SPATIAL AND TEMPORAL VARIATION OF BENTHIC MACROINVERTEBRATE COMMUNITIES IN THREE WESTSLOPE CUTTHROAT TROUT TRIBUTARIES IN THE ROCKY MOUNTAINS

MARTA REE 126133
SUPERVISORS: DR. MARIOLA JANOWICZ & Dr. XIN CHEN

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CONCORDIA UNIVERSITY COLLEGE OF ALBERTA

Abstract

Westslope cutthroat trout (*Oncorhynchus clarkii lewisi*) is an important subspecies to Alberta, as a representative of a rich and diverse fish taxon that intrinsically holds many local adaptations to their native range. The reduction of westslope cutthroat trout from their historical range to isolated headwaters is mainly a result of the introduction of nonnative salmonids (ex. *Oncorhynchus mykiss*) and consequent genetic introgression. This study aims to describe and determine factors affecting benthic communities in three Rocky Mountain tributaries in Alberta associated with pure strains of westslope cutthroat trout. Benthic macroinvertebrate collection and environmental analysis of streams were conducted every two weeks from June to August of 2014 and once in September, 2014. Macroinvertebrates were collected along 5 different transects at 50 m intervals in each stream. The samples were sorted, preserved, identified to the genus level, and weighed. The results of taxonomic diversity, macroinvertebrate density, and biomass show temporal and spatial differences in the three streams. Further, the relationship between macroinvertebrate taxa and environmental parameters are discussed. The three tributaries studied (Blairmore Creek, Gold Creek, and Daisy Creek) represent critical habitat for remaining populations of westslope cutthroat trout. Describing the macroinvertebrate communities and the environmental factors that influence these communities is essential in establishing criteria for suitable habitat available for westslope cutthroat trout.

Introduction

Oncorhynchus clarkii lewisi (Westslope cutthroat trout) represents an extremely diverse subspecies of cutthroat trout, with populations that contain alleles that are limited in high frequencies to individual populations (Allendorf and Leary 1988). Therefore, a loss of one population is a prodigious loss of genetic diversity, local adaptations, and life histories. *O. c. lewisi* is the only native subspecies of cutthroat trout in Alberta and therefore represents a long-standing history of local adaptations. Anthropogenic introduction of nonnative salmonids (ex. *Oncorhynchus mykiss*), along with other factors, has limited Southern Alberta populations to isolated headwaters of the Oldman and Bow river drainage basins (Behnke 1992) and threatens to eliminate *O. c. lewisi* from Alberta (Allendorf and Leary 1988; Rasmussen et al. 2010). Therefore, conservation efforts and management of remaining pure populations are warranted (Fisheries and Oceans Canada 2014).

In order to successfully extend existing populations, habitat requirements are necessary to increase this management area. The Alberta Westslope Cutthroat Trout Recovery Plan (Fisheries and Oceans Canada 2014) aims to re-establish *O. c. lewisi* within their historical range, as well as increase population levels, distribution, and connectivity. There are several research gaps, however, that limit these efforts; this includes baseline information on the biophysical and chemical parameters, and secondary production of *O. c. lewisi* habitat. Therefore, a description of the macroinvertebrate communities in relation to habitat diversity will be beneficial to determine the amount of suitable habitat available for the re-establishment of native *O. c. lewisi* populations.

Benthic macroinvertebrate communities are extremely important for fish communities and food abundance can affect habitat distribution of cutthroat trout species (Wilzbach 1985; Luecke 1986). *O. c. lewisi* are rarely piscivorous and they are mainly invertebrate feeders (Liknes and Graham 1988). Cutthroat trout fry have been shown to consume over 30 different taxa, which includes mainly chironomids, ephemeropterans, and ostracods (Moore and Gregory 1988). However, the specific diet of *O. c. lewisi* in Alberta is relatively unknown.

Stream ecosystems are continuous and dynamic networks. The movement of water in floods and droughts, spatial variation, and geomorphological characteristics of the catchment can change the flow of energy and create a variety of habitats that vary in physiochemical properties (Curry et al. 2012; Vannote et al. 1980). The study of changes in macroinvertebrate assemblages along chemical and physical gradients is fairly well understood (Koetsier et al. 1996; Curry et al. 2012; Gill et al. 2014).

Canonical correspondence analysis (CCA) is an important multivariate tool used to explain ecological relationships of species to environmental variables (Braak 1987). The ordination diagram produced by CCA visualizes the pattern of community variation and the main trends of species along the environmental gradients. This analysis, along with other direct gradient analyses, have been used to detect species-environment relations and investigate specific questions about the effect on species distribution of environmental variables (Braak 1987; Curry et al. 2012; Li et al. 2012).

The purpose of this study is to describe the spatial and temporal variation of benthic macroinvertebrate communities associated with pure strains of *O. c. lewisi*. In addition, it aims to describe the relationship of this variation with the diversity of different *O. c. lewisi* habitats.

Methods

Study Area and Sites

The Oldman River sub-basin accounts for approximately 22% of the South Saskatchewan River Basin, and has a drainage area of 26, 700 km^2. Gold and Blairmore Creek are tributaries of Crowsnest River which drains into the Oldman River. Daisy Creek is proximate to Gold and Blairmore Creek, but drains into the Livingstone River near the confluence of Livingstone with the Oldman River (Table 1; Figure 1). Data for each stream was collected along a 200 m reach that has been associated with pure native *O. c. lewisi* (Rasmussen 2010; Janowicz 2005).

Habitat Assessment

Water chemistry was measured at the middle of each 200 m reach and hydrologic variables were measured at each transect every two weeks from June 15, 2014-August 10, 2014

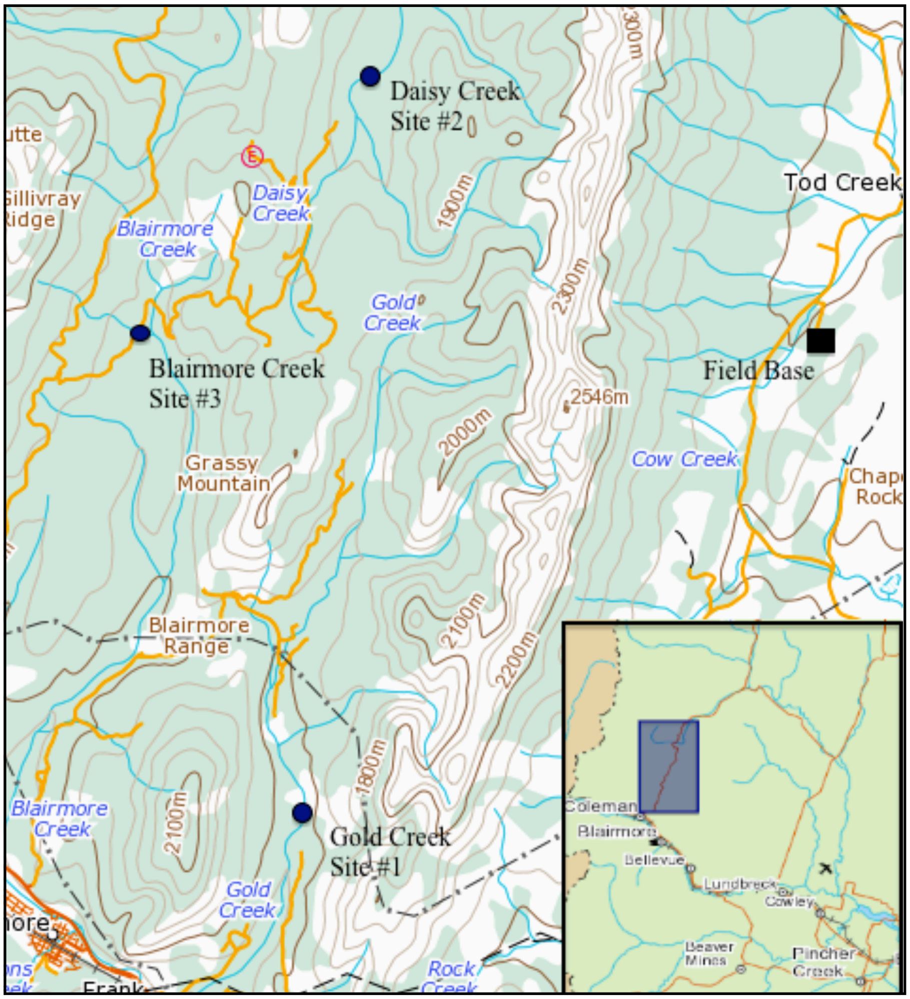

Figure 1 Sampling sites located at the head waters of the Livingstone River (Daisy, *elev=1634m*) and Crowsnest River (Blairmore, *elev=1655m*; Gold, *elev=1436m*)

and September 20, 2014 in accordance with benthic macroinvertebrate collection (Table 2). Riparian vegetation, cover, and substrate composition, and stream morphology were visually assessed for each stream site on June 15, 2014 according to Johnson et al. (1998) and are summarized in Table 2.

<u>Macroinvertebrate Collection</u>

Benthic macroinvertebrate samples were collected using a 20cm D-shaped frame kick-net. Collection took place at each stream every two weeks from June 15, 2014-August 10, 2014 and September 20, 2014. A 20 cm x 20 cm area of substrate was dislodged in 3 different areas at

Table 1 Location, elevation, drainage, and *O. c. lewisi* X *O. mykiss* hybridization of sampling sites (modified from Janowicz 2005)

Site	Stream location	Latitude	Longitude	Elevation (m)	Gradient (°)	Stream Order	Tributary to	Degree of hybridization
1	Gold Creek	49.641129	-114.381984	1436	3.3	4	Crowsnest	0.00
2	Daisy Creek	49.757477	-114.430054	1634	2.1	1	Livingstone	0.00
3	Blairmore Creek	49.723541	-114.465154	1655	3.6	3	Crowsnest	0.00

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different transects that were spaced 50 m apart. Samples were retained, sorted, and preserved in 70% ethanol. Specimens were identified to the family or genus level using the dichotomous key of Clifford (1991) and weighed using a top loading balance.

<u>Data Analysis</u>

Two-way ANOVA (α=0.05) using Excel 2010 was used to determine significant differences in taxa richness (genus-level), macroinvertebrate density, and biomass between stream sites and over the study period. Detrending Correspondence Analysis (DCA) of macroinvertebrate taxa and environmental data using CANOCO version 4.5 was used to determine unimodality of correlations. Therefore, Canonical Correspondence analysis (CCA) using CANOCO version 4.5 was used to illustrate taxa ordination and environmental vectors to describe relationships between environmental variables and macroinvertebrate community composition.

The length of gradients for the first and second axis in the DCA output was higher than 4SD, indicating a strong unimodal response. Therefore, Canonical Correspondence Analysis (CCA) was used to analyze the macroinvertebrate genera and environmental data (Braak and Smilauer 2002). In CCA taxa data was transformed logarithmically to normalize data, and rare species were down-weighed to decrease their influence on the model. An initial CCA using forward selection was used to select 15 environmental variables that account for the majority of the explained taxa variation. A second CCA with forward selection was run with these 15 variables to determine the significance of each of the variables in the model and taxa-environment correlations.

Results

Environmental Variables and Habitat Diversity

Table 2 summarizes the water quality, hydrologic variables, riparian vegetation, cover, substrate composition, and stream morphology of Gold, Daisy, and Blairmore Creek. Over the study period, Daisy Creek was the highest average temperature by 1°C compared to Blairmore and Gold (8.4°C ; Table 2). Conductivity, pH, channel width, wetted width, and rooted width decreased according to stream order (Table 1; Table 2); similarly, Daisy and Gold had the highest flow rate (0.84, 0.80 m/s; Table 2) compared to Blairmore (0.50m/s; Table 2). Average residual pool depth was 0.4 m for all three streams, varying from 0.25 - 0.90 m (Table 2).

The riparian vegetation of Gold Creek is characterized by young, mixed wood forest with 15% canopy closure. Shrubs and young forest dominate Daisy Creek's riparian zone with a canopy closure of 30% and Blairmore Creek's riparian vegetation consists of young, coniferous forest with a high percentage of canopy closure at 70% (Table 2). Total coverage is highest at the Blairmore site with a high percentage of small woody debris (Table 2). Daisy Creek has a high percentage of deep pools and overhanging vegetation, where Gold Creek has a low overall cover percentage (Table 2).

Table 2 Environmental variables measured for habitat diversity-community analysis of Gold (G), Daisy (D), and Blairmore (B) Creek

Category	Variable measured	Units	Methods	Stream	Mean, range (min, max)
Water chemistry	Water Temperature	°C	mercury thermometer	G	7.4 (5, 9.5)
				D	8.4 (6.5, 12)
				B	7.4 (6, 8.5)
	pH	$[H^+]$	HANNA pHep 1 pH meter	G	8.4 (7.5, 10.1)
				D	8.3 (6.5, 9.9)
				B	8.0 (7.1, 9)
	Conductivity	µS/cm	TDSTEstr 3: Cond. meter	G	204 (140, 260)
				D	137.5 (80, 165)
				B	130 (90, 180)
Hydrologic variables	Flow	m/s	Xplorer GLX: Flow rate/temp. sensor	G	0.80 (0.21, 1.15)
				D	0.84 (0.47, 1.25)
				B	0.50 (0.22, 0.90)
	Channel width	m		G	10.1 (6.3, 16.2)
				D	8.8 (4.9, 14.8)
				B	7.3 (5.1, 13.7)
	Wetted width	m		G	6.7 (5.6, 10.5)
				D	4.9 (2.9, 8.8)
				B	3.2 (1.6, 6.4)
	Rooted width	m		G	7.1 (5.9, 10.2)
				D	4.8 (3.3, 6.8)
				B	3.7 (1.9, 6.8)
	Residual pool depth	m		G	0.4 (0.25, 0.7)
				D	0.4 (0.1, 0.8)
				B	0.4 (01, 0.9)

Table 2 Continued

Category	Variable measured	Units	Methods	Stream	Mean, range (min, max)
Riparian Vegetation	Maturity/structure of riparian zone		visually assessed	G	mixed wood; young forest
				D	shrub, young forest
				B	coniferous; young forest
	Canopy closure	%	visually estimated	G	15%
				D	30%
				B	70%
Cover	Total coverage	%	visually estimated	G	25%
				D	40%
				B	45%
	Small woody debris	%		G	<5%
				D	<5%
				B	20%
	Large woody debris	%		G	<5%
				D	<5%
				B	15%
	Undercut banks	%		G	10%
				D	5%
				B	10%
	Deep pool	%		G	10%
				D	20%
				B	5%
	Overhanging vegetation	%		G	5%
				D	15%
				B	5%

Table 2 Continued

Category	Variable measured	Units	Methods	Stream	Mean, range (min, max)
Substrate Composition	Size of dominant bed material		visually assessed; percent of substrate estimated	G	boulders
				D	cobbles
				B	cobbles
	Size of subdominant bed material	%		G	cobbles
				D	large gravel
				B	boulders
	D_{95}	cm	Diameter of particles larger	G	10
			than 95% of bed material	D	8
				B	8
Stream Morphology	Channel morphology		visually assessed	G	riffle pool
				D	riffle pool
				B	riffle pool
	Left bank shape			G	sloped
				D	open
				B	open
	Right bank shape			G	v-shaped
				D	open
				B	sloped
	Left bank texture			G	boulders
				D	fines
				B	riparian
	Right bank texture			G	fines
				D	fines
				B	riparian

The bed material of Gold and Blairmore Creek is dominated by boulders and cobbles and Daisy Creek consists of cobbles and large gravel (Table 2). All three streams are characterized by a series of riffles and pools. Gold Creek is frequently confined by v-shaped and sloped banks; Daisy Creek is unconfined with sloped with open banks and Blairmore Creek is occasionally confined with open and sloped banks (Table 2). The bank texture of Gold Creek consists of boulders and fines; Daisy Creek's bank texture is mainly fines, and Blairmore Creek has banks with riparian vegetation (Table 2).

Macroinvertebrate Taxa Richness, Density, and Biomass

The total number of taxa observed over the study period and across all sites was 40 invertebrate genera including 37 insect invertebrate genera and 3 non-insect genera. Samples taken on June 29, 2014 were not properly stored and were severely decomposed before they were able to be sorted. Therefore, these samples were excluded from the study as they did not represent macroinvertebrate density, richness, and biomass of the sites. Macroinvertebrate genera varied between the three sites; many taxa were limited to one site over the study period (Figure 2). For example, genera of the family Tipulidae exclusively occurred in Daisy Creek. Taxa richness at each site appears to follow the same trend with only marginally significant variation between sites ($F(2,8)=9.72$, $p,0.10$). Variation of taxa richness over the study period was highly significant ($F(4,8)=3.29$, $p<0.005$) with an average maximum of 16 genera and an average minimum of 5.67 genera (Figure 3).

All three sites were consistently dominated, although unequally ($F(2,8)=4.18$, $p<0.10$), by the insect orders Plecoptera, Diptera, Trichoptera, and Ephemeroptera (Figure 2.2). Each dominant insect order exhibited different relationships with other streams and over the study

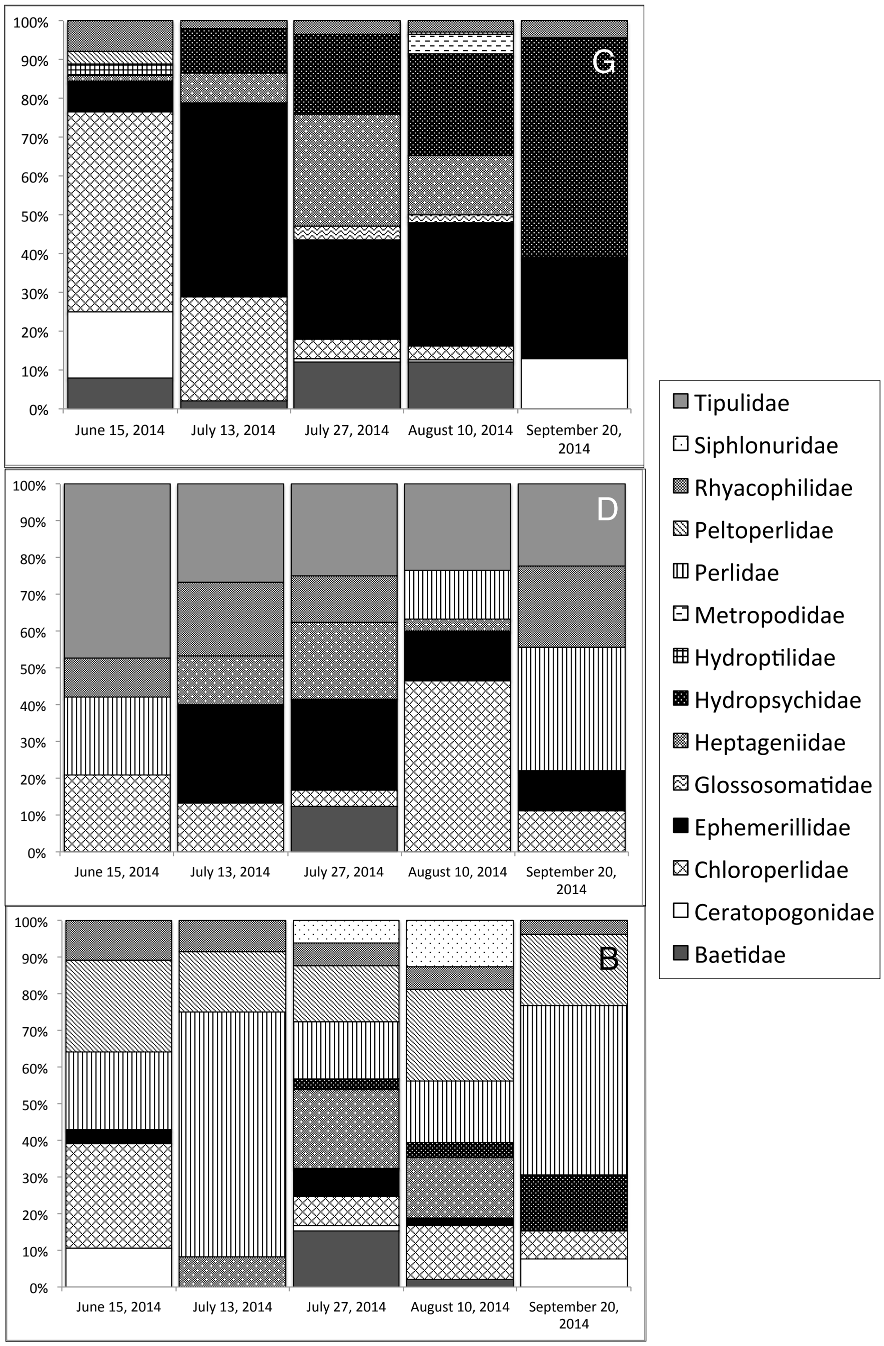

Figure 2 Benthic macroinvertebrate family percent composition over the study period of numerically abundant taxa (>2 individuals in total study period) of Gold (G), Daisy (D), and Blairmore (B) Creek.

period (Figure 2.2). Plecoptera and Trichoptera density did not vary over the study period ($F(4,8)=1.72$, $p>0.10$; $F(4,8)=1.25$, $p>0.10$) and Ephemeroptera had slight variation over the

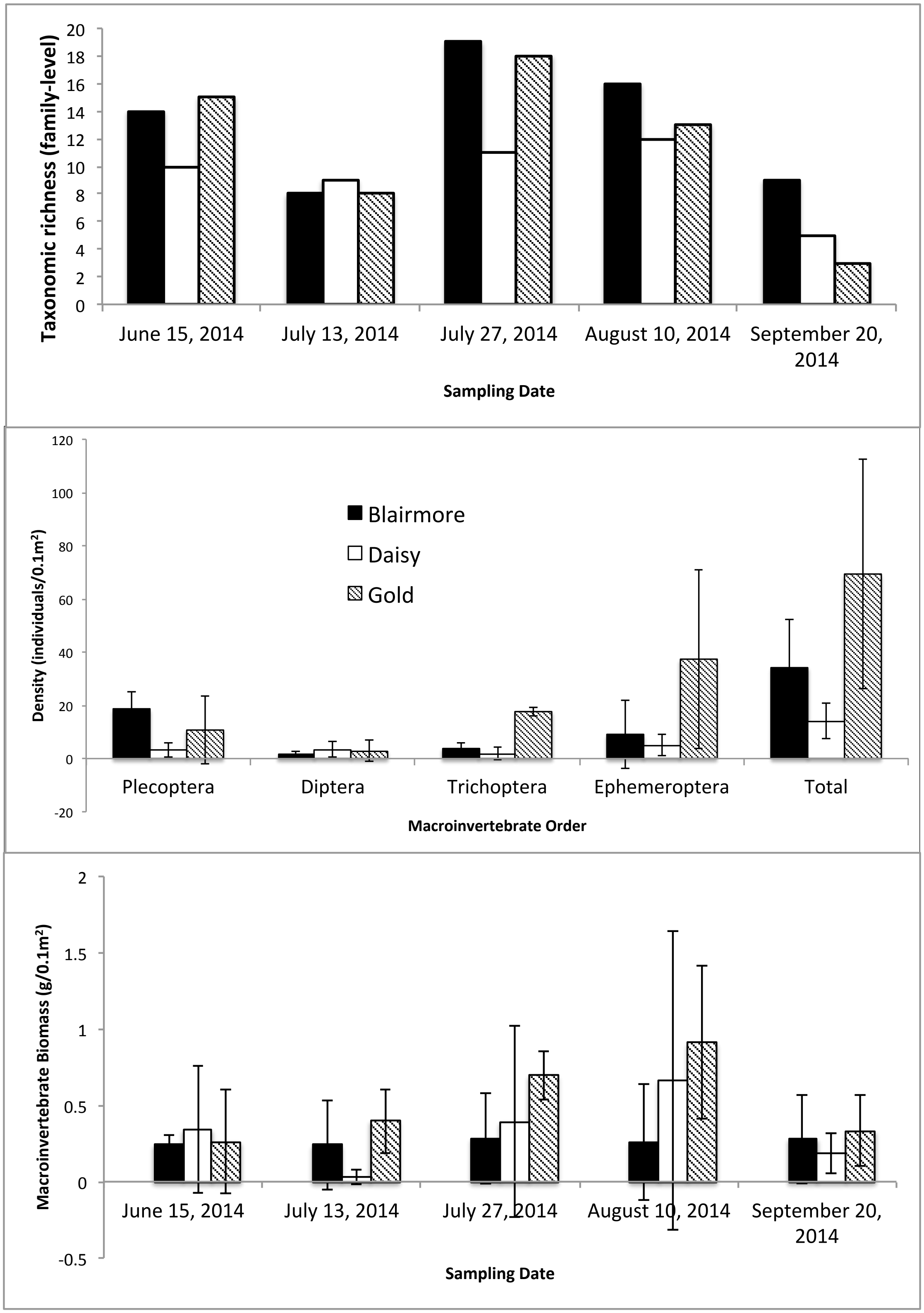

Figure 3 Taxonomic richness, density, and biomass of Blairmore, Daisy, and Gold Creek over the study period. Error bars indicate variation between sampling times for macroinvertebrate density of numerically abundant taxa (> 1 individual/0.1m^2) and variation between transects for biomass (SD=1).

study

period (F(4,8)=2.90, p<0.10). In contrast, Diptera varied significantly (F(4,8)=4.49, p< 0.05). Plecoptera, Trichoptera, and Ephemeroptera showed significant variation between sites (F(2,8)=5.53, p<0.05; F(2,8)=5.94, p<0.05, F(2,8)=5.81, p<0.05). Biomass had marginally significant variation over the study period (F(4,8)=2.43, p<0.10) and between sites (F(2,8)=2.80, p<0.10;Figure 2.2).

Canonical Correspondence Analysis (CCA)

The eigenvalues of the first and second axes are 0.044 and 0.018, indicating that the gradients of these axes are very weak (Table 3). The taxa-environment relations for the first two axes are very high with taxa-environment correlations of 0.912 and 0.788 (Table 3). The percentage of variance of the taxa data explained by axis 1 is 14.7% and axis 2 explains 5.9%; in total, both axes explain 20.6% of the percentage of variance (Table 3). 12% of the total variance of the taxa data is significantly explained by the model, as indicated by the sum of all canonical eigenvalues and the Monte Carlo test for significance (F=2.738, p<0.005; (Table 3). Of this 12%,

Table 3 Summary of CCA of benthic macroinvertebrate taxa and environmental variables data of three Rocky Mountain tributaries after forward selection

Axes	1	2	3	4
Eigenvalues	0.044	0.018	0.013	0.010
Taxa-environment correlations	0.912	0.788	0.778	0.772
Cumulative percentage variance of taxa data	14.7	20.6	24.9	28.3
Cumulative percentage variance of taxa-environment relation	37.8	52.8	63.9	72.6
Sum of all eigenvalues/total inertia				0.301
Sum of all canonical eigenvalues				0.117

52.8% is explained by the first two axes (Table 3). The total variance in the species data is 0.301 and is also equal to the sum of all eigenvalues (Table 3).

Environmental variables appear to form two axis dependent from each other on the ordination plot (Figure 4). The first axis includes stream order, conductivity, flow rate, and elevation; the second axis includes temperature, pH, and sampling date (Figure 4). This is supported by a strong negative correlation of -0.8226 between order and elevation; similarly, elevation and conductivity have a negative correlation of -0.7261. Stream order is also highly correlated with wetted width, and rooted width. Temperature, pH, and sampling date are not highly correlated with the variables that represent the first 'axis', but sampling date and pH have a strong correlation of -0.7433 and temperature and pH have a correlation of 0.5526.

<u>Relationships between Macroinvertebrates and Environmental Variables</u>

Fifteen variables were selected using forward selection in Canonical Correspondence Analysis that account for the variation explained by the model discussed above . The fifteen variables selected were elevation, stream order, conductivity, rooted width, wetted width, height

Table 4 Conditional effects of seven environmental variables that significantly account for the taxa variation explained by the model produced by CCA

Variable	LambdaA	P-value	F
Elevation	0.04	0.002	9.86
Stream order	0.01	0.002	4.02
Temperature	0.01	0.002	3.55
Conductivity	0.01	0.002	2.95
Sampling date	0.01	0.004	2.37
Flow rate	0.01	0.002	2.16
pH	0	0.006	1.89

of banks, average gradient, flow, temperature, date, time, pH, transect, and pool depth. However, not all of these factors significantly explain the variation of the model. Elevation, stream order, temperature, conductivity, date, flow, and pH significantly ($\alpha = 0.05$) account for the variation explained by the model according to the conditional effects of each variable (Figure 4; Table 4).

Elevation accounts for 4% of the total variation in taxa data; stream order, temperature, conductivity, sampling date, and flow rate each account for 1% of the total variation and pH explains less than 1% of the variation (Table 4).

Figure 4 illustrates the relationships of numerically abundant macroinvertebrate genera with fifteen forward selection environmental variables. In general, the Plecopterans *Calineuria* and *Yoraperla* are associated with high elevation, low stream order, low flow, and low conductivity and *Drunella, Triznaka* and *Parapsyche* are associated with low elevation, high stream order, high flow, and high conductivity (Figure 4). *Hesperoperla*, *Claasenia,* and *Ameletus* are more likely to be found in the mid-high elevation and stream order range, where as *Baetis* and *Agapetus* are associated with the mid-low elevation range. All genera from the Tipulidae family, *Neotrichia*, *Bezzia, Doroneuria, Siphloplecton, Seretella, Heptagenia, and Ephemerella* are found associated with mid-elevation.

Tipula is associated with an earlier sampling date during the summer, and low pH and temperature (Figure 4). The other Tipulidae genera, *Bezzia, Neotrichia,* and *Rhyacophila* follow this same trend, but to a lesser extent (Figure 4). *Cinygmula*, *Parapsyche, Drunella, Calineuria*, *Yoraperla*, *Pseudocloeon,* and *Ameletus* are associated with a later sampling date during the summer. However, *Calineuria* and *Yoraperla* are associated with a lower temperature and pH.

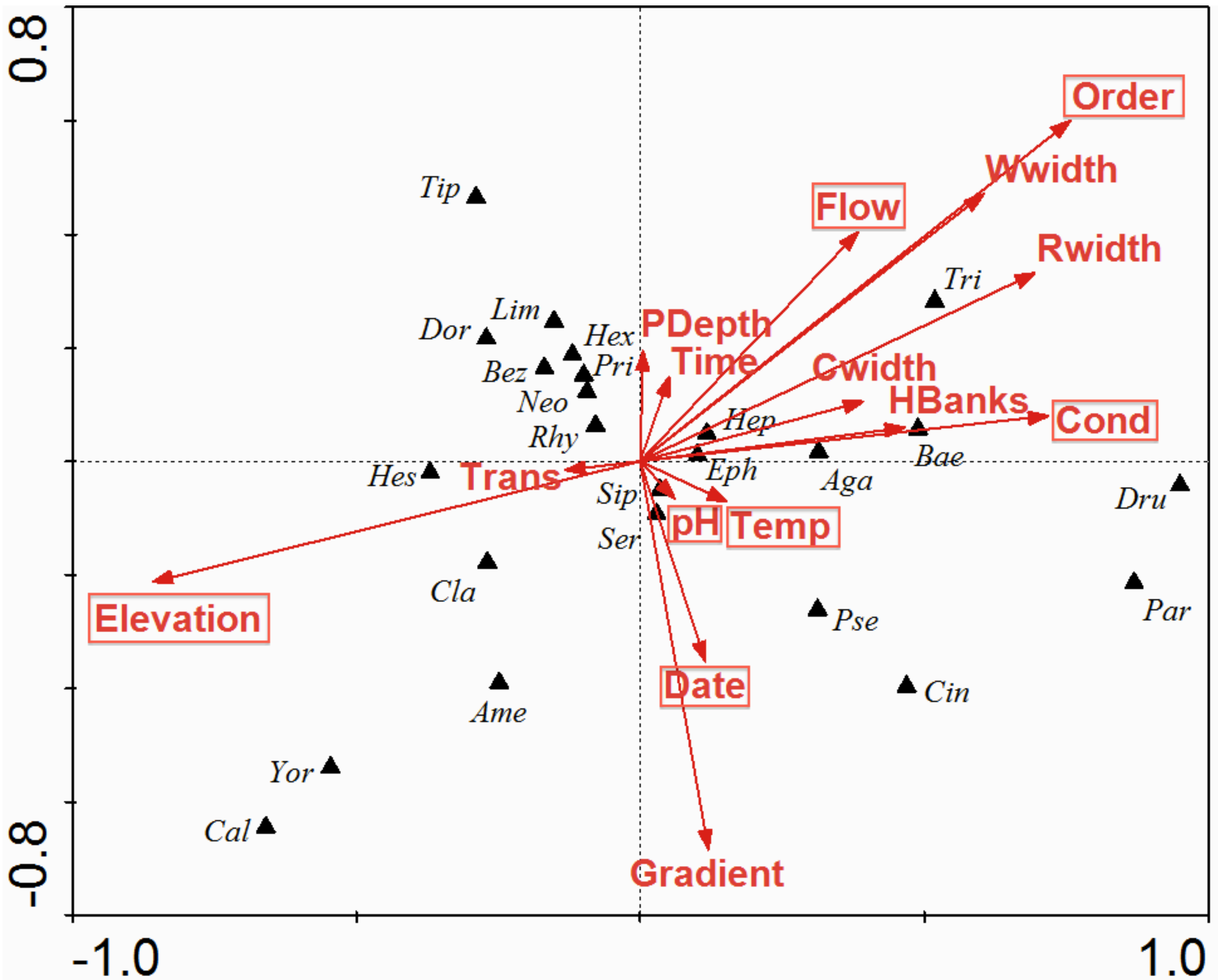

Figure 4 The distribution of numerically abundant genera (>2 ind. over the study period) in benthic samples of three Rocky Mountain tributaries. Canonical correspondence analysis (CCA) ordination diagram with macroinvertebrate genera (▲), and environmental variables identified using forward selection (arrows). First axis is horizontal and second axis is vertical. Macroinvertebrate genera are: *Aga = Agapetus*, *Ame = Ameletus, Bae = Baetis, Bez = Bezzia, Cal = Calineuria, Cla = Claasenia, Cin = Cinygmula, Dor = Doroneuria, Dru = Drunella, Eph = Ephemerella, Hep = Heptagenia, Hes = Hesperoperla, Hex = Hexatoma, Lim = Limoniinae, Neo = Neotricia, Par = Parapsyche, Pri = Prionocera, Pse = Pseudocloeon, Rhy = Rhyacophila, Ser = Seretella, Sip = Siphloplecton, Tip = Tipula, Tri = Triznaka, Yor = Yoraperla.* The environmental variables are: Cond = conductivity, Cwidth = channel width, Date = sampling date, Flow = flow rate, Gradient = average gradient, HBanks = height of banks, Order = stream order, PDepth = residual pool depth, Rwidth = rooted width, Temp = stream temperature, Time = time of day sampled, Trans = distance upstream from first transect, Wwidth = wetted width. Red boxes indicate environmental variables accounting for a significant amount of the explained variation using forward selection in CCA.

Alternatively, *Pseudocloeon, Cinygmula, Paraspyche,* and *Drunella* are associated with higher temperatures and pH (Figure 4).

<u>Relationships between Streams and Environmental Variables</u>

Figure 5 illustrates the relationship of the three tributaries sampled throughout the summer to the environmental variables measured. Gold Creek is associated with high stream order, low elevation and high flow, conductivity, banks, channel width, rooted width, wetted width, and temperature. There are five sampling data points that appear to be associated with mid-elevation, low temperature and pH, and early sampling time (Figure 5). Daisy Creek is associated with mid-elevation, and associated conductivity, and flow, as well as low pH and temperature (Figure 5). Blairmore Creek is associated with high elevation, low conductivity, low flow, low channel width, and varying pH and temperature (Figure 5).

Discussion

Habitat Diversity and Environmental Variables of the Streams

Daisy Creek was the highest average temperature over the study period compared to Blairmore and Gold. It would be expected that Gold would be a higher temperature due to its reduced crown closure and lower elevation. However, compared to Daisy's open banks, Gold is confined by tall embankments which may provide shading during early and late times of the day.

On average, conductivity, rooted width, wetted width, channel width, and pH decreased with increasing stream order. This is supported by high correlations between stream order and conductivity, rooted width, and wetted width determined by the CCA. In addition, these relationships are supported by the River Continuum Concept presented by Vannote et al. (1980), that describes the relationship between stream order, elevation, and physical variables of a stream. However, pH is not correlated with stream order in the CCA. It is not clear why the pH varies between the streams; it could be due to small-scale calcite mineralization, land use,

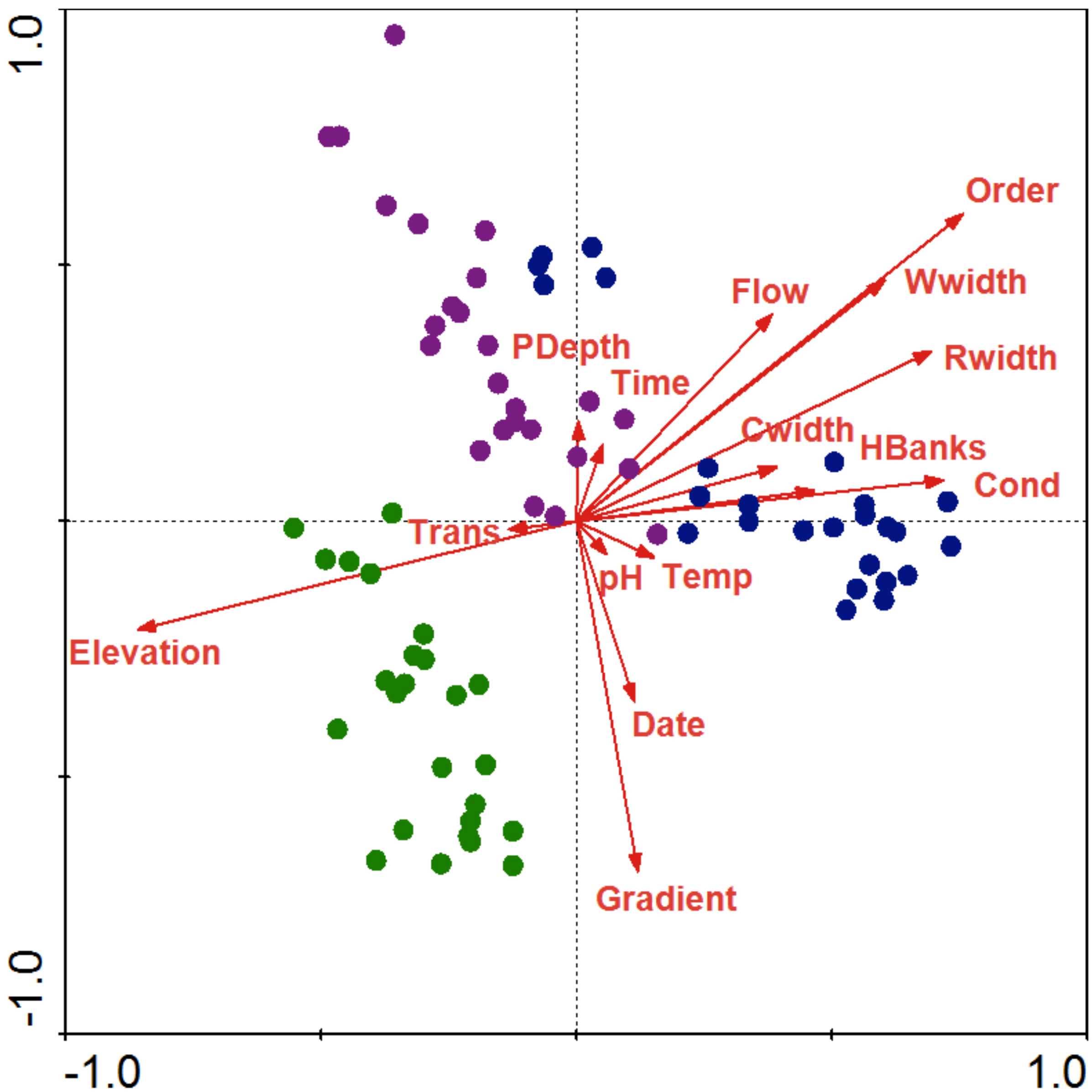

Figure 5 Sample distribution of three Rocky Mountain tributaries. Canonical coordination analysis (CCA) diagram with samples from Gold (blue circles), Daisy (purple circles), and Blairmore Creek (green circles), and environment variables identified using forward selection (arrows). The environmental variables are: Cond = conductivity, Cwidth = channel width, Date = sampling date, Flow = flow rate, Gradient = average gradient, HBanks = height of banks, Order = stream order, PDepth = residual pool depth, Rwidth = rooted width, Temp = stream temperature, Time = time of day sampled, Trans = distance upstream from first transect, Wwidth = wetted width.

addition of fertilizer, proportions of peat in the catchment, or other unknown factors (Hornung et al. 1985).

The ordination of the variables and stream samples illustrates the relationships described in Table 2. Each stream has a cluster of data points that represent its unique habitat, with little area that overlaps in between the three sites. The five samples from Gold that are apart from the other data points represent the first sampling day on June 15, 2014. This early sampling day may explain why these points are associated with an earlier date and therefore a low temperature on the ordination (Liu et al. 2005).

Taxa Richness

The relatively low diversity (40 genera) observed across all sites over the study period may be attributable to a few different factors. Taxonomic diversity will favour environmental heterogeneity and therefore headwater and large river reaches are expected to have reduced taxonomic diversity (Vannote et al. 1980; Cummins et al. 1983). However, Blairmore (stream order = 1) had comparable taxa richness to Gold (stream order = 4), which does not support this hypothesis. Blairmore actually exhibited the highest taxa richness with marginal significance (Figure 3), and this may be due to the high percentage of cover and consequent microhabitats that promote habitat heterogeneity (Ligeiro et al. 2013). Gold exhibited the second highest taxa richness, which may be attributable to its lower elevation (Cummins et al. 1983) and high percentage of boulder/cobble substrate (Negishi and Richardson 2011).

Macroinvertebrate Density and Biomass

As with taxa richness, macroinvertebrate densities were also relatively low. These low macroinvertebrate numbers may be an effect of the catastrophic flooding that occurred in June of 2013 in Southern Alberta. Flood disturbances can disturb debris flows, mobilize sediment and wood, and move riparian vegetation from headwater streams. These effects may last for over 10 years from the flooding event (Cover et al. 2010; Short and Ward 1980). This may explain the lower macroinvertebrate densities in Blairmore and Daisy which occur at higher elevations and would be greatly affected by the removal of allochthonous carbon.

High numbers of Ephemeropterans are common in headwater streams and high elevations usually results in higher numbers of Plecopterans compared to Trichopterans (Andrews and Minshall 1979; Bergey and Ward 1989; Short and Ward 1980). This shift can be seen from Blairmore, where Plecopterans are more abundant than Trichopterans, to Gold where the opposite is true.

Macroinvertebrate biomass did not significantly vary over the study period or between sites (Figure 3). This may indicate that food resources were consistent throughout the study period and between sites (Straka et al. 2012). This relationship can only be conjectured due to unknown factors such as assimilation efficiency, variations in ecosystem function, and changes in primary productivity, and organic matter input. Koetsier et al. (1996) found that biomass increased with alkalinity and specific conductance; however, either there is other factors affecting biomass or there is not a sufficient conductance gradient to elicit this response. In addition, biomass may not be a good indicator of the productivity of a macroinvertebrate community, since higher temperatures and higher alkalinity can increase turnover rates in lower elevation streams (Koetsier et al. 1996).

Relationships Between Macroinvertebrates and Environmental Variables

Elevation accounted for the majority of variation explained by the model. Gill et al. (2014) also found that elevation had a significant effect on taxa richness, turnover, and lateral β-diversity. This may be a result of the collinearity of elevation with other abiotic factors (Gill et al.

2014) that increases the significance of this variable. The model used to describe taxa variation only explained approximately 12% of the total variation. Although there were strong correlations between environmental variables and taxa data, there may be other factors not accounted for by this model that explain the remaining 88% of variation. Lujan et al. (2013) concluded that elevational gradient and environmental parameters are not sufficient in describing taxonomic and functional structure. Trophic interaction and turnover in food resources may have an influential effect on the composition of macroinvertebrates (Creed 2006). For example, fish abundances were not observed for this study and this may have a large effect on taxa composition, densities, and biomass. However, Chinnayakanahalli et al. (2011) found that that taxa composition was strongly associated with streamflow and temperature.

In addition, community ordination may not reveal relationships of ecosystem structure and function; categorizing genera into functional groups may prove to describe important links between habitat diversity and macroinvertebrate function (Cummins et al. 1989; Vannote et al. 1980). CCA ordination illustrates important correlations between specific macroinvertebrate genera and environmental variables. Elevation appears to be the most important variable, explaining 4% of the species variation. Other relationships may also exist; for example, *Cinygmula* was shown to be associated with late summer which is supported by Andrews and Minshall (1979). In addition, the genera from Tipulidae were consistently found at the mid-elevation range on the ordination. This is most likely attributable to the isolation of this family to Daisy Creek which has an elevation between Blairmore and Gold Creek. Although some correlations such as sampling date to genera is less clear. Therefore, these specific taxa-environmental interactions need to be further investigated using specific correlations between taxa and variables. Error can than be reduced when reading the ordination bi-plot and assumed correlations can be confirmed using a genera-environmental variable correlation matrix.

Conclusion

The habitat of *Oncorhynchus clarkii lewisi* is diverse in water chemistry, hydrologic variables, riparian vegetation, cover, substrate composition, and stream morphology. This habitat also has varied macroinvertebrate assemblages. Using Canonical Correspondence Analysis, elevation was determined to account for 4% of the taxa variation of three Rocky Mountain tributaries. This is important when considering changes in macroinvertebrate availability as *O. c. lewisi* continues to be isolated to high-elevation streams. Biomass is not significantly different at a lower elevation, which suggests that food availability may be suitable in high-elevation reaches. However, analysis of trophic relations including predator-prey interactions may be needed to fully understand the relationship between *O. c. lewisi* habitat and macroinvertebrate community composition.

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