The impacts of cattle grazing on stream ecosystems in Grasslands National Park of
Canada, Saskatchewan

by

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Abstract

    Cattle are responsible for the deterioration of aquatic and riparian ecosystems throughout the North American prairies. Marked preference for riparian areas has resulted in vegetation loss, stream bank destabilization, changes in sediment particle size, and increased nutrient loads in the streams. A grazing experiment in Grasslands National Park of Canada manipulated the density of cattle to represent a range of grazing intensities (from no grazing to very heavy grazing, 70% forage utilization). This experiment provided the opportunity to study how streams in the semi-arid mixed-grass prairie environment respond to a range of grazing pressure. Nine experimental pastures located on previously ungrazed land within the park boundary and four located within the adjacent community pastures were created, each subjected to a specified grazing treatment. Sampling occurred in the autumn from 2007 to 2009 and included the measurement of 33 physical, chemical, and biological habitat metrics and the characterization of the aquatic invertebrate community. Linear regressions were performed to determine if the habitat variables had a significant relationship to grazing intensity ($P < 0.05$). Of the habitat variables, two sediment particle size categories were significantly related to grazing intensity: per cent of fine gravel (4-8 mm diameter) ($P = 0.003$) and per cent of medium gravel (8-16 mm diameter) ($P = 0.007$). The only other habitat variable with a significant linear relationship to grazing intensity was the concentration of suspended carbon in the stream water ($P = 0.050$). Three invertebrate community metrics were focused on for their expected response to changes associated with cattle impacts:

per cent Chironomidae (%Chiron), per cent Ephemeroptera, Odonata, and Trichoptera (%EOT), and taxa richness. There was a significant non-linear relationship between %Chiron ($P = 0.005$) and grazing intensity, no linear or non-linear relationship between %EOT and grazing intensity, and a non-linear trend between richness and grazing intensity ($P = 0.083$). A Reference Condition Approach was used to test for the effects of grazing on the invertebrate community. Multiple regression was used to create a model predicting the invertebrate community from habitat metrics. Of the three community metrics, only richness resulted in a model with acceptable predictive ability. The predicted richness values for each test site were calculated and their residuals were determined and compared to the distribution of residuals observed in the reference sites. Using this technique, I determined that 73.3% of the sites subjected to grazing deviated significantly for the reference condition and were therefore deemed to be impacted. There was no significant relationship between the test site residuals and grazing intensity. The macroinvertebrate community in this semi-arid environment is already under a lot of stress, the addition of cattle to the environment, even at low intensities, pushed the community beyond the reference condition.

<u>Dedication</u>

To my parents, Kim and Gary, who have given me unwavering support for as long

as I can remember. Thank you for being there for me every step of the way.

I love you.

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Before European settlement, the grassland ecosystem of North America consisted of about 162 million ha (Samson and Knopf 1994), with the northern portion extending from the Canadian Rockies in Alberta to eastern Manitoba and the southern portion extending into the central, midwest, and southern United States and even further south into Mexico (Gauthier et al. 2003). Grasslands are characterized by periods of drought, fire, and the dominance of short, mixed, and tall grasses (Anderson 2006). Short-grass, mixed-grass, and tall-grass prairies receive approximately 260 to 375 mm, 375 to 625 mm, and 625 to 1200 cm of annual precipitation, respectively (Shorthouse and Larson 2010). Although streams and the adjacent riparian environments make up less than 5% of the grassland ecosystem (National Research Council 2002), they are an important source of food, water, and shelter for biota. With the arrival of European settlers, the spatial extent of grasslands diminished. These ecosystems have been converted into agricultural lands, roads, cities, and homesteads, resulting in fragmentation of the remaining grasslands (Gauthier et al. 2003; Shorthouse and Larson 2010). Less than 20% of native prairies remain in western Canada (Samson and Knopf 1994).

Disturbance is an important component that maintains the heterogeneity of the grassland flora. Historically, this disturbance was provided by bison, grazing and fire (Shorthouse and Larson 2010). These disturbance regimes have changed over time. Bison have been extirpated from the majority of the grasslands in North America and fires are suppressed (Shorthouse and Larson 2010). Fires are

important forces that shape grassland ecosystems by removing woody vegetation and minimizing their spread (Shorthouse 2010). The loss of these disturbances have led to concerns about the integrity of grassland ecosystems (Henderson 2006).

Grasslands National Park of Canada, located in southern Saskatchewan, was established to help preserve the native, mixed-grass prairie ecosystem. Within the park, the loss of disturbance has resulted in a more homogeneous landscape and an increase in the prevalence of invasive species (Henderson 2006). To assess the effectiveness of reintroducing disturbance into the landscape as a means to reduce invasive species, Parks Canada initiated an experiment in which a gradient of cattle grazing intensity was applied to large experimental pastures. The overall objective of the experiment was to determine how "grazing intensity affects heterogeneity in the multi-scale structure and function of mixed-grass prairie communities" (Henderson 2006). The study was established with a terrestrial focus on songbirds, grasshoppers, carabid beetles, and vegetation. As a smaller component, annual riparian assessments were also planned by Parks Canada staff following the methods outlined by Fitch et al. (2001). This was the major extent of their focus on the streams and riparian ecosystems until Dr. Cheryl Podemski was invited to join the study.

The design of the grazing experiment favoured the use of the reference condition approach (e.g., Bailey *et al.* 1998) to study the impacts of cattle on the stream environment. Streams in grasslands are complex ecosystems and cattle can impact them through in variety of ways. The reference condition approach integrates

changes in the biological, physical, and chemical environment in the creation of a model which captures changes to the invertebrate community, regardless of the route through which the streams are influenced.

Prairie streams have not been well studied, and although overall diversity in these systems may not that great (e.g., Stagliano and Whiles 2002), they are unique and important features of the landscape. My study contributes to the overall understanding of prairie streams and how these already stressed ecosystems respond to the addition of cattle stressors.

Streams in semi-arid environments

The Northern Great Plains in Canada extend from the foothills of the Canadian Rocky Mountains to the Manitoba/Saskatchewan border and reach north to 52°N, approximately the latitude of Saskatoon and Edmonton (Wilms and Jefferson 1993). The driest portion of the Northern Great Plains region is the mixed-grass prairie (Matthews 1988; Wilms and Jefferson 1993), which accounts for 6.5 million ha in Alberta and Saskatchewan (Wilms and Jefferson 1993). The terrestrial environment in this area has been heavily modified for agricultural use (Armour *et al.* 1991; Wilms and Jefferson 1993) and this development has impacted lentic and lotic ecosystems (Armour *et al.* 1991; Belsky *et al.* 1999).

Streams in the mixed-grass prairies have seasonal water flow. The streams swell in the spring from snowmelt, and then water levels decrease throughout the summer and into the autumn, which can lead to a cessation of surface flow in low order streams (Matthews 1988). The streams are low gradient (<2%) and follow a riffle-pool sequence (Rosgen and Silvey 1998). Riffles are shallower, with faster water and coarser substrate than pools, which are deeper, have slower moving water, and have finer substrate (Gordon *et al.* 1993). As the summer dry season progresses, water levels may decrease to the point where the riffles no longer have surface water and the stream becomes a series of isolated pools (Stanley *et al.* 1997). Sediment in pools of mixed-grass prairie streams often consists of fine glacial till, including clay and other small particles (Galat *et al.* 2005). Streams with clay

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bottoms have less subsurface flow than those with more porous substrates (e.g., sand, gravel), further isolating pools (Matthews 1988).

Discontinuity between pools can lead to high variability in the biophysicochemical characteristics among pools (Acuña *et al.* 2005). Nutrient concentrations in pools change as organic matter accumulates and decomposes in the pool instead of moving downstream with flow (Acuña *et al.* 2005), and as the relative contribution of groundwater to surface water changes. Decomposition of organic material and the loss of turbulent water flow can lead to reduced oxygen concentrations (Caruso 2002; Makowecki 1980; Stanley *et al.* 1997; Towns 1985). Nutrients and ions leach from decomposing organic matter and will also become more concentrated as water volume decreases, resulting in high seasonal and inter-pool variability (Caruso 2002).

Worldwide, salinity in freshwater streams ranges from less than 0.1 g L^{-1} to more than 10 g L^{-1}, with semi-arid and arid streams represented at the higher end of the spectrum (Allan and Castillo 2008c; Hynes 1970). Lower water volumes in semi-arid and arid environments contribute to higher ionic concentrations in these areas as compared to regions with more moist climates (Allan and Castillo 2008c). Salinity concentrations in small streams within a watershed have been demonstrated to show high variability (25-650 mg L^{-1}) (Allan and Castillo 2008c). Elevated salinity concentrations and variability between pools are stressors that require organisms to have a wide tolerance range in order to survive (Stanley *et al.* 1997).

As streams dry, the composition of the invertebrate community changes. Flowing water supports filter feeders that capture downstream moving suspended particles (e.g., members of the Trichoptera suborder Annulipalpa, including families Hydropsychidae and Polycentropodidae) and those that have external gills and high oxygen requirements (e.g., Ephemeridae, Heptageniidae, and most Plecoptera) (Brown and Brussock 1991; Merritt *et al.* 2008). These lotic taxa are primarily found in the erosional portions of the streams (i.e., riffles) (Merritt *et al.* 2008; Stanley *et al.* 1997), where species richness is highest just before flow ceases (Boulton and Lake 1992b). When riffles dry, this ecological niche is lost (Stanley *et al.* 1997) and the stream is transformed into a series of surface-flow isolated pools. Species richness peaks in the pools soon after flow stops because of coexisting riffle and pool taxa (Boulton and Lake 1992b). As the dry season continues, a greater proportion of the invertebrate community becomes comprised of lentic species, similar to those found in ponds (Acuña *et al.* 2005; Stanley *et al.* 1997). As streams dry and pools further decrease in volume, competition between species also increases, and in some cases, predator density may exceed prey density (Acuña *et al.* 2005; Boulton and Lake 1992b; Reice 1985; Stanley *et al.* 1997).

The changes in flow that occur in intermittent streams require invertebrates to have behavioural or physiological adaptations that are resistant to these changes or to be resilient in their ability to recolonize habitats when flow resumes (Doeg *et al.* 1989; Fritz and Dodds 2004; Stanley *et al.* 1994). Hynes (1970) identified six main faunal groups of intermittent streams: taxa that are able to survive high water

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temperatures and low oxygen concentrations; can burrow into the substrate; have a drought-resistant egg stage; are reintroduced to an area through recolonization from other water sources; are terrestrial and move into the stream mainly during the dry periods; and/or, are highly specialized to live in intermittent streams, including physiological adaptations (e.g., using mucus to seal off shells in snails) or behavioural adaptations (e.g., have an aerial life stage that coincides with the dry stage). Studies from Australia, Canada, and Spain support the faunal groupings provided by Hynes (1970) (Table 1). In an ecosystem where taxa already require specific adaptations to survive low flow or drought conditions, the addition of any anthropogenic disturbance has the potential to cause measurable changes in the aquatic invertebrate community.

Cattle and streams

Across most of North America, cattle have free access to riparian and stream environments (Trimble 1994). This has resulted in an estimated 50-80% of the riparian zones in western United States being adversely impacted (Armour *et al.* 1994; Belsky *et al.* 1999). With no federal legislation in place in Canada to protect riparian areas from cattle grazing (Trimble 1994), it is reasonable to assume that the streams and riparian areas of the Canadian mixed-grass prairie rangeland have been similarly affected (Wilms and Jefferson 1993). Riparian and aquatic areas are especially prone to changes caused by cattle activity because of the disproportionate time the animals spend in these areas and the multiple pathways through which cattle can impact the ecosystems.

Cattle distribution on rangelands

Mixed-grass prairies provide a variety of habitats for cattle to use, including drier uplands, wetter lowland valleys, and the aquatic and riparian areas within the valleys (Henderson 2006). Riparian areas are estimated to account for less than five per cent of the total land in the United States (National Research Council 2002), although estimates on provincial land in British Columbia reach about 10% (Banner and MacKenzie 1998). Cattle do not use riparian areas in proportion to their availability. Clary and Webster (1989) suggest that riparian area use is five to thirty times greater than expected based on area alone. Similarly, Howery *et al.* (1998) found that only three to eight per cent of the rangeland was riparian area, but cattle spent 48 to 75% of their time there. A number of factors contribute towards the disproportionate use of riparian areas, including the availability of water (Kauffman Pinchak *et al.* 1991; Platts 1991; Roath and Krueger 1982), forage (Ames 1977; Platts 1991), and land with a low slope (Bryant 1982; Hart *et al.* 1991b; Martin and Ward 1973; Pinchak *et al.* 1991).

Water attracts cattle to the riparian areas (Kauffman and Krueger 1984; Patten 1998; Pinchak *et al.* 1991; Platts 1991; Roath and Krueger 1982; Trimble and Mendel 1995). Cattle use water to fulfill their dietary requirements and to reduce elevated body temperatures (Bryant 1982; National Research Council 1996; Trimble and Mendel 1995). Growing steers weighing approximately 182-364 kg require 16.3-25.7 L day^{-1} when temperatures reach 10°C. This requirement more than doubles when temperatures reach 32°C (Winchester and Morris 1956). Water is

limited in semi-arid environments and cattle usually stay within one kilometre of water sources (Henderson 2006). Pinchak *et al.* (1991) found that cattle stayed even closer to water, typically remaining within 225 m of the source, although this distance increased during the grazing season as preferred forage decreased. This trend was also seen by Howery *et al.* (1996), but Platts (1991) reported that continuous growth in the riparian area limited animal dispersal. In general, it can be assumed that cattle will travel no further than they must from the water source to meet their dietary requirements (Martin and Ward 1973).

Cattle prefer the succulent vegetation found in riparian areas (Ames 1977; Platts 1991), resulting in greater use of this landscape (Armour *et al.* 1991; Bryant 1982; Chamberlain and Doverspike 2001; Kauffman and Krueger 1984; Patten 1998; Platts 1991; Roath and Krueger 1982). The higher soil moisture in riparian areas contributes to greater plant biomass and diversity than in surrounding uplands (Trimble and Mendel 1995). As an example, in a grazing area in eastern Oregon, the riparian area made up only 1.9% of the total area but produced 21% of the available forage in the pasture and 81% of the forage consumed by cattle (Roath and Krueger 1982).

Slope of land is another factor affecting cattle distribution on rangelands. Cattle typically prefer landscapes with less than a 35% slope (Bryant 1982; Hart *et al.* 1991b; Martin and Ward 1973; Pinchak *et al.* 1991). As slope increases, the relative amount of time cattle spend on that slope and the distance travelled up that slope

decreases (Mueggler 1965). This translates to less use of the uplands and greater use of lowland and riparian areas (Platts 1981, 1991).

The combination of available water and preferred vegetation in the riparian areas and low slopes in the lowlands results in the high use of riparian areas by cattle (Mosley 1983). This can result in a cascade of impacts on the riparian and aquatic ecosystems.

Analysis of pathways of effect

Cattle grazing has damaged aquatic ecosystems across North America (Belsky *et al.* 1999; Clary and Kinney 2002). Livestock can influence the water table, nutrient availability, and the suspended and deposited sediment loads in streams. They can affect riparian zones, altering the connection between terrestrial and aquatic environments and affecting energy flow within the stream. Changes to these components can cause a change in the aquatic invertebrate community.

<u>Water availability and riparian vegetation</u>

Depth of the water table is influenced by the amount of precipitation that infiltrates the soil (Williams 2006). In intermittent streams, the water table is naturally shallow (Williams 2006). Any changes to inputs to the water table can influence how long an intermittent stream will flow, and once surface flow ceases, how much groundwater will be available in the isolated pools (Williams 2006).

Livestock affect the rate of overland water flow and the ability of the soil to absorb and retain water by consuming and trampling vegetation, and by compacting soil (Li *et al.* 2000; 2006). Along tributaries of the Murrumbidgee River in

southeastern Australia, riparian biomass was an order of magnitude lower in areas

with grazing (Robertson and Rowling 2000). These findings are supported by

Scrimgeour and Kendall (2002), who determined that riparian biomass was three to

five times higher, and riparian vegetation cover was two times greater after two

years of grazing exclusion in the Cypress Hills grasslands plateau in southern

Alberta. Herbst *et al.* (2012) and McIver and McInnis (2007) also reported

significantly lower bank vegetation per cent cover in grazed than ungrazed sites.

Riparian vegetation helps to reduce overland flow velocity and volume in a number

of ways, the most obvious of which is that riparian plants act as a physical

obstruction to overland flow, slowing water movement (Dosskey *et al.* 2010; Fitch

and Adams 1998; Henley *et al.* 2000; Trimble and Mendel 1995). Slower runoff

(velocity) means there is more time for water to infiltrate the soil. Also, riparian

plant root systems help soils resist compaction and the greater interstitial pore

space facilitates water movement into the water table and then through subsurface

flow into the stream (Clary and Kinney 2002; Williams 2006). Increased soil

compaction can lead to decreased infiltration (Meek *et al.* 1992). In southern and

central Alberta, soil compaction was 25 to 50% higher in grazed sites compared to

ungrazed controls (Naeth *et al.* 1990). Tufekcioglu (2010) found a positive,

significant correlation between riparian soil bulk density and stocking rates in

southern Iowa. In a review of the effects of grazing on infiltration rates, Gifford and

Hawkins (1978) summarized that, compared to ungrazed conditions, light-medium

grazing and heavy grazing resulted in about a 25% and 50% decrease in infiltration

rates, respectively. The combined effects of riparian vegetation loss, alterations to overland flow, and soil compaction can have a profound impact on the ecosystem.

Vegetation loss causes a feedback loop in which topsoil is exposed to wind erosion, further decreasing the ability of the soil to support vegetation, resulting in further vegetation loss. This effect, combined with increased soil compaction and decreased permeability, can result in desertification (Huang *et al.* 2007; Li *et al.* 2000; 2006; Yong-Zhong *et al.* 2005). Geist and Lambin (2004) reported that extensive livestock grazing was a main causal factor in 74 of 132 cases of anthropogenically-induced desertification from around the world.

The energy balance

Stream energy flow is shaped by the amount of allochthonous input (heterotrophic) and autochthonous (autotrophic) production. The River Continuum Concept is generally used to describe the longitudinal gradient in streams with forested headwaters (Vannote *et al.* 1980). In prairie streams, this relationship is roughly reversed. Headwaters are located within prairie grasslands. They are generally shallow and have minimal shading, allowing for more autotrophic production and higher water temperatures than seen in areas further downstream (Dodds *et al.* 2004; Wiley *et al.* 1990). There is usually a larger proportion of autochthonous production than allochthonous inputs due to the absence of leaf material and high light availability (Webster and Benfield 1986; Webster and Meyer 1997; Wiley *et al.* 1990). However, Whiting *et al.* (2011) found that grass and grass roots represented 53% of coarse particulate organic matter (CPOM) in the grass

headwater reaches of Kings Creek, Kansas. In upper reaches dominated by autochthonous production, grazers typically outnumber shredders (Dodds *et al.* 2004).

As stream order increases, grass riparian zones transition to shrubs (Dodds *et al.* 2004; Whiting *et al.* 2011). Streams become wider, deeper, and cooler than the upstream reaches. Grazers and shredders become more equal in numbers as deeper water and more shading decreases available light and the emphasis on primary production while the addition of leaf litter provides more food for shredders (Dodds *et al.* 2004; Wiley *et al.* 1990).

In high order streams, trees become the dominant riparian vegetation and streams become even wider, deeper and cooler (Dodds *et al.* 2004; Whiting *et al.* 2011). Less light penetrates into the water because of shade from the forested banks, and deeper, more turbid, water (Wiley *et al.* 1990). In Kings Creek, the composition of coarse particulate organic matter (CPOM) in the higher order streams with forested stream banks was similar to that of forested headwater streams, although the abundance of CPOM was much lower (Whiting *et al.* 2011). This was attributed to low contribution from upland streams and floods which scour the stream and move larger material downstream (Whiting *et al.* 2011). Allochthonous inputs associated with the riparian vegetation tend to dominate the system (Dodds *et al.* 2004). Typically, the high abundance of CPOM in the form of leaf litter would result in a greater abundance of shredders than grazers (Dodds *et al.* 2004). However, Whiting *et al.* (2011) found that the macroinvertebrate

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functional groups did not shift between the grass, shrub, and forested riparian zone stream sections; collector-gatherers and predators were always the most abundant and scrapers were low, even in the grassy upland streams. Predators, including crayfish, may be responsible for this low abundance of scrappers, which, in Kings Creek, is primarily the snail *Physa* sp., a known prey of crayfish (Whiting *et al.* 2011).

Livestock can alter the energy balance in a stream by altering nutrient concentrations and the form of organic matter input (manure inputs versus riparian vegetation input). In turn, these changes can influence the invertebrate community composition and structure. Cattle use only about 30% of the energy available in mature forage ingested; the remainder is excreted as liquid and solid waste (National Research Council 1996). Agricultural engineers in the United States estimate that one beef cow excretes 13–22 kg of urine and eliminates 13–33 kg of faecal matter daily, with the precise value depending on the animals' size, production status (i.e., growth, lactation, or conception), air temperature, water consumption, daily forage consumption, and vegetation digestibility (Brenner and Mondok 1995; Nader *et al.* 1998; National Research Council 1996). Manure enters the aquatic environment by direct deposition and through movement with overland flow (Belsky *et al.* 1999; Bilotta *et al.* 2007; Braccia and Voshell 2006b; Caruso 2002; Godwin and Miner 1996; Scrimgeour and Kendall 2002). Manure provides additional nutrients to streams, is a source of food for aquatic invertebrates as particulate organic matter, and is a substrate for algae (del Rosario *et al.* 2002).

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After four weeks of manure enrichment in a coastal Californian stream, densities of different chiromid larvae were over 11 times (*Microtendipes* sp., *Micropsectra* sp., and *Parametriocnemus* sp.), 30 times (*Rheocricotopus* sp., *Thienemannimyia* sp., and *Brillia* sp.), and 80 times (*Polypedilum* sp.) greater than in the control site (del Rosario *et al.* 2002). Between 75-90% of the nitrogen consumed by cattle is excreted in waste (Azevedo and Stout 1974). Approximately half of all nitrogenous wastes are eliminated in faecal matter, with the remainder in urine (Azevedo and Stout 1974). The amount of nitrogen in solid waste varies depending on diet and type of cattle, but values range from five to 19 grams of nitrogen per kilogram of manure (Brenner and Mondok 1995; Chadwick *et al.* 2000; Malley *et al.* 2005; Sharpley and Moyer 2000). Organic nitrogen accounts for over 70% of the total nitrogen in manure (Malley *et al.* 2005), and can exceed 90% (Chadwick *et al.* 2000). The remainder is inorganic nitrogen, over 90% of which is ammonium (Chadwick *et al.* 2000). High concentrations of ammonia in water may harm invertebrate taxa living in the streams (Menzel 1983; Strand and Merritt 1999). Chronic exposure may damage the gills of some aquatic insect taxa (Strand and Merritt 1999), and at concentrations of 0.525 mg L^{-1} can cause death (Canadian Council of Ministers of the Environment 2010). Concentrations causing acute ammonia toxicity (1.2 to 8.0 mg/L) are possible, especially in arid regions where cattle spend a lot of time in stagnant streams (Strand and Merritt 1999).

Menzel (1983) suggested that livestock waste runoff may contain high levels of nitrogen, but reports on the effects of grazing on nitrogen concentrations in streams

have not been consistent. While Lemly (1982) found nitrate concentrations to be an order of magnitude higher in a stream adjacent to a livestock pasture (mean = 1.16 mg L^{-1}) compared to the ungrazed counterpart (mean = 0.13 g L^{-1}), Brenner and Mondok (1995) found nitrate concentrations were more strongly influenced by groundwater inputs than livestock. Scrimgeour and Kendall (2002) found no detectable effects of livestock grazing on total nitrogen, nitrate, and nitrite concentrations in water.

Estimates for the percentage of consumed phosphorus that is excreted in cattle waste vary from 22% (Nader *et al.* 1998; National Research Council 1996) to 85%, depending on food type and the growth stage of the cattle. Of the phosphorus eliminated, over 97% is in the faeces (Azevedo and Stout 1974), totalling approximately four grams of phosphorus for every kilogram of manure (Brenner and Mondok 1995; Malley *et al.* 2005; Sharpley and Moyer 2000). Approximately half of this (2 g) is dissolved inorganic phosphorus and one eighth (0.5 g) is dissolved organic phosphorus (Sharpley and Moyer 2000). Phosphorus forms complexes with soil particles and is transported with overland flow into streams (Henley *et al.* 2000; Menzel 1983; Nader *et al.* 1998).

The results of studies relating cattle grazing to phosphorus concentrations in streams are ambiguous. In Cullowhee Creek (North Carolina), Lemly (1982) measured higher phosphate concentrations in the portion of the stream accessed by cattle than in cattle-absent areas. In Samsonville Brook (Vermont), Meals and Hopkins (2002) restricted cattle access to streams and protected a two to five metre

wide riparian buffer zone. They compared the stream water total phosphorus concentration and discharge volume over three years prior to riparian protection with the two years after treatment. Although they observed a 21% decrease in total phosphorus concentrations, there was also significantly higher stream discharge resulting in significantly higher export of total phosphorus after treatment. This suggests that increased discharge and not cattle removal was responsible for reduced total phosphorus concentrations. The importance of water flow is supported by Scrimgeour and Kendall (2002), who determined there was no discernible difference in stream phosphorus concentrations between grazed and ungrazed sites in Cypress Hills, Alberta, and suggested that low rainfall and runoff during the study period was responsible. Periods of high precipitation can greatly increase the nutrient loading of water bodies (Chapra and Dolan 2012).

Precipitation and riparian buffers can explain why nitrogen and phosphorus concentrations are not always elevated in streams influenced by cattle grazing. Under conditions where precipitation is low, overland flow, and therefore nutrient movement, is limited (Caruso 2002; Scrimgeour and Kendall 2002). There have been many studies on the nutrient retention capability of riparian zones (e.g., Beck and Young 1975; Daniels and Gilliam 1996; Dillaha *et al.* 1988; 1989; Gharabaghi *et al.* 2000; Line *et al.* 2000; Magette *et al.* 1989; McKergow *et al.* 2003; Young *et al.* 1980). Although modified by zone width, slope, and vegetation type, riparian zones have been proven to be excellent at capturing nutrients from overland flow. Decreases in nitrogen concentrations range from 40-91% for total nitrogen (Dillaha

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et al. 1988; 1989; Kuusemets *et al.* 2000; Young *et al.* 1980), 50-93% for total

Kjeldahl nitrogen (Daniels and Gilliam 1996; Dillaha *et al.* 1989; Lim *et al.* 1998; Line

et al. 2000), 9-89% for ammonium (Dillaha *et al.* 1988; 1989; Young *et al.* 1980), and

2-78% for nitrate (Daniels and Gilliam 1996; Dillaha *et al.* 1989). Phosphorus

concentrations can also be reduced by riparian buffers, with decreases of 27-93%

reported (Daniels and Gilliam 1996; Dillaha *et al.* 1988; 1989; Kuusemets *et al.*

2000; Lim *et al.* 1998; Line *et al.* 2000; Magette *et al.* 1989; Young *et al.* 1980). While

it is clear that cattle wastes have the potential to add a lot of nutrients to the

riparian and aquatic environment, climate and riparian characteristics will strongly

influence how much of those nutrients reach the stream.

Riparian vegetation loss resulting from cattle grazing (e.g., Herbst *et al.* 2012;

McIver and McInnis 2007; Robertson and Rowling 2000; Scrimgeour and Kendall

2002), means the loss of an important natural source of organic matter input to

streams (Makowecki 1980; Robertson and Rowling 2000). Leaf litter, twigs, and

other plant material from riparian vegetation can provide over 90% of the total

organic matter input required to maintain headwater stream invertebrate

communities (Cummins and Spengler 1978; Fisher and Likens 1973). Grasses and

grassroots may contribute a large amount of organic matter to tallgrass prairie

streams. Many aquatic invertebrates depend on organic matter for food and

decreases in the quantity of this material could cause changes in invertebrate

community biomass and/or composition (Makowecki 1980; Platts 1991).

Livestock can also influence the amount of vegetation in a stream. Platts (1981) reported proportionally lower macrophyte coverage in heavily grazed stream sites compared to lightly grazed sites. These findings were supported by Scrimgeour and Kendall (2002, 2003), who made visual estimates of instream vegetation coverage and reported a 200% increase after two years of livestock exclusion. Taxa richness in streams has been shown to be increased by the presence of macrophytes (Gregg and Rose 1985; Shupryt and Stelzer 2009).

Sediment additions and the response of biota

Increased sediment transport into streams is thought to be one of the most influential impacts of cattle grazing on the stream environment (Armour *et al.* 1991; Lemly 1982; Sidle and Sharma 1996). Braccia and Voshell (2006b) named sedimentation as the factor having the greatest effect on aquatic invertebrate species density and diversity, an idea supported by Lemly (1982) and Menzel (1983). Cattle increase sediment transport into streams by reducing riparian vegetation and by modifying bank structure.

Stream banks are a major contributor of sediment to streams, but the amount of sediment added is highly influenced by the presence of riparian vegetation. Willett (2010) reported that bank sediment accounted for 47-71% of in-stream sediment in two Missouri Creeks (Willett 2010). Riparian vegetation above-ground biomass slows overland flow and roots help to stabilize stream banks. Vegetated banks, which were 16-18% root by volume, showed significantly lower erosion rates than non-vegetated portions of the Alexandra River (Alberta) (Smith 1976). Non-

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vegetated banks on streams in southern British Columbia were five times more likely to undergo detectable erosion than vegetated banks (Beeson and Doyle 1995).

Riparian buffers remove a large percentage of sediment from overland runoff, resulting in decreased sediment loads in streams. Buffer widths of three to six meters are estimated to remove from 50-85% of the sediment in runoff (Dillaha *et al.* 1988; 1989; Lim *et al.* 1998; Magette *et al.* 1989; Robinson *et al.* 1996; Young *et al.* 1980). Although wider buffers can remove more sediment (Dillaha *et al.* 1988; 1989; Gharabaghi *et al.* 2000; Line *et al.* 2000), Gharabaghi *et al.* (2000) determined that effectiveness plateaued after 10 m, with about 90% of all sediment particle sizes being captured. If cattle decrease the amount of riparian vegetation, the sediment retention capacity of this area may be reduced.

Grazing has been shown to cause increased stream bank erosion. Trimble (1994) reported erosion rates three to six times greater in areas grazed by cattle than in ungrazed control sites on Jenkins Creek, Tennessee. This was attributed to ramp formation, where cattle re-use the same access points, wearing down a path from the top of the stream bank to the water. These ramps lack vegetation and act to increase overland flow velocity by focusing water flow (Trimble 1994). Tufekcioglu (2010) also found significantly higher erosion rates at grazed pastures sites compared to ungrazed sites.

Livestock can increase the frequency of bank failure (Armour *et al.* 1991; Kauffman and Krueger 1984; Laubel *et al.* 1999; Makowecki 1980; Robertson and Rowling 2000), changing bank and channel morphology and increasing sediment

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load to streams. Bank failure (i.e., collapse or sloughing) occurs when gravitational

force exceeds the shear strength of the bank (Easterbrook 1999; Pizzuto 2008;

Simon *et al.* 1999). The balance between these two opposing forces is influenced by

soil characteristics. Smaller particles (i.e., clay and silt) have higher cohesive forces

than larger particles (i.e., sand and gravel) because of their greater surface area to

volume ratio, making fine sediment more resistant to bank erosion (Pizzuto 2008).

High soil moisture reduces the cohesive forces of soil particles, decreasing the force

required to overcome the shear strength (Fox *et al.* 2007; Pizzuto 2008; Richards

1982; Rinaldi and Casagli 1999). This results in accelerated collapse when the

stream banks are moist, making them most vulnerable after snowmelt or heavy

rains (Hooke 1979; Platts 1991; Simon *et al.* 1999; Trimble and Mendel 1995).

Gravitational forces can be increased by the added weight of livestock, with

undercut banks especially prone to collapse (Richards 1982). McInnis and McIver

(2001) observed a significant decrease in bank stability in grazed pastures after just

42 days during the summer in Oregon and Platts (1981) observed that undercut

banks were almost completely eliminated in the heavily grazed portion of a stream

tributary in Idaho. Over longer periods, increased frequency of bank collapse results

in changes to channel morphology; heavy grazing regimes in New Zealand resulted

in channel widths up to four times greater than lightly grazed or ungrazed areas

(Platts 1981).

    The fate of slumped bank material, whether it is transported downstream or

remains as part of the channel bottom, is determined by flow and particle size

(Pizzuto 2008; Simon *et al.* 1999). Sediment can be transported either as suspended sediment or as part of bedflow. The velocity required to move water downstream is dependent on particle size and cohesiveness of the sediment. Suspended sediments usually consist of silt and clay particles, ranging from 2-60 μm in diameter, although faster velocities can keep larger particles suspended (Waters 1995). Generally, the larger the particle size, the higher the water velocity required to move it downstream (Simon *et al.* 1999) but this only holds true for non-cohesive particles (e.g., sand and gravel). Cohesive particles (e.g., clay and silt) do not typically act as individual grains; aggregates are formed which act as larger particles (Pizzuto 2008; Richards 1982). This means that even though a bank slump may be made of fine materials, erosion and downstream transport of it may require equal or greater water flow velocity than what is required for non-cohesive particles (Pizzuto 2008). The variability in bank materials and water velocity makes the residence time of collapsed segments variable. In streams where flow has ceased, the collapsed segments may remain intact until flow returns (Simon *et al.* 1999).

Suspended sediment load can decrease the primary productivity of a stream by decreasing visible light penetration (Armour *et al.* 1991; Ellis 1936; Hynes 1973). Lloyd *et al.* (1987) determined that an increase in turbidity of only 5 NTU decreased primary productivity by 3-13% and an increase of 25 NTU resulted in a 50% decrease. A decrease in primary productivity of a stream can cause changes to the invertebrate community, through the loss of a food source and habitat (Henley *et al.* 2000).

Suspended sediment may directly impact invertebrate populations by reducing the effectiveness of filter-feeding mechanisms (Lemly 1982) and by settling on gills, causing asphyxiation in taxa with high oxygen demands (e.g., Plecoptera, some Ephemeroptera) (Lemly 1982). The majority of filter feeders, including trichopteran nymphs, ingest fine particulate organic matter (FPOM) much smaller than 1 mm in diameter that is collected on filtering mechanisms (e.g., Trichoptera nymph nets) (Merritt *et al.* 2008; Wallace and Merritt 1980). Many filter-feeding invertebrates are unable to selectively filter particles in the water and are, therefore, unable to avoid capturing inorganic particles in favour of organic matter (Broekhuizen and Miller 2001). This dilutes the concentration of FPOM that is available to and consumed by filterers (Broekhuizen and Miller 2001). This, as well as the clogging of filtering apparatuses by inorganic sediment (Gard 2002; Shaw and Richardson 2001), can contribute to decreased filtering efficiency, lower rates of assimilation (Euliss and Mushet 1999; Hynes 1970; Lemly 1982; Newcombe and Macdonald 1991), and/or population declines (Hynes 1970; Rabeni *et al.* 2005).

Abrasion by suspended sediment particles may indirectly affect invertebrates by damaging food sources such as algal cells (Newcombe and Macdonald 1991; Shaver *et al.* 1997), increasing algal biomass removal by scour (Francoeur and Biggs 2006), or by accelerating leaf litter decomposition (Welsh 2007). Although direct impacts of abrasion caused by suspended sediments have been suggested as a reason for changes to aquatic invertebrate populations, there is little substantial support in the literature for this idea (Waters 1995).

Sediment deposition can influence invertebrate populations by changing the physical and chemical characteristics of the substratum. Spaces on and around larger sediment particles (e.g., pebbles, cobbles) are used as habitat by Ephemeroptera, Odonata, and Trichoptera nymphs (Waters 1995). As fine particles settle onto the streambed, they infiltrate, filling interstitial spaces and changing the characteristics of the substrate (Beschta and Jackson 1979). This change in the sediment characteristics of a stream can lead to changes to the invertebrate community (Hynes 1973; Lenat *et al.* 1981; Menzel 1983). Herbst *et al.* (2012) and Braccia and Voshell (2006a) measured significantly higher proportions of fine particle sizes at grazed sites than ungrazed sites. With enough sedimentation, surface water can become isolated from groundwater, limiting the chemical exchange between surface water and groundwater in the hyporheic zone (Chen *et al.* 2013; Hancock 2002; Henley *et al.* 2000). Less chemical exchange in this zone can cause decreased oxygen concentrations (Henley *et al.* 2000), further affecting invertebrate populations.

Reports of the relationship between proportion of fine sediment and invertebrate communities are varied. An increase of 12-17% of fine sediment in the interstitial spaces of a New Zealand streambed resulted in a 16-40% decrease in invertebrate abundance (Ryder 1989). Rabeni *et al.* (2005) found that the densities of gatherers, predators, scrapers, and filter-feeders decreased with increasing sediment deposition whereas there was no significant change in shredder richness. In

contrast, when 0.2 mm sand was added to experimental stream channels to increase fine sediment bottom cover from 2.5% to over 81%, Piggott *et al.* (2012) observed an increase in total invertebrate abundance. They also reported decreases in EPT abundance and the mayfly nymph *Delatidium*, and increases in *Hydora* (an elmid beetle), nematodes, and Tanypodinae larvae. In contrast, Cover *et al.* (2006) found no significant relationship between taxa richness, total abundance, EPT richness, and EPT abundance with the proportion of fine sediment bottom cover. The proportion of Chironomidae larvae that belonged to the Chironominae subfamily was significantly negatively correlated with the amount of fine material whereas the proportion of Orthocladiinae larvae was significantly positively correlated with the amount of fine material. In Spain, there were more multivoltine taxa in areas with more fine sediment (Buendia *et al.* 2013). Von Bertrab *et al.* (2013) determined that while the deposition of fine sediment is important, the nutrient composition of that sediment is even more important in determining the invertebrate community. These studies demonstrate that the responses of stream invertebrates to sedimentation are not consistent. When sedimentation in a stream is increased from anthropogenic activities, the response in the invertebrate community will depend on the natural state of the system. Streams with naturally coarse substrate, such as portions of the Isábena River in Spain (Buendia *et al.* 2013) and Elk Creek and Little North Fork Salmon River in northern California (Cover *et al.* 2006), may see a greater shift in the invertebrate community than streams with a naturally higher proportion of finer sediment.

Invertebrate drift may be influenced by suspended sediment and sedimentation. Ciborowski *et al.* (1977) observed that the drift density of the mayfly *Ephemerella subvaria* McDunnough 1931 increased with increased water turbidity and sediment deposition. Fairchild *et al.* (1987) conducted an experiment in experimental streams and observed increased drift, mainly by the amphipod crustacean *Hyalella azteca* Saussure 1858, immediately after the first addition of sediment. Sand (0.25–0.5 mm) was added to two replicate streams in sufficient volume to correspond to about 25-35% bed cover. This addition resulted in a 45% increase in drift density, and a 30% decrease in total benthic invertebrate density, with *Baetis* sp. decreasing by 63% (Ryder 1989). Invertebrates may drift because of sediment coating their food or habitat (Ciborowski *et al.* 1977; Waters 1995) or because increased turbidity decreases light penetration. A decrease in light may signal the start of nocturnal drifting, a natural periodicity seen in some lotic invertebrates (Waters 1995).

Grazing and aquatic invertebrate communities

The reports of the influence of elevated sedimentation on invertebrate community are inconsistent. Of five studies that examined the influence of grazing on aquatic macroinvertebrates, four focused on comparing the communities between grazed sites and ungrazed or more lightly grazed sites (Herbst *et al.* 2012; McIver and McInnis 2007; Quinn *et al.* 1992; Scrimgeour and Kendall 2003) and one (Braccia and Voshell 2006b; 2007) investigated the response along a range of grazing intensities. Total taxa richness was significantly lower at grazed sites in the study by Herbst *et al.* (2012), but there was no significant difference in the three

other studies (McIver and McInnis 2007; Quinn *et al.* 1992; Scrimgeour and Kendall 2003). Density (or abundance) was significantly lower at grazed sites in one study (McIver and McInnis 2007) but showed no significant difference in the other three studies (Herbst *et al.* 2012; Quinn *et al.* 1992; Scrimgeour and Kendall 2003). Total invertebrate biomass was significantly higher at grazed sites in the study by Scrimgeour and Kendall (2003), but this metric was not measured in Herbst *et al.* (2012), McIver and McInnis (2007), and Quinn *et al.* (1992). The sensitivity of Ephemeroptera, Plecoptera, and Trichoptera (EPT) and the tolerance of chironomids to sedimentation resulted in these group receiving particular attention in several studies. Richness of the EPTs was significantly reduced in the grazing sites of the Herbst *et al.* (2012) study but this metric was not mentioned specifically in the other studies. EPT density and biomass were not statistically different between the grazed and ungrazed sites in the Scrimgeour and Kendall (2003) study, and although McIver and McInnis (2007) reported a "strong trend" of lower EPT density in the grazed sites, it was not statistically significant. The relative density (as % of total density) of EPT nymphs decreased about 30% in the study by Quinn *et al.* (1992), but this may be attributed to an increase in the density of dipteran larvae. Quinn *et al.* (1992) found that densities of Chironomidae larvae were significantly higher at the heavy grazing site whereas Chironomidae larval density and biomass were not significantly different between grazed and ungrazed sites in the study by Scrimgeour and Kendall (2003). Although McIver and McInnis (2007) did observe significantly lower densities of two chironomid larvae species at grazed sites,

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richness and abundance at the family level were not specifically addressed. There were few other community metrics reported. Quinn *et al.* (1992) determined that Naididae were significantly less abundant at the heavy grazing sites and McIver and McInnis (2007) found a significant decrease in abundance of Coleoptera at grazed sites. The inconsistent results when comparing grazed sites with ungrazed or lightly grazed sites using these various metrics underlines the complicated nature of streams.

Braccia and Voshell (2006b, 2007) looked at the influence of five different grazing intensities (ranging from 0 to 2.85 cattle per hectare) on stream invertebrate communities in the Little River drainage basin, Virginia. About 50% of the species found in the streams either decreased in abundance or disappeared along the grazing gradient (Braccia and Voshell 2006b), but relationships were not always linear. Total richness, Simpson's diversity, per cent collector-filterers, per cent scrapers, and per cent clingers all showed significant non-linear relationships for each of the two years of fall sampling, peaking around the intermediate grazing intensities (Braccia and Voshell 2007). Per cent Plecoptera was significantly negatively correlated with grazing intensity and per cent burrowers were significantly positively correlated with grazing intensity in the two fall sampling periods. Other metrics, including per cent Coleoptera, per cent collector-filterers, per cent shredders, and per cent crawlers, had different responses to grazing intensity between the two fall sampling periods (Braccia and Voshell 2007). These differences were attributed by the authors to lower flow in one of the sampling

years (Braccia and Voshell 2007). In summary, a number of community metrics were significantly related to grazing density. These metrics reflected changes occurring to the stream environment, such as changes to food sources and available habitat types (Braccia and Voshell 2007). The presence of non-linear relationships with some variables, including species richness and Simpson's diversity, suggests that stream community heterogeneity can be increased by grazing, but there exists a threshold that if exceeded, will result in the loss of taxa. Non-linear relationships may also describe the varying responses of community metrics seen in the literature; without a gradient, the observation may fall on any portion of the non-linear relationship.

<u>Study overview</u>

Unrestricted access to streams in the United States has resulted in damage to over 80% of streams in the western United States (Belsky *et al.* 1999). With a similar lack of regulations for restricting stream access, Canada's waterways are similarly susceptible to habitat degradation. This is especially true in the prairies, where over 70% of Canada's cattle are found (Statistics Canada: Agriculture Division 2010).

Ranchers can employ several management practices to protect stream banks from cattle. The most successful means of keeping cattle out of riparian areas is to fence them out (Godwin and Miner 1996). Unfortunately, fences are expensive to construct and maintain and they separate livestock from valuable water and forage resources (Bryant 1982). Watering troughs (Ganskopp 2001; Godwin and Miner 1996; McInnis and McIver 2001) and salt blocks (Bryant 1982; Ganskopp 2001; McInnis and McIver 2001; Nader *et al.* 1998) have been used to draw cattle away from, and reduce the time spent in the riparian zone, with mixed results. Another method to reduce impacts to the riparian areas is to prohibit cattle access during the spring, providing time required for stream banks to become more robust, with drier soil and stabilizing vegetation (Marlow *et al.* 1987). An alternative potential approach to decrease impacts in the riparian and aquatic ecosystem is to identify the threshold of grazing intensity below which cattle can access the streams without causing a significant change.

A large scale experiment in Grasslands National Park, Saskatchewan provided the opportunity to determine if there is a density of cattle that could be allowed access to prairie streams without causing significant changes to the ecosystem as measured by changes to the invertebrate community. Nine enclosures were constructed within the park to which a gradient of grazing intensities was applied. In combination with four long-term grazing pastures, these sites were used to assess the impacts cattle have on the aquatic ecosystems.

Grasslands National Park of Canada

Grasslands National Park of Canada (GNPC) encompasses 900 square kilometers of southern Saskatchewan along the Canada-United States border (Parks Canada 2013). The park is divided into an East and a West Block. In 2008, a large scale experiment manipulating the intensity of cattle grazing was initiated in a 100 square kilometre portion of the East Block, designated the Biodiversity and Grazing Management Area (BAGMA) (Henderson 2006) (Figure 1). This experiment also used community pasture land just outside the park boundary that had been subjected to grazing for over 20 years (Henderson 2006).

The East Block of GNPC is semi-arid, northern mixed-grass prairie, as characterized by a mean annual precipitation of 350 mm and dominant upland grass vegetation (Henderson 2006). The riparian plant communities are composed primarily of three main shrub species: *Symphoricarpos occidentalis* Hook. (western snowberry), *Rosa acicularis* Lindl. (prairie rose), and *Artemisia cana* Pursh (sagebrush). Herbaceous vegetation includes *Solidago canadensis* L. (Canada

goldenrod), *Glycyrrhiza lepidota* Pursh (wild licorice), and *Poa compressa* L. (blue

grass) (Henderson 2006). Alluvial deposits and glacial till characterize the soil, with

saline, unconsolidated, fine-grained regosols, dense saline clayey solonetz, and

organic-rich chernozems (Henderson 2006; International Union of Soil Sciences

Working Group World Reference Base 2006).

Land purchased for GNPC began in 1987 and the strategy upon purchase was

complete grazing exclusion (Henderson 2006). Between the 1930s and their

acquisition by Parks Canada, lands that make up the BAGMA were inconsistently

grazed by cattle in the summer with a few exceptions of year-round grazing

(Henderson 2006). In the early 1980s, infrared aerial photographs were used to

assess the extensiveness of grazing in GNPC. When compared to pastures located

nearby, the lands that now comprise GNPC were determined to be lightly grazed.

This land was never used for growing crops (Henderson 2006).

Grazing treatments

The BAGMA project was the largest-scale grazing experiment conducted to date

(Henderson 2006). The BAGMA experimental design consisted of seven different

levels of grazing distributed over 13 pastures (Figure 1). Each pasture was

approximately 300 ha in size and contained roughly equal proportions of riparian,

valley, and upland landscapes. The nominal levels of grazing utilization for the study

were approximately 0%, 20%, 33%, 45%, 50%, 57%, and 70%, where the

percentages are the proportions of usable forage to be consumed by cattle within

the grazing season (Table 2) (Henderson 2006). A maximum of 70% utilization was

chosen to allow for annual variation in vegetation growth. If too much vegetation was lost during the grazing season, cattle would have to be removed from those experimental pastures earlier than planned, resulting in inconsistencies in grazing duration between the different experimental pastures (Henderson 2006). Replicate pastures were not evenly distributed among the treatments (Table 3). While the experimental pastures within the park boundaries had cattle placed in them for the first time in 2008, all of the 50% grazing intensity pastures were community pastures and represented a condition of long-term grazing in the mixed grass prairies.

Yearling steers were grazed in the experimental pastures from late spring to early autumn, following a conventional grazing regime as discussed in the project proposal by Henderson (2006). In the summer of 2007, cattle were present only in the long-term community pastures. Cattle were placed in the experimental enclosures for the first time in early June of 2008 and were removed from the pastures in late September. In 2009, cattle were moved into the pastures in mid-May and removed in mid-September.

The realized grazing intensity for each pasture varied between years. Table 2 provides a list of estimated grazing intensity in each pasture, measured as animal unit months per hectare (AUMs/ha), where one AUM is equivalent to the forage required by one 455 kg cow and her suckling calf per month (Alberta Agriculture and Food 2007). One yearling steer is an estimated 0.67 AUMs (Alberta Agriculture and Rural Development 1998).

Historically, heterogeneity within the prairies was maintained by fire and bison disturbance (Hamilton 2007; Hulbert 1986; Knapp *et al.* 1999). In Kings Creek at the KPBS, fire only temporarily reduced total nitrogen and total phosphorus concentrations and had no significant impact on TSS concentrations. After burning, above-ground biomass continued to increase until the sixth year (Hulbert 1986). Regrowth of grasses occurs within the first growing season post-burn, and shrubs reappear within the next few grazing seasons (Gibson and Hulbert 1987; Hulbert 1986; Larson *et al.* 2013). Bison use the landscape differently than cattle, spending less time close to water (Allred *et al.* 2011; England and DeVos 1969; Fortin *et al.* 2003; Larson *et al.* 2013). Bison spent less than 6% of their time within 10 m of the stream and only influenced TSS when they were standing directly in the water (Larson *et al.* 2013). The removal of these disturbances in GNPC has contributed to decreased heterogeneity across the terrestrial landscape (Henderson 2006).

Within the BAGMA, researchers lead by Dr. Nicola Kopera (University of Manitoba, Winnipeg, MB) studied the terrestrial landscape. This included studying the fauna, including determining the richness and relative abundance of songbirds, grasshoppers (Orthoptera: Acrididae), and carabid beetles (Coleoptera: Carabidae). They also studied the vegetation, focusing on plant community richness, vascular plant species relative abundance, litter cover, vegetation height, and above-ground vegetation biomass(Henderson 2006).

<u>Study objectives and hypotheses</u>

The main objective of my study was to determine the relationship between the intensity of grazing by cattle and aquatic habitat quality using benthic macroinvertebrates as bioindicators. I wanted to determine if there was a threshold for grazing below which cattle can be allowed access to streams without causing significant changes in the ecosystem. To accomplish this objective, I sampled streams along a gradient of grazing intensity that included the experimental pastures, long-term community pastures and control sites located away from anthropogenic disturbances, including grazing. Sites were sampled for water chemistry, physical sediment properties, stream and stream bank morphology, and the invertebrate community in the autumn of 2007, 2008, and 2009.

Regression analysis was used to assess the relationships of different habitat and invertebrate metrics with grazing intensity. My expectations were as follows:

1. Increased amounts of faecal waste and urine would result in higher stream nitrogen and phosphorus loading as grazing intensity increased. At the time of sampling there was no discharge and, therefore, concentrations were measured and not loading.

2. Consumption and trampling of streambank vegetation by cattle would result in an increased proportion of streambank exposed in higher grazing intensities.

3. Increased bank erosion and less riparian vegetation would combine to result in increased sediment transport into the stream, which would be measured as increased total suspended solids as grazing intensity increased.

4. Increased sediment transport into the low gradient streams of GNPC would result in increased proportion of fine particle sizes in the stream sediments as grazing intensity increased.

I used simple regressions and the multiple regression variant of the reference condition approach (Bailey *et al.* 1998) to describe changes in the stream ecosystem as indicated through the macroinvertebrate community. Three community metrics were used: per cent Chironomidae (%Chiron), per cent of the invertebrate community that consisted of Ephemeroptera, Odonata, and Trichoptera (%EOT), and taxa richness. The %Chiron and %EOT metrics were expected to respond in opposite ways. Larvae of many species of chironomids are adapted to environments high in organic matter and low in dissolved oxygen (Rosenberg and Resh 1993). Increased density can occur when sedimentation and organic enrichment increases (Braccia and Voshell 2006b; del Rosario *et al.* 2002; Lemly 1982), so I expected an increase in the proportion of chironomid larvae in the invertebrate community as grazing intensity increased. Ephemeroptera, Odonata, and Trichoptera nymphs are sensitive to changes in their environment, including sedimentation, (Braccia and Voshell 2006b; Lemly 1982) and were, therefore, expected to decrease with increased cattle intensity. Richness can show curvilinear response to disturbance (Pearson and Rosenberg 1978). How the invertebrate community reacts to new

stressors in the ecosystem will be a function of how much stress the system is already under. Elevated richness can occur due to the coexistence of taxa at the edge of their tolerance to the disturbance with colonizing species that are adapted to survive under those conditions (Pearson and Rosenberg 1978). When the tolerance ranges for many taxa are exceeded, only the disturbance-tolerant taxa remain (Lemly 1982; Pearson and Rosenberg 1978). Along the curvilinear response, the peak in richness has passed and is in a decline. To summarize, I expected to see an increased proportion of Chironomidae larvae, and a decreased proportion of Trichoptera, Odonata, and Ephemeroptera nymphs with increasing grazing intensity. I also expected to see a non-linear relationship between taxa richness and the grazing gradient.

The reference condition approach

The history of the reference condition approach (RCA) is well summarized in Bowman and Somers (2005) and Bailey *et al.* (2004). Initially, the use of reference distributions represented a departure from more conventional means of hypothesis testing by comparing a test site community metric to the actual distribution of the same metric as measured at a large number of reference (unimpacted) sites rather than to a theoretical distribution. This method has since evolved to use multiple regression or multivariate statistical models that incorporate habitat characteristics to describe the reference condition (Bailey *et al.* 1998; 2004).

The first large scale application of the RCA began in the 1980s in the United Kingdom, with the aim of using habitat variables to predict the invertebrate

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community in unpolluted waters (Bailey *et al.* 2004; Bowman and Somers 2005). This evolved into a national biomonitoring program, termed the River InVertebrate Prediction and Classification System (RIVPACS), which uses a multivariate approach to provide site-specific predictions of the macroinvertebrate community in the reference condition. These predictions are compared to the observed communities at test sites in order to establish the status of the test site (Wright *et al.* 1984; 2000).

A similar national biomonitoring program was established in Australia in the 1990s (Australian River Assessment System or AusRivAS) (Bailey *et al.* 2004; Bowman and Somers 2005). In Canada, RIVPACS was used as a basis for a model, the BEnthic Assessment of SedimenT (BEAST), to assess the status of the near-shore environment in the Great Lakes in the 1990s (Reynoldson *et al.* 1995). The BEAST methodology was later applied to streams in the Fraser River Valley in British Columbia (Reynoldson *et al.* 1997; 2001). The development of BEAST in Canada led to the creation of the Canadian Aquatic Biomonitoring Network (CABIN). Maintained by Environment Canada, CABIN provides the protocols to implement a RCA study and is home to a national database which is added to by researchers (Environment Canada).

Bailey *et al.* (1998) simplified the RCA to use multiple regression, incorporating the descriptive capability associated with using habitat variables but removing the need to construct and use complicated multivariate models. This method involved creating a predictive multiple regression model that used habitat variables from the reference sites to explain a community metric at those same sites. The model was

then used to predict the expected value of the metric at test sites based on the habitat conditions observed at those sites. Test site residual values were calculated from the observed and expected values and compared to the residuals from the reference sites. If they fell outside a predetermined pass/fail boundary, then the sites were considered impacted. I used this version of the RCA to determine whether streams were being influenced by cattle, and to see if the magnitude of the residuals and, thus, the severity of effect was related to grazing intensity.

<u>Materials and methods</u>

Sampling sites

There are three main creeks within my study area: Horse Creek, Wetherall Creek, and Dunn Creek. Horse Creek and Wetherall Creek are the two main creeks located within the BAGMA. Both are third order streams in the northern portion of the BAGMA and become fourth order streams in the southern portion. They feed into Rock Creek, a perennial stream found in the most southern extent of GNPC. Dunn Creek is an additional stream (fourth order) used for the study that is outside the GNPC boundaries (Figure 1). The study streams are intermittent streams, and in most years, surface flow ceases by midsummer, resulting in a series of isolated pools. This drainage basin belongs to the larger Missouri River Drainage Basin, and water flows south to the Gulf of Mexico (Galat *et al.* 2005; Henderson 2006).

The number of reference and test sites that were sampled varied each year (Table 4, Figure 2, and Figure 3). In total, 85 sites were sampled; 52 of the sites were unimpacted reference sites at the time of sampling and the remaining 33 were test sites. Reference sites were considered unimpacted if they had not been grazed by cattle for more than 10 years and were not adjacent to stream crossings, dug outs, or any other sources of anthropogenic disturbance. Test sites were largely located within the experimental pastures found within GNPC, although some sites were located in the long-term grazing community pastures outside of the park boundaries (pastures 10-13 in Figure 3). Pastures 11-13 were not individually fenced and were located within larger community pastures. Therefore, sampling sites may not have

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occurred directly within the boundaries outlined. No sites in pasture 1 were sampled because the characteristics of the stream in this area were very different from the other steams, including very low stream banks or even the loss of a defined channel in some locations. In 2007, there were no cattle in the experimental pastures and all sites were designated reference except pasture 10 which was within a larger community pasture and, therefore, a test site (Figure 2). Cattle were placed into the remaining pastures in 2008 and 2009, except for pastures 5 and 9 which were the BAGMA project control pastures (Figure 3).

Each sampling site consisted of a single stream reach. The length of the stream reach was either approximately eight times the bankfull width (Bailey *et al.* 2004) or the entire pool, whichever was less. Although riffles are typically sampled in stream studies (e.g., Caruso 2002; Death and Winterbourn 1995; Delong and Brusven 1998; Doeg *et al.* 1989; Scrimgeour and Kendall 2003; Stagliano and Whiles 2002), this was not possible in my study as they were dry during the sampling period. Bailey *et al.* (2004) suggested, as a rough guideline, that if streams are limited and multiple sites must be placed on the same stream, then sites should be separated by approximately 100 times the average stream widths. As pools in the streams had maximum width usually less than 10 m wide, this would be accomplished by using a distance of 1 km between sites. This guideline is based on flowing streams; since my streams consisted of surface-flow isolated pools, it was not always followed. Within a sampling year, sites outside of the experimental pastures were always separated by at least a kilometre. Within the experimental pastures, sites closer together were

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sometimes sampled as there were only a few available pools per pasture. Prior to sampling, each pool within each pasture was assigned a number and a random number generator was used to select the pool to be sampled.

Data collection and laboratory analysis

At each site, habitat components were measured (e.g., stream and stream bank width, water depth), estimated (e.g., bank vegetation cover and type), or collected (sediment and water samples). In addition, macroinvertebrates were collected at each of the sites. Sampling occurred from mid- to late October in 2007 and to early November in 2008 and 2009. Sampling in autumn is ideal, as it ensures that data are being collected after a full season of grazing (Kauffman *et al.* 2004) and that insects with aerial forms were present in their aquatic immature stages (Merritt *et al.* 2008).

Physical Habitat

Methods used to collect data for the 33 habitat variables (Table 5) are described below.

Stream bank vegetation

Estimates of stream bank cover and vegetation type were made by two observers standing on one stream bank and independently estimating the percentage of the opposite bank with exposed soil versus the percentage that was covered in vegetation. The two observers then discussed their estimates until a consensus was reached. These estimates were performed for both stream banks at a site and the

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values were averaged. The same two observers also estimated the proportion of vegetation that was grass, shrubs, and trees using the same method.

Stream substrate

Two observers independently estimated the per cent macrophyte coverage of the stream site based on area and then reached a consensus. Where macrophytes could not be clearly observed due to water depth or poor clarity, an estimate of cover was determined during the collection of sediment.

A sediment sample was collected from each site. To accomplish this, one or two people traversed the site, taking a scoop of sediment using a small folding shovel at more than 10 locations, ensuring that the length and width of the site were sampled. Each scoop, approximately 10 cm deep and 250 ml by volume, was combined in a plastic bag and then refrigerated. This collection method is similar to the method used by Knapp and Matthews (1996). Upon return to Winnipeg, the sediment samples were dried and then processed to determine the particle size distribution. Laboratory analysis of the sediment included preparing the sample by mechanically breaking all the clumps (Johnson 1996); dry sieving to separate out particles greater than 2mm in diameter (Johnson 1996); carbonate, organic matter, and iron removal from the fraction that was less than 2 mm in diameter (Johnson 1996; Soukup *et al.* 2008); and wet sieving to separate the less than 2 mm fraction into different size categories (Johnson 1996). The particle size diameter categories are shown in the first 10 rows of Table 5 along with the associated size name categories as described by Allan and Castillo (2008a). Four subsamples were analyzed for each whole

sample, and data from these subsamples were averaged to produce a mean per cent of total weight in that category.

The organic matter content of the sediment, measured as loss on ignition (LOI), was determined by placing twenty approximately 1 g portions of dried sediment from each sample in a muffle furnace at 550°C for five hours (Dean 1974; Reiners and Reiners 1972). The difference in weight pre- and post-treatment, was determined, then averaged, providing an estimate of the organic content in the sediment (%LOI).

Site morphology

Stream morphological measurements at each site included measuring the widest part of the stream (wetted width), the bankfull width at the widest part of the stream (bankfull width at maximum wetted width), and the deepest water depth (maximum water depth).

Water chemistry

At each site, one water sample was collected using a 500 mL acid-washed high-density polyethylene bottle. The bottle was rinsed three times in the stream site water and then was filled and capped underwater. The sample was brought back to the field station where a 100 mL sample was filtered through a pre-combusted 0.47 μm GF/C filter. This was repeated for a total of four filters. These filters were placed in a desiccator, and placed in a dark place to dry. The filters were analyzed by the Freshwater Institute Analytical Chemistry Laboratory (Winnipeg, MB), using the procedures outlined in Stainton *et al.* (1977), for suspended phosphorus, suspended

carbon, suspended nitrogen, and chlorophyll *a*. About half of the filtrate was frozen while the remainder, along with the remaining original sample, was refrigerated. The filtrate was later analyzed by the Freshwater Institute Analytical Chemistry Laboratory for the remainder of the water chemistry variables shown in Table 5 by the procedures outlined in Stainton *et al.* (1977).

Dissolved oxygen, water temperature, conductivity, pH, and oxidation-reduction potential were measured *in situ* using a YSI® 556 MPS multi-parameter instrument. Unfortunately, the data from 2008 were lost and therefore these variables for all sites (2007-1009) were not used in the analyses.

Invertebrates

The macroinvertebrate community was sampled by kick sampling with a 250 μm mesh D-net. Before sampling began, the site was divided into six different microhabitats. These microhabitats represented the proportions of the site that were edge habitat versus the proportion that was not located along the edge, area with macrophytes versus macrophyte-absent area, and portion with gravel bottom versus proportion with finer sediment bottom. Kick sampling was conducted for three minutes, with the time spent sampling each microhabitat proportional to the representation of that habitat type present in the site. The samples were immediately preserved in 5-10% formalin. Upon return to the lab, the samples were initially transferred into 90% ethanol. Because ethanol replaces the water in the organic material in the sample, samples can become diluted and, therefore, after 2-3 weeks, they were drained through a 250 μm sieve and then placed in 70% ethanol.

The large size of the collected samples meant that subsampling was required. Subsampling was conducted by first separating samples into different fractions: large (> 6.35 mm), small (<6.35 mm and >250 μm), sand, and coarse sand. The first separation step involved dividing the sample into 2 size fractions, > 6.5 mm and 250 μm - 6.36 mm. This was accomplished by placing a 6.35 mm sieve over a bucket with 250 μm mesh bottom. The whole sample was rinsed thoroughly through the sieves, resulting in the two different size classes. The material that remained on the 6.35 mm sieve was considered the large fraction. Large and rare organisms were removed when seen and preserved for later identification. Large stones from the 6.35 mm sieve were removed and placed in a separate container after being well washed. As the large fraction was later subsampled by weight (discussed below), rocks were removed to prevent them from influencing the total weight of the fraction. The material retained on the 250 μm sieve was further separated by elutriation into an organic fraction and a heavier sand fraction. The material that had been retained on the 250 μm sieve was scooped into a 1 L bucket a couple of spoonfuls at a time. This bucket was placed into a larger 250 μm mesh-bottom bucket. A tube was attached to the sink faucet so that the direction and strength of water flow could be controlled. Warm water (approximately 25-35 °C) was directed into the bucket, making the water and sample material spin around the perimeter of the bucket. This motion of the water caused the lighter, mainly organic, material to be pulled to the top of the inner bucket, overflowing onto the 250 μm mesh bottom of the larger, outer bucket. The heavier, mainly inorganic, material remained behind

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in the bottom of the inner bucket. The lighter, organic material was the small fraction (<6.35 mm and > 250 μm). The heavier, inorganic portion mainly consisted of sand. It was placed in a bowl (approximately 30 cm in diameter) and swirled until the material sorted with the coarser material on top of the finer material. This material was scraped off into a separate container to become the coarse sand fraction, leaving the remaining, finer sand as the sand fraction.

The coarse sand fraction was completely processed to find organisms that did not float during elutriation (i.e., molluscs). The volume of the fine sand fraction was estimated and 20% of that fraction was examined under a stereoscopic microscope; if any organisms (other than molluscs) were found in this portion, it meant that the sample had not been completely elutriated or that the coarse sand fraction had been incompletely separated (if molluscs were found) and the appropriate technique was repeated on the whole sand fraction until no organisms were found in the 20% subsample.

The large fraction was subsampled by weight using a method similar to Sebastien *et al.* (1988). The large fraction was placed into one or more pre-weighed, 250 μm sieves. The sieves were left to drain for 15 minutes and then reweighed to determine the total wet weight of the large fraction. Ten, 5 ± 0.20 g subsamples were taken from the total large fraction and the weight for each was recorded. Each subsample was processed separately; aquatic invertebrates were removed, counted, and identified. The portion of the large fraction that was not sorted was placed back into sample jars and kept. The total number of invertebrates for the large fraction

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was calculated by taking the average of the number of invertebrates found per gram in each subsample. This number was then extrapolated to the entire sample by multiplying the number per gram by the weight of the large fraction.

The small fraction was subsampled by area, following the United States Geological Society subsampling protocol (Moulton *et al.* 2000). The protocol essentially goes though several steps to estimate the number of animals in subsamples and guides you to subsample a sufficient proportion of the samples to reach or exceed a fixed count of 300 individuals. Samples were spread out over a rectangular 12" by 14" tray with mesh bottom (mesh size < 250 μm) divided into 1" grids. Five randomly selected grids from this tray were removed and each put into a separate nine-by-nine estimation tray (0.5" grid size). The invertebrates from three randomly selected grids per estimation tray were counted. For each estimation tray, the number of organisms found in the three grids was summed and averaged. This provided an average number of invertebrates per grid for each estimation tray. The averages were summed for all five estimation trays and then divided by five to get the average per grid for the five estimation trays. This value was multiplied by 81 to estimate the number of invertebrates in a nine-by-nine estimation tray (i.e., the number of invertebrates in a single 1" by 1" grid from the 12" by 14" tray) (D). If D was less than 120, the number of grids to process was equal to 300/D so that a minimum of three grids was required to reach the target count of at least 300 individuals. If, however, D was greater than 120, there were too many invertebrates to efficiently process and additional subsampling was required.

To determine how many grids from the 12" by 14" tray needed to be transferred to a smaller six-by-eight tray (48 girds), D was multiplied by the three (G_3), four (G_4), and five (G_5).These G values represent the number of grids to transfer from the large to the small tray. Each of three G values were divided by 48, and then 300 was divided by each of these numbers. These values were rounded up. This resulted in three different values that represented the number of grids to process from the six-by-eight subsampling tray. Of the three values, the one closest to three without going under dictated how many grids from the 12" by 14" tray to transfer to the six-by-eight tray, and how many of the grids from the smaller tray to process. Each subsample from the six-by-eight tray was maintained individually, with separate counts, vials, and bench sheets. The invertebrate counts were then extrapolated to the entire small fraction.

Finally, the large and rare organisms that had been removed during sample separation were counted and identified.

All fractions were examined under a stereoscopic microscope and invertebrates were identified to family, except for Acari and Nemata which were identified to order. Taxonomic references used were Merritt *et al.* (2008), Kathman and Brinkhurst (1999), Wiggins (1996), and Pennak (1989).

The invertebrate totals for each fraction were added together, resulting in the total taxa counts for the entire sample. These values were then used to calculate taxa richness, the per cent of the total community that was chironomid larvae

(%Chiron) and the per cent of the community that was Ephemeroptera, Odonata, and Trichoptera (%EOT).

Data analysis

The invertebrate community in Grasslands National Park

I summarized the invertebrate taxa by per cent occurrence and per cent relative abundance. This was done for reference and test sites separately. To determine per cent occurrence, I divided the number of reference sites that had at least one specimen from a specific taxon by the total number of reference sites and multiplied by 100. This was also done for the test sites. The reference site taxon per cent relative abundance was calculated by summing the counts for each taxon at the reference sites, dividing it by the total number of invertebrates collected at the reference sites, and multiplying it by 100. This was completed for the test sites using the same method.

Invertebrate community response to grazing – simple metric

Regressions analysis was conducted to determine if invertebrate metrics (%Chiron, %EOT, and richness) calculated for the experimental sites showed significant linear or non-linear relationships with grazing intensity.

Reference condition approach: creating and using the model to classify test sites

I used a multiple regression form of reference condition analysis in which I created individual multiple regression models to predict the value of each of the three community metrics (per cent Chironomidae (%Chiron), per cent Ephemeroptera, Odonata, and Trichoptera (%EOT), and taxa richness($\log_{10}$)) from

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habitat variables. These three community metrics were chosen based on their potential to respond to sedimentation, organic enrichment, and/or disturbance: the abundance of many species of chironomid larvae will increase with sedimentation and organic enrichment (Braccia and Voshell 2006b; del Rosario *et al.* 2002; Lemly 1982; Rosenberg and Resh 1993), EOT nymph abundance decreases with increased sedimentation and disturbance (Braccia and Voshell 2006b; Lemly 1982), and taxa richness can increase or decrease depending on the magnitude of the disturbance (Lemly 1982; Pearson and Rosenberg 1978).

The reference condition predictive model predicts the expected invertebrate metric of a test site if the site was in the "reference condition". It is therefore important that the habitat variables in the reference condition model not be influenced by the stressor of interest (i.e., cattle grazing) (Bailey *et al.* 2004). If the habitat variables being used in the predictive model are influenced by the stressor of interest, then the predicted community metric is not based on the reference condition. To screen the potential habitat variables to ensure independence from influence of grazing, linear regressions using the test sites were conducted to determine if a significant relationship ($P < 0.05$) existed between the different habitat variables and grazing intensity. Any habitat variables that were significantly related to grazing intensity were omitted from the list of possible reference model components.

If the water concentration of an analyte was below the minimum detection limit, a value of half of the detection limit was used (Antweiler and Taylor 2008; Glass and

Gray 2001). Only nitrite (NO_2^-), nitrate (NO_3^-), ammonium (NH_4), and iron required a few substitutions of half the detection limit. Manganese was removed from the data set as over 80% of the sites had concentrations below the minimum detection limit and substitutions should only be used when this occurs in less than 20% of sites (Sanford *et al.* 1993).

As NO_2^-, NO_3^-, and NH_4^+ are the most common reactive forms of nitrogen (Camargo *et al.* 2005), they were added together as dissolved inorganic nitrogen (DIN) (Lenat and Crawford 1994). Collinearity was avoided by combining them into one measurement. Dissolved organic nitrogen (DON) was calculated by subtracting DIN from TDN.

A correlation matrix for all of the habitat variables was constructed. This information was used to guide the process of model building. As a variable was added into the predictive model, any correlated variables (|Pearson correlation coefficient| > 0.275, *P* < 0.05) were no longer candidates for addition at the next step.

Each habitat variable was regressed against the community metric to determine if a linear or quadratic relationship existed. The quadratic relationship was chosen when the regression *P*-value was significant (*P* < 0.05) and was lower than the linear regression *P*-value (Braccia and Voshell 2007). Note that the adjusted r^2 ($r^2_{(adj)}$) was used over r^2 because it accounts for the number of terms in the regression, and the quadratic models includes an additional term, x^2.

The predictive model was created using a forward step-wise approach where each habitat variable and the related interaction terms were added in turn, using the appropriate linear or quadratic relationship. The starting model (first step) contained the habitat variable that produced the best predicted r^2 ($r^2_{(pred)}$) when regressed with the community metric.

The $r^2_{(pred)}$ term was used to identify the best model because it helps prevent model overfitting by determining how capable the model is of predicting the responses for new observations. The $r^2_{(pred)}$ is calculated from the predicted residual sum of squares (PRESS statistic) for the regression (Minitab 2013). The PRESS statistic is similar to the errors sum of squares (SSE), with one major difference. While both the PRESS and SSE statistics are calculated by summing the squared residuals (observed minus the expected value of the community metric), the expected values for the PRESS statistic are calculated from a model that removes each site in turn and calculates the expected value without that site in the model (Frost 2013; Minitab 2013). The $r^2_{(pred)}$ is calculated by subtracting the PRESS from one, dividing by the total sum of squares, and multiplying by 100 (Minitab 2013).

The "first step" model was used as the base for the next habitat variable addition (the next step), with any terms that were not significant ($p>0.15$) removed from the model. If the main effect of a habitat variable was not significant but any of the related interactions were, then it remained in the model. Model creation ceased when no additional habitat variables increased the models effectiveness, as described by the $r^2_{(pred)}$.

I used three model diagnostics to determine if any sites were disproportionately influencing the model: leverage values, Cook's distance, and the difference in fit statistics (DFITS). Leverage values are a measure of the difference between the x-value for a site and the mean. Large leverages (values greater than 2p/n where p equals the number of terms in an equation and n is the number of observations) suggest that the site may be disproportionately affecting the model (Myers *et al.* 2010). Cook's distance uses leverage values and standardized residuals to determine if the site x- and y-values are unusual. Values of 1 or greater indicate that an observation may need further investigation (Myers *et al.* 2010). The final diagnostic used was DFITS. This represents roughly the number of standard deviations by which the fitted value of a site changes when changes when that site is removed from the model (Kutner *et al.* 2005). More specifically, DFITS is calculated by taking the fitted value of a site when that site is included in regression model and subtracting the predicted value of that site when the site is not included in the regression model, and then standardizing the result using the mean square error (Kutner *et al.* 2005). Any sites with an absolute value greater than 1 were identified as sites requiring closer examination (Kutner *et al.* 2005). Once potential problem sites were identified, the model was rerun without each site in turn to examine if the $r^2_{(pred)}$ was negatively influenced. If the removal of a site increased the the $r^2_{(pred)}$ value by at least 3%, then that site was permanently removed from the model.

I also looked at the residuals of the reference sites from the created model to ensure that they met the assumptions of a multiple regression model, including a normal distribution and equal variance of the error terms.

Once the model was created and tested as above, the equation was used to calculate the expected (fitted) value of the community metric for each reference and test site. These values were then subtracted from the observed community metric to determine the residual values for each site.

The distribution of reference site residuals was used to identify test boundaries for determining whether a test site passed (was unimpacted) or failed (was impacted). Bailey *et al.* (1998) suggested placement of the boundary so that it contains 75% of the reference sites. In the case of one-tailed hypotheses, the pass-fail decision boundary was therefore set at either the first quartile or third quartile, depending on whether the metric was expected to respond to cattle grazing with a negative or positive change, respectively. For two-tailed hypotheses, the decision boundaries were the 12.5 and the 87.5 percentiles.

The three metrics each used a different pass-fail boundary because the hypothesized responses differed (Figure 4). For %Chiron, any test site where the residual value was greater than the third quartile (i.e., 75%) of the reference sites, meaning abundance of chironomid larvae was greater than expected, was categorized as a site that failed. Increased chironomid abundance is expected in areas subjected to increased disturbance, organic enrichment, and sedimentation (Braccia and Voshell 2006b; del Rosario *et al.* 2002; Lemly 1982), all of which are

possible results of cattle grazing. Since %EOT taxa react negatively to sedimentation

(Braccia and Voshell 2006b; Lemly 1982), their abundance was expected to

decrease if a site was impacted. This means if the test site residual was less than the

first quartile value (i.e., 25%) of the reference sites, it was considered impacted. I

expected to see both an increase and a decrease in taxa richness, relative to the

reference conditions (a two-tailed response). Therefore, any test sites that fell below

the 12.5 percentile or above the 87.5 percentile of the reference site residual

distribution were deemed impacted.

The greater the residual of the test site, the greater the deviation from the

reference condition. A regression of the test site residuals with grazing intensity was

used to determine if greater grazing intensity resulted in greater impact to the

streams. A visual comparison of the residual distribution over grazing intensity was

used to determine if the site status (i.e., whether the site was a long-term grazing

site or an experimental pasture), year of sampling, and/or stream the site was

located on were influencing the community metrics.

To assess if there was an effect of site location along the length of each stream, a

regression was conducted of reference site residuals with stream distance from the

most upstream site. Stream habitat changes along the longitudinal profile of a

stream as headwaters and tributaries combine to form larger and larger streams.

These changes often include greater water volume, changes to riparian vegetation

cover and stream shading, and differences in stream substrate (Allan and Castillo

2008b; Wiley *et al.* 1990). With changes to the stream habitat, it is probable that the aquatic invertebrate community will also change.

All data analysis was performed using Minitab 16. Graphs were created in SigmaPlot 11.

Results

The invertebrate community in Grasslands National Park

Of the 44 aquatic invertebrate taxa collected at the reference and the test sites, seven taxa (Candonidae, Caenidae, Chironomidae, Ceratopogonidae, Cyprididae, Naididae, and Nemata) accounted for over 97% of the total abundance (Table 6). Chironomidae was the most abundant taxon, representing 67.9% and 60.2% of the invertebrate abundance at the reference and test sites, respectively. Candonidae and Cyprididae were the next most abundant taxa; the two families combined represented 18.4% and 20.1% of the taxa at the reference and test sites, respectively.

Most taxa were found at both the reference and the test sites (Table 6). Chironomidae and Ceratopogonidae larvae were found at every reference and test site. Naididae worms were found at each test site and 94% of the reference sites. Taxa found only at the reference sites were the coleopteran Curculionidae (3.9%), the dipteran familes Ephydridae (5.9%) and Psychodidae (3.9%), and the aquatic lepidopteran Crambidae (2%). Combined, these taxa accounted for less than 0.03% of the total abundance in the reference sites. Only Gomphidae (Odonata: Anisoptera) was found at test and not the reference sites; one specimen was found at just one site (6.5% of test sites), representing less than 0.001% of the total abundance at the test sites.

Community response to grazing

I performed linear and non-linear regressions for each of the three community metrics (%Chiron, %EOT, and taxa richness) with grazing intensity. None of the metrics showed a significant linear relationship with grazing intensity (Figure 5) with P–values of 0.175, 0.320, and 0.772, respectively. The %Chiron metric was significantly related to grazing intensity when using a quadratic model ($P = 0.005$) (Figure 6 (a)). The non-linear regression of %EOT with grazing intensity was not significant ($P = 0.606$) (Figure 6 (b)). Taxa richness did not show a significant non-linear relationship to grazing intensity, although it was suggestive ($P = 0.083$) (Figure 6 (c)).

Effects of grazing on physicochemical characteristics

The relationships between the test site habitat variables and grazing intensity were explored using regression analysis. Of the 33 test sites sampled, two test sites were not used when performing the regressions: the data set of one site was missing more than one variable, and the other site had SuspN, SuspP, SuspC, and Chla concentrations that were five to seven times greater than other test sites subjected to similar levels of grazing. The r^2 and P-values from regression analysis for each habitat variable versus grazing intensity are summarized in Appendix 1.

The regressions of per cent composition of the different sediment particle sizes and %LOI with grazing intensity are shown in Figure 7. Two of the sediment variables showed significant positive correlations with grazing intensity ($P < 0.05$): %FGravel (Figure 7 (h)) and %MGravel (Figure 7 (i)). These two habitat variables

also showed higher variability at higher grazing intensities compared to lower grazing intensities. The means, standard deviations, medians, and ranges of the sediment variables for the reference and test sites are listed in Appendix 2 and Appendix 3, respectively.

The distributions of the physical descriptors of the stream and streambanks (%Macro, %Eroding, %Grass, WetWidth, Wet Depth, and BFWidth) with grazing intensity are shown in Figure 8. No variables were significantly related to grazing intensity ($P > 0.05$). The means, standard deviations, medians, and ranges for the reference and tests sites are presented in Appendix 4 and Appendix 5, respectively.

Of the 16 water chemistry variables (Figure 9), only SuspC was significantly related to grazing intensity ($P = 0.05$) (Figure 9 (f)). Concentrations of SuspC also became more variable at higher grazing intensities than lower grazing intensities. Although the relationships of SuspN (Figure 9 (c)), SuspP (Figure 9 (d)), and Chla (Figure 9 (g)) concentrations with grazing were not significant ($P > 0.05$), they showed higher variability at higher grazing intensity sites compared to lower grazing intensity sites. The means, standard deviations, medians, and ranges of the reference and test sites can be found in Appendix 6 and Appendix 7, respectively.

Reference condition approach: creating and using the model to classify test sites

Correlated variables

A correlation matrix was created using only the habitat variable values from the reference sites. Variables were considered to be correlated if the absolute value of

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the Pearson correlation coefficient was greater than 0.275 ($P < 0.05$). Three main groupings of correlated habitat variables were found: particle size, nutrients, and ions. The different particle sizes (%VFSand, %FSand, %MSand, %CSand, %VCSand, %VFGravel, %FGravel, %MGravel, and %CVCGravel) were generally correlated with each other, although exceptions did occur. Nutrients (DIN, DON, SuspN, SuspP, TDP, and SuspC) were typically correlated with each other, but again, there were exceptions. Ions (SO_4, Na, Cl, K, Mg, Ca, and Fe) were usually correlated with each other, with some exceptions. The full correlation matrix with the significant Pearson correlation coefficients identified is provided in Appendix 8.

%Chiron

The linear and non-linear regressions of %Chiron with each habitat variable (Table 7) indicated that %LOI should each be treated as a quadratic polynomial function when using them in the creation of the predictive model.

I was unable to create a model that would predict a significant portion of the variability in %Chiron. The best model I was able to construct was: %Chiron = 0.2706 + 0.2848%LOI – 0.045%LOI^2. It used only one habitat variable (%LOI) and had an $r^2_{(pred)}$ of 0.88% *(P = 0.016)*. The inability of this model to predict %Chiron values accurately for new sites made it an ineffective metric for the reference condition approach.

%EOT

A linear relationship was used for each of the habitat variables when creating the %EOT predictive model since none of the habitat variables showed a quadratic polynomial relationship with grazing intensity (Table 7).

The best four models created for the %EOT metric, as determined by the $r^2_{(pred)}$, are shown in Table 8. Of the four, the best model (Model #4 in Table 8) used %Silt and %Macro as main effects plus their interaction term. The $r^2_{(pred)}$ was only 17.09% (P = 0.003). The low $r^2_{(pred)}$ of this model prevented any further data analysis using this metric for the reference condition approach.

Richness(log$_{10}$)

The $r^2_{(adj)}$ values for the linear and quadratic regressions of Richness(log$_{10}$) with each habitat variables are shown in Table 7. Of the habitat variables, %VFSand, %LOI, and %Macro showed a better quadratic than linear relationship with richness(log$_{10}$).

The final Richness(log$_{10}$) model had an r^2 of 90.48%, $r^2_{(adj)}$ of 85.56%, and $r^2_{(pred)}$ of 69.19% (P < 0.001). Each model addition, after the removal of insignificant terms, is shown as a step in Table 9. The six main habitat variables included in the model were %LOI, BFWidth, SRSi, Chla, %Eroding, and WetWidth.

After the final main effect habitat variable and related significant interactions were added to the model (the equation for the model is shown as Step 6 in Table 9), the leverage, Cook's distance, and DFIT values for each reference site were calculated. There were 14 sites that were potentially influencing the model more

than other sites; some of which were identified under more than one of the model

diagnostics. Seven sites had high leverage scores (i.e., were greater than 2p/n =

(2)(17)/49 = 0.694), one site had a Cook's distance value greater than 1, and 11

sites had absolute DFIT values greater than 1. After removing each suspect site in

turn and rerunning the model, only six of the 14 sites resulted in an increased $r^2_{(pred)}$

from the $r^2_{(pred)}$ in step 6 of Table 9 (65.95%). Of those six sites, only three increased

the model $r^2_{(pred)}$ by more than 2%, with one site increasing more than 3%. I tried

removing different combinations of those three sites but no combination resulted in

a further increase greater than 1%. The only site permanently removed from the

model was the one that resulted in an increase in the $r^2_{(pred)}$ from 65.95% (step 6 in

Table 9) to 69.19% ('Final Model' in Table 9). The final model residuals follow a

normal distribution (Anderson-Darling Normality Test, A^2 = 0.21, P = 0.842) and

have equal variances.

The coefficients for all of the final model terms and the associated P – values are

listed in Table 10. The final Richness(log_{10}) model used 48 of the 52 reference sites

sampled. Two sites were removed because they had missing data points, one site

because it had some water chemistry values an order of magnitude greater than the

next highest concentration observed anywhere in the study area, and one site was

removed during the model diagnostics step described above.

Test sites were determined to be impacted if the residual value fell outside of the

pass-fail boundary determined by the distribution of the reference site residuals.

For the Richness(log_{10}) model, the lower boundary was -0.03995 (12.5 percentile)

and the upper boundary was 0.04395 (87.5 percentile) (Table 11). The failure rate

for the test sites was 73.3%, with 22 of the 30 sites falling outside of the pass-fail

boundary. The test site residuals are shown in Figure 10 where failed sites fall

outside of the grey bar. There was no significant relationship between the

Richness($\log_{10}$) test site residuals and grazing intensity (r^2 = 11.3%, $r^2_{(adj)}$ = 4.7%, P

= 0.198). The three parts of Figure 10 contain the same data points but have

different symbols to highlight different characteristics of the sites. The residuals of

the long-term grazing sites are comparable to those of the experimental pasture

sites at similar grazing intensities (Figure 10 (a)). The year of sampling was not

important as the sites sampled in different years have comparable residuals at

similar grazing intensities (Figure 10 (b)). There were no inter-stream differences

between Horse Creek and Wetherall Creek as the residuals are comparable at

similar levels of grazing intensities (Figure 10 (c)).

There was no significant effect of site location along the longitudinal axis of the

stream on taxa richness. The reference site residuals for Horse Creek were not

significantly related to downstream distance (r^2 = 8.5, P = 0.084) (Figure 11). There

was also no significant relationship between reference site residuals and Wetherall

Creek site location (r^2 = 0%, P = 0.929) (Figure 12).

Discussion

The invertebrate community in Grasslands National Park

The list of taxa collected in Horse and Wetherall Creeks is typical of intermittent streams with most of the major groups found on the list of taxa for temporary waters produced by Williams (2006), including: Nemata, Planorbidae, Naididae, Enchytraeidae, Corixidae, Elmidae, Dytsicidae, Hydrophilidae, Tipulidae, Ceratopogonidae, and Chironomidae. Williams summarized the characteristics of taxa found in four studies of intermittent streams in California (del Rosario and Resh 2000), Canada (Williams and Hynes 1976), Brazil (Alkins-Koo 1989/1990; Heckman 1998), and Australia (Boulton and Lake 1992c). They shared adaptations to temporary habitats such as avoidance of drying through burrowing (Nemata, Oligochaeta, and some Diptera), tolerance to low oxygen concentrations (Nemata, Oligochaeta, Hemiptera, Coleoptera, and some Diptera), prolonged diapause through periods of high temperature or drying (Planorbidae, some Diptera), and aerial stages that provide for rapid recolonization and/or avoidance of dry periods (Hemiptera, Coleoptera, and Diptera) (Hynes 1970). Noticeably absent from my study were Plecoptera nymphs, although Williams (2006) listed Plecoptera as a group commonly occurring in intermittent streams. Plecoptera are typically found in running, oligotrophic water, and there are species that are adapted to intermittent streams. However, they are more typically reported from streams that are intermittent because they are headwater, low order streams rather than being ephemeral because they are located in semi-arid areas. These taxa may enter a

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prolonged egg diapause to withstand periods of adverse conditions (Merritt *et al.*

2008). Plecoptera were likely absent from the GNPC creeks because of the

combination of long periods with no flow, the predominance of fine particle sizes,

high summer temperatures and high salinity, making the environment inhospitable.

The general absence of Plecoptera in southcentral Saskatchewan is supported by

Dosdall and Lehmkuhl (1979), who found no specimens in sampling trips to this

region.

The composition of the invertebrate community collected in the GNPC creeks is

similar to that collected by Stagliano and Whiles (2002) in Kings Creek, Konza

Prairie Biological Station (KPBS), Kansas, the only study of a prairie stream in which

comparable data had been provided. Although total taxa richness in GNPC (44) was

similar to KPBS (50), Stagliano and Whiles (2002) identified their taxa to genus and

species (where possible). My estimates of taxa richness in GNPC are underestimates

because of the use of family or higher level identification. Their estimates may also

be an underestimate as they sampled only in riffles, using a stovepipe corer. In both

locations, the six most abundant invertebrate taxa (excluding Copepoda),

representing over 85% of total abundance, were: Chironomidae, Ostracoda,

Oligochaeta, Ceratopogonidae, Nemata, and Ephemeroptera. Chironomidae was the

most abundant taxon in both studies, comprising an average of 64% and 49.5% of

the total invertebrate abundance in GNPC and KPBS, respectively. The relative

abundance of Ceratopogonidae, Nemata, and Ephemeroptera was similar in the two

study locations. The main differences in the invertebrate communities were the

higher relative abundance of ostracods (19.3% versus 3.5%) and lower relative

abundance of oligochaetes (4.6% versus 20.7%) in GNPC, and a difference in the

ephemeropteran families. Ephemeropteran nymphs were mainly represented by

Caenidae in GNPC streams, while in the KPBS, the baetid, *Fallceon quilleri* (Dodds,

1923), was the primary taxon present. This difference can be attributed to the

difference in substrate particle size distributions. In GNPC, fine material (< 2 mm)

made up 78% of the stream substrate while in the KPBS study, reaches were gravel

and cobble-dominated, with 85% of the sediment composed of particles greater

than 2 mm in diameter (Stagliano and Whiles 2002). Caenidae are collector-

gatherers that sprawl on the surface of fine sediment (Merritt *et al.* 2008) while the

baerid *F. quilleri* is a scraper (Merritt *et al.* 2008), and would, therefore, be

associated with larger particle sizes. Although *F. quilleri* is present in Saskatchewan

(Webb 2002), its presence in GNPC is not confirmed and nymphs are likely

outcompeted by taxa better adapted to the conditions found there. The ratio of

relative abundance of Oligochaeta and Ostracoda is opposite between the two

studies, with Ostracoda predominant in GNPC and Oligochaeta in KPBS. This

reversal could be related to sediment particle size, and differences in salinity and

freezing tolerances. Ostracods are more abundant in finer particle sizes (< 1 mm)

than larger ones (Barker 1983/84), which could explain their lower abundance in

KPBS. Both taxa can be found over a wide range of salinity concentrations (Berezina

2003; Chapman and Brinkhurst 1980; Hart *et al.* 1991a), including in intermittent

saline habitats (Bunn and Davies 1992; Williams 2006). In two Australian saline

streams, one intermittent and one perennial, Bunn and Davies (1992) found the

relative abundance of ostracods to be, on average, 13% higher than that of

oligochaetes. The conductivity of sites in GNPC was only about 0.25 of the Australian

streams, but was four times higher than in Kings Creek. The more saline habitat in

GNPC could be a contributing factor to the higher proportion of ostracods than

oligochaetes in the benthic community. Another factor that could be influencing the

ratio is winter freezing. The streams in GNPC are shallow (< 1.06 m) and likely

freeze to the bottom. In one *in situ* experiment, both ostracods and oligochaetes

survived in frozen substrate, but ostracods showed higher survival (Andrews and

Rigler 1985). Fine sediment, salinity, and freezing are all possible contributing

factors to the higher ratio of ostracods to oligochaetes seen in GNPC.

Effects of grazing on the physicochemical characteristics

Literature reports on effects of grazing on stream substrate particle size

distribution are variable due to the interaction of stream hydraulics with local soil

characteristics. The interpretation of results is further complicated by the

differences in grazing intensity between the studies and because authors have not

reported comparable substrate size fractions. A significant increase in fine

sediments as a result of grazing has been reported in two studies. Herbst *et al.*

(2012) compared grazed (unknown intensity) with ungrazed conditions, whereas

Clary (1999) found no effect of light grazing (20-25% utilization) but an effect of

moderate intensity grazing (35-50% utilization). The definition of "fine sediment"

was broader in Clary (1999) (< 4.7 mm in diameter) than in Herbst *et al.* (2012) (< 2

mm). In comparison, two studies reported that grazing resulted in a significant reduction in the proportion of fine sediment (< 4.8 mm). Platts (1981) compared heavily grazed versus lightly grazed streams while Platts and Nelson (1985) compared areas with riparian grazing versus areas where cattle had been fenced out of the riparian area. In GNPC, I observed that two sediment fractions, %FGravel and %MGravel, were significantly affected by grazing intensity, both increasing as grazing intensity was increased. Conversely, Quinn *et al.* (1992) and Williamson *et al.* (1992) reported no effect of grazing on sediment particle size distribution. It may seem as though there is no consistent information available to support any conclusion regarding the impacts of grazing on sediment particle size distribution. Inter-study differences in the characteristics of the study streams and their surrounding environments (e.g., water flow velocity, precipitation, time of year, water depth, stream width, grazing intensity, grazing management practices, bank sediment characteristics, frequency of riparian use, upstream land use, and surrounding vegetation type) will control how the stream substrate responds to erosion caused by cattle grazing.

Grazing results in addition of sediment to streams both as fine particles suspended in overland flow and as mass erosion of chunks of bank material. In arid areas with limited rainfall, or in situations with well-developed riparian filters, the addition of suspended particulates in overland flow is reduced (Dillaha *et al.* 1988; 1989; Lim *et al.* 1998; Magette *et al.* 1989; Robinson *et al.* 1996; Scrimgeour and Kendall 2002; Young *et al.* 1980). The effects of mass erosion events on substrate

particle size distribution will be determined both by the initial composition of the

bank material and by stream hydraulics. The fate of the added sediment

(downstream transport or deposition) is determined by flow velocity and

turbulence. The size of particles that can be transported by flow is directly related to

velocity (Graf 1971; Hjulström 1935) and particles smaller than 20 μm will remain

suspended even at the lowest velocities. As sediment size increases beyond 20 μm,

there is a linear increase in the velocity required to keep the material suspended.

However, once material has been deposited, the relationship between particle size

and the velocity necessary to transport the particles downstream becomes more

complicated. Particles will erode when the velocity of water provides sufficient

shear stress to overcome the forces keeping the sediment on the stream bottom,

such as mass and cohesion (Zhen-Gang 2008). Resuspension of deposited sediment

or the erosion of sediment from mass deposits requires higher water velocities than

maintaining the same particle size in the stream water column. The velocity

required to resuspend a particle is not linearly related to particle size because

particles smaller than 63 μm are more cohesive than larger particles. Clay particles

(<2 μm) are more difficult to resuspend than larger particles up to 3 mm in

diameter. Small clay particles are flat with large surface area to volume ratios and

strong electrochemical attractive forces, making them resistant to resuspension

(Richards 1982; Zhen-Gang 2008). The flat shape also makes them more

hydrodynamic, increasing the critical velocity needed to resuspend them (Hsü

2004). As particle size increases from clay to the upper boundary of silt (0.063 mm),

the cohesive forces between particles decrease and the particles become rougher, decreasing the velocity required to entrain them in the flow. Resuspension of particles larger than silt require increasing velocity to counteract their greater mass. The effect of grazing on particle size distribution starts first with the particle size distribution that is deposited into the streams and this is then modified by the hydraulic conditions that are encountered in the stream, which can also exhibit considerable temporal variation. Systems that show an increase in smaller particle size fractions as a result of grazing must start with fine particle sizes entering either with overland flow or in mass deposits and then have instream flow velocities that are too slow to maintain the transport of those particles or to erode cohesive fine material. Systems that show an increase in larger particle sizes with grazing must have bank soils that contain as a component those particle sizes. Flow must at some point during the annual stream hydrograph be sufficient to remove and transport any finer material in the deposited soil downstream, with this threshold velocity dependant on the presence or absence of material that can form cohesive deposits. In Horse and Wetherall Creeks, silt (< 0.63μm) was the primary component of pool sediment (35%), indicating that flow velocity during any part of the year must not have been sufficient to entrain these cohesive materials and move them downstream. Inter-annual differences in the relative proportions of particle sizes could be an indicator of yearly variation in the stream hydrograph.

The variability I observed in the %FGravel and %MGravel measurements was greater at higher grazing intensity sites than lower grazing intensity sites (Figure 7

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(h) and (i)). Minimum measurements of these metrics at the higher grazing intensities are the same as seen at the low grazing intensities, but maxima are much higher. Similar minima suggest that these sites were not used by cattle. Higher maxima indicate heterogeneity; those with higher maxima may have been used more by cattle.

Low rainfall and the presence of riparian vegetation could explain why there were no significant relationships between DIN, DON, SuspN, SuspP, TDP, or TSS and grazing intensity. Rainfall is required to move nutrients and sediment from the terrestrial ecosystem into the streams; if there is low rainfall, then there will also be low transport (Caruso 2002; Scrimgeour and Kendall 2002). If there is sufficient rainfall to cause overland flow, a well vegetated riparian zone will remove a high proportion of nutrients (Beck and Young 1975; Daniels and Gilliam 1996; Dillaha *et al.* 1988; 1989; Gharabaghi *et al.* 2000; Line *et al.* 2000; Magette *et al.* 1989; McKergow *et al.* 2003; Young *et al.* 1980) and sediments (Dillaha *et al.* 1988; 1989; Lim *et al.* 1998; Magette *et al.* 1989; Robinson *et al.* 1996; Young *et al.* 1980) from the runoff before it enters the stream. In GNPC, the vegetated riparian zone was not disrupted by grazing, as evidenced by the lack of a significant relationship in the proportion of the bank that was vegetated versus exposed with grazing intensity (Figure 8 (b)).

Heterogeneous use of the stream pools by cattle may also have contributed to non-significant relationships between water chemistry variables and cattle grazing intensity. Cattle are herding animals that do not distribute themselves uniformly

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across rangelands. Therefore, all pools in each pasture may not have been used equally, and when one pool was used, it was probably used by a number of cattle at once. Distributions of cattle across the rangeland are influenced by water availability (Kauffman and Krueger 1984; Patten 1998; Pinchak *et al.* 1991; Platts 1991; Roath and Krueger 1982; Trimble and Mendel 1995) and vegetation (Ames 1977; Platts 1991). Water troughs attract cattle, reducing the amount of time they spend in riparian areas (Bagshaw *et al.* 2008; Miner *et al.* 1992). In the GNPC experimental enclosures, two water troughs reduced the time cattle spent within 20 m of streams by 27% (Ghamoushi-Ramandi and Fitzsimmons 2009). It is possible that pools closer to these troughs would be more often frequented than ones further away. It is also possible that when cattle have moved away from troughs, they are more dependent on the stream pools and therefore those pools are used more. Cattle may also use pools that are closer to their preferred forage more often. Assuming heterogeneous vegetation distribution, this could contribute to patchy pool use. There were on average 55 pools per pasture, and a maximum of three randomly-selected pools per pasture were sampled each year. Therefore, I may have missed pools that were more commonly used, and since the water flow was discontinuous, impacts would have remained localized.

Natural inter-pool variability may have masked the effects of grazing in GNPC. All pools may not have the same baseline chemical compositions (due to differences in groundwater input, riparian vegetation, wildlife use, etc.), and, therefore, any differences as a result of grazing would be more difficult to detect. The change in

concentrations, and comparisons of this change between the different grazing treatments, may be a better indicator of the influence of grazing intensity.

The only measured water quality variable to show a significant relationship with grazing intensity was suspended carbon. Forms of suspended carbon include detrital particles, algae, and bacteria, and the methods I used when processing the collected water samples would also have allowed zooplankton to be on the filter used for suspended carbon analysis. Grazing could have increased the amount of all of these suspended carbon components. Increased detritus in the streams could occur from waste deposition and bank sloughing and algal density may increase with increased nutrients in the streams. Since there were no significant relationships between TSS or any of the phosphorus and nitrogen forms with grazing intensity, this explanation is not supported. Cattle deposit their wastes in and around streams (Gary *et al.* 1983), resulting in higher faecal coliform (Gary *et al.* 1983; Myers and Kane 2011; Roche *et al.* 2013), faecal streptococci (Gary *et al.* 1983), and *Escherichia coli* (Migula 1995) (Myers and Kane 2011; Roche *et al.* 2013) counts than found in ungrazed comparisons. No response in TSS suggests that bacteria were not responsible for the increase in SuspC, but since bacteria concentrations were not measured in GNPC, this conclusion, although plausible, cannot be confirmed. Zooplankton was not removed from the filters when filtering the water samples. As such, the presence of zooplankton on a filter could have significantly increased the carbon concentrations. Zooplankton abundance can be related to nutrient concentration (Stemberger and Lazorchak 1994). There was no

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significant difference in nutrient concentrations along the grazing gradient, suggesting that zooplankton were not responsible for increased suspended carbon concentrations. Without zooplankton abundance data, this cannot be known for certain. Since there was no significant change in nitrogen, phosphorus, Chla, and TSS concentrations with grazing, I hypothesize that a change in the organic content of the total suspended solids is responsible for increased suspended carbon concentrations.

There was no relationship between grazing intensity and the proportion of the bank that was vegetated or the proportion of the bank that was grasses versus shrubs. This could be because of the short period over which most of these sites had been subjected to grazing. The majority of the test sites had been grazed for only one or two seasons at the time of sampling and it may take longer for measureable changes to the stream bank to appear. Even after just a few months of grazing, there were locations along the stream where shrubs in the riparian zone were trampled; trails running parallel with the stream through the riparian zone had been formed; and there were ramps running down the stream bank to the stream water. The impacts of these alterations were apparently not captured by the methods that I used to assess the banks. Canada conducted riparian assessments in the GNPC experimental pastures following the methods of Fitch *et al.* (2001). These methods create riparian health scores based on 11 different observations, including per cent vegetation cover, proportion of invasive plant species, proportion of disturbance-caused vegetation, presence of self-maintaining woody vegetation, whether woody

vegetation is being used, amount of dead wood, amount of deep-rooted vegetation, proportion of bare ground due to human activity, per cent of bank altered by human activity, whether the reach is compacted, bumpy, or rutted from use, and if the stream has access to the floodplain. Scores for each of these observations are applied and combined using different weighting. The final score classifies the site as healthy, healthy but with problems, or unhealthy. Parks Canada made four assessments per experimental pasture (pastures 1-9) in 2006 before any grazing and again at the same sites in 2009 after almost two complete grazing seasons. Ghamoushi-Ramandi and Fitzsimmons (2009) reported a decrease in riparian health after grazing had been implemented, with a 45% decrease, 42% increase, and 3% increase in the number of healthy sites categorized as healthy, healthy but with problems, and unhealthy, respectively. There was a negative trend between the difference in the median riparian health scores (2006 minus 2009) as grazing intensity increased (Ramandi and Wruth 2010). This riparian assessment method was obviously far more sensitive than the %Eroding estimates that I made but could not be used with my data set since estimates were not conducted for each of my reference and test sampling sites.

Community response to grazing

Two main hypotheses that were considered when looking at invertebrate community responses to grazing are the Pearson-Rosenberg model (Pearson and Rosenberg 1978) and the intermediate disturbance hypothesis (IDH) (Connell 1978). The Pearson-Rosenberg model describes the responses of abundance,

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richness, and biomass along a gradient of organic enrichment (Figure 13) (Pearson and Rosenberg 1978). As organic enrichment increases, there is an initial peak in taxa richness due to the coexistence of species that were present at lower levels of organic matter and species that are tolerant of high organic enrichment. A peak in biomass is associated with this peak in richness as organic enrichment encourages increased size of individuals. Richness and biomass decrease as organic enrichment further increases as sensitive taxa are sequentially lost. As organic enrichment continues to increase, there is a peak in abundance associated with large numbers of the few taxa adapted to high organic content and low oxygen concentrations. This peak in abundance is associated with a secondary smaller peak in biomass. At even higher levels of organic enrichment, there is a decline in all invertebrate taxa (Pearson and Rosenberg 1978). In freshwater environments, the taxa commonly associated with peak abundance are Chironomidae larvae (Armitage *et al.* 1983; Hynes 1960; Simião-Ferreira *et al.* 2009) and Tubificinae (Armitage *et al.* 1983; Aston 1973; Hynes 1960) that feed on fine particulate organic matter and can survive in areas with low oxygen (Hershey and Lamberti 2001; Hynes 1960). For example, Simião-Ferreira *et al.* (2009) sampled upstream and 200 m and 500 m downstream of a sewage treatment plant. Although they did not observe a change in taxa richness, there was an increase in mean abundance of chironomid larvae from 422 individuals collected at the upstream site to 11767 at 200 m and 9319 at 500 m. The increase was attributed to four genera that are usually considered resistant to

organic pollution: *Chironomus* spp., *Polypedilum* spp., *Thienemanniella* spp., and *Rheotanytarsus* spp.

A response in the chironomid community to grazing intensity is seen in some studies. Quinn *et al.* (1992) found higher densities of chironomid larvae at the more heavily grazed site (2.5 cattle ha^{-1}) than the lightly grazed site. This increase in Chironomidae abundance under "heavy" grazing matches the findings of Braccia and Voshell (2007), the only authors I have found who conducted a study on macroinvertebrates in streams affected by a gradient of cattle grazing. They found a significant positive linear relationship between density of Chironomidae and grazing intensity, with the highest density at the "very heavy" grazing intensity (2.85 cattle ha^{-1}). The peak in abundances at heavily grazing sites in these studies supports the Pearson-Rosenberg model. In GNPC, I did observe a significant non-linear relationship between the relative abundance of chironomid larvae and grazing. However, relative abundance peaked at 0.4 – 0.5 AUMs ha^{-1}, equivalent to an intermediate grazing level of 30-50% forage utilization (Figure 6 (a)) and there was no peak in %LOI of the sediment at this grazing intensity. In addition, %LOI was low overall, indicating that the Pearson-Rosenberg model does not explain the chironomid response that I observed.

The other concept to consider when looking at the response of the invertebrate community to grazing intensity is the IDH, a hypothesis based on a competitive hierarchy among species; where disturbances in the ecosystem are of an intermediate magnitude and frequency, maximum species diversity can be reached

through the coexistence of *K*-selected and *r*-selected taxa (Figure 14) (Connell 1978; Resh *et al.* 1988). Traits of *K*-selected species can include smaller tolerance ranges, slower development to maturity, larger body size, more than one set of offspring, and longer life spans (Begon *et al.* 1986; Pianka 1970). Traits of *r-s*elected species are almost the opposite of K selected, including wider tolerances ranges, rapid development, smaller body size, one set of offspring, and shorter life spans. At one extreme of the disturbance gradient, where there are only rare and/or small disturbance events, *K*-selected taxa will outcompete *r*-selected taxa because they can more efficiently use available resources, and are better adapted to the stable ecological niche. This results in low species diversity (Begon *et al.* 1986; Connell 1978; Pianka 1970; Resh *et al.* 1988). At the other extreme of the disturbance gradient, where frequent and/or large scale disturbances occur, *r*-selected taxa will dominate the system because of characteristics that favour rapid reproduction and wide tolerance ranges. The *K*-selected taxa are unable to survive the magnitude and frequency of the disturbance at this extreme, resulting in their absence in the environment (Connell 1978; Resh *et al.* 1988). The IDH states that where disturbances occur at moderate intensity and/or medium frequency intervals, species diversity will be maximized because the community will be composed of both the *K*-selected and *r*-selected taxa (Figure 14)(Connell 1978; Resh *et al.* 1988).

In my study, 17.5% of variability in taxa richness was described by grazing intensity and although the relationship was not significant, it was suggestive of a non-linear relationship (P = 0.083). This relationship indicates a peak in richness at

medium grazing intensity (0.5 to 0.6 AUMs ha^{-1}) and suggests support of the IDH.

The work of Townsend and Scarsbrook (1997), who studied the gradient of bed

disturbance caused by high flow events on the invertebrate richness in streams, and

Braccia and Voshell (2007), who examine the influence of a grazing gradient on the

invertebrate richness in streams, support the IDH. There was a significant quadratic

relationship between richness and the magnitude of the disturbance in both studies.

However, the majority of studies on the response of aquatic invertebrate diversity to

disturbance in lotic environments do not support the IDH (e.g., Death and

Winterbourn 1995; Doeg *et al.* 1989; Lake *et al.* 1989; Malmqvist and Otto 1987;

McCabe and Gotelli 2000; Reice 1985; Resh *et al.* 1988; Robinson and Minshall

1986; Thorp and Bergey 1981). The IDH curve may not have been seen in many lotic

studies because the stream may already be at intermediate or high disturbance level

prior to the start of study, rather than in an undisturbed state as investigators

assume (Ward and Stanford 1983). In perennial streams, changes in flow regime

through the year provide a frequently changing source of disturbance (Holomuzki

and Biggs 1999; Jellyman *et al.* 2013; O'Connor *et al.* 2012), but in the study streams

in GNPC, there is no flow for most of the year. This may allow for a more stable

invertebrate community composition in the absence of a grazing disturbance and

the appearance of the IDH curve after the introduction of grazing.

Reference condition approach: creating and using the model to classify test sites

%Chiron

There was a significant non-linear relationship between %LOI and the relative abundance of chironomid larvae in the reference sites. However, it explained only 16.2% of the variability and had low predictive capability ($r^2_{(pred)}$ = 0.88%). Many chironomid species feed on organic detritus (Armitage *et al.* 1995; Merritt *et al.* 2008; Pinder 1986), so an increase in %LOI could represent increased food availability. The observed relationship was non-linear with peak relative abundance at approximately 3% LOI, and declining at higher values. The Pearson-Rosenberg model suggests peak in abundance at some level of organic enrichment (Pearson and Rosenberg 1978); but 3% is not high. Chironomid larvae are often abundant in profundal lake sediments (Brinkhurst 1974; Jyväsjärvi *et al.* 2013) and profundal sediment organic content often exceeds 10% (Rowan *et al.* 1992). The decrease in relative abundance of chironomid larvae could be related to increased abundance of another taxon.

Taxonomic resolution at the family level has likely reduced my ability to relate differences in community composition to environmental conditions. Different species of chironomid larvae vary in their environmental preferences (Saether 1979) and thus respond differently to the same habitat metrics (Merritt *et al.* 2008). Under the Chironomidae umbrella, these taxa-specific responses are lost.

%EOT

The Ephemeroptera, Odonata, and Trichoptera predictive model used %Silt and %Macro as the main effects but these variables accounted for only 20.9% of the variability in the reference sites. Although there were 11 families within the EOT group, Caenidae was the main taxon, representing 4.1% of all individuals in the reference sites. In comparison, the rest of the taxa combined contributed less than 1% to the total invertebrate abundance. Caenid nymphs are typically found in the depositional zones of streams or in lentic environments and feed on detritus (Merritt *et al.* 2008). Caenidae larvae are classified as sprawlers and climbers by Merritt *et al.* (2008), associated with fine sediments and macrophytes. The absence of flow, prevalence of fine sediment, and abundance of macrophytes (33.7% mean coverage in pools), make these streams favourable habitat for caenid nymphs. Therefore, it is not surprising that the best %EOT model consisted of %Macro and %Silt main effects. Unfortunately, the model was not capable of accurately predicting new responses for the %EOT metric ($r^2_{(pred)}$ = 17.09%); therefore, it was not used for reference condition analysis.

The stream environment in GNPC is a stressful one, with large ranges in salinity, temperature, and water volume. The EOT community may not have responded to the stressors associated with cattle because sensitive taxa with low tolerance ranges were already absent due to the challenging environmental conditions.

Richness(log$_{10}$)

I was able to construct a model that accounted for almost 70% of the taxa richness variability in the reference sites ($r^2_{(pred)}$ = 69.19%). The main effects variables (%LOI, BFWidth, SRSi, Chla, %Eroding, and WetWidth) in the predictive model could be influencing taxa richness through a variety of pathways. Sediment organic content can influence species richness because it is a food source for many invertebrates (e.g., Ostracoda, Chironomidae, Oligochaeta) (Armitage *et al.* 1995; Delorme 2001; Merritt *et al.* 2008; Pinder 1986). The bankfull width of a stream could be an indication of the width of a riparian zone, and this could be related to the amount of riparian vegetation. The amount of allochthonous input into streams is determined by the type and amount of riparian vegetation (Afonso *et al.* 2000; Delong and Brusven 1994), and in turn influences the invertebrate community (Menninger and Palmer 2007). A direct relationship between BFWidth and %Eroding (i.e., per cent vegetated) does not exist though, as the two were not significantly correlated. Another possibility is that BFWidth could be an indicator of the slope of the stream bank. Slope can influence how much plant material and sediment from the riparian zone enters the stream (Wallace *et al.* 1992), thereby influencing the invertebrate taxa. Bankfull width may also be a surrogate for the amount of stored energy and organic matter in the riparian area. Bankfull water levels may only be reached once every few years and during the interim organic material can accumulate. During high water flows, this organic matter can be transported into the portions of the stream that remain when water levels decline.

This addition of allochthonous material could influence the invertebrate community. Slope was not measured at the sampling sites, so this relationship is unknown for these streams. Bankful width Silica could be an indicator of diatom abundance, and therefore food abundance for some invertebrates (Wang 1969). Chlorophyll concentrations in streams are significantly correlated with the concentrations of SuspN, SuspP, TDP, SuspC, and TSS. These variables can be an indicator of food availability in the stream, and, in the case of TSS, sediment in the water column. Suspended sediment can influence collector-filterer feeding efficiency and survival (Broekhuizen and Miller 2001; Euliss and Mushet 1999; Gard 2002; Hynes 1970; Lemly 1982; Newcombe and Macdonald 1991; Shaw and Richardson 2001) and can influence light penetration and associated primary productivity in the stream (Armour *et al.* 1991; Ellis 1936; Hynes 1973; Lloyd *et al.* 1987). The proportion of the bank that is unvegetated (%Eroding) could influence taxa richness by influencing bank stability and the rate of sediment transport into the streams from the riparian areas. Channel width has been shown to be positively correlated with taxa richness (Malmqvist and Hoffsten 2000). Wider stream widths could result in greater microhabitat availability and heterogeneity within a site. Increased surface area could allow for more macrophytes growth, which had been linked to taxa richness. Taxa richness is higher in the presence of macrophytes (Gregg and Rose 1985; Shupryt and Stelzer 2009), although this relationship may break down in slower flow velocities as there is less need for a refuge (Gregg and Rose 1985).

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Wider pools also could influence flow dynamics within the site during the spring, causing patchy particle size distribution.

Using the richness predictive model, 22 of the 30 test sites (73.3%) failed, with failure not obviously related to whether the site was located within a long-term grazing pasture or the experimental pastures; whether the site was sampled in 2007, 2008, or 2009; or whether the site was located on Horse Creek, Wetherall Creek, or Dunn Creek. Therefore, grazing-induced changes to the stream characteristics that are the most responsible for the presence of taxa (e.g., the proportion of fine sediment, organic matter content, and nutrients concentrations) in this study were not cumulative from one year to the next, even after decades of grazing. In the mixed-grass prairies, the majority of overland and stream flow occurs during snowmelt and spring runoff (Jensen *et al.* 2011; Matthews 1988). Under high flow velocity, a large range of particle sizes can be entrained (Graf 1971; Hjulström 1935; Simon *et al.* 1999), increasing bed roughness. This water, with the associated suspended material, can scour the stream bed, abrading macrophytes (Henley *et al.* 2000) and invertebrates (Shaver *et al.* 1997; Waters 1995), and transporting downstream invertebrates, nutrients, organic matter, and sediment that had accumulated in the pools since flow stopped the previous summer (Williams 2006). High flows and scour can cause over 94% mortality in invertebrates that lack suitable adaptations or that are unable to find a refuge (Cobb et al. 1992; Lytle and White 2007; Tate and Gurtz 1986) and can induce drift (Waters 1995). Spring flows (assuming sufficient volume, velocity, and duration) in

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intermittent streams can essentially "reset" the stream, returning nutrients, organic matter, sediment, and invertebrates to a similar starting point each year. Cattle spend time in the riparian areas, depositing waste and consuming vegetation and water. With decreased riparian vegetation, soil and associated particles are more susceptible to erosion and the buffering capacity of the riparian area is decreased (Dillaha *et al.* 1988; 1989; Lim *et al.* 1998; Magette *et al.* 1989; Robinson *et al.* 1996; Young *et al.* 1980). This potential for increased nutrient, organic matter, and sediment input into the stream is realized only if there is overland flow to transport the material. The majority of cattle-induced changes to the stream environment probably start after most of surface runoff has stopped and water velocity has significantly slowed. After surface flow has stopped, cattle will influence the stream by direct means, including bank slumping, waste deposition directly into the water, and physically disturbing the stream bottom. If effects are not cumulative, it suggests that streams could return to the reference condition within a short time period.

Richness metrics are relatively stable between years (Robinson *et al.* 2000; Sánchez-Montoya *et al.* 2009). High seasonal and/or annual species turnover in a stream due to varying environmental conditions can be counteracted by the presence of the same adaptive traits in different taxa, resulting in similar richness values (Bêche and Resh 2007; Mesa 2012; Resh *et al.* 2013).

Dunn Creek, Horse Creek and Wetherall Creek are all third and fourth order streams within the study area. The distances between Dunn Creek and Horse Creek,

and Horse Creek and Wetherall Creek are approximately 5 kilometres. There was no difference between the residuals for the taxa richness model between the streams, indicating that inter-stream community differences are not significant. Differences between streams were not expected as they share similar landscapes, stream orders, and are close together.

There was no significant relationship (either linear or non-linear) between the richness model residuals and grazing intensity, but a large percentage of the test sites were categorized as impacted, with their residuals falling outside of the pass-fail boundary. When taxa richness was directly compared to grazing intensity (without using the predictive model), the relationship was suggestive of a non-linear trend. After incorporating habitat variables to account for the underlying environmental factors that affect invertebrate communities, a significant predictive model was created. There was no relationship between the residuals and the grazing gradient, and sites that were subjected to low grazing intensities were just as likely to fail as sites in higher grazing intensity pastures. This suggests that even low grazing intensity by cattle in the southern Saskatchewan mixed-grass prairies will cause changes to the stream invertebrate community. The stream communities in the semi-arid, mixed-grass prairies are already under a lot of stress associated with temperature (Piggott *et al.* 2012; Storey and Quinn 2013), water flow (Hynes 1970), and salinity (Stanley *et al.* 1997). The introduction of another stressor to the environment (cattle grazing) resulted in pushing the test sites beyond the reference condition.

<u>Conclusion</u>

    The BAGMA provided a unique opportunity to study the effects of a gradient in grazing intensity on the mixed-grass prairie ecosystem; no other studies have included such a wide range of grazing intensities applied at such a large scale. Other projects within the BAGMA focused on changes to the terrestrial ecosystem, including song bird (e.g., Bleho 2009; Pipher 2011; Sliwinski 2011), terrestrial insect (e.g., Selinger 2010), and vegetation (e.g., Lwiwiski 2013; Tastad 2013) diversity and density. While Parks Canada performed annual riparian assessments, this study was the only one to look intensively at changes to the aquatic system, including the use of aquatic invertebrates. My results indicate that cattle are influencing some of the physical, biological, and chemical characteristics of the study streams.

    In GNPC, cattle influenced the stream physicochemical and biological characteristics after only one or two grazing seasons. This is illustrated by the significant increase in the proportion of fine gravel, the proportion of medium gravel, and the concentration of suspended carbon as grazing intensity increased. The biological community reflects changes in the stream environment. The relative abundance of Chironomidae had a significant, non-linear relationship to grazing intensity. This relationship indicates a shift in invertebrate community composition, i.e., an indicator of grazing impacts on the stream. Although not significant when directly compared to the grazing gradient, taxa richness did show a non-linear trend, suggesting that the intermediate disturbance hypothesis may be applicable in

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these streams. Using multiple regression to model the reference condition for taxa richness, I classified 73.3% of the sites with grazing as impacted. There was no significant relationship between the residuals and grazing intensity; low and high grazing intensities were causing similar changes to taxa richness.

There are different management practices aimed at reducing the impacts to the riparian and aquatic environments while still permitting cattle access to these resource-rich areas. Some common practices include incorporating off-stream water sources (e.g., Bagshaw *et al.* 2008; Miner *et al.* 1992), using salt blocks to lure cattle away from riparian areas (e.g., Ganskopp 2001; Martin and Ward 1973; McDowell 1996), and implementing late-season grazing only (e.g., Kauffman *et al.* 1983; Marlow *et al.* 1987; Trimble and Mendel 1995). Water troughs were used in the BAGMA experimental pastures and reduced the amount of time cattle spent close to steams by 27% (Ghamoushi-Ramandi and Fitzsimmons 2009). Salt blocks were also used in the pastures, but their effectiveness in drawing cattle away from streams in this study is unknown. Naturally high concentrations of salt in the soil and water in GNPC may reduce the effectiveness of this mitigation measure, as suggested in other studies (e.g., Martin and Ward 1973; McDowell 1996). Despite the measures implemented to reduce riparian and stream use by cattle, even low grazing intensities changed the stream environment, as evidenced by changes in taxa richness and the relative abundance of Chironomidae.

Only 27% of the mixed grass prairies in the Northern Great Plains of Alberta and Saskatchewan have retained their native characteristics (Wilms and Jefferson 1993)

and Grasslands National Park is the only national park representing the prairie biome in Canada (Parks Canada 2010). With little natural mixed-grass prairie remaining, this ecosystem should be protected, especially within the confines of a national park. Even the lowest cattle grazing intensities caused streams to be altered beyond the natural range of variability found within Grasslands National Park. The best method to mitigate these effects is to prevent cattle from accessing stream and riparian areas by fencing them out of the area. From a ranchers perspective, this may not be desirable since streams and riparian areas provide important sources of water and forage. Another possibility is to limit access to the streams to a few locations by making fenced corridors. This localizes cattle impacts to a few locations along the stream. Unfortunately, it may also influence cattle distribution across the entire rangeland, as cattle typically stay close to water sources (Howery *et al.* 1998). The cost associated with fencing construction and maintenance can also be prohibitive (Godwin and Miner 1996). Cattle were used within the BAGMA to determine if they could reintroduce heterogeneity into the terrestrial floral and faunal communities through disturbance. Historically, this was done by fire and bison (Hamilton 2007; Hulbert 1986; Knapp *et al.* 1999). Fire has the benefit of only temporarily affecting nutrient concentrations in the streams while protecting the stability of the banks (Hulbert 1986) and bison spend much less time close to water than cattle (Allred *et al.* 2011; England and DeVos 1969; Fortin *et al.* 2003; Larson *et al.* 2013). If the goal is to reintroduce heterogeneity and

reduce invasive species in the terrestrial ecosystem, fire and bison provide an

attractive alternative to cattle that preserves the riparian and aquatic ecosystem.

Tables

Table 1. Intermittent stream faunal groups as described by Hynes (1970) with example taxa from Australia (Boulton and Lake 1992a), Canada (Williams and Hynes 1976), and Spain (Díaz *et al.* 2008).

Intermittent stream faunal groups	Werribee River basin, southeastern Australia	Grand River basin, southern Ontario, Canada	Segura River basin, southeastern Spain
High thermal and/or hypoxia tolerance.	*Chironomus* sp.	*Micropsectra* sp., *Chironomus* sp.	
Burrow into substrate to avoid drying		*Micropsectra* sp., *Chironomus* sp. , *Allocapnia vivipara* (Claassen 1924)	*Berosus* sp. (Hydrophilidae), *Procambarus clarkii* (Girard 1852)
Drought-resistant eggs		*Paraleptophlebia ontario* (McDonnough 1926), *Symbiocladius* sp., *Diplocadius* sp., *Orthocladius* sp., Naididae	
Can recolonize from other water sources	*Dinotoperla thwaitesi* Kimmins 1951, *Riekoperla* spp., *Podonomopsis* spp.	*Aquarius remigis* (Say 1832), *Helophorus orientalis* Motschulsky 1860	Heteroptera, Coleoptera
Are terrestrial, move into stream during dry periods		Enchytraeidae	
Physiological (e.g., aestivation), life-cycle, or behavioural adaptations	*Austrocerca tasmanica* (Tillyard 1924, *Leptorussa darlingtoni* (Banks 1939)	Coleoptera, Hemiptera	

Table 2. The planned and actual grazing intensities for the experimental pastures from 2007-2009 expressed as animal unit months per hectare (AUMs/ha). Pastures 10-13 are the long-term grazing treatments located within the community pastures.

Pasture	Planned Utilitzation Rates %	Planned Grazing Intensity (AUMs ha^{-1})	2007 Actual Grazing Intensity (AUMs ha^{-1})*	2008 Actual Grazing Intensity (AUMs ha^{-1})	2009 Actual Grazing Intensity (AUMs ha^{-1})
1	0	0	0	0	0
2	20	0.25	0	0.23	0.25
3	57	0.71	0	0.66	0.71
4	70	0.88	0	0.74	0.82
5	0	0	0	0	0
6	33	0.4	0	0.36	0.39
7	45	0.56	0	0.54	0.57
8	70	0.88	0	0.81	0.83
9	0	0	0	0	0
10	50	0.5	0.25	0.29	0**
11	50	0.5	0.48	0.47	0.53
12	50	0.5	0.53	0.51	0.38
13	50	0.5	0.54	0.56	0.42

* Cattle were not placed into experimental pastures until 2008.
** Although there were no cattle placed into pasture 10 in 2009, it is a long-term grazing pasture. For purposes of data analysis, the average of the 2007 and 2008 grazing intensities (0.27 AUMs ha^{-1}) was used instead of 0.

Table 3. The number of replicate pastures within each planned grazing treatment for the grazing experiment. A range of targeted vegetation utilization targets were decided on for the experimental design, with replicate pastures unevenly divided amongst the treatments.

Nominal Grazing Intensity (%)	Number of Replicate Pastures
0	3
20	1
33	1
45	1
50*	4
57	1
70	2

*These are community pastures and had no ungrazed period prior to the start of the BAGMA study. All other experimental pastures were ungrazed for at least 16 years prior to 2008.

Table 4. The total number of reference and test sites sampled from 2007 to 2009. For the reference condition approach, a large number of unimpacted, reference, sites were sampled in addition to the sites located within the different grazing treatments (test sites).

Year	Number of Reference Sites	Number of Test Sites
2007	16	2
2008	20	7
2009	16	24
TOTAL	52	33

Table 5. The units and abbreviations of all habitat variables that were measured or collected at the sampling sites from 2007-2009.

Analyte	Unit	Abbreviation
Silt: <0.063 mm	%	%Silt
Very fine sand: 0.063 – 0.125 mm	%	%VFSand
Fine sand: 0.125 – 0.25 mm	%	%FSand
Medium sand: 0.25 – 0.5 mm	%	%MSand
Coarse sand: 0.5 – 1 mm	%	%CSand
Very coarse sand: 1 – 2 mm	%	%VCSand
Very fine gravel: 2 – 4 mm	%	%VFGravel
Fine gravel: 4 – 8 mm	%	%FGravel
Medium gravel : 8 – 16 mm	%	%MGravel
Coarse and very coarse gravel: > 16 mm	%	%CVCGravel
Organic matter in sediment	%	%LOI
Macrophyte cover	%	%Macro
Eroding area of streambank	%	%Eroding
Bank vegetation that consists of grasses	%	%Grass
Maximum wetted depth	m	WetDepth
Maximum wetted width	m	WetWidth
Bankfull width at maximum wetted width	m	BFWidth
Dissolved inorganic nitrogen	$\mu g\ L^{-1}$	DIN
Dissolved organic nitrogen	$\mu g\ L^{-1}$	DON
Suspended nitrogen	$\mu g\ L^{-1}$	SuspN
Suspended phosphorus	$\mu g\ L^{-1}$	SuspP
Total dissolved phosphorus	$\mu g\ L^{-1}$	TDP
Suspended carbon	$\mu g\ L^{-1}$	SuspC
Chlorophyll α	$\mu g\ L^{-1}$	Chla
Soluble reactive silicon	$mg\ L^{-1}$	SRSi
Sulphate	$mg\ L^{-1}$	SO_4
Total suspended solids	$mg\ L^{-1}$	TSS
Sodium	$mg\ L^{-1}$	Na
Chloride	$mg\ L^{-1}$	Cl
Potassium	$mg\ L^{-1}$	K
Magnesium	$mg\ L^{-1}$	Mg
Calcium	$mg\ L^{-1}$	Ca
Iron	$mg\ L^{-1}$	Fe

Table 6. The invertebrate taxa collected at the reference and test sites. The proportion of sites that at least one individual was collected at (% Occurrence) and the relative abundance (%) of each taxon as a proportion of all the invertebrates collected at the reference or test sites. Where taxa are fully aquatic, adult and immature counts were combined (e.g., Coleoptera).

Taxon	% Occurence		Relative Abundance (%)	
	Reference	Test	Reference	Test
Acari	68.6	67.7	0.2	0.4
Amphipoda: Gammaridae	29.4	29.0	0	0
Amphipoda: Talitridae	68.6	71.0	0.8	1.1
Coleoptera: Curculionidae	3.9	0	0	0
Coleoptera: Dytiscidae	90.2	74.2	0	0
Coleoptera: Elmidae	78.4	83.9	0.4	0.4
Coleoptera: Haliplidae	60.8	54.8	0.1	0.1
Coleoptera: Hydrophilidae	11.8	6.5	0	0
Decapoda: Cambaridae	17.6	25.8	0	0
Diptera: Ceratopogonidae	100	100	2.4	3.3
Diptera: Chaoboridae	21.6	32.3	0	0.1
Diptera: Chironomidae	100	100	67.9	60.2
Diptera: Ephydridae	5.9	0	0	0
Diptera: Psychodidae	3.9	0	0	0
Diptera: Stratiomyidae	3.9	19.4	0	0
Diptera: Tabanidae	78.4	71.0	0	0
Diptera: Tipulidae	9.8	12.9	0	0
Ephemeroptera: Baetidae	49.0	54.8	0.5	0.8
Ephemeroptera: Caenidae	98.0	96.8	4.1	4.7
Ephemeroptera: Ephemeridae	2.0	6.5	0	0
Gastropoda: Lymnaeidae	25.5	16.1	0	0
Gastropoda: Physidae	47.1	41.9	0.1	0
Gastropoda: Pisidiidae	25.5	41.9	0	0
Gastropoda: Planorbidae	54.9	80.6	0.2	0.2
Hemiptera: Corixidae	86.3	67.7	0.2	0.1
Hemiptera: Nepidae	11.8	3.2	0	0
Hemiptera: Notonectidae	70.6	61.3	0	0
Hirudinea: Erpobdellidae	54.9	67.7	0	0
Hirudinea: Glossophoniidae	27.5	41.9	0	0
Lepidoptera: Crambidae	2.0	0	0	0
Megaloptera: Sialidae	3.9	3.2	0	0

(continued)

Table 6(continued).

Taxon	% Occurence		Relative Abundance (%)	
	Reference	Test	Reference	Test
Nemata	74.5	80.6	1.2	1.5
Odonata: Aeshnidae	47.1	35.5	0	0
Odonata: Corduliidae	15.7	35.5	0	0
Odonata: Gomphidae	0	6.5	0	0
Odonata: Libellulidae	31.4	16.1	0	0
Odonata: Coenagrionidae	86.3	83.9	0.7	0.5
Oligochaeta: Enchytraeidae	13.7	19.4	0	0.1
Oligochaeta: Naididae	94.1	100	2.8	6.2
Ostracoda: Candonidae	94.1	93.5	11.3	10.8
Ostracoda: Cyprididae	90.2	90.3	7.1	9.3
Trichoptera: Hydroptilidae	2.0	3.2	0	0
Trichoptera: Leptoceridae	35.3	38.7	0	0
Trichoptera: Phryganeidae	58.8	77.4	0	0

Table 7. The adjusted r² (%) values for linear (L) and non-linear (NL) regressions of each habitat variable with each community descriptor using only the reference sites from Horse and Wetherall Creek. **Bold** indicates whether the linear or non-linear relationship was used when constructing the predictive models.

Habitat Variable	%Chiron		%EOT		Richness($\log_{10}$)	
	L	NL	L	NL	L	NL
%Silt	**5.5**	4	**11.1**	9.3	**4.4**	4.5
%VFSand	**3**	3.3	**4.5**	2.7	14	**24**
%FSand	**0**	2.5	**0**	0	**0.7**	1.2
%MSand	**0**	0	**0.5**	4	**12.2**	15.5
%CSand	**0**	0	**13.8**	13.4	**3.1**	4.1
%VCSand	**2**	0.1	**5.8**	3.8	**0**	0
%VFGravel	**0**	0.9	**0**	0	**0**	0
%FGravel	**2.2**	0.2	**1**	0	**0**	0
%MGravel	**6.5**	5.6	**0**	0.1	**0**	0
%CVCGravel	**0.7**	0	**0**	0.5	**0**	0
%LOI	4.2	**12.7**	**17.3**	15.6	18.9	**28.7**
%Macro	**0**	0	**9.8**	12.2	11.6	**27**
%Eroding	**0**	0	**0**	0	**0**	0
%Grass	**0**	0	**0**	0	**1.2**	0
WetDepth	**0**	0	**7**	5.8	**0.1**	3.4
WetWidth	**0.6**	0	**0**	0	**7.4**	6.9
BFWidth	**0**	0	**0**	0	**0**	0
DIN	**0**	1.5	**1.8**	0.2	**0**	0
DON	**0**	0	**0.2**	0	**0**	0
SuspN	**0**	4.3	**0**	0	**0**	1.3
SuspP	**0**	4.6	**0**	0	**5.6**	8.2
TDP	**0**	0	**0.7**	0	**0**	0
SuspC	**0**	0	**0**	0	**0**	0
Chla	**0**	1.1	**0**	0	**0**	0
SRSi	**0**	0	**11.4**	9.6	**0**	0
SO₄	**2.1**	1.9	**0**	0	**0**	0
TSS	**0**	0	**0**	0	**0**	0
Na	**1.2**	0	**0.7**	0	**0**	0
Cl	**3**	1	**0**	0	**0**	0
K	**0**	0	**0**	3.7	**6.4**	5.6
Mg	**2.6**	2.1	**0.4**	0	**3.5**	1.5
Ca	**0**	0	**0.7**	0	**0**	0
Fe	**1.2**	0	**0**	0	**0**	0

Table 8. The %EOT model regression equations and associated predicted r^2 ($r^2_{(pred)}$) values for the different variable combinations. No additional habitat variables could increase the $r^2_{(pred)}$ of the models. Habitat variable abbreviations are expanded in Table 5.

Model #	Model Regression Equation	$r^2_{(pred)}$ (%)	*P*-value
1	%EOT = 0.067 – 0.015(%SRSi)	8.07	0.009
2	%EOT = 0.157 – 0.0395(%LOI) – 0.00003(DON) + 0.00002(%LOI)(DON)	14.37	0.003
3	%EOT = 0.211 – 0.051(%LOI) – 0.011(%FSand) + 0.005(%LOI)(%FSand)	15.97	0.002
4	%EOT = 0.092 – 0.001(%Silt) – 0.0003(%Macro) + 0.00002(%Silt)(%Macro)	17.09	0.003

Table 9. The progressive steps to create the Richness($\log_{10}$) predictive model, including the regression equation and the final predicted r^2 ($r^2_{(pred)}$) for that step in the model. Habitat variable is the new main variable that was incorporated into the model at that step. The regression equation excludes any interactions that were insignificant. The final model is after the removal of a site with a high DFIT value. This resulted in an increase in the $r^2_{(pred)}$ of the model. Habitat variable abbreviations are expanded in Table 5.

Step Number	Habitat Variable	Regression Equation	$r^2_{(pred)}$ (%)	*P*-value
1	%LOI	$\text{Richlog}_{10} = 1.040 + 0.209(\%LOI) - 0.040(\%LOI^2)$	15.27	0.000
2	BFWidth	$\text{Richlog}_{10} = 0.080 + 0.903(\%LOI) + 0.062(BFWidth) - 0.151(\%LOI^2) - 0.044(\%LOI)(BFWidth) + 0.007(\%LOI^2)(BFWidth)$	36.39	0.000
3	SRSi	$\text{Richlog}_{10} = 0.254 + 0.745(\%LOI) + 0.049(BFWidth) + 0.172(SRSi) - 0.130(\%LOI^2) - 0.034(\%LOI)(BFWidth) - 0.008(BFWidth)(SRSi) + 0.006(\%LOI^2)(BFWidth)$	47.32	0.000
4	Chla	$\text{Richlog}_{10} = 0.026 + 0.929(\%LOI) + 0.048(BFWidth) + 0.203(SRSi) + 0.020(Chla) - 0.168(\%LOI^2) - 0.033(\%LOI)(BFWidth) - 0.015(\%LOI)(Chla) - 0.010(BFWidth)(SRSi) + 0.006(\%LOI^2)(BFWidth) + 0.003(\%LOI^2)(Chla)$	58.33	0.000
5	%Eroding	$\text{Richlog}_{10} = 0.609 + 0.641(\%LOI) + 0.036(BFWidth) + 0.217(SRSi) - 0.005(Chla) - 0.007(\%Eroding) - 0.128(\%LOI^2) - 0.030(\%LOI)(BFWidth) + 0.002(\%LOI)(Chla) - 0.010(BFWidth)(SRSi) + 0.0004(BFWidth)(\%Eroding) + 0.005(\%LOI^2)(BFWidth)$	61.80	0.000

(continued)

Table 9 (continued).

Step Number	Habitat Variable	Regression Equation	$r^2_{(pred)}$ (%)	P-value
6	WetWidth	$Richlog_{10}$ = -0.045 + 1.100(%LOI) + 0.019(BFWidth) + 0.104(SRSi) – 0.011(Chla) – 0.006(%Eroding) + 0.161(WetWidth) – 0.180(%LOI²) – 0.016(%LOI)(BFWidth) + 0.002(%LOI)(Chla) - 0.122(%LOI)(WetWidth) – 0.011(BFwidth)(SRSi) + 0.0003(BFWidth)(%Eroding) + 0.023(SRSi)(WetWidth) + 0.001(Chla)(WetWidth) + 0.003(%LOI²)(BFWidth) + 0.016(%LOI²)(WetWidth)	65.95	0.000
Final Model		$Richlog_{10}$ = -0.111 + 1.141(%LOI) + 0.019(BFWidth) + 0.0.090(SRSi) – 0.012(Chla) – 0.005(%Eroding) + 0.173(WetWidth) – 0.185(%LOI²) – 0.015(%LOI)(BFWidth) + 0.002(%LOI)(Chla) - 0.131(%LOI)(WetWidth) – 0.010(BFwidth)(SRSi) + 0.0002(BFWidth)(%Eroding) + 0.022(SRSi)(WetWidth) + 0.001(Chla)(WetWidth) + 0.003(%LOI²)(BFWidth) + 0.017(%LOI²)(WetWidth)	69.19	0.000

Table 10. Richness ($\log_{10}$) predictive model terms, coefficients, and p-values. Habitat variable abbreviations are expanded in Table 5.

Model Variables	Coefficicent	p-value
Constant	-0.11070	0.826
%LOI	1.14144	0.002
BFWidth	0.01864	0.136
SRSi	0.08967	0.045
Chla	-0.01169	0.001
%Eroding	-0.00525	0.011
WetWidth	0.17343	0.017
%LOI^2	-0.18544	0.002
(%LOI)(BFWidth)	-0.01517	0.071
(%LOI)(Chla)	0.00198	0.005
(%LOI)(WetWidth)	-0.13058	0.006
(BFWidth)(SRSi)	-0.01000	0.000
(BFWidth)(%Eroding)	0.00024	0.032
(SRSi)(WetWidth)	0.02191	0.000
(Chla)(WetWidth)	0.00128	0.001
(%LOI)^2(BFWidth)	0.00295	0.028
(%LOI)^2(WetWidth)	0.01697	0.020

Table 11. The distributions of the residuals for the reference sites calculated from the Richness($\log_{10}$) predictive model. The decision criteria (*) determine the upper and lower boundaries for test sites being categorized as impacted or unimpacted. Test site residuals lower than the 12.5 percentile and greater than then 87.5 percentile of the reference sites are impacted.

Descriptor	Reference Site Residual
Minimum	-0.08466
12.5 Percentile	-0.03995*
25th Percentile	-0.01906
Median	0.00203
75th Percentile	0.01899
87.5 Percentile	0.04395*
Maximum	0.06779
Range	0.15245
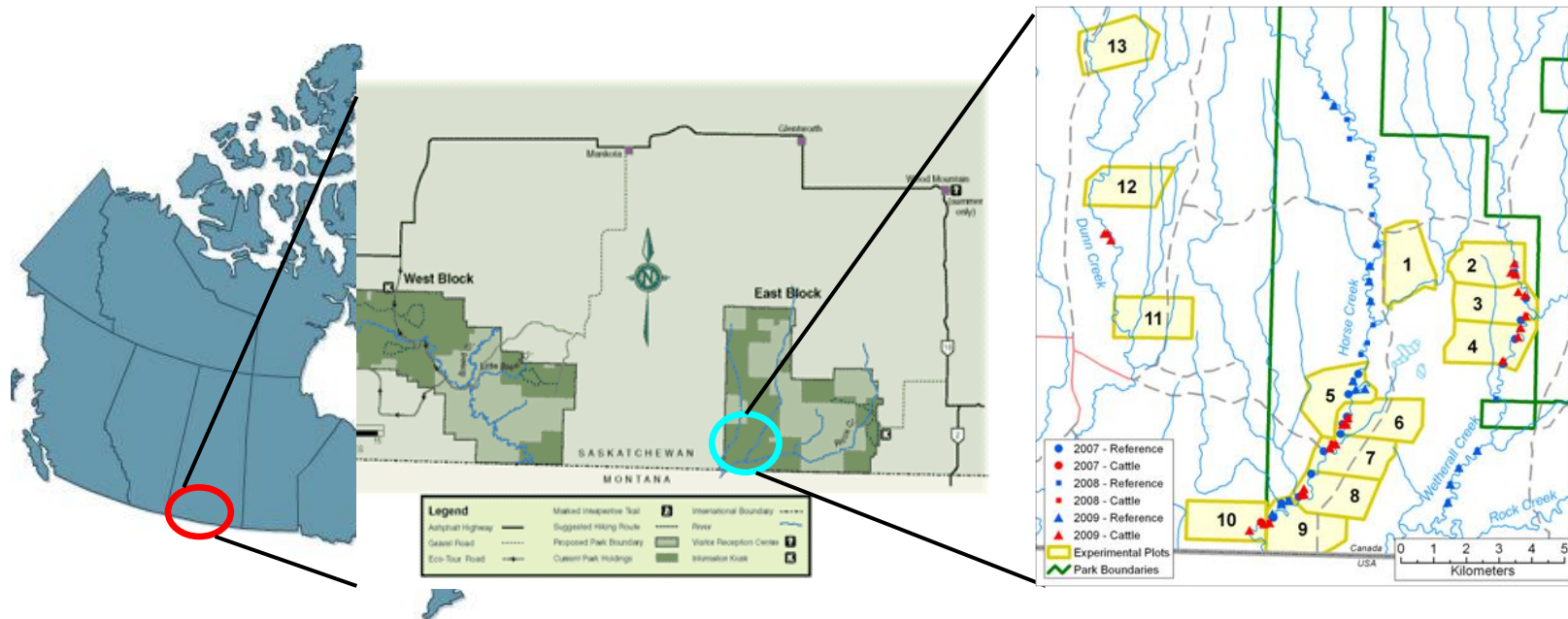
Figure 1. Grasslands National Park of Canada is circled in red, with the magnified map of the park showing the Biodiversity and Grazing Management Area circled in blue. Sampling site locations are identified in the right panel. *Map adapted from Parks Canada. Permission to use map was granted August 14, 2104 by Parks Canada. Accessed August 14, 2014 at http://www.pc.gc.ca/eng/pn-np/sk/grasslands/visit/visit9/b.aspx*
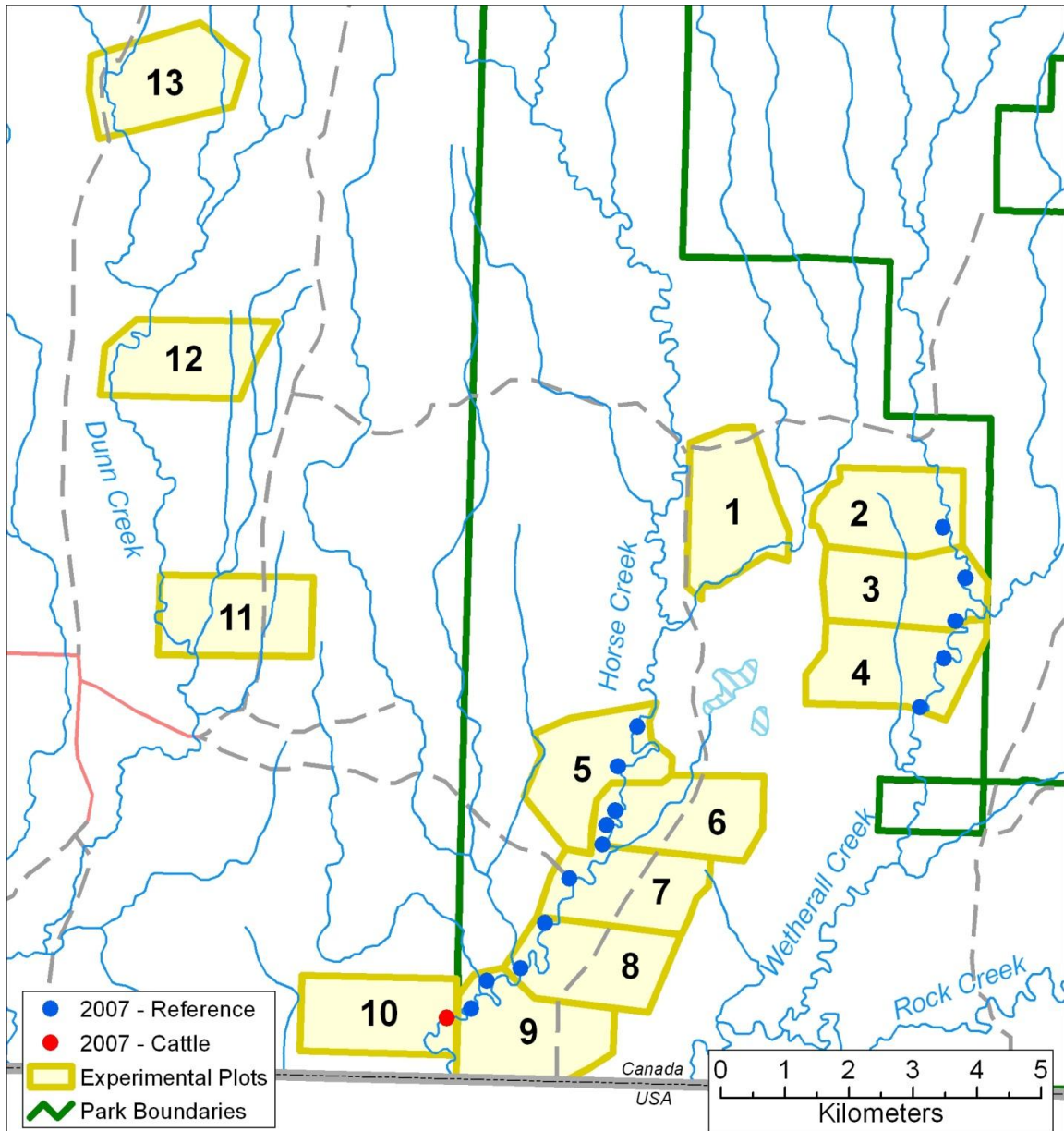
Figure 2. The 2007 reference (●) and test (●) sites. In 2007, cattle were present only in the community pasture (pasture 10).
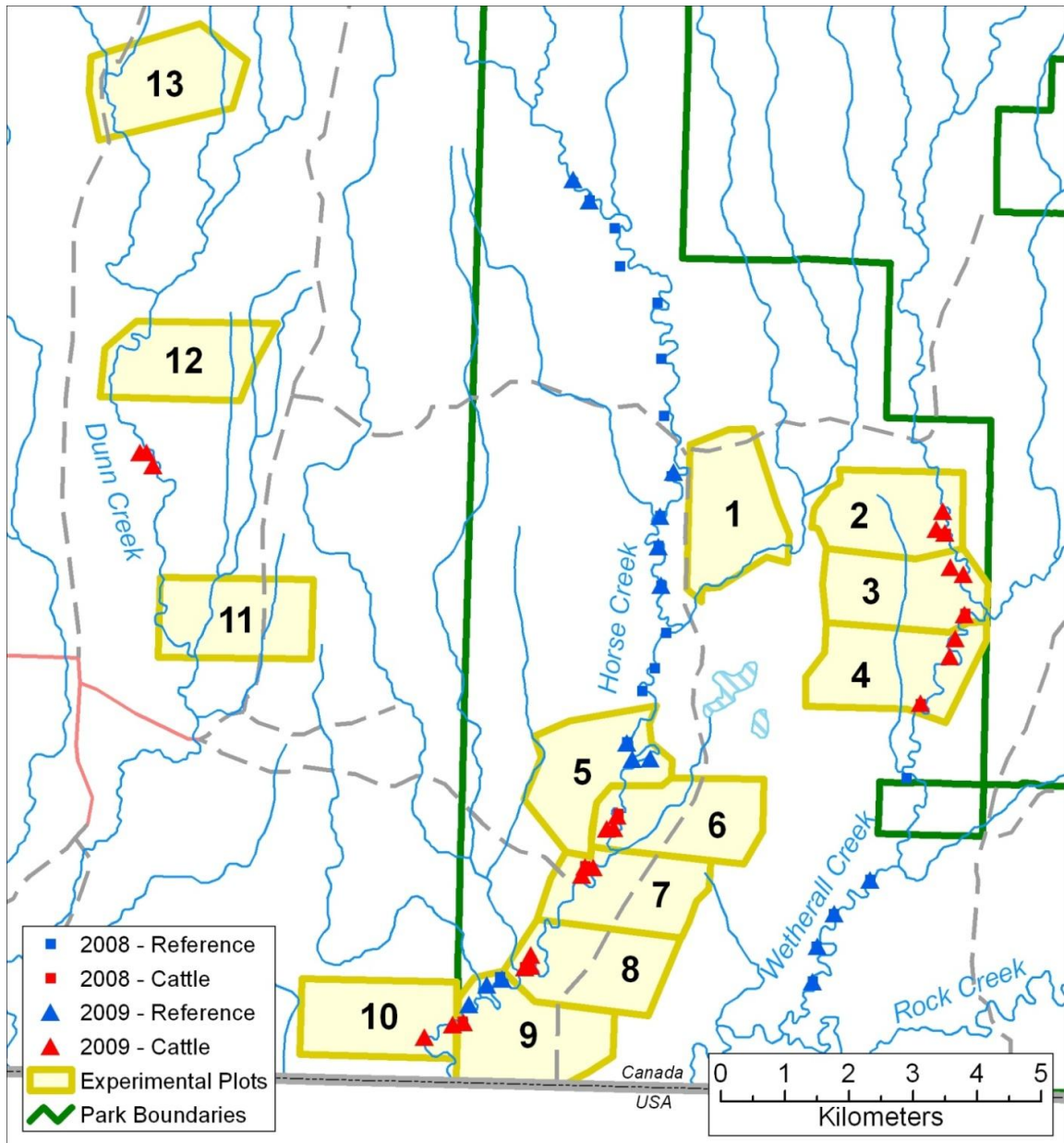
Figure 3. The 2008 (■) and 2009 (▲) reference (blue) and test (red) sites. Cattle were placed in the experimental pastures in 2008 and 2009, except for pastures 5 and 9, which remained ungrazed.
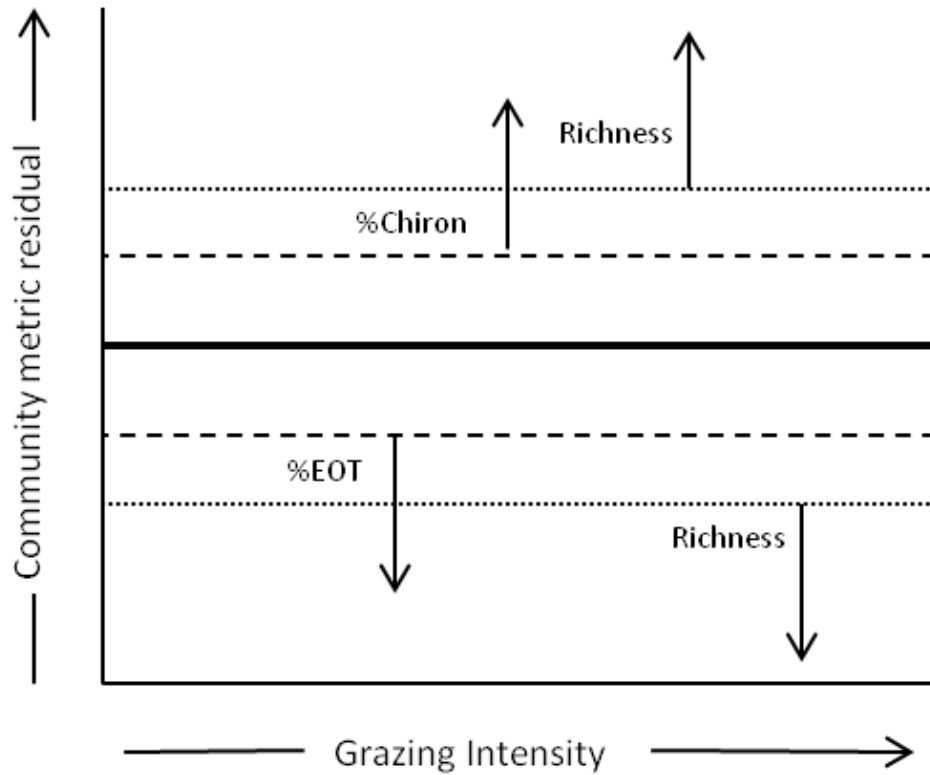
Figure 4. The median (―――), first and third quartiles (‒ ‒ ‒), and 12.5 and 87.5 percentiles (………) of the reference site residuals act as the pass-fail boundaries for the test sites. Test sites failed if: (1) %Chiron residuals were greater than the third quartile; (2) %EOT residuals were less than the first quartile; or (3) richness residuals were less than the 12.5 percentile or greater than the 87.5 percentile.
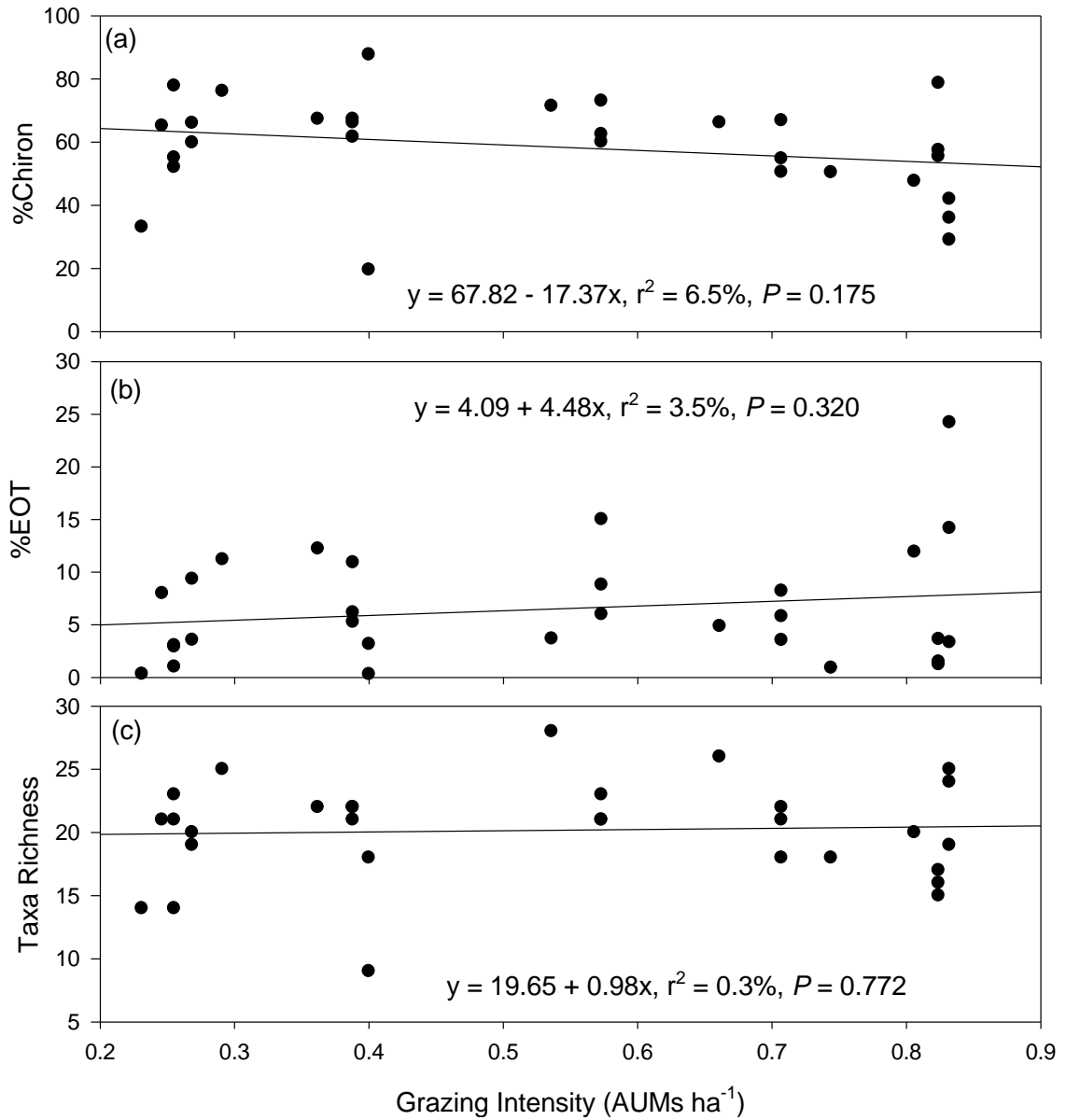
Figure 5. Linear regressions of %Chiron (a), %EOT (b), and taxa richness (c) with grazing intensity. Habitat variable abbreviations are expanded in Table 5.
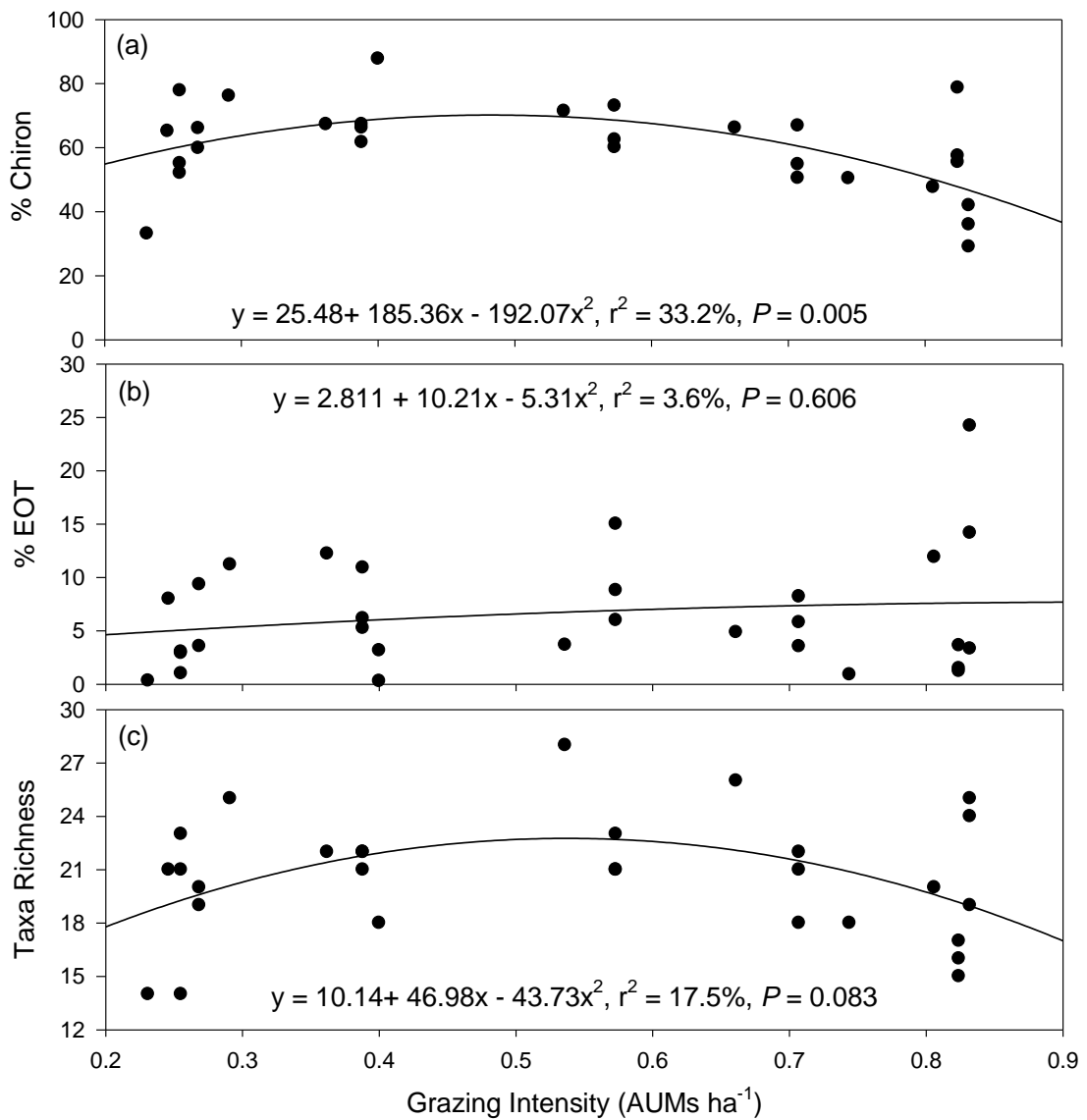
Figure 6. Non-linear regressions of %Chiron (a), %EOT (b), and taxa richness (c) with grazing intensity. Habitat variable abbreviations are expanded in Table 5.
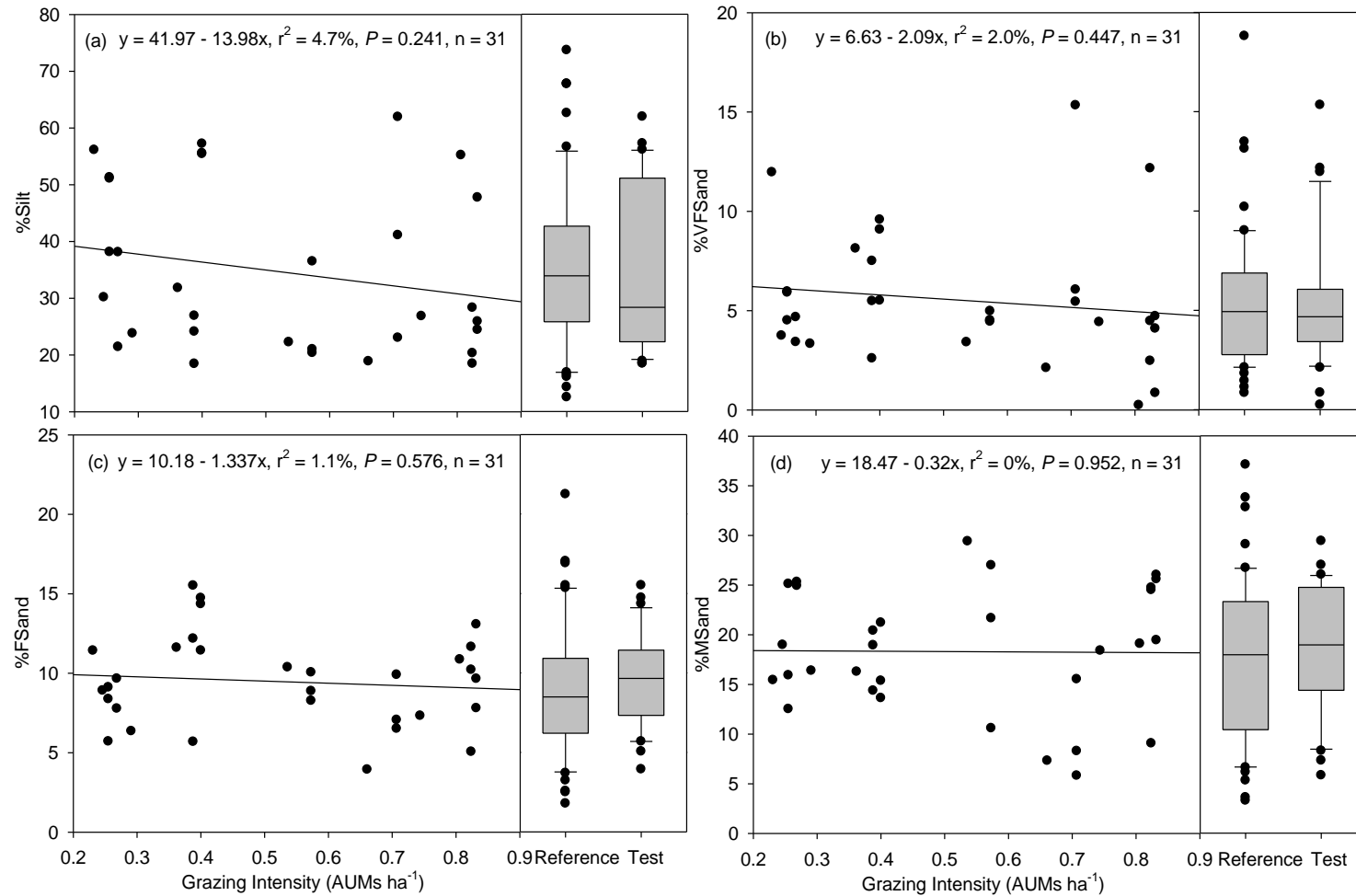
Figure 7. Regression of sediment variables with grazing intensity and boxplots depicting the distribution of these variables found at the reference and test sites for %Silt (a), %VFSand (b), %FSand (c), and %MSand (d). Habitat variable abbreviations are expanded in Table 5.
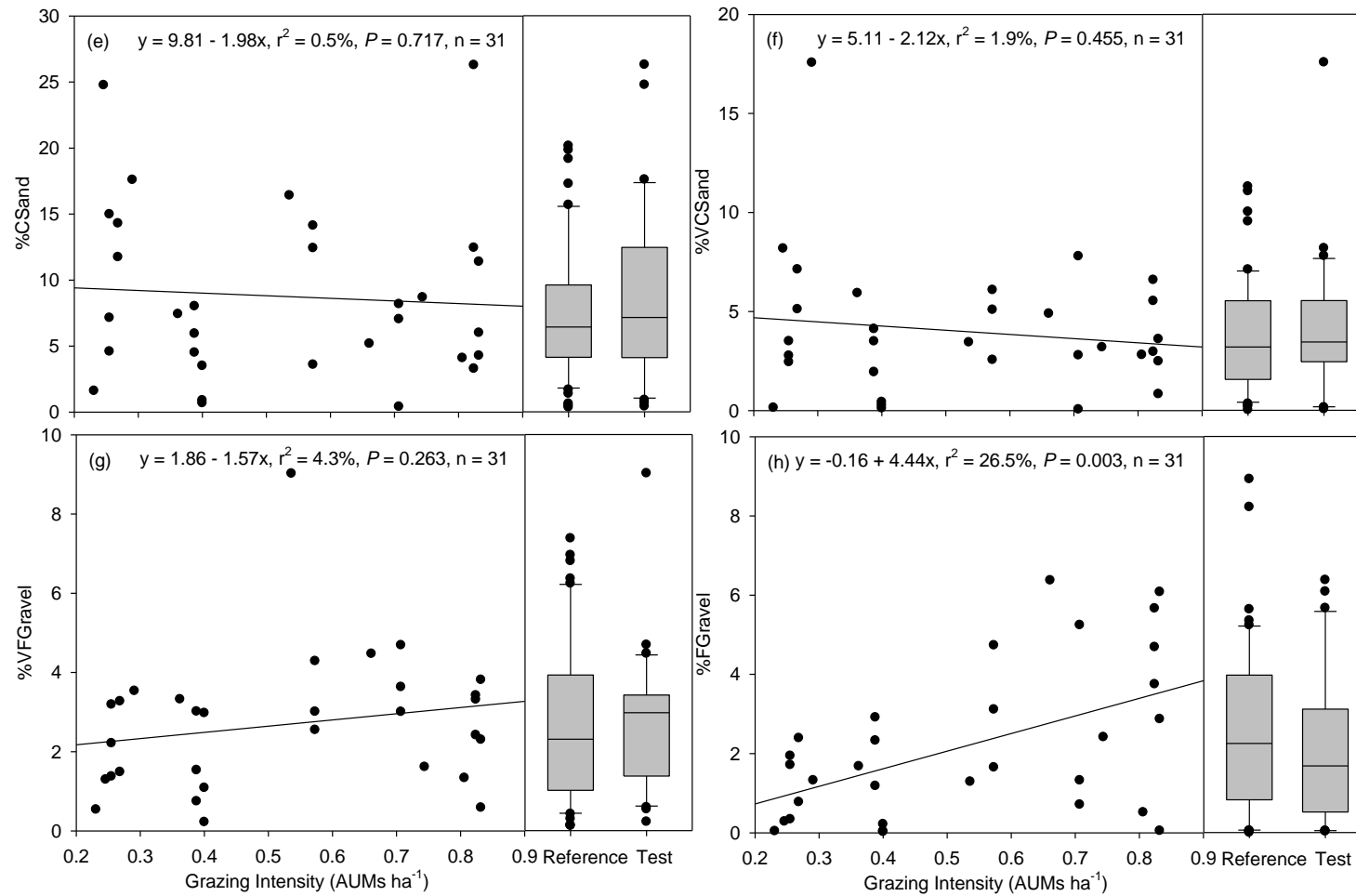
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Figure 7 (continued). Regression of sediment variables with grazing intensity and boxplots depicting the distribution of these variables found at the reference and test sites for %CSand (e), %VCSand (f), %VFGravel (g), and %FGravel (h). Habitat variable abbreviations are expanded in Table 5.
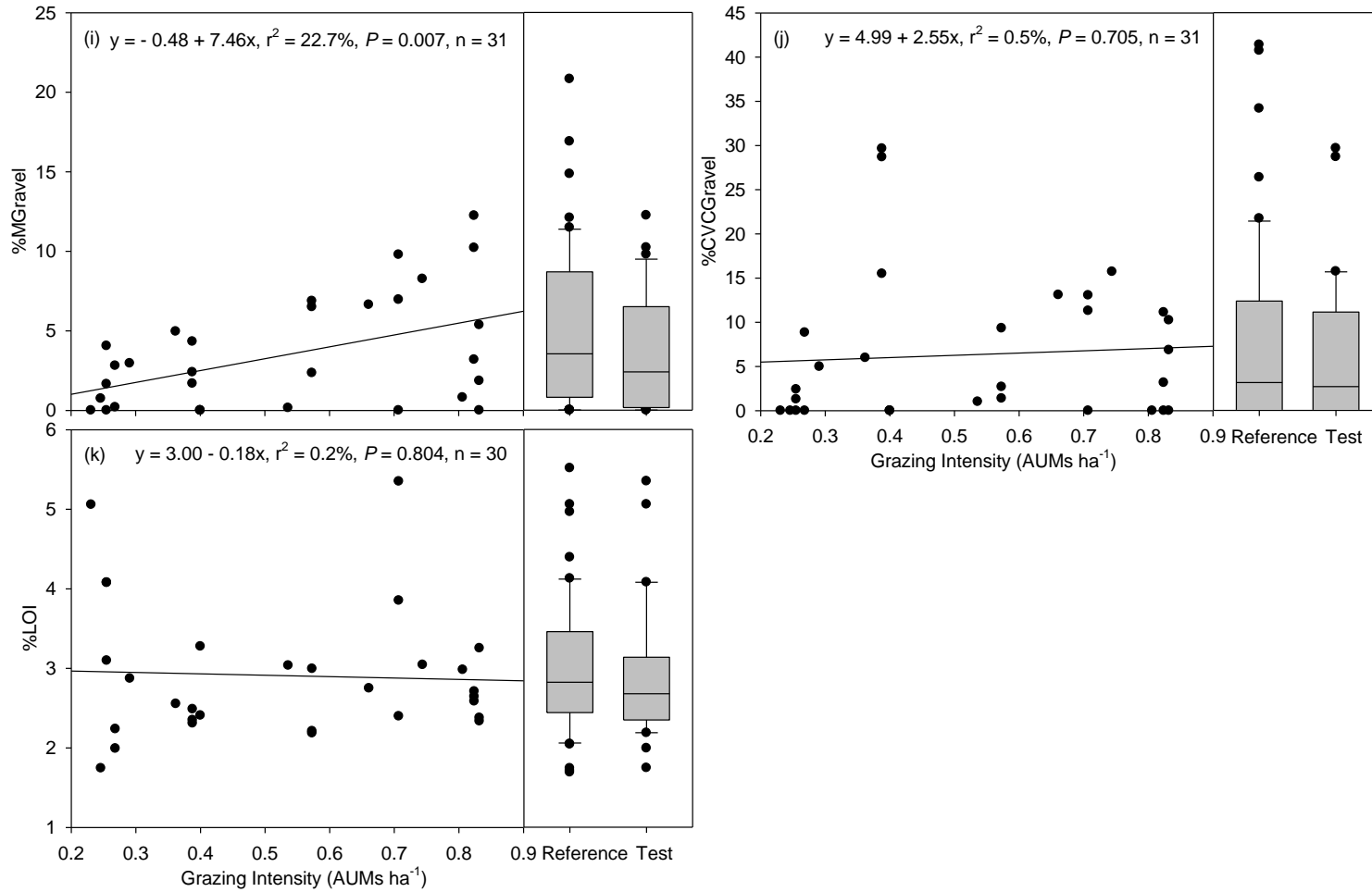
(i) $y = -0.48 + 7.46x$, $r^2 = 22.7\%$, $P = 0.007$, $n = 31$

(j) $y = 4.99 + 2.55x$, $r^2 = 0.5\%$, $P = 0.705$, $n = 31$

(k) $y = 3.00 - 0.18x$, $r^2 = 0.2\%$, $P = 0.804$, $n = 30$

Figure 7 (continued). Regression of sediment variables with grazing intensity and boxplots depicting the distribution of these variables found at the reference and test sites for %MGravel (i), %CVCGravel (j), and %LOI (k). Habitat variable abbreviations are expanded in Table 5.
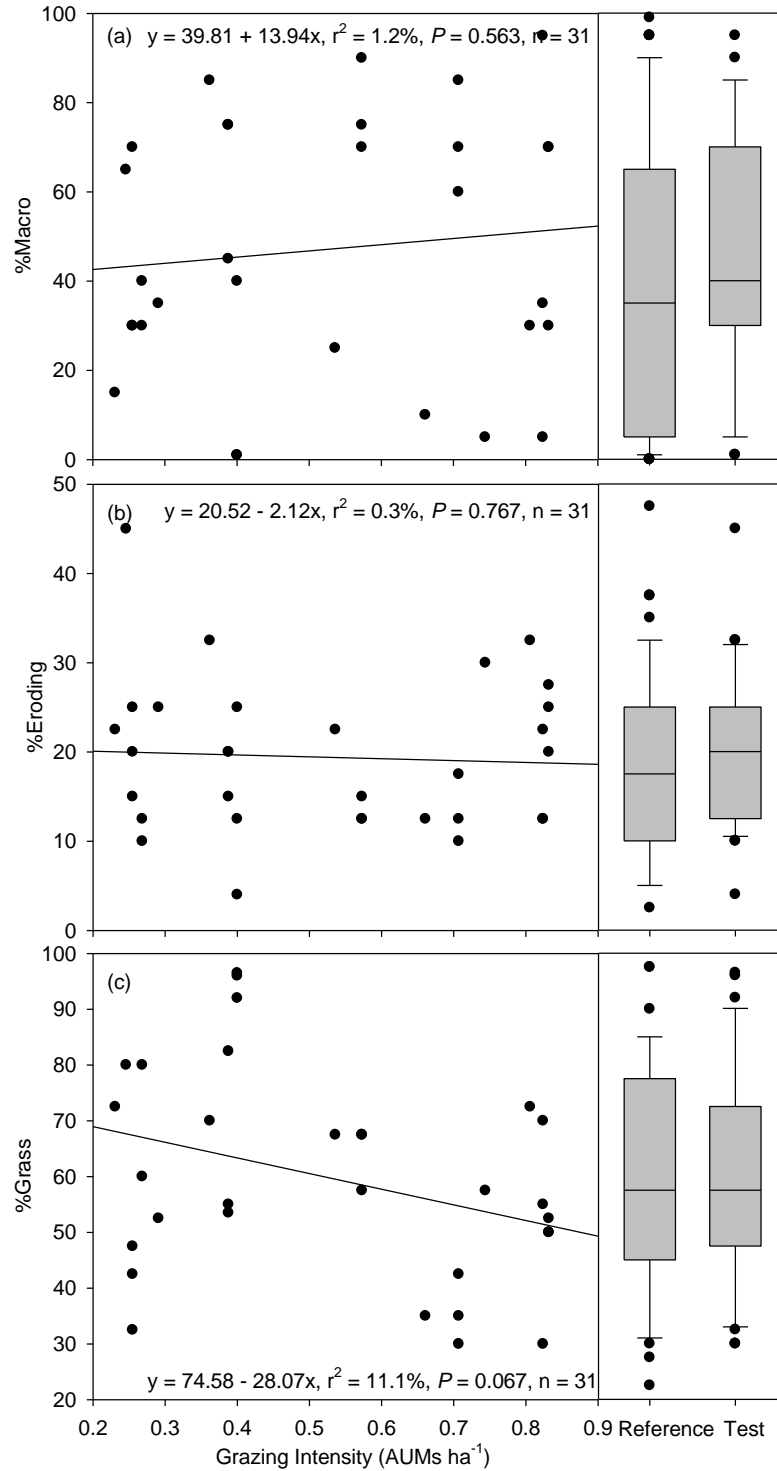
Figure 8. Regression of streambank and stream variables with grazing intensity and boxplots depicting the distribution of these variables found at the reference and test sites for %Macro (a), %Eroding (b), and %Grass (c). Habitat variable abbreviations are expanded in Table 5.
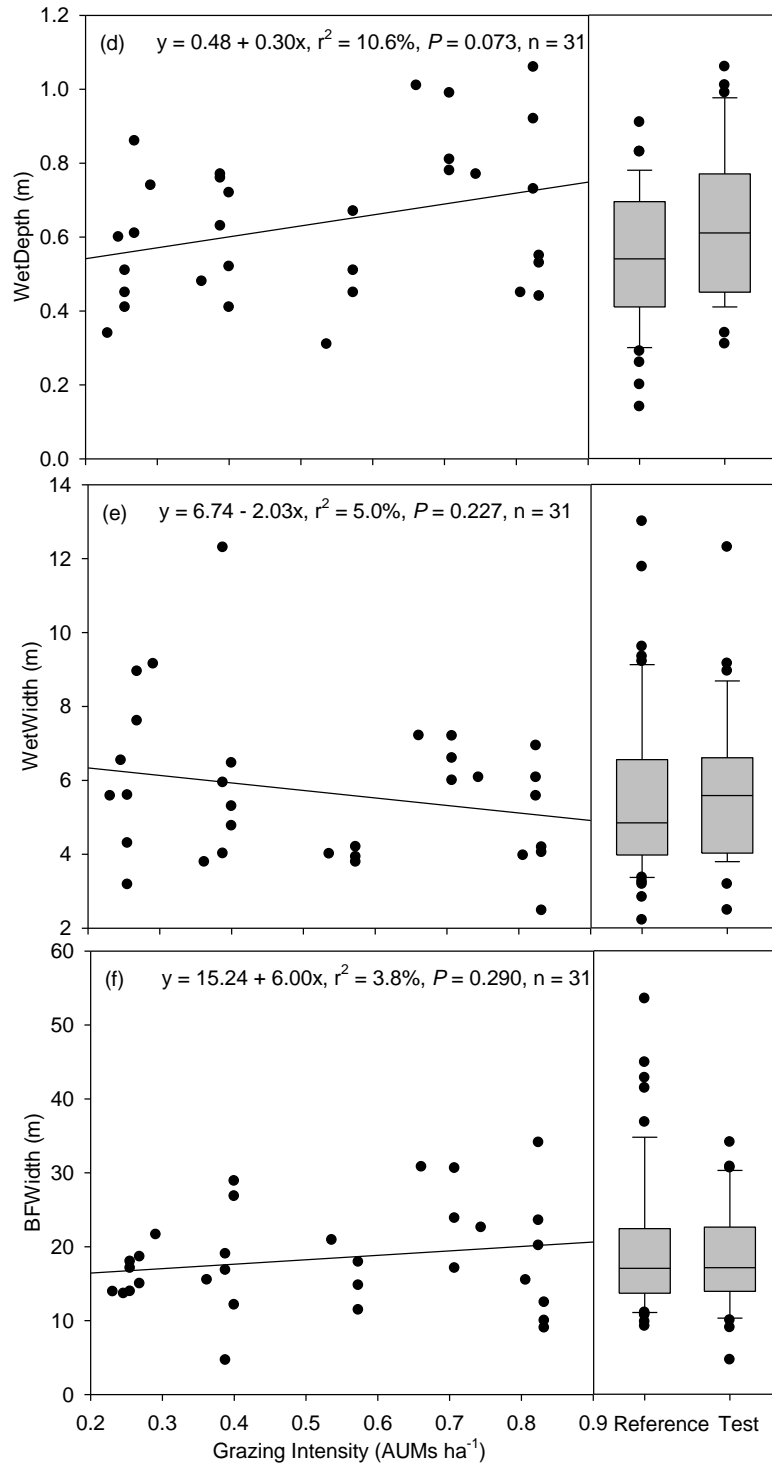
Figure 8 (continued). Regression of streambank and stream variables with grazing intensity and boxplots depicting the distribution of these variables found at the reference and test sites (d), WetWidth (e), and BFWidth (f). Habitat variable abbreviations are expanded in Table 5.

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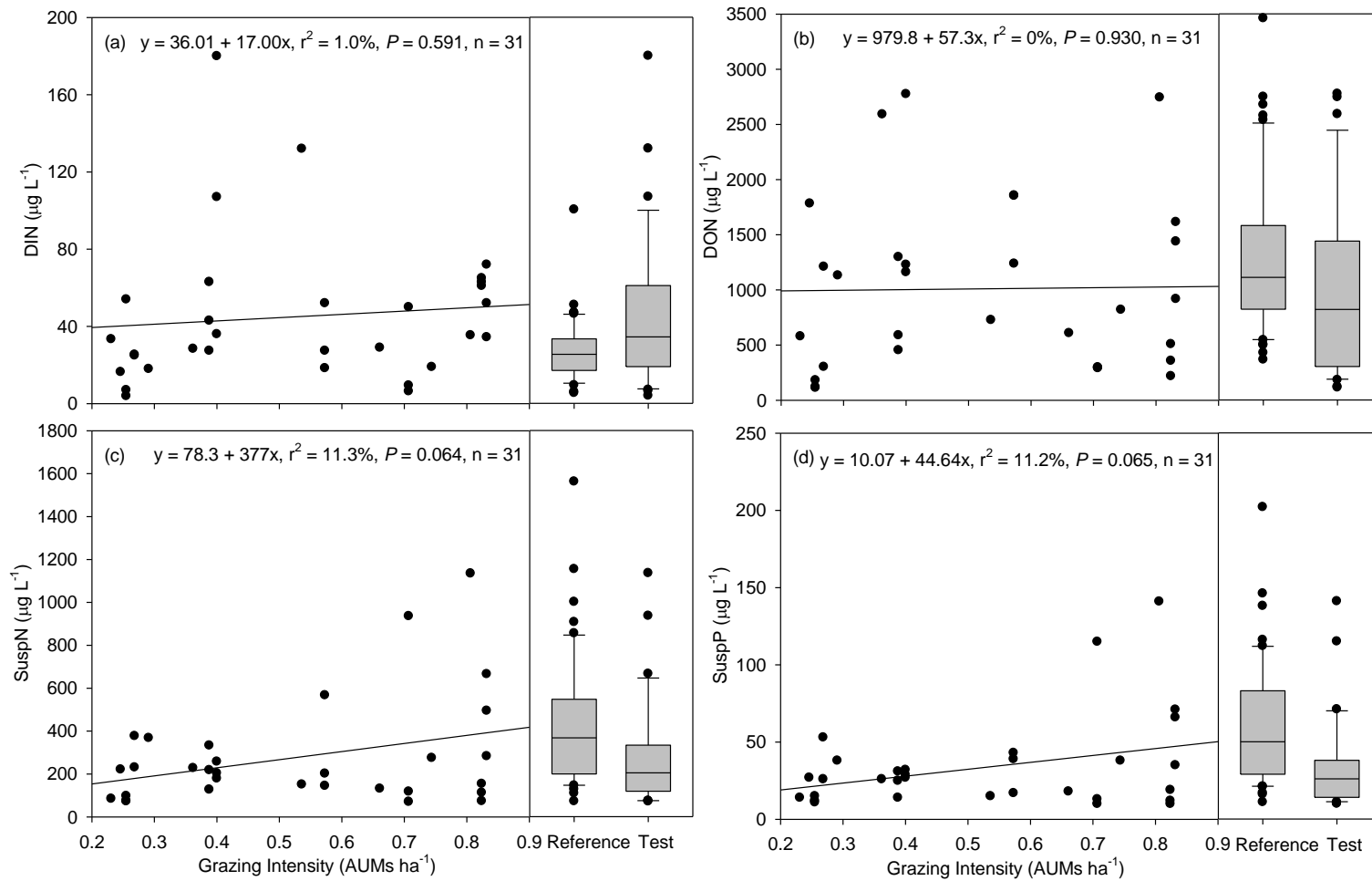
Figure 9. Regression of water chemistry variables with grazing intensity and boxplots depicting the distribution of the concentrations found at the reference and test sites for DIN (a), DON (b), SuspN (c), and SuspP (d). Habitat variable abbreviations are expanded in Table 5.
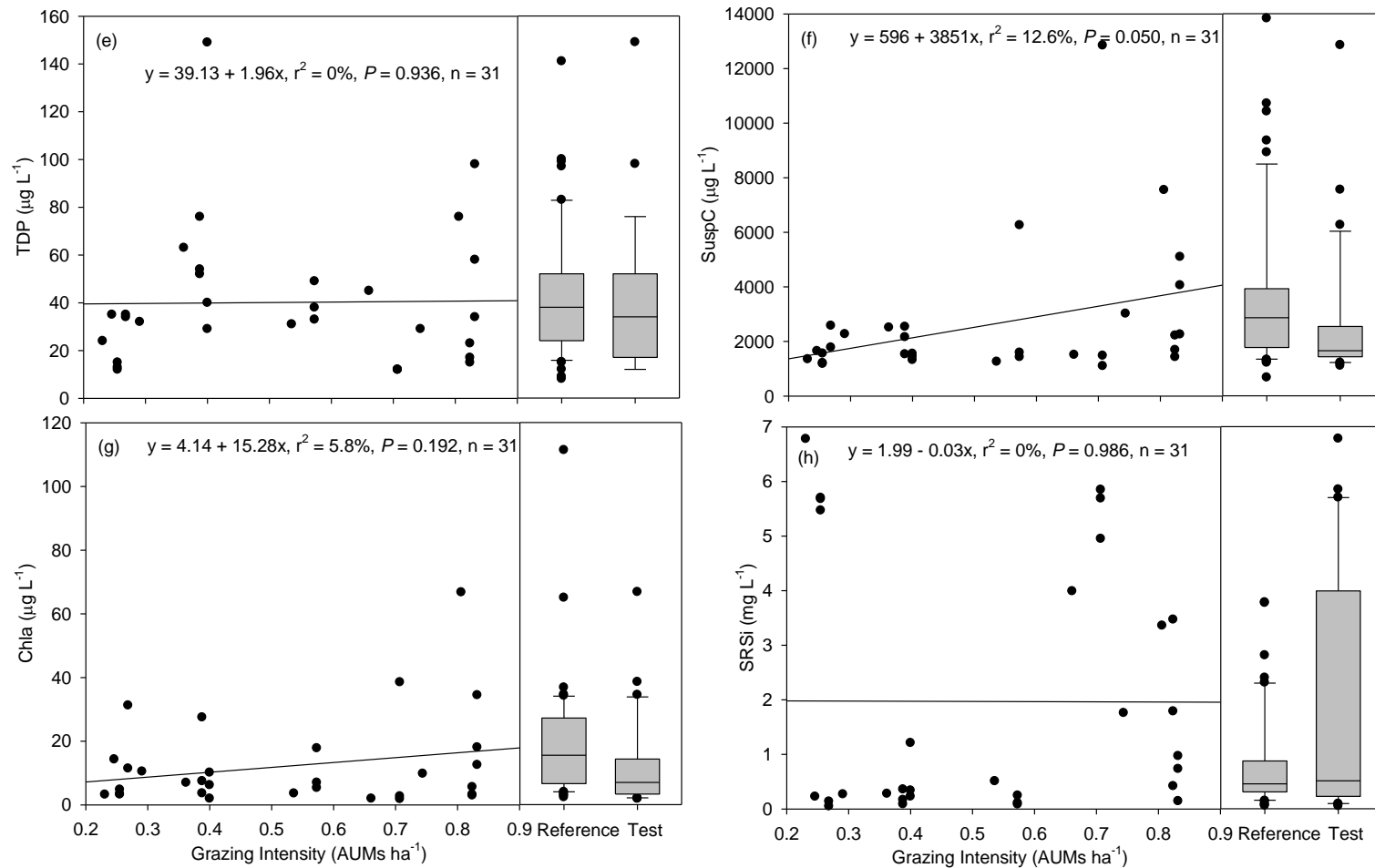
Figure 9 (continued). Regression of water chemistry variables with grazing intensity and boxplots depicting the distribution of the concentrations found at the reference and test sites for TDP (e), SuspC (f), Chla (g), and SRSi (h). Habitat variable abbreviations are expanded in Table 5.
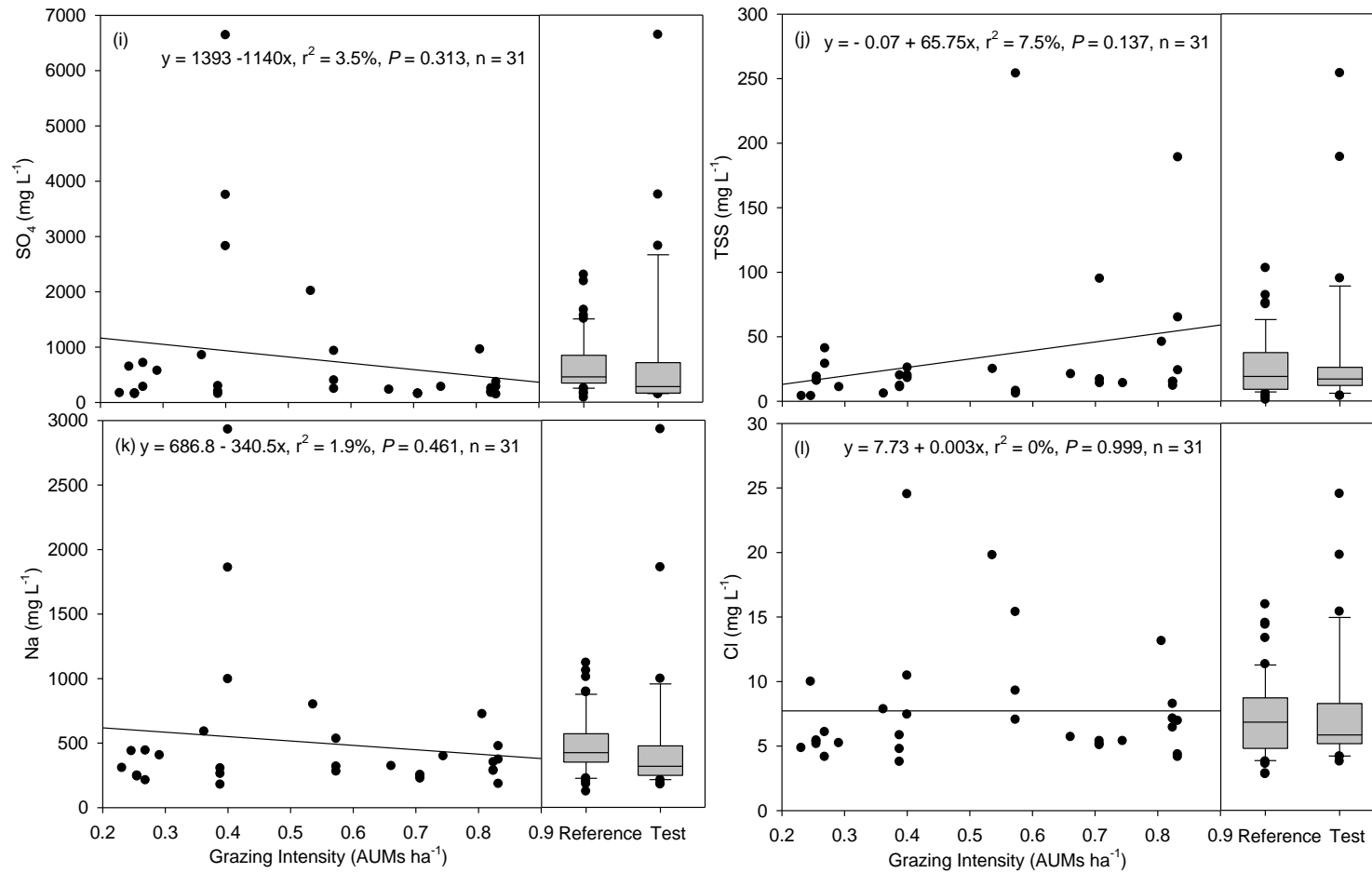
Figure 9 (continued). Regression of water chemistry variables with grazing intensity and boxplots depicting the distribution of the concentrations found at the reference and test sites for SO_4 (i), TSS (j), Na (k), and Cl (l). Habitat variable abbreviations are expanded in Table 5.
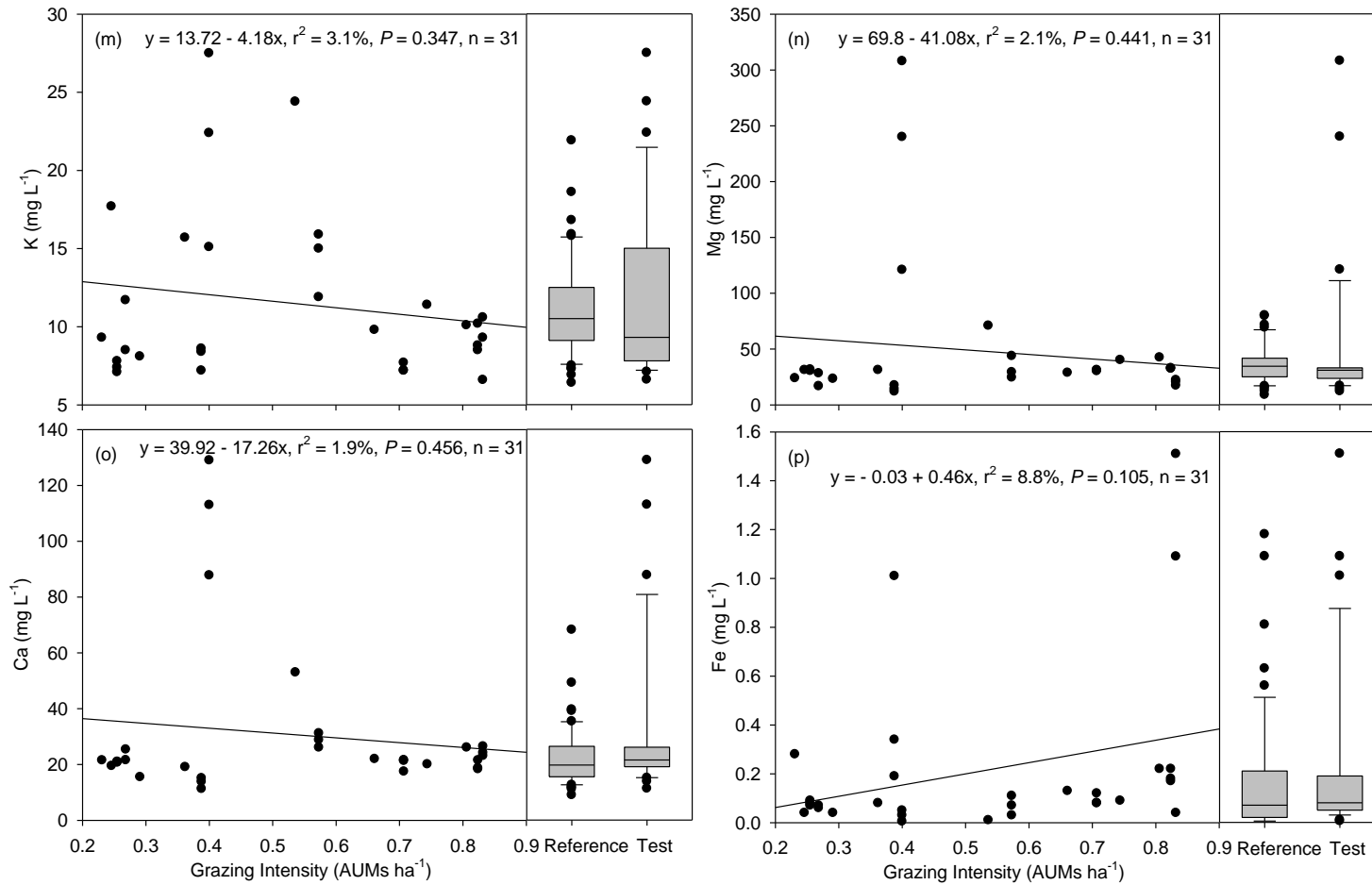
Figure 9 (continued). Regression of water chemistry variables with grazing intensity and boxplots depicting the distribution of the concentrations found at the reference and test sites for K (m), Mg (n), Ca (o), and Fe (p). Habitat variable abbreviations are expanded in Table 5.
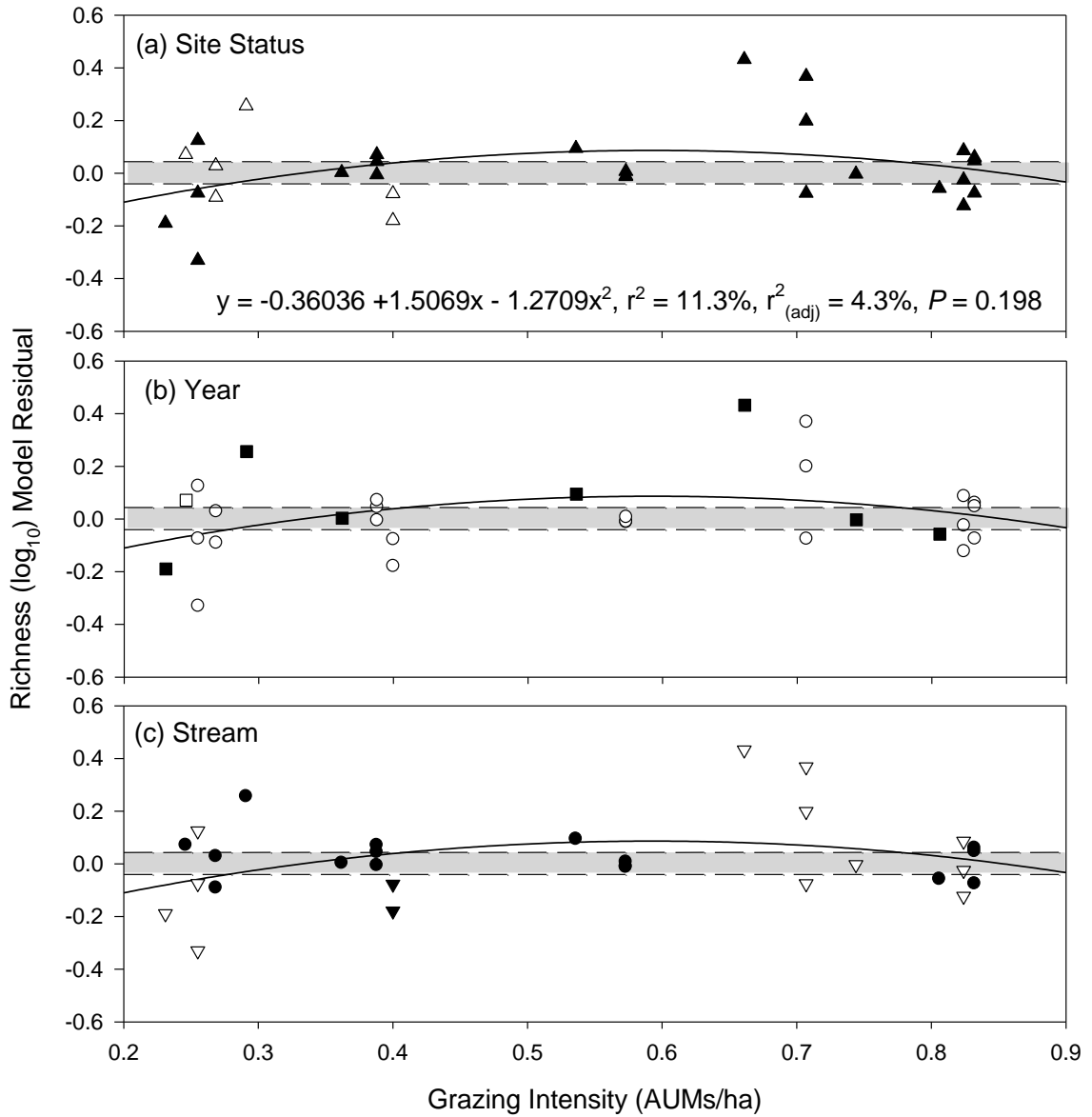
Figure 10. The passing (within grey bar) and failing (outside grey bar) test site Richness($\log_{10}$) residuals in relation to grazing intensity (AUMs ha^{-1}). The 12.5 and 87.5 percentiles of the reference site distributions represent the pass-fail boundary (‒ ‒ ‒ ‒). The site status graph (a) differentiates long term grazing sites ($\triangle$) from experimental pastures ($\blacktriangle$). The year of sampling graph (b) separates 2007 ($\square$), 2008($\blacksquare$), and 2009 ($\circ$). The stream on which each site is located is identified in (c) as Horse Creek ($\bullet$), Wetherall Creek ($\triangledown$), or Dunn Creek($\blacktriangledown$).
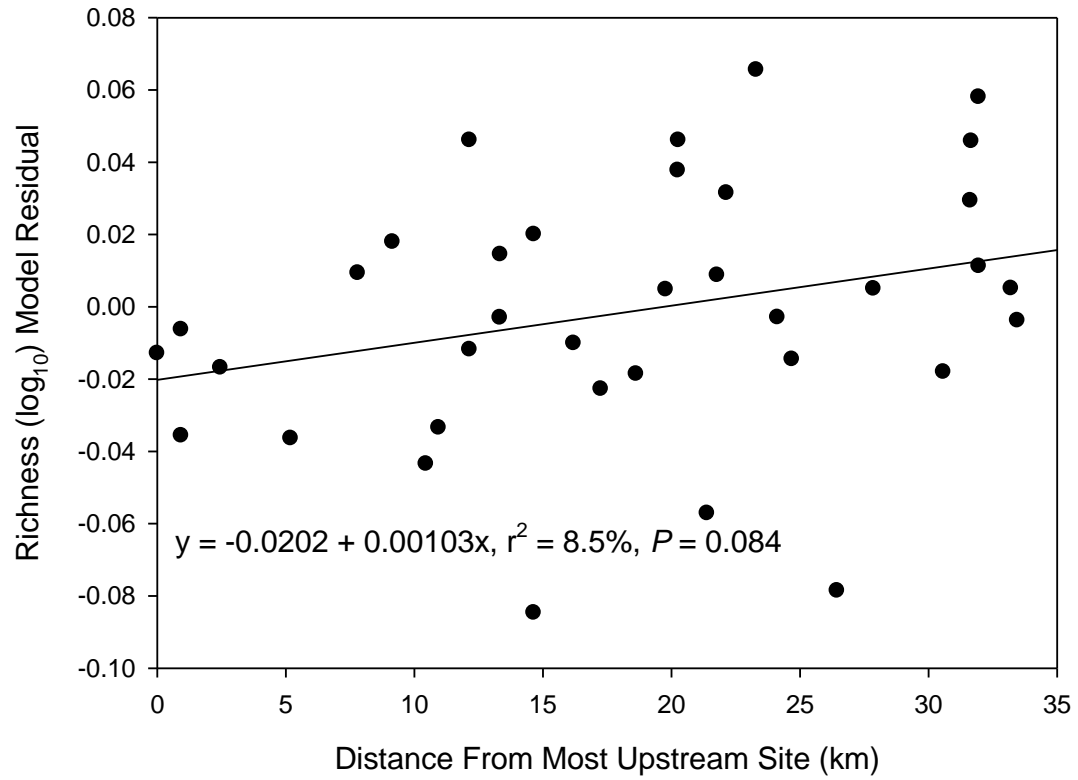
Figure 11. Regression (———) of the Horse Creek reference sites residuals from the Richness(log$_{10}$) model with distance from the most upstream site on Horse Creek (km).
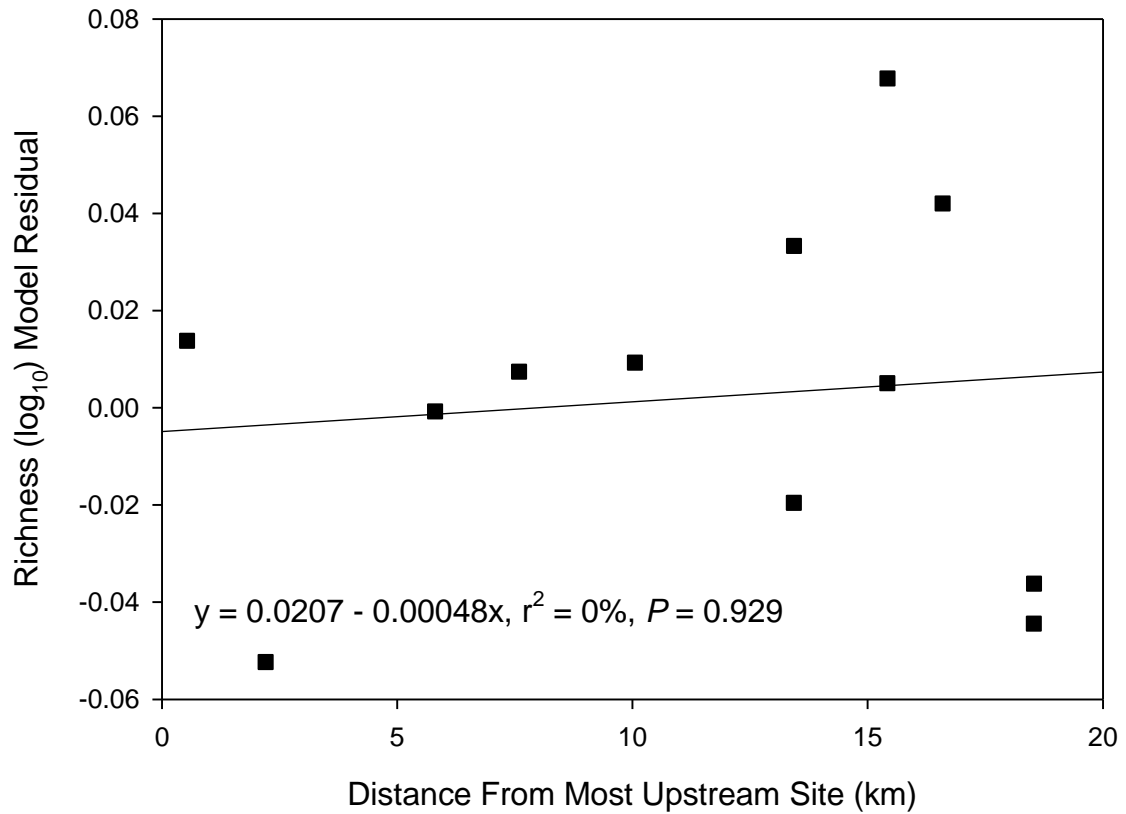
Figure 12. Regression (———) of the Wetherall Creek reference sites residuals from the Richness($\log_{10}$) model with distance from the most upstream site on Wetherall Creek (km).
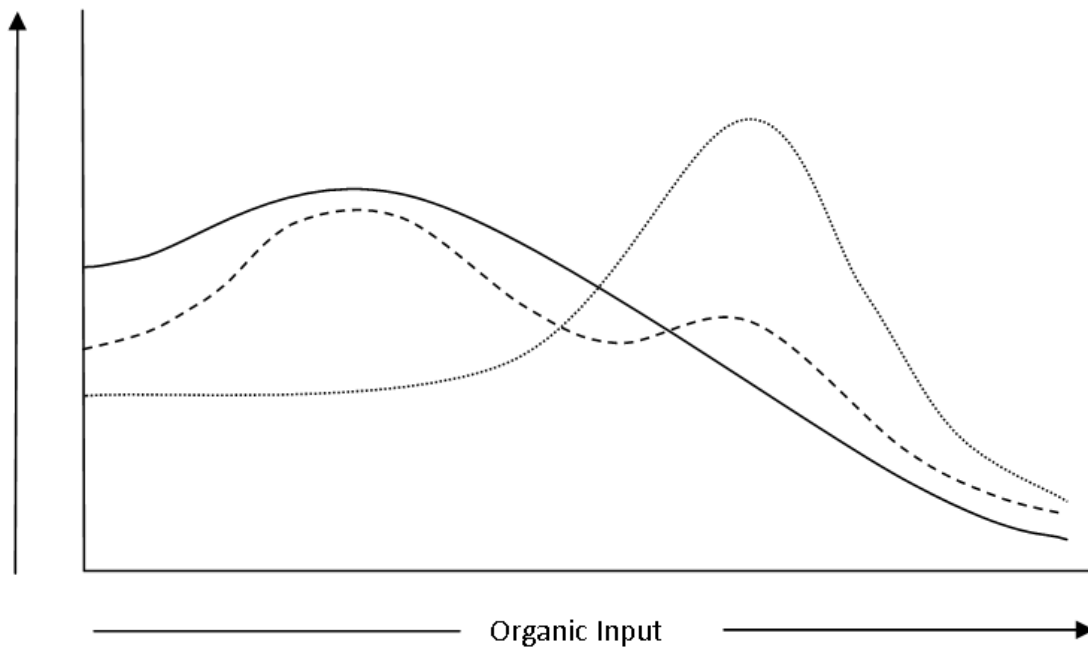
Figure 13. The Pearson-Rosenberg model for organic enrichment depicting the expected response of taxa richness (___), abundance (..........) and biomass(- - - - -). Based on Pearson and Rosenberg (1978).
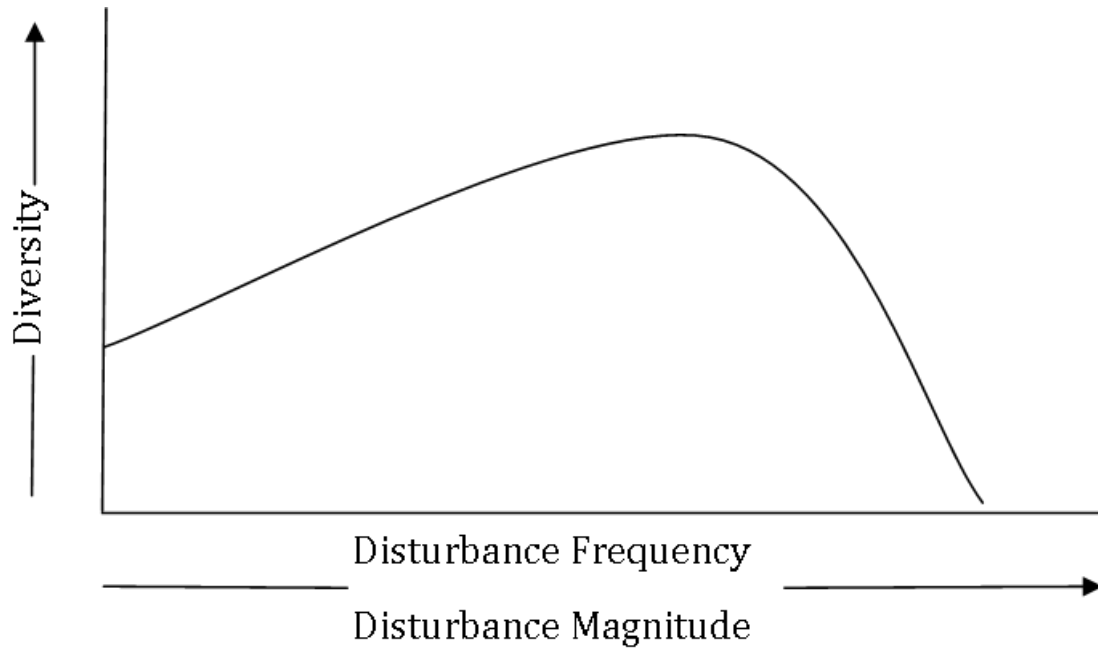
Figure 14. The intermediate disturbance hypothesis indicates that as the frequency or magnitude of a disturbance increases, there will be a peak in taxa diversity, followed by a steep decline. This peak occurs because of the presence of resident and colonizing taxa. The decline occurs when the resident taxa can no longer persist against the stressor. Based on Connell (1978).

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Appendices

Appendix 1. The r^2 and P-values from regressing each habitat descriptor with grazing intensity for the test sites. Significant values ($P < 0.05$) are indicated by an asterisk (*). Habitat variable abbreviations are expanded in Table 5.

Habitat Variable	r^2 (%)	P-value
%Silt	4.7	0.241
%VFSand	2.0	0.447
%FSand	1.1	0.576
%MSand	0	0.952
%CSand	0.5	0.717
%VCSand	1.9	0.455
%VFGravel	4.3	0.263
%FGravel	26.5	0.003*
%MGravel	22.7	0.007*
%CVCGravel	0.5	0.705
%LOI	0.2	0.804
%Macro	1.2	0.563
%Eroding	0.3	0.767
%Grass	11.1	0.067
WetDepth	10.6	0.073
WetWidth	5.0	0.227
BFWidth	3.8	0.290
DIN	1.0	0.591
DON	0	0.930
SuspN	11.3	0.064
SuspP	11.2	0.065
TDP	0	0.936
SuspC	12.6	0.050*
Chla	5.8	0.192
SRSi	0	0.986
SO_4	3.5	0.313
TSS	3.9	0.151
Na	1.9	0.461
Cl	0	0.999
K	3.1	0.347
Mg	2.1	0.441
Ca	1.9	0.456
Fe	8.8	0.105

Appendix 2. The descriptors of the reference site sediment variables, including the mean, median, range, standard deviation (StDev), and the number of sites (n). For full name of habitat variables, see Table 5.

Year	Descriptor	%Silt	%VFSand	%FSand	%MSand	%CSand	%VCSand	%VFGravel	%FGravel
2007	Mean	36.21	5.25	9.18	16.71	7.3	2.84	1.79	1.94
	StDev	16.21	3.06	3.49	7.27	5.14	2.39	1.14	1.67
	Median	36.99	5.093	8.81	16.43	5.92	2.38	1.72	1.58
	Range	59.36	12.00	13.72	22.88	19.21	9.78	3.43	5.2
	n	16	16	16	16	16	16	16	16
2008	Mean	34.82	5.21	8.82	17.99	8.01	4.23	3.52	2.43
	StDev	12.94	3.06	4.81	9.34	5.14	3.00	2.19	1.74
	Median	32.9	5.00	7.4	18.51	6.96	4.55	3.66	2.36
	Range	51.61	12.64	18.68	31.79	18.68	11.04	6.68	5.61
	n	19	19	19	19	19	19	19	19
2009	Mean	33.78	5.85	8.95	16.63	7.45	4.24	2.83	3.10
	StDev	15.26	4.14	3.89	8.11	4.76	2.91	2.27	2.79
	Median	31.03	4.75	9.18	16.15	6.32	4.50	2.00	2.32
	Range	55.22	16.69	14.42	25.79	19.79	11.10	6.87	8.92
	n	16	16	16	16	16	16	16	16
All	Mean	34.93	5.42	8.97	17.16	7.61	3.80	2.76	2.49
	StDev	14.49	3.38	4.07	8.21	4.93	2.82	2.05	2.12
	Median	33.89	4.94	8.50	17.96	6.43	3.20	2.31	2.25
	Range	61.15	17.96	19.46	33.82	19.79	11.27	7.25	8.92
	n	51	51	51	51	51	51	51	51

(continued)

Appendix 2 (continued).

Year	Descriptor	%MGravel	%CVCGravel	%LOI
2007	Mean	5.5	8.21	2.82
	StDev	5.84	13.25	0.75
	Median	3.26	0.86	2.69
	Range	16.88	41.36	2.36
	n	16	16	15
2008	Mean	3.78	6.58	3.05
	StDev	3.18	8.35	0.94
	Median	4.12	2.91	2.80
	Range	9.46	26.36	3.02
	n	19	19	19
2009	Mean	5.72	9.35	3.12
	StDev	5.52	10.26	0.88
	Median	4.11	6.83	2.96
	Range	20.81	40.69	3.77
	n	16	16	16
All	Mean	4.93	7.96	3.00
	StDev	4.88	10.52	0.86
	Median	3.54	3.14	2.82
	Range	20.81	41.36	3.82
	n	51	51	50

Appendix 3. The descriptors of the test site sediment variables, including the mean, median, range, standard deviation (StDev), and the number of sites (n). For full name of habitat variables, see Table 5.

Year	Descriptor	%Silt	%VFSand	%FSand	%MSand	%CSand	%VCSand	%VFGravel	%FGravel
2007	Mean	30.18	3.74	8.92	18.99	24.77	8.18	1.30	0.29
	StDev	-	-	-	-	-	-	-	-
	Median	30.18	3.74	8.92	18.99	24.77	8.18	1.30	0.29
	Range	-	-	-	-	-	-	-	-
	n	1	1	1	1	1	1	1	1
2008	Mean	33.58	4.81	8.85	17.48	8.72	5.43	3.41	1.95
	StDev	15.62	3.97	2.98	6.53	6.11	5.65	2.84	2.10
	Median	26.86	3.42	10.38	16.38	7.43	3.44	3.32	1.32
	Range	37.24	11.73	7.67	22.09	15.98	17.42	8.48	6.33
	n	7	7	7	7	7	7	7	7
2009	Mean	35.10	5.82	9.68	18.53	8.08	3.38	2.53	2.34
	StDev	14.66	3.19	2.93	6.49	6.01	2.28	1.20	1.92
	Median	28.35	4.98	9.66	19.46	7.04	2.97	2.98	1.94
	Range	43.48	14.48	10.45	21.18	25.87	7.74	4.46	6.06
	n	23	23	23	23	23	23	23	23
All	Mean	34.60	5.52	9.47	18.31	8.76	4.00	2.69	2.18
	StDev	14.40	3.30	2.86	6.29	6.55	3.40	1.70	1.93
	Median	28.35	4.68	9.66	18.95	7.14	3.44	2.98	1.68
	Range	43.48	15.10	11.58	23.59	25.87	17.51	8.80	6.35
	n	31	31	31	31	31	31	31	31

(continued)

Appendix 3 (continued).

Year	Descriptor	%MGravel	%CVCGravel	%LOI
2007	Mean	0.74	0.00	1.74
	StDev	-	-	-
	Median	0.74	0.00	1.74
	Range	-	-	-
	n	1	1	1
2008	Mean	3.40	5.83	3.18
	StDev	3.31	6.36	0.84
	Median	2.96	4.99	2.98
	Range	8.26	15.72	2.50
	n	7	7	7.0
2009	Mean	3.59	6.76	2.87
	StDev	3.66	8.68	0.83
	Median	2.40	2.68	2.54
	Range	12.25	29.64	3.36
	n	23	23	22
All	Mean	3.46	6.33	2.91
	StDev	3.50	8.05	0.84
	Median	2.40	2.68	2.68
	Range	12.25	29.64	3.61
	n	31	31	30

Appendix 4. The descriptors of the reference site stream and streambank variables, including the mean, median, range, standard deviation (StDev), and the number of sites (n). For full name and units of habitat variables, see Table 5.

Year	Descriptor	%Macro	%Eroding	%Grass	WetDepth	WetWidth	BFWidth
2007	Mean	30.0	18.0	50.8	0.56	5.30	17.49
	StDev	30.1	12.4	18.5	0.16	2.42	5.41
	Median	15.0	13.8	50.0	0.58	4.73	16.40
	Range	85.0	45.0	67.5	0.63	8.94	15.90
	n	16	16	16	16	16	16
2008	Mean	37.9	22.1	68.2	0.44	5.68	21.51
	StDev	29.4	8.3	21.1	0.16	2.00	11.53
	Median	40.0	22.5	77.5	0.39	5.60	17.69
	Range	95.0	30.0	79.5	0.61	6.43	43.73
	n	19	19	19	17	19	19
2009	Mean	48.9	13.6	55.3	0.66	5.76	19.85
	StDev	40.6	8.8	15.3	0.17	2.55	9.70
	Median	57.5	15.0	56.3	0.69	4.96	17.65
	Range	99.0	32.5	57.5	0.62	10.79	33.60
	n	16	16	16	16	16	16
All	Mean	38.9	18.2	58.7	0.55	5.59	19.73
	StDev	33.7	10.4	19.8	0.18	2.28	9.36
	Median	35.0	17.5	57.5	0.54	4.84	17.02
	Range	99.0	45.0	80.0	0.77	10.79	44.33
	n	51	51	51	49	51	51

Appendix 5. The descriptors of the test site stream and streambank variables, including the mean, median, range, standard deviation (StDev), and the number of sites (n). For full name and units of habitat variables, see Table 5.

Year	Descriptor	%Macro	%Eroding	%Grass	WetDepth	WetWidth	BFWidth
2007	Mean	65.0	45.0	80.0	0.60	6.54	13.65
	StDev	-	-	-	-	-	-
	Median	65.0	45.0	80.0	0.60	6.54	13.65
	Range	-	-	-	-	-	-
	n	1	1	1	1	1	1
2008	Mean	29.3	25.4	61.1	0.59	5.68	20.12
	StDev	26.8	7.1	13.8	0.26	1.99	5.84
	Median	25.0	25.0	67.5	0.48	5.58	20.91
	Range	80.0	20.0	37.5	0.70	5.36	16.88
	n	7	7	7	7	7	7
2009	Mean	51.8	16.5	58.5	0.66	5.63	18.09
	StDev	28.3	6.0	20.3	0.19	2.13	7.25
	Median	60.0	15.0	55.0	0.63	5.58	17.12
	Range	94.0	23.5	66.5	0.65	9.82	29.45
	n	23	23	23	23	23	23
All	Mean	47.2	19.4	59.8	0.64	5.67	18.41
	StDev	28.9	8.5	18.8	0.20	2.04	6.85
	Median	40.0	20.0	57.5	0.61	5.58	17.12
	Range	94.0	41.0	66.5	0.75	9.82	29.45
	n	31	31	31	31	31	31

Appendix 6. The descriptors of the reference site water chemistry variables, including the mean, median, range, standard deviation (StDev), and the number of sites (n). For full name and units of habitat variables, see Table 5.

Year	Descriptor	DIN	DON	SuspN	SuspP	TDP	SuspC	Chla	SRSi	TSS	SO_4
2007	Mean	18.8	1268	306	49	36	2263	16.40	1.118	10	686.3
	StDev	10.0	740	164	30	21	968	10.72	1.085	7	368.3
	Median	17.5	1096	316	43	30	2380	15.98	0.643	9	602.5
	Range	41.0	2382	540	90	87	2980	31.29	3.501	26	1331.8
	N	16	16	16	16	16	16	16	16	14	16
2008	Mean	30.8	1500	414	59	44	3613	14.96	0.816	26	791.0
	StDev	20.1	824	266	37	24	2914	15.41	0.903	21	665.0
	Median	29.0	1315	336	50	44	2860	9.80	0.437	15	470.0
	Range	95.0	3031	892	130	89	12620	62.66	3.692	69	2152.0
	N	19	19	19	19	19	19	18	19	19	19
2009	Mean	29.8	1097	583	78	49	4859	27.91	0.447	41	476.0
	StDev	11.4	466	364	47	32	3168	24.20	0.247	26	361.1
	Median	29.0	934	437	68	39	3755	25.23	0.420	34	349.9
	Range	36.6	1610	1396	181	122	9210	105.59	0.817	88	1402.8
	N	16	16	16	16	16	16	16	16	16	16
All	Mean	26.7	1301	433	62	43	3580	19.56	0.795	26	659.5
	StDev	15.6	709	293	39	26	2726	18.18	0.859	23	506.6
	Median	25.4	1111	367	50	38	2860	15.50	0.455	19	457.3
	Range	95.0	3094	1490	191	133	13160	109.03	3.722	102	2219.8
	N	51	51	51	51	51	51	50	51	49	51

(continued)

Appendix 6 (continued).

Year	Descriptor	Na	Cl	K	Mg	Ca	Fe
2007	Mean	463	8.403	13.0	42.5	31.5	0.08
	StDev	185	3.224	3.8	14.0	13.1	0.14
	Median	460	7.805	12.4	39.2	26.3	0.02
	Range	687	12.18	14.4	52.0	49.6	0.56
	N	16	16	16	16	16	16
2008	Mean	550	6.559	10.4	34.7	16.9	0.23
	StDev	268	2.911	2.3	22.0	6.0	0.37
	Median	434	5.32	10.2	26.8	15.1	0.07
	Range	896	11.71	8.9	71.3	21.4	1.18
	N	19	19	19	19	19	19
2009	Mean	410	6.804	10.2	33.5	20.3	0.17
	StDev	183	2.852	2.2	14.9	4.9	0.16
	Median	371	6.145	10.0	31.3	18.7	0.11
	Range	696	9.75	9.0	54.7	17.0	0.59
	N	16	16	16	16	16	16
All	Mean	479	7.215	11.1	36.7	22.6	0.17
	StDev	223	3.046	3.1	17.7	10.5	0.26
	Median	424	6.83	10.5	34.3	19.7	0.07
	Range	997	13.15	15.5	71.3	59.3	1.18
	N	51	51	51	51	51	51

Appendix 7. The descriptors of the test site water chemistry variables, including the mean, median, range, standard deviation (StDev), and the number of sites (n). For full name and units of habitat variables, see Table 5.

Year	Descriptor	DIN	DON	SuspN	SuspP	TDP	SuspC	Chla	SRSi	TSS	SO$_4$
2007	Mean	16.4	1783.6	221	27	35	1650	14.30	0.226	4	647.6
	StDev	-	-	-	-	-	-	-	-	-	-
	Median	16.4	1783.6	221	27	35	1650	14.30	0.226	4	647.6
	Range	-	-	-	-	-	-	-	-	-	-
	n	1	1	1	1	1	1	1	1	1	1
2008	Mean	42.2	1315.0	339	41	43	2781	14.69	2.421	18	726.3
	StDev	40.2	943.0	364	45	20	2203	23.20	2.438	14	646.7
	Median	29.0	820.0	228	26	32	2270	6.97	1.760	14	572.5
	Range	114.0	2165.0	1050	127	52	6290	64.81	6.511	42	1846.1
	n	7	7	7	7	7	7	7	7	7	7
2009	Mean	47.1	883.7	261	32	40	2623	11.35	1.908	41	818.1
	StDev	38.4	710.0	219	25	33	2579	11.21	2.344	61	1554.1
	Median	43.0	589.0	202	26	34	1590	6.23	0.420	18	246.9
	Range	176.0	2662.0	865	105	137	11750	36.66	5.802	248	6497.2
	n	23	23	23	23	23	23	23	23	23	23
All	Mean	45.0	1010.1	277	34	40	2627	12.20	1.970	35	791.9
	StDev	37.9	775.5	251	30	29	2426	14.21	2.317	54	1362.7
	Median	34.4	820.0	204	26	34	1650	6.97	0.510	17	281.6
	Range	176.0	2662.0	1064	131	137	11750	64.93	6.732	250	6497.2
	n	31	31	31	31	31	31	31	31	31	31

(continued)

Appendix 7 (continued)

Year	Descriptor	Na	Cl	K	Mg	Ca	Fe
2007	Mean	438	9.99	17.7	31.3	19.5	0.04
	StDev	-	-	-	-	-	-
	Median	438	9.99	17.7	31.3	19.5	0.04
	Range	-	-	-	-	-	-
	n	1	1	1	1	1	1
2008	Mean	507	8.85	12.7	37.3	25.3	0.12
	StDev	198	5.63	5.7	16.6	12.6	0.10
	Median	406	5.70	10.1	31.3	21.5	0.09
	n	7	7	7	7	7	7
2009	Mean	510	7.29	10.9	52.1	33.0	0.25
	StDev	639	4.54	5.2	73.8	31.4	0.39
	Median	286	5.84	8.6	30.5	21.6	0.08
	Range	2752	20.74	20.9	296.0	117.7	1.51
	n	23	23	23	23	23	23
All	Mean	507	7.73	11.5	48.1	30.8	0.21
	StDev	555	4.70	5.3	64.0	27.8	0.35
	Median	318	5.84	9.3	30.8	21.5	0.08
	Range	2752	20.74	20.9	296.0	117.7	1.51
	n	31	31	31	31	31	31

Appendix 8. The correlation matrix with the Pearson correlation coefficients for the habitat variables. Significant values ($P < 0.05$) are marked with an asterisk (*). Habitat variable abbreviations are expanded in Table 5.

	%Silt	%VFSand	%FSand	%MSand	%CSand	%VCSand
%VFSand	0.567*					
%FSand	0.027	0.506*				
%MSand	-0.401*	-0.107	0.593*			
%CSand	-0.466*	-0.470*	-0.042	0.332*		
%VCSand	-0.555*	-0.554*	-0.471*	0.032	0.587*	
%VFGravel	-0.351*	-0.489*	-0.555*	-0.231	0.219	0.676*
%FGravel	-0.526*	-0.530*	-0.570*	-0.273	0.127	0.675*
%Mgravel	-0.528*	-0.480*	-0.491*	-0.282*	0.023	0.392*
%CVCGravel	-0.492*	-0.389*	-0.453*	-0.362*	-0.197	0.123
%LOI	0.738*	0.496*	-0.266	-0.588*	-0.531*	-0.406*
%Macro	-0.176	-0.154	0.140	0.316*	0.176	0.130
%Eroding	0.154	-0.036	-0.085	0.011	-0.056	0.113
%Grass	-0.017	0.102	-0.083	-0.173	-0.077	0.075
WetDepth	-0.497*	-0.495*	-0.130	0.102	0.352*	0.420*
WetWidth	-0.037	0.849	-0.168	-0.220	-0.151	0.127
BFWidth	-0.323	-0.157	-0.054	0.044	0.112	0.333*
DIN	-0.099	0.028	0.136	0.238	0.029	0.046
DON	-0.076	0.027	0.115	0.059	-0.107	-0.163
SuspN	0.069	0.059	0.024	-0.221	-0.070	-0.087
SuspP	0.172	0.161	0.118	-0.178	-0.113	-0.152
TDP	0.098	-0.053	-0.100	-0.150	-0.115	-0.023
SuspC	-0.043	0.044	0.020	-0.137	-0.049	-0.004
Chla	0.190	0.139	0.155	-0.146	0.015	-0.146
SRSi	0.213	0.126	0.047	0.122	-0.128	-0.261
SO_4	0.105	0.230	0.327*	0.157	-0.071	-0.268
TSS	-0.065	0.043	0.100	0.011	-0.084	0.018
Na	0.059	0.270	0.332*	0.131	-0.131	-0.325*
Cl	0.183	0.329*	0.445*	0.237	-0.118	-0.424*
K	0.154	0.390*	0.172	0.005	-0.176	-0.205
Mg	0.196	0.431*	0.280*	0.083	-0.267	-0.415*
Ca	0.173	-0.055	0.005	0.075	0.195	-0.022
Fe	-0.094	-0.015	0.050	0.143	-0.005	0.120

(continued)

Appendix 8 (continued).

	%VFGravel	%FGravel	%Mgravel	%CVCGravel	%LOI	%Macro
%VFSand						
%FSand						
%MSand						
%CSand						
%VCSand						
%VFGravel						
%FGravel	0.732*					
%Mgravel	0.370*	0.802*				
%CVCGravel	0.237	0.417*	0.564*			
%LOI	-0.113	-0.199	-0.210	-0.076		
%Macro	-0.060	-0.146	-0.203	0.026	-0.343*	
%Eroding	0.242	-0.007	-0.198	-0.145	0.015	-0.014
%Grass	0.106	0.069	0.044	0.104	0.066	0.086
WetDepth	0.260	0.462*	0.421*	0.270	-0.430*	0.090
WetWidth	0.262	0.290*	0.183	0.150	0.054	-0.304*
BFWidth	0.194	0.300*	0.287*	0.140	-0.098	0.017
DIN	0.075	0.069	-0.027	-0.142	-0.130	0.236
DON	-0.090	-0.111	-0.039	0.142	0.026	0.038
SuspN	-0.127	-0.118	-0.012	0.202	0.036	0.203
SuspP	-0.223	-0.146	-0.051	0.025	0.100	0
TDP	-0.018	0.028	0.017	0.113	0.111	0.070
SuspC	-0.137	-0.091	-0.011	0.253	-0.028	0.357*
Chla	-0.305*	-0.236	-0.128	-0.036	0.083	0.058
SRSi	-0.188	-0.208	-0.085	-0.269	0.123	-0.190
SO_4	-0.255	-0.195	-0.098	-0.214	-0.062	-0.301*
TSS	-0.047	-0.012	0.043	0.135	-0.111	0.380*
Na	-0.261	-0.182	-0.089	-0.127	0.032	-0.393*
Cl	-0.388	-0.337*	-0.234	-0.299*	0.108	-0.346*
K	-0.248	-0.169	-0.091	-0.129	0.153	-0.458*
Mg	-0.351*	-0.207	-0.090	-0.169	0.158	-0.477*
Ca	-0.168	-0.135	-0.070	-0.278*	-0.166	0.021
Fe	0.104	-0.014	-0.097	-0.014	-0.129	0.322*

(continued)

Appendix 8 (continued).

	%Eroding	%Grass	WetDepth	WetWidth	BFWidth	DIN
%VFSand						
%FSand						
%MSand						
%CSand						
%VCSand						
%VFGravel						
%FGravel						
%Mgravel						
%CVCGravel						
%LOI						
%Macro						
%Eroding						
%Grass	0.177					
WetDepth	-0.255	-0.154				
WetWidth	0.071	0.099	0.208			
BFWidth	-0.221	-0.262	0.054	0.224		
DIN	0.028	0.030	0.024	-0.124	0.074	
DON	0.259	-0.015	-0.241	-0.254	-0.010	0.503*
SuspN	-0.016	0.101	0.14	-0.038	-0.095	0.213
SuspP	-0.019	0.011	0.126	-0.040	-0.058	0.142
TDP	0.201	0	0.000	-0.274	-0.133	0.456*
SuspC	-0.034	0.142	0.046	-0.088	0.016	0.166
Chla	-0.062	0.048	0.179	-0.040	-0.154	0.112
SRSi	-0.083	0.072	-0.135	0.094	-0.016	-0.176
SO_4	0.025	0.202	-0.137	0.283*	-0.054	0
TSS	-0.157	0.062	0.266	-0.051	-0.055	0.271
Na	0.001	0.167	-0.205	0.284*	-0.052	0.022
Cl	-0.035	-0.275*	-0.214	0.079	-0.140	-0.060
K	0.264	-0.234	0.238	0.066	-0.029	-0.140
Mg	-0.113	-0.064	0.237	0.250	-0.031	-0.141
Ca	-0.147	-0.186	0.165	-0.047	-0.186	-0.198
Fe	0.173	0.134	-0.068	-0.280*	-0.156	0.385*

(continued)

Appendix 8 (continued).

	DON	SuspN	SuspP	TDP	SuspC	Chla
%VFSand						
%FSand						
%MSand						
%CSand						
%VCSand						
%VFGravel						
%FGravel						
%Mgravel						
%CVCGravel						
%LOI						
%Macro						
%Eroding						
%Grass						
WetDepth						
WetWidth						
BFWidth						
DIN						
DON						
SuspN	0.364*					
SuspP	0.199	0.736*				
TDP	0.595*	0.480*	0.470*			
SuspC	0.328*	0.878*	0.652*	0.305*		
Chla	0.258	0.830*	0.718*	0.521*	0.655*	
SRSi	-0.298*	-0.184	-0.143	-0.378*	-0.193	-0.119
SO$_4$	-0.100	-0.048	0.059	-0.250	-0.107	0.001
TSS	0.073	0.660*	0.559*	0.181	0.635	0.437*
Na	0.040	0.016	0.116	-0.156	-0.045	0.029
Cl	0.117	-0.067	-0.037	-0.163	-0.203	0.010
K	0.228	-0.131	0.116	-0.048	-0.163	-0.060
Mg	-0.197	-0.208	0.011	-0.335*	-0.234	-0.152
Ca	0.392*	-0.298	-0.226	-0.343*	-0.349*	-0.163
Fe	0.246	0.074	0.031	0.439*	0.042	0.060

(continued)

Appendix 8 (continued).

	SRSi	SO$_4$	TSS	Na	Cl
%VFSand					
%FSand					
%MSand					
%CSand					
%VCSand					
%VFGravel					
%FGravel					
%Mgravel					
%CVCGravel					
%LOI					
%Macro					
%Eroding					
%Grass					
WetDepth					
WetWidth					
BFWidth					
DIN					
DON					
SuspN					
SuspP					
TDP					
SuspC					
Chla					
SRSi					
SO$_4$	0.371*				
TSS	-0.178	0.011			
Na	0.309*	0.934*	0.036		
Cl	0.160	0.594*	-0.082	0.676*	
K	0.153	0.303*	-0.179	0.348*	0.541*
Mg	0.437*	0.782*	-0.111	0.792*	0.697*
Ca	0.359*	0.228	-0.260	0.017	0.213
Fe	-0.197	-0.299*	0.150	-0.245	-0.235

(continued)

Appendix 8(continued).

	K	Mg	Ca
%VFSand			
%FSand			
%MSand			
%CSand			
%VCSand			
%VFGravel			
%FGravel			
%Mgravel			
%CVCGravel			
%LOI			
%Macro			
%Eroding			
%Grass			
WetDepth			
WetWidth			
BFWidth			
DIN			
DON			
SuspN			
SuspP			
TDP			
SuspC			
Chla			
SRSi			
SO_4			
TSS			
Na			
Cl			
K			
Mg	0.578*		
Ca	0.158	0.308*	
Fe	-0.212	-0.430*	-0.339*