Shapiro-Wilk test was used to check if data was normally distributed and Levene's Statistic was used to test the homogeneity of variances (SPSS 1999, Zar 1999). Repeated measures analyses of variance (or Friedman's tests for non-normal data) were performed to account for seasonal changes in water chemistry, water quality, and odonate fauna. I used Newman-Keuls *post hoc* multiple range tests to determine which means differed when significant differences were found with Friedman's tests. Moristia-Horn indices (i.e. 0 – 1) were arcsine transformed before testing for significant differences between treatments. An alpha of 0.10 was selected *a priori* for treatment impacts on water quality and larval odonates due to the difficulties in obtaining a representative measurement for an attribute as inherently variable of water quality, and the limited number of water samples. An alpha of 0.05 was selected *a priori* for all other analysis.

2.5 Results

2.5.1 Wetland Water Chemistry and Grazing Treatment

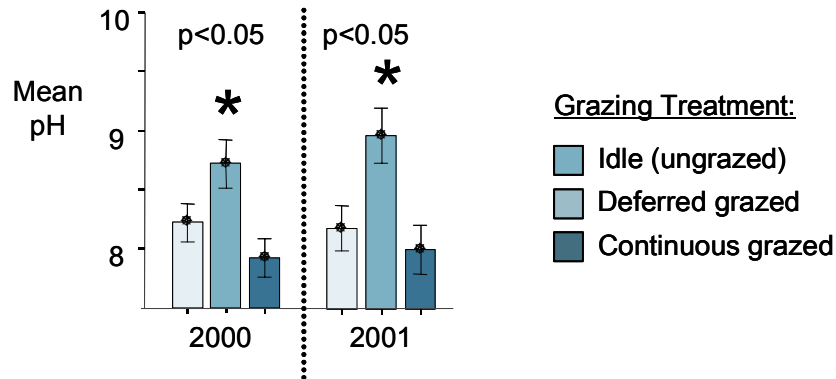
Wetland pH was significantly higher at deferred grazed compared to idle or continuously grazed sites in both 2000 and 2001, when accounting for sampling round (significant treatment effects, sampling period effects and interactions not significant; Table 2-1; Figure 2-3).

Salinity (g/L, temperature corrected) was significantly higher at wetlands with deferred grazed compared to continuously grazed regimes during 2001, accounting for round (significant treatment effects, sampling period effects and interactions not significant; no significant differences in 2000; Table 2-2; Figure 2-4).

Table 2-1: Results for differences in wetland pH using Friedman’s test.
(χ^2 values from Table B.1 in Zar 1999)

Year	Source	df	H	χ^2_{crit}	p-value
2000	Treatment	2	8.829	5.991	<0.05
	Round	2	0.041	5.991	not significant
	Interaction	4	0.582	9.488	not significant
2001	Treatment	2	10.633	5.991	<0.05
	Round	2	5.537	5.991	not significant
	Interaction	4	<1	9.488	not significant

Figure 2-3: Wetland pH and grazing treatment.
(Bars represent ± 1 SE; *= significant differences)



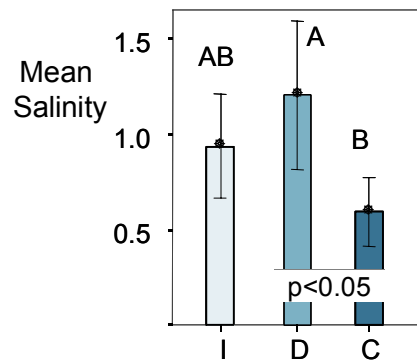
Grazing treatment did not significantly affect wetland temperature ($p=0.154$), or dissolved oxygen ($p=0.258$) in either year of data collection, accounting for sampling round (i.e. repeated measures ANOVA).

Table 2-2: Results for differences in wetland salinity Friedman’s test.
(χ^2 values from Table B.1 in Zar 1999)

Year	Source	df	H	χ^2_{crit}	p-value
2001	Treatment	2	6.338	5.991	<0.05
	Round	2	<1	5.991	not significant
	Interaction	4	<1	9.488	not significant

Figure 2-4: Wetland salinity and grazing treatments, 2001.

(Bars represent ± 1 SE; *= significant differences; I=idle, D=deferred, C=continuous)



2.5.2 Cattle Impacts on Wetland Water Quality

The impact of cattle urine on water quality as a result of cattle grazing at a wetland was measured via total dissolved solids (TDS). No significant differences in TDS were found between grazing treatments when accounting for time (Friedman's test).

Organic inputs from cattle feces in and around a grazed wetland were assessed by analysing differences in fecal coliform, nitrogen, phosphorus, and chlorophyll-a concentrations. No significant differences in fecal coliforms ($p=0.861$), chlorophyll-a, total dissolved phosphorus, total phosphorus, total Kjeldahl nitrogen (TKN), total nitrogen (i.e. $\text{NO}_2+\text{NO}_3 + \text{TKN}$), or ammonium were found between grazing treatments when accounting for time (Friedman's test for all); time was not significant for any water quality parameter.

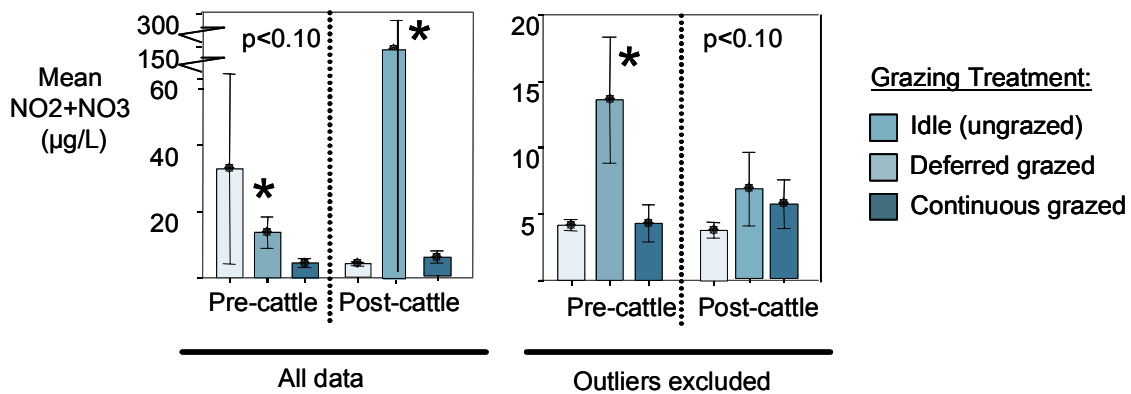
Significantly higher concentrations in NO_2+NO_3 ($\mu\text{g/L}$) were present at deferred grazed sites, as compared to continuously grazed or idle sites, for both pre- and post-cattle samples (power=0.50; Table 2-3; Figure 2-5). However, even when outliers were omitted (i.e. one site from both idle and deferred grazed wetlands) to reduce the high

level of within-treatment variation and maintain homoscedasticity, significantly higher NO₂+NO₃ concentrations were found only within the pre-cattle sample, and therefore can not be associated with cattle grazing (power=0.35; Table 2-3; Figure 2-5).

Table 2-3: Results for differences in wetland NO₂+NO₃ levels using Friedman’s test. (χ^2 values from Table B.1 in Zar 1999)

Outliers present	Source	df	H	χ^2_{crit}	p-value
Yes	Treatment	2	6.402	4.605	<0.10
	Round	1	2.822	2.706	<0.10
	Interaction	2	2.764	4.605	not significant
No	Treatment	2	5.460	4.605	<0.10
	Round	1	3.035	2.706	<0.10
	Interaction	2	4.263	4.605	not significant

Figure 2-5: Wetland NO₂+NO₃ levels between grazing treatments. (Bars represent ± 1 SE; *= significant differences)



2.5.3 Cattle Grazing and Larval Odonates

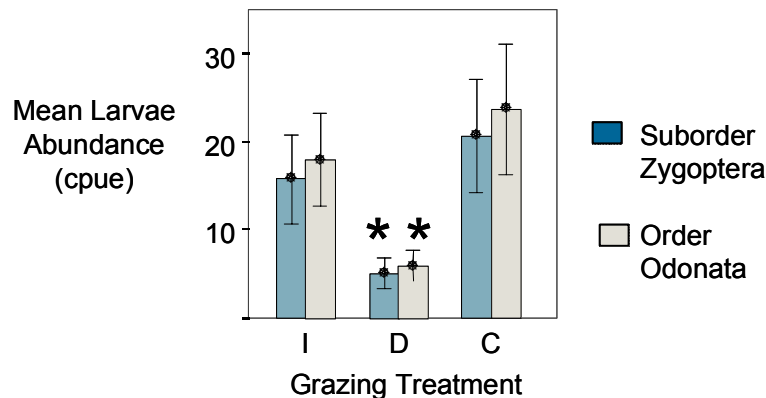
Five genera of larval odonates were collected from the study site including the dragonflies *Aeshna*, *Anax*, and *Sympetrum*, and the damselflies *Enallagma* and *Lestes*. Analysis was done per genus as well as by taxonomic groups (i.e. Suborder Anisoptera

(dragonflies), Suborder Zygoptera (damselflies), and Order Odonata). Only data from 2000 was analysed to coincide with the year water quality samples were collected. No significant treatment effect was found for Anisoptera, Zygoptera, or overall genus richness, when accounting for sampling round (Friedman's test). Zygoptera and overall Odonata abundance was significantly lower at deferred grazed wetlands compared to idle or continuously grazed wetlands (significant treatment effects, sampling period effects and interaction not significant; Table 2-4; Figure 2-6). No treatment effect was found regarding differences in Anisoptera abundance.

Table 2-4: Results for differences in larval abundance using Friedman's test. (χ^2 values from Table B.1 in Zar 1999)

Group	Source	df	H	χ^2_{crit}	p-value
Zygoptera	Treatment	2	8.413	4.605	<0.10
	Round	5	5.853	9.236	not significant
	Interaction	10	4.625	15.987	not significant
Odonata	Treatment	2	6.560	4.605	<0.10
	Round	5	3.286	9.236	not significant
	Interaction	10	4.854	15.987	not significant

Figure 2-6: Larval Odonate Abundance between Grazing Treatments. (Bars represent ± 1 SE; *=significant difference; I=idle, D=deferred, C=continuous)



The Morisita-Horn index is a measure of the similarity in community composition between different communities, ranging from completely different to identical (i.e. score 0–1, respectively) that is relatively independent of sample size or diversity (Wolda 1981). This similarity index was calculated for the larval odonate fauna per round for each site, and analysed for significant differences between treatments (i.e. arcsine transformed, Friedman’s tests). Significant differences in larval odonate composition were found between wetlands with idle and continuous grazing regimes, when accounting for sampling round (significant treatment effects, sampling period effects and interaction not significant; Table 2-5; Figure 2-7).

Table 2-5: Results for differences in larval odonate composition using Friedman’s test.
(χ^2 values from Table B.1 in Zar 1999)

Source	df	H	χ^2_{crit}	p-value
Treatment	2	7.620	4.605	<0.10
Round	5	7.803	9.236	not significant
Interaction	10	9.931	15.987	not significant

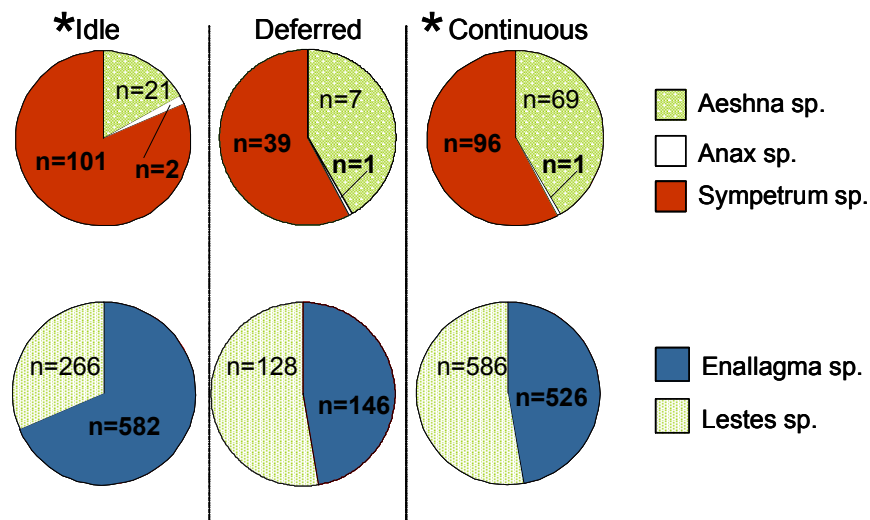
Figure 2-7: Differences in larval odonate genus composition between treatments.
(Bars represent ± 1 SE; I=idle, D=deferred, C=continuous; plots of untransformed data)



Closer examination of the genus composition of larval odonates between grazing treatments reveals differences in the relative abundance of both Anisoptera and Zygoptera larvae at idle and continuously grazed wetlands (Figure 2–8). *Sympetrum* sp. (5 possible species) and *Enallagma* sp. (5 possible species) dominate at ungrazed sites, but near equal numbers of *Sympetrum* sp. and *Aeshna* sp. (2 possible species), as well as *Enallagma* sp. and *Lestes* sp. (3 possible species) exists at continuously grazed sites.

Figure 2-8: Larval odonate composition and relative abundance between grazing treatments.

(* = significant differences)



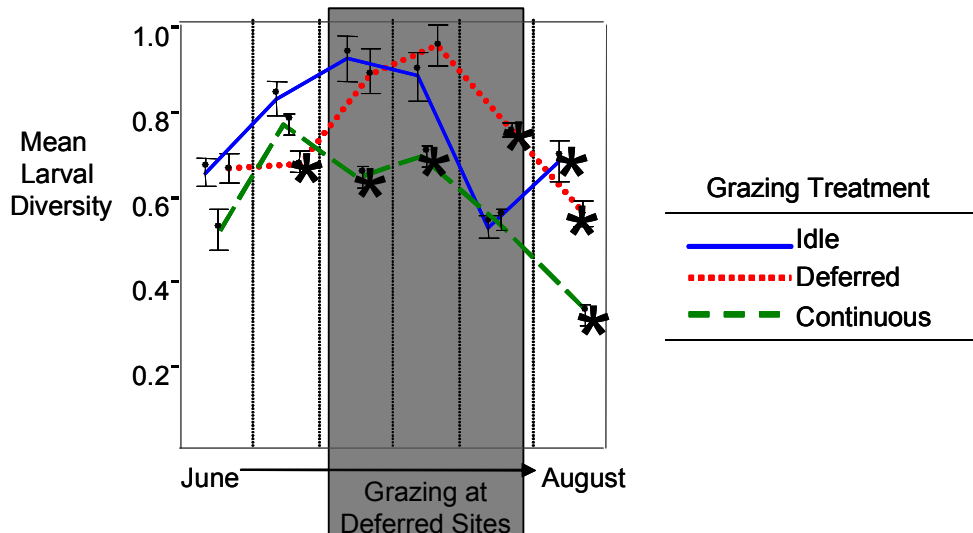
Shannon diversity indices measure the number and evenness of species in a community and were used to assess differences in larval odonate diversity between treatments. Both treatment and round were found to significantly affect larval odonate diversity (significant treatment and sampling period effects, and significant interaction; Table 2-6; Figure 2–9). Overall larval odonate diversity tended to increase over summer,

peaking in July. Larval diversity was significantly lower at continuously grazed wetlands compared to idle and deferred grazed wetlands during Round 2-6. Larval diversity at idle and deferred grazed sites was relatively comparable and high for rounds one to four. Larval odonate diversity was highest at deferred sites in round five (i.e. immediately after grazing stopped), and at idle sites in round six.

Table 2-6: Results for differences in larval odonate diversity using Friedman’s test.
(χ^2 values from Table B.1 in Zar 1999)

Source	df	H	χ^2_{crit}	p-value
Treatment	2	7.269	4.605	<0.10
Round	5	86.587	9.236	<0.10
Interaction	10	30.061	15.987	<0.10

Figure 2–9: Differences in Larval Odonate Genus Diversity at Wetlands with Different Grazing Treatments
(Bars represent ± 1 SE, * = significant differences when $\alpha=0.10$)



2.5.4 *Fish Presence*

No significant differences in the presence/absence of fish at study wetlands were found between treatments ($p=0.538$). Fish were found at all but six of the twenty-seven study ponds.

2.6 Discussion

Wetland hydrology is the primary source of influence for wetland properties such as pH, oxygen content, and nutrient availability (Mitsch & Gosselink 2000). The hydrogeomorphology of a wetland refers to its hydrology (e.g. flooding period, water source), geomorphology (e.g. landscape topography, degree of isolation), and encompassing climate (Brinson 1993). Initial site selection attempted to select template wetlands whose hydrogeomorphology was as similar as possible, with cattle grazing regime being the only major difference between sites.

2.6.1 *Wetland Water Chemistry*

Basic wetland water chemistry data were collected to characterize study sites, as well as to compare treatments and account for any consequent effects on the odonate community. Dissolved oxygen fluctuates widely both spatially and temporally within wetlands, and therefore, was not a useful parameter for characterising study ponds. Redox potential is a more reliable quantitative measure for wetland oxygen (i.e. decreases in a predictable fashion as a sequence of reducing reactions occur) (Mitsch & Gosselink 2000), and is recommended in place of dissolved oxygen for future studies. Additionally, macroinvertebrates that respire via gills (i.e. odonates) are indicative of

well-oxygenated wetlands since they can not survive long periods of anoxia (Murkin et al. 2000).

Wetland temperature fluctuates diurnally, but critical thresholds occur during the winter (not summer) months (Murkin et al. 2000). Treatments were compared to see if the removal of wetland vegetation via grazing affects water temperature. No significant difference was detected suggesting the presence of emergent vegetation has little impact on water temperature; however, the scale of measurement was not appropriate to detect micro-habitat changes. Water temperatures were recorded 1m into the open water zone, rather than within the emergent vegetation zone where cattle typically cluster (Jansen & Robertson 2001). Grazing may impact the micro-habitat within the littoral zone, but does not appear to affect overall wetland temperature.

Wetland pH is primarily determined by soil type; mineral soils are usually alkaline while organic soils are typically acidic (Mitsch & Gooselink 2000). The pH data for all study sites, including the significantly higher deferred grazed sites (i.e. $\text{mean}_{2000} = 8.72$, $\text{SD} = 1.02$; $\text{mean}_{2001} = 8.96$, $\text{SD} = 1.22$), are well within the expected range of pH 6-10 for prairie wetlands and likely not biologically significant to aquatic organisms (Mitsch & Gosselink 1993). A slight increase in pH could alter the availability of wetland nutrients by increasing their solubility (National Science Foundation 2003). The link between higher pH and intensive cattle use (i.e. deferred grazed sites) is most likely the wetland soil; trampling of the shoreline by grazing/drinking cattle may disturb wetland sediments and introduce more of the alkaline parental material to the water, thereby increasing wetland pH.

Salinity levels range from nearly fresh to near sea water concentrations on the prairies and are primarily determined by the interaction of groundwater, evapotranspiration, and soil type (Mitsch & Gosselink 2000). Although statistically higher salinity was detected at deferred grazed sites in 2001, these wetlands are relatively fresh (i.e. $\text{mean}_{\text{deferred 2001}}=1.20\text{g/L}$, $\text{SD}=2.02$; $\text{mean}_{\text{others 2001}}=0.899\text{g/L}$, $\text{SD}=1.54$). It is unlikely this increase is biologically significant since salinity levels are well within known tolerance limits of aquatic invertebrates (i.e. most sensitive species thrive in waters 0-5g/L), and within the salinity range required for most wetland vegetation (Murkin et al. 2000).

2.6.2 Wetland Water Quality

Cattle tend to concentrate around prairie wetlands and loiter in the water while they drink and graze, as observed in this study and others (Nader et al. 1998, Jansen & Robertson 2001). Wetland water samples were analysed for substances present in cattle excrement. Nitrogen (i.e. TKN, NO_2+NO_3 , NH_4^+), phosphorus (i.e. TP and TDP), and fecal coliforms are found in cattle manure, while the ions in TDS are found in cattle urine (pers. comm. Richard Casey 2000). Chlorophyll-a was measured to test for differences in wetland productivity with respect to algae as a result of increased nutrients.

Significantly higher nitrate and nitrite (NO_2+NO_3) concentrations were detected within the deferred grazing treatment (Figure 2-5), however these elevated nitrogen levels can not be attributed to cattle since they were recorded before cattle began grazing these sites (i.e. prior to July 15th). Nitrogen differences cannot be attributed to dissimilarities in nitrogen fixing blue-green algae concentrations since chlorophyll-a data (i.e. index for

algae) between treatments were similar. It is probable that the higher pH of deferred grazed sites (Figure 2-3) is affecting NO_2+NO_3 concentrations by increasing nutrient solubility (National Science Foundation 2003). Sources of NO_2+NO_3 other than livestock manure include rainfall, agricultural fertilizers, and decomposing organic matter (Mitsch & Gosselink 2000), but it is unlikely that rainfall or agricultural pollutants are driving this difference in NO_2+NO_3 since all study sites are influenced by the same general weather pattern, and wetlands surrounded by croplands were not selected for study. Higher pH may increase the rate of organic matter decomposition (Mitsch & Gosselink 2000) and thereby indirectly increase wetland NO_2+NO_3 concentrations. Toxicity levels for NO_2+NO_3 are at least two orders of magnitude larger than the maximum concentrations recorded in this study (i.e. 2 500 – 100 000 $\mu\text{g}/\text{L}$ for amphibians and >100 000 $\mu\text{g}/\text{L}$ for cattle and humans), therefore these elevated levels likely have little impact on vertebrate wetland biota. Little is known regarding the ecological impact of nutrient loading on aquatic invertebrates in freshwater wetlands; however preliminary research shows a positive relationship between nutrient loading and overall aquatic invertebrate abundance due to an increase in overall wetland productivity (Murkin et al 1991, Campeau et al. 1994). The seasonal drop in NO_2+NO_3 concentrations due to plant uptake in early summer may explain the lack of a significant difference within the second (post-cattle) sample (Bender 1976).

It is unclear whether the lack of significant difference in water quality between treatments is due to the high buffering capacity of prairie wetlands, or the inherently variable nature of water quality data exacerbated by an insufficient sampling regime; however the former is most likely. Prairie wetlands are known to function as nitrogen

and phosphorus sinks, and consequently nutrient-rich in-flows have minimal effects on the concentration of available nutrients and no impact of long-term nutrient budgets (Mitsch & Gosselink 2000). It is likely that the level of disturbance and nutrient inputs from unrestricted cattle grazing is too low to biologically impact prairie wetlands, which have evolved with, and are dependent on, disturbance (i.e. periodic drought and grazing from bison herds). Conversely, environmental data alone may be insufficient to properly make this assessment. Nutrient inputs to nitrogen-limited systems (i.e. prairie wetlands) will be quickly absorbed by wetland vegetation (Mitsch and Gosselink 1993) and will be difficult to detect via environmental data. Cattle activity (i.e. defecation, trampling) at the wetland shore is an inherently intermittent event, and it is therefore unfeasible to sample water immediately after a disturbance event to measure maximum impact. Sampling efforts were conservative and may have been too infrequent to detect the impact of an intermittent disturbance such as cattle defecation, urination, and trampling (i.e. 50% chance of committing Type II error with outliers included, 35% chance with outliers excluded). Analysis of the impact of grazing treatment on wetland biota (i.e. adult odonates and other aquatic macro-invertebrates) is required before the effect of cattle grazing on wetland habitat quality can be fully assessed.

2.6.3 Larvae and Grazing Treatment

Odonates are predacious macro-invertebrates that occupy both aquatic and terrestrial habitats at different stages of their life cycle. Long-lived aquatic larvae require unpolluted, oxygenated, fresh water, submerged vegetation as cover for avoiding predation and detection by prey, emergent vegetation as emergence structures, and other

aquatic invertebrates as prey (Corbet 1999). Terrestrial adults concentrate around water bodies and depend on aquatic vegetation as a territorial perches, oviposition cues, and shelter from winds (Corbet 1999). Different species emerge and mature throughout the season causing a shift in adult species composition throughout the season (Figure 2–10).

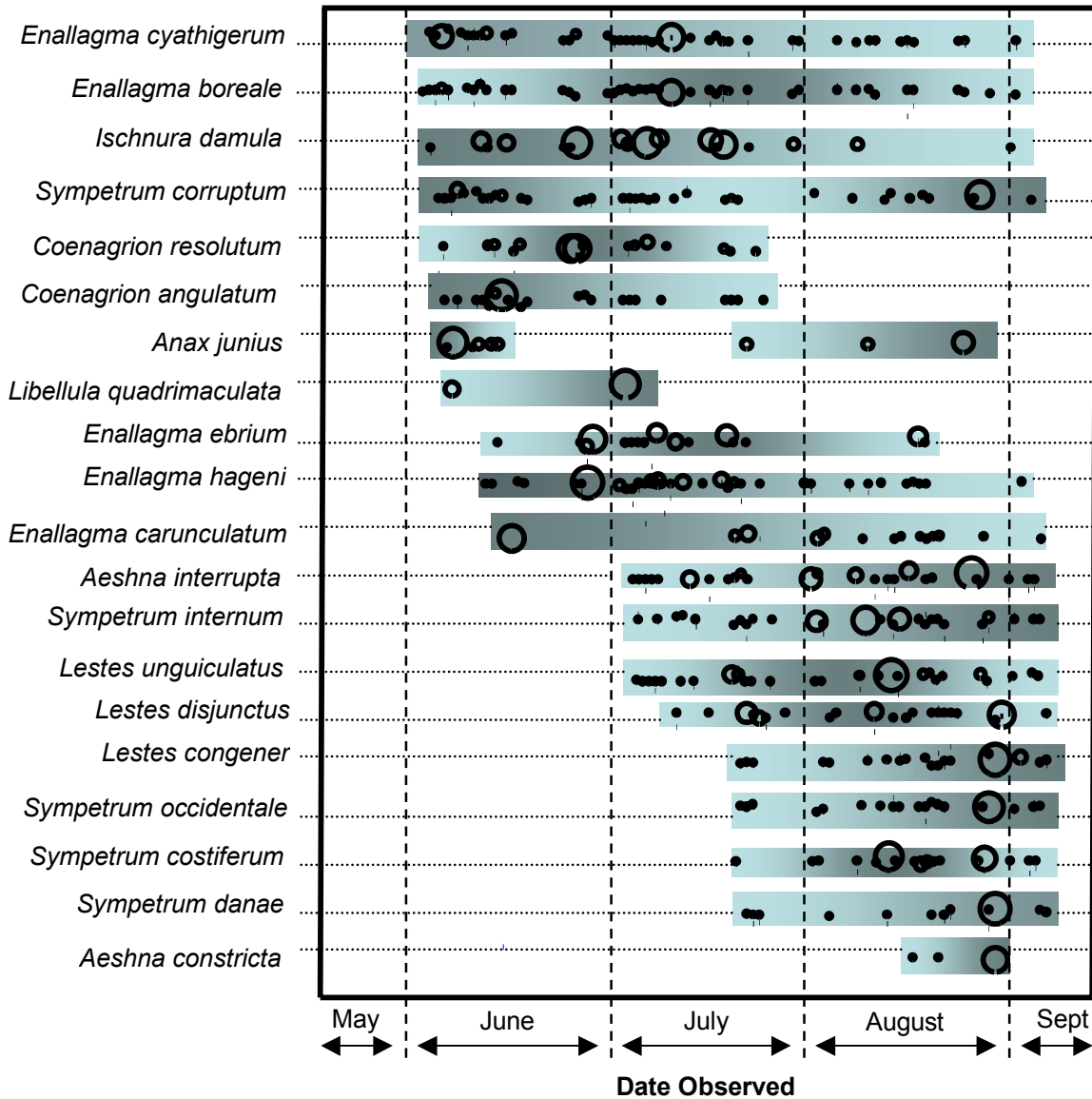
Fish and the consequent predatory pressure on larval odonates are essentially ubiquitous, present at wetlands regardless of treatment, and likely not responsible for any differences in larval odonate communities.

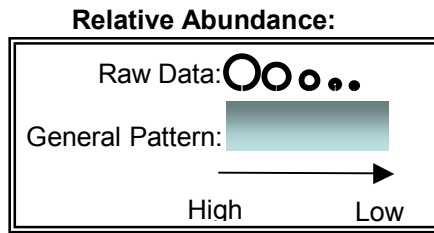
Surveying wetland biota, unlike environmental parameters, incorporates time since the presence and condition of relatively long-lived organisms reflects past events (i.e. intermittent pollution) rather than simply conditions at the time of measurement (Rosenburg and Resh 1996). Larval odonate communities were analysed to test for significant differences between treatments that may not have been statistically attributable to any of the specific environmental parameters measured in this study, reflecting antecedent conditions instead.

Significantly fewer larvae were collected at deferred grazed wetlands rather than continuously grazed wetlands as initially predicted (Figure 2–6). Deferred grazing regimes involve combining many herds of cattle into one “super herd” (i.e. hundreds of cattle) whereas continuously grazed wetlands involve smaller grazing operations. The number of cattle rather than simply the duration of their presence may be more important than initially recognized, making deferred grazing a more acute grazing disturbance. This difference is largely reflected in the Suborder Zygoptera due a larger sample size since seven times more Zygoptera vs. Anisoptera larvae were collected (i.e. $\text{mean}_{\text{Total Zygoptera}}=2235$, $\text{mean}_{\text{Total Anisoptera}}=337$). This relative abundance was also observed in

adult odonates as adult damselflies were observed to be far more abundant in the field than adult dragonflies, especially with respect to the family Aeshnidae.

Figure 2–10: Seasonal changes in adult odonate composition and abundance, 2000.





Habitat quality must be evaluated in terms of survival and reproductive success in concert with abundance data (van Horne 1983). Although the abundance of odonate larvae at idle and continuously grazed wetlands is similar, the proportion of larvae that successfully emerge and reproduce is unknown. Odonates are very vulnerable to physical disturbance during emergence (Corbet 1999), and presumably cattle activity along shorelines (i.e. walking through vegetation) will disturb emerging odonates and decrease emergence success, making continuously grazed habitat more hostile environments for odonates than ungrazed sites, despite similar numbers of larvae.

Deferred grazing is associated with decreased abundance of odonate larvae at wetlands. The littoral zone is the link between cows and odonates; this is where most odonate larvae exist and where most of the physical disturbance from foraging and loitering cattle occurs (i.e. cows were observed trampling and defecating on shorelines while browsing submergent and emergent vegetation). Analysis of water samples found no link between grazing and water quality, however significantly lower larvae abundances suggests that large numbers of cattle (i.e. deferred grazed sites) compromise littoral habitat enough to impact the odonate community of a wetland. Larval odonate fauna represent previous adult odonate fauna via generational lag and therefore fewer larvae may reflect a decrease in ovipositing adults, which cue into wetland vegetation for ovipositing sites (Corbet 1999). Figure 2–10 shows that the majority of the mature odonate fauna at the time of deferred grazing (i.e. mid-July to mid-August) are

damselflies of the genus *Enallagma*, which is consistent with the decline in damselfly larvae.

The larval odonate community of each treatment is compared using the Morisita-Horn similarity index which incorporates presence/absence, abundance, and genus richness, and ranges from 0 to 1 (i.e. 1 most similar). Larval odonate communities were significantly less similar among idle (ungrazed) compared to continuously grazed wetlands (Figure 2–7). There are relatively more *Enallagma sp.* and *Sympetrum sp.* and less *Lestes sp.* and *Aeshna sp.* larvae at idle compared to continuously grazed wetlands (Figure 2–8). No significant difference in wetland water quality was detected between grazing treatments to explain this shift in larval odonate composition. Larvae from the two Zygoptera genera have similar foraging behaviours (i.e. they crawl amongst submerged vegetation as larvae, and glean vegetation as adults) and ovipositing strategies (i.e. endophytic) but differ in over-wintering strategies (Corbet 1999). Over-wintering strategies are more related to wetland hydrology than habitat quality (i.e. *Lestes sp.* Over-winter as diapause eggs as an adaptation to ephemeral prairie wetlands; Corbet 1999), and do not explain differences in larval composition since all wetland water levels are similarly controlled.

Both Anisoptera genera over-winter as diapause eggs and depend on wetland vegetation during ovipositing (i.e. all *Aeshna sp.* and most *Sympetrum sp.*). *Aeshna sp.* larvae are ‘climbers’ that crawl along submerged vegetation and other substrates, whereas larval *Sympetrum sp.* are considered ‘sprawlers’ that crawl along wetland sediments (Corbet 1999). More submerged vegetation exists at continuously grazed sites and cattle chronically trample wetland sediments in the littoral zone, therefore conditions at

continuously grazed sites may favour the climbers *Aeshna sp.* larvae over *Sympetrum sp.* larvae (Buskirk and Sherman 1985). Larvae odonate populations reflect previous adult odonate populations via generational lag and therefore indicate at least minimal prior habitat suitability for adult odonates. *Aeshna sp.* adults are ‘flyers’ that hunt and defend territories on the wing, while *Sympetrum sp.* adults are ‘perchers’ and use riparian vegetation as a perch from which they hunt or defend (Corbet 1999). When chronic grazing decreases wetland vegetation structure (i.e. height; refer to Chapter 3) then there are fewer perches and therefore less suitable habitat for *Sympetrum sp.*, resulting in lower occurrence of oviposition by *Sympetrum sp.* at that wetland.

The Shannon diversity index is perhaps the most widely accepted diversity index since it incorporates evenness and is relatively independent of sample size (Wilhm and Dorris 1968). The seasonal pattern of diversity observed in larval odonates in Figure 2–9 (peaked in July) was also observed in adult odonates throughout the study area (Figure 2–10). Continuously grazed wetlands consistently had significantly lower larval odonate diversity, whereas the larval diversity between deferred and idle sites remains similar until the end of the summer. Once cattle were removed from deferred grazed pastures the larval odonate diversity in those wetlands was significantly higher than at wetlands with any other grazing treatment. These results are consistent with the Intermediate Disturbance Hypothesis (IDH) which suggests both extreme and trivial disturbance decrease diversity (Connell 1978). The chronically disturbed continuously grazed sites represent sufficient disturbance to suppress odonate diversity (Figure 2–10). Idle wetlands represent minimal disturbance according to the IDH since idle and ungrazed deferred sites (i.e. pre-July 15th) are similar until grazing ended in late July, at which time

odonate diversity temporarily increases at deferred sites. Similar patterns in odonate diversity with respect to habitat disturbance have been found in lotic habitats (Stewart & Samways 1998).

Significantly higher community similarity and lower diversity indices at continuously grazed sites suggest that these sites are inhabited by generalist species or adaptive species that can withstand and thrive under chronic disturbance. Taxonomic limitations restrict the identification of larval odonates to the species level however adult odonates are easily identified to species and therefore analysis of adult odonate data is needed to further support this hypothesis (see Chapter 3 and 4 this thesis).

2.7 Summary and Recommendations

Water chemistry data support that all twenty-seven study wetlands share similar hydrogeomorphology and differ predominantly according to grazing regime. No significant difference in water quality due cattle presence was detected between grazing treatments therefore I reject the hypothesis that nutrient inputs from cattle excrement measurably impacts wetland water quality (i.e. reject Hypothesis #1 and consequently invalidate Hypothesis #2). It is important to note that the cattle stocking rates are uncharacteristically low within the study area compared to other regions due to the small amount of precipitation and low carrying capacity of the pastures. Higher stocking rates may have a greater influence on wetland water quality. Furthermore, the opportunity exists for future research to study the impact of other non-point source agricultural pollutants such as pesticide or fertilizer run-off at wetlands greatly influenced by cropland.

Deferred grazing can be a more substantial disturbance than initially thought due to large number of cattle on these pastures. Fewer odonate larvae inhabit deferred grazed wetlands, presumably due to the acute disturbance resulting from hundreds of cattle focusing their grazing efforts on the littoral zone of one wetland (i.e. trampling and vegetation removal). Deferred grazing may be conceptualized as a brief bottleneck treatment. Although odonate larvae are more abundant at continuously grazed sites, the odonate communities at these wetlands are significantly less diverse than communities at either idle or deferred grazed wetlands. Further analysis involving vegetation and adult odonate fauna (identifiable to species rather than genera level; presented in Chapters 3 and 4) will help clarify the sensitivity of odonates to cattle grazing at prairie wetlands, and their subsequent suitability as bioindicators of this disturbance.

Grazing treatments were not contrived for this research but are current management regimes employed on the prairies of Alberta. For the purposes of this study, grazing was simplistically measured based on its duration (i.e. all summer, mid-summer, or not at all), but future research should be more sensitive to its complexity and the myriad opportunities associated with grazing impacts and timing. On-site livestock watering technology using renewable energy (i.e. solar powered water pumps) is available and offers wetland managers an option other than simply unlimited or prohibited cattle access. Grazing duration, timing (i.e. early, mid-, or late summer), frequency (i.e. annually or less frequent), and intensity (i.e. large vs. small herds) are all important variables that interact synergistically, that have not been addressed with respect to their impact on wetland flora and fauna.

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