AN ANALYSIS OF WHIRLING DISEASE RISK IN WESTERN MONTANA

by

Stephanie Ann McGinnis

A thesis submitted in partial fulfillment
of the requirements for the degree

of

Master of Science

in

Biological Sciences

MONTANA STATE UNIVERSITY
Bozeman, Montana

April 2007

APPROVAL

of a thesis submitted by

Stephanie Ann McGinnis

This thesis has been read by each member of the thesis committee and has been found to be satisfactory regarding content, English usage, format, citations, bibliographic style, and consistency, and is ready for submission to the Division of Graduate Education.

Dr. Billie Kerans

Approved for the Department of Ecology

Dr. Dave Roberts

Approved for the Division of Graduate Education

Dr. Carl A. Fox

STATEMENT OF PERMISSION TO USE

Stephanie McGinnis

April 2007

ACKNOWLEDGEMENTS

I would like to thank my committee members, Dave Roberts and Tom McMahon, for their advice and suggestions. I would like to especially thank my advisor, Billie Kerans, for her continual support, guidance and encouragement throughout this process. Thanks to Julie Alexander, Chelsea Cada, Leah Steinbach Elwell, Jody Hupka, Silvia Murcia, Jeremy Smith, and Dick Vincent for their field, laboratory and overall contributions to this research. Special thanks to Linda Phillips for her invaluable GIS assistance. I am grateful to Chris Forristal, Claire Gower, Jamin Grigg, and Julia Nelson for providing moral support and much needed comic relief in the basement. Thanks to the Whirling Disease Initiative for their monetary contribution. Thanks to my mom for always being there for me. Finally, I dedicate this thesis to Amy Buchanan, who had the pleasure of editing numerous drafts of this document, listening to scores of presentations, and putting up with me for the last two years.

TABLE OF CONTENTS

TABLE OF CONTENTS – CONTINUED

TABLE OF CONTENTS – CONTINUED

TABLE OF CONTENTS – CONTINUED

LIST OF TABLES

LIST OF TABLES – CONTINUED

Table Page

LIST OF FIGURES

LIST OF FIGURES – CONTINUED

ABSTRACT

Myxobolus cerebralis, the causative agent of whirling disease, has been a major contributor to the loss of young trout in numerous streams within the Intermountain West (Colorado, Idaho, Montana, Utah, Wyoming). Currently there are no effective management procedures for mitigating the effects of this disease because it is not fully known why the parasite has severe effects in some trout populations while remaining fairly benign in others. Characteristics of the parasite, hosts, environment, and their interactions, may partially explain varying responses of wild rainbow trout populations to whirling disease. The goal of this study was to examine possible contributors to, and indicators of, stream degradation and their relationship to whirling disease risk. Specifically, the objectives were to quantify relationships between whirling disease risk and 1) land use 2) biological stream integrity and 3) physicochemical parameters within four major drainages in western Montana. The hypothesis was that whirling disease risk is influenced by anthropogenic land use practices that create favorable habitat for the oligochaete worm host, *Tubifex tubifex*, which is reflected in the biological integrity and physicochemical features of the stream. Whirling disease risk was quantified by the severity and prevalence of infection in caged sentinel trout. A geographic information system (GIS) was used to model land use (e.g., agriculture, mines) within drainages. Bioassessment metrics specific to Montana (e.g., total taxa richness, sensitive taxa richness) and those directly related to whirling disease (e.g., density of *T. tubifex*) were used to assess biological stream integrity. Physicochemical characteristics included those associated with favorable *T. tubifex* habitat (e.g., substrate), and those that have been associated with an increased incidence of whirling disease (e.g., temperature). Importance of predictor variables was assessed using Spearman's rank correlation and regression tree analyses. Final regression trees identified the proportion of riparian forest, road density, oligochaete density, *Limnodrilus hoffmeisteri* density, Plecoptera taxa richness, and the proportion of Plecoptera as the most important predictors of whirling disease risk among drainages. A greater understanding of the linkage between land use, biological stream integrity, physicochemical features and whirling disease risk is needed before effective management techniques can be implemented in Montana watersheds and elsewhere.

INTRODUCTION

Myxobolus cerebralis, the causative agent of whirling disease, has been detected in 24 states where salmonid populations exist (K. Stromberg, Whirling Disease Initiative, Montana Water Center, Bozeman, MT, personal communication). Many infected salmonid populations have suffered little or no effect from this parasite (e.g., California: Modin 1998; Oregon: Sandell et al. 2001), whereas others have been decimated (e.g., Montana and Colorado: Nehring and Walker 1996; Vincent 1996). Within the Intermountain West (Colorado, Idaho, Montana, Utah, Wyoming), whirling disease has been identified as the main contributor to a decline in many wild rainbow trout *Oncorhynchus mykiss* populations (Nehring and Walker 1996; Vincent 1996; Baldwin et al. 1998) although not all populations have been severely affected (Baldwin et al. 1998; Hiner and Moffitt 2001; Sandell et al. 2001). For example, some tributaries in Montana have consistently tested negative for the parasite, or disease severity has remained low, despite their proximity to infected streams where rainbow trout populations have declined dramatically (E. R. Vincent, Montana Department of Fish, Wildlife and Parks, Bozeman, MT, personal communication). Characteristics of the parasite, hosts, environment, and their interactions, may partially explain the varying response of wild rainbow trout populations to whirling disease (Kerans and Zale 2002).

Identification of the factors that exacerbate whirling disease is confounded by the complex life cycle of *M. cerebralis*, which involves two hosts and two spore stages. The two spores, the myxospore and the triactinomyxon (TAM), are produced within the salmonid host and the oligochaete worm host, *Tubifex tubifex*, respectively (Markiw and Wolf 1983; Wolf and Markiw 1984). The myxospore can lie dormant for an extended period of time, possibly years, and is able to withstand freezing and passage through avian and fish digestive systems (El-Matbouli and Hoffmann 1991), whereas the TAM remains viable for only a few days to weeks (El-Matbouli et al. 1999). The success of the parasite in one or more of its life cycle stages may be influenced by biotic and abiotic features within the environment, which further adds to the complexity of understanding this organism.

Several features of the hosts and environment may influence whirling disease. Factors such as rainbow trout life history traits (Downing et al. 2002), the ability to propagate the parasite by different genetically defined strains of *T. tubifex* (Beauchamp et al. 2002, 2005; DuBey and Caldwell 2004), and environmental characteristics (e.g., water temperature, conductivity, velocity, and substrate composition) (Sandell et al. 2001; Hiner and Moffitt 2001, 2002; McConnell and Vincent 2002; Krueger et al. 2006) have been correlated to whirling disease severity. Additionally, anthropogenic disturbances within watersheds that alter these biotic and abiotic characteristics may influence whirling disease risk (Zendt and Bergersen 2000; Allendorf et al. 2001).

It has been suggested that many streams where *M. cerebralis* is present have altered or impaired habitat (Zendt and Bergersen 2000; Allendorf et al. 2001; Kaeser and Sharpe 2006). Anthropogenic alterations of the landscape (e.g., agricultural practices and mining) can change the environmental characteristics of a stream, modifying physical, chemical, and biological processes (Karr 1991). These processes define the biological integrity of the stream and when altered, reveal relationships between human disturbances and biological attributes (Kerans and Karr 1994; Karr and Chu 1999). The cumulative effects of human activities within watersheds lower biological stream integrity by altering water quality and flow, habitat structure, food sources, and biotic interactions.

The Montana Department of Environmental Quality (MTDEQ) lists 9,826 miles of the state's streams as impaired, unable to fully or partially support at least one designated beneficial use as defined by the state's Water-Use Classification System (i.e., agriculture, industrial, recreation, drinking water, aquatic life, and fisheries (cold and warm) water supply), due to anthropogenic activities (MTDEQ 2004). The primary impairment sources cited by MTDEQ (2004) are agricultural and silvicultural practices, construction, mining and milling processes, hydromodification, and habitat modification such as removal of riparian vegetation and/or streambank alteration. As a result of these land use practices, increased nutrients and siltation, thermal modifications, and alteration of flow regimes have been cited as some of the major causes of impairment (MTDEQ 2004). All of these may influence whirling disease risk by affecting salmonid susceptibility and creating favorable habitat for increased *T. tubifex* abundance, thus providing increased host density for *M. cerebralis* (Bartholomew et al. 2005). Focusing efforts on maintaining and restoring habitats may be an integral component to alleviating the effects of this disease on wild trout populations (Zendt and Bergersen 2000; Allendorf et al. 2001).

The goal of this study was to examine possible contributors to, and indicators of, stream degradation and their relationship to whirling disease risk. The purpose was not to determine the specific causes and/or sources of impairment, nor the biological stream integrity indicators of that impairment, but rather to identify broad patterns within and among drainages. Specifically, the objectives were to quantify relationships between whirling disease risk and 1) land use; 2) measures of biological stream integrity; and 3) physicochemical parameters within four major drainages in western Montana. The hypothesis was that whirling disease risk is influenced by anthropogenic land use practices that create favorable habitat for *T. tubifex*, which is reflected in the biological integrity and physicochemical features of the stream. Catchments that have more anthropogenic land use (i.e., agriculture, residential, disturbance, mines and roads), low proportions of forest, and streams with low biological integrity will have a higher risk of whirling disease than those with less anthropogenic land use, high proportions of forest, and high biological stream integrity. Furthermore, those streams with physicochemical features that provide favorable *T. tubifex* habitat will have a higher risk of whirling disease than those streams that do not provide favorable habitat.

The a priori hypothesis was tested by the following predictions: 1) whirling disease risk is positively correlated to anthropogenic land use; 2) whirling disease risk is negatively correlated to the proportion of forest; 3) whirling disease risk is negatively correlated to bioassessment metrics that indicate high biological stream integrity; 4) whirling disease risk is positively correlated to bioassessment metrics that indicate low biological stream integrity and 5) whirling disease risk is positively associated with physicochemical features of the habitat that reflect land use disturbance, low biological stream integrity, and suitable *T. tubifex* habitat (e.g., low gradient, high organic content in sediment, high conductivity, and increased temperatures). A clearer understanding of the relationship between whirling disease risk and land use practices, measures of biological stream integrity, and physicochemical features is needed to help explain the broad scale variation in whirling disease risk and help contribute to watershed management policies by identifying characteristics of the stream and watershed that support the establishment and proliferation of *M. cerebralis*.

BACKGROUND

The myxozoan parasite *M. cerebralis* is the causative agent of whirling disease. Affecting only salmonids, whirling disease is characterized by blackened tails, skeletal deformities, and whirling behavior for which it is named (Hedrick et al. 1998). *Myxobolus cerebralis* is believed to have evolved in Europe as a parasite of brown trout *Salmo trutta* (Hoffman 1970). The parasite was documented in Germany over 100 years ago after nonnative rainbow trout, introduced from North America, displayed signs of the disease (Hőfer 1903). The parasite was first detected in the United States in 1958 at the Benner Springs fish hatchery in Pennsylvania (Hoffmann et al. 1962). The suspected vector of infection was the import from Europe of frozen trout infected with the parasite (Hoffman 1990). Nearly 50 years later the parasite has spread to 24 states (K. Stromberg, Whirling Disease Initiative, Montana Water Center, Bozeman, MT, personal communication) and is of particular concern within the Intermountain West (Colorado, Montana, Idaho, Wyoming, Utah).

Major declines of rainbow trout populations, one of the most vulnerable salmonid species, have been documented in many rivers within the Intermountain West where *M. cerebralis* is present (Nehring and Walker 1996; Vincent 1996; Baldwin et al. 1998). The Colorado, Delores, Cache la Poudre, Fryingpan, Gunnison, South Platte, Rio Grande, and Roaring Fork rivers in Colorado (Bartholomew and Reno 2002) and the Madison River (Vincent 1996), Rock Creek, Willow Creek, and Little Prickly Pear Creek in Montana have all documented rainbow trout population declines within the last decade

(Baldwin et al. 1998). However, not all rainbow trout populations within and across drainages in Montana and elsewhere have been severely affected (Baldwin et al. 1998; Hiner and Moffitt 2001; Sandell et al. 2001). For example, some tributaries in Montana have consistently tested negative for the parasite, or salmonid disease severity has remained low, despite their proximity to infected streams where rainbow trout populations have declined dramatically (E. R. Vincent, Montana Department of Fish, Wildlife and Parks, Bozeman, MT, personal communication). What causes the parasite to be prevalent and have severe effects in some populations while remaining fairly benign in others is unknown. Rainbow trout life history traits (Downing et al. 2002), the differential ability to propagate the parasite by genetically distinct strains of *T. tubifex* (Beauchamp et al. 2002, 2005; DuBey and Caldwell 2004), and the environmental parameters within the stream (Schisler et al. 2000) are factors that potentially contribute to the variation in whirling disease severity. Additionally, anthropogenic disturbances within watersheds that alter these factors may influence whirling disease risk (Zendt and Bergersen 2000; Allendorf et al. 2001).

Identification of the factors that exacerbate whirling disease is further confounded by the complex life cycle of the parasite. Although whirling disease was first described over 100 years ago, the life cycle of the parasite was not fully understood until the alternate host was identified (Wolf and Markiw 1984; Wolf et al. 1986). The discovery revealed that there are two distinct spore stages and two hosts in the life cycle of the parasite. This led to the nullification of an entire class (Class Actinosporea) within the

Phylum Myxozoa (Kent et al. 2001) and sparked many studies to better understand the ecology and life cycle of the myxozoans.

The life cycle of *M. cerebralis* alternates between a salmonid fish and the oligochaete worm host, *T. tubifex*, from which the myxospore and TAM are released, respectively (El-Matbouli et al. 1992; El-Matbouli et al. 1999). The myxospore is released into the sediment upon decomposition of an infected salmonid or through the fecal matter of a predator which consumes the infected salmonid. The myxospore can lie dormant for an extended period of time, possibly years, and is able to withstand freezing and passage through avian and fish digestive systems until ingested by *T. tubifex* (El-Matbouli and Hoffmann 1991). Once inside the worm, the parasite attaches to, and penetrates between the intestinal epithelium cells where *M. cerebralis* multiplies and matures into TAMs (Wolf and Markiw 1984; El-Matbouli and Hoffmann 1989). TAMs are expelled into the water column through fecal matter (Gilbert and Granath 2001) or upon death of the tubificid (El-Matbouli et al. 1992). Unlike the myxospore, the TAM remains viable for only a few days to weeks (El-Matbouli et al. 1999). TAMs float in the water column until they come into contact with a fish. Upon contact, the TAM attaches to the epidermis of the trout using polar filaments and injects the sporoplasm of the parasite. The infective germ cells of the parasite penetrate the epidermis of the fish host, then travel via the nervous system to the cartilage where myxospores form and the life cycle is completed (El-Matbouli et al. 1995).

Rainbow trout life history traits such as size and age during early rearing and timing of emergence could significantly increase risk of exposure to the parasite and disease severity. Although rainbow trout can become infected at any point during their lifespan, manifestations of clinical signs in Erwin strain rainbow trout were significantly greater for rainbow trout fry smaller than 40 mm forklength during the first 9 weeks posthatch (Ryce et al. 2004, 2005). A study conducted on the Madison River, Montana by Downing et al. (2002) found that the emergence of rainbow trout occurred primarily from late June to early July, coinciding with an increase in stream temperature, decline in flow, and an increase in disease risk (E. R. Vincent, Montana Department of Fish, Wildlife and Parks, Bozeman, MT, personal communication). Emergence of rainbow trout fry is closely associated with temperature (Kwain 1975), and seasonal changes in water temperature are closely linked to seasonal changes in infection (El-Matbouli et al. 1999). Furthermore, low flow could concentrate TAMs, effectively increasing the risk of exposure to the parasite (MacConnell and Vincent 2002).

Recent studies suggest that some genetically distinct strains of *T. tubifex* are more resistant to *M. cerebralis* than others (Kerans et al. 2004) and are found in different stream habitats (Beauchamp et al. 2002, 2005; DuBey and Caldwell 2004). In the San Juan River TAM-producing lineage III *T. tubifex* were found in pool habitats, whereas the parasite was not detected in lineage VI, which were dominant in riffles (DuBey and Caldwell 2004). In rivers of central Colorado, susceptible lineage III *T. Tubifex* were more prominent in artificial habitat (e.g., reservoirs), whereas waters less affected by anthropogenic activities were dominantd by the less susceptible lineage V (Beauchamp et al. 2005). Therefore, stream channel morphology may further influence disease severity by selecting for or against various *T. tubifex* lineages (Anlauf and Neumann 1997).

Resistant strains of *T. tubifex* may possibly limit prevalence of infection, which is dependent upon myxospore abundance (Steinbach Elwell et al. in preparation), by ingesting myxospores and in doing so act as biological filters (El-Matbouli et al. 1999; Beauchamp et al. 2006). Beauchamp et al. (2006) found that TAM production was reduced by 70% when resistant and susceptible strains of *T. tubifex* were cultured together. However, Steinbach Elwell et al. (2006) found that neither infection prevalence nor parasite proliferation in susceptible *T. tubifex* was reduced when a susceptible *T. tubifex* strain was cultured with a resistant *T. tubifex* strain, although they did note a trend for infection prevalence to decrease as *T. tubifex* abundance increased. Steinbach Elwell et al. (2006) employed a response-surface competition design that controlled for both increases in density and composition, whereas the design of Beauchamp et al. (2006) confounded density and relative abundance. Key differences in experimental design likely contributed to the conflicting results of these studies; however, the effect of the lineage of *T. tubifex* on the disease in salmonids warrants further investigation.

Environmental factors may exacerbate disease severity by influencing parasite success within both hosts and in the spore stages. Water temperature affects development of the parasite within *T. tubifex* (El-Matbouli et al. 1999; Blazer et al. 2003; Kerans et al. 2005), TAM viability (El-Matbouli et al. 1999), and disease severity within the salmonid host (Schisler et al. 2000). Higher temperatures (13-20°C) can induce an earlier TAM release and amplify the number of TAMs as compared to lower temperatures (5-10°C), which delay the development of the TAM; however, at increased temperatures (>20°C), TAM viability decreases (El-Matbouli et al. 1999; Blazer et al. 2003). Schisler et al.

(2000) found that mortality among infected rainbow trout was much higher at 17°C than at 12.5°C. In addition, Halliday (1973) found that whirling disease symptoms (whirling behavior and blackened tails) were more prevalent and developed sooner at higher temperatures (17°C) than at lower temperatures (12°C and 7°C). Although these laboratory studies show a tendency for the parasite to flourish at higher temperatures (13-20°C), a field study of spring creeks in Montana detected the highest infection in rainbow trout at temperatures ranging from 6°C to 12°C (Anderson 2004). These differences among laboratory and field studies suggest that additional factors influence the effect of water temperature on infection of *T. tubifex* and severity of disease in rainbow trout.

Other environmental characteristics such as conductivity, substrate composition, and water velocity have also been correlated to whirling disease severity. Conductivity, a measurement of ion concentration, was positively correlated to infection of rainbow trout (Sandell et al. 2001). Conductivity generally increases with nutrient enrichment of the water (e.g., NO_2^-, NO_3^-), which can create ideal habitat for *T. tubifex* (Lestochova 1994). *Tubifex tubifex* thrives not only in nutrient enriched waters but also in areas of increased siltation (Juget and Lafont 1994). Such areas are indicative of low velocity waters that are generally inhabited by young trout. This is also where dead fish tend to be deposited by the current and where myxospores are most likely to settle into the sediment (Kerans and Zale 2002). Areas possessing one or more of these environmental characteristics, such as impoundments and backwater/pool areas where water velocities are low and silt can accumulate, may provide favorable habitat for the parasite and its oligochaete host (Krueger et al. 2006), in turn creating possible point sources of infection for trout

(Nehring et al. 2003). Disturbances or changes within the environment that influence these habitat characteristics may possibly influence whirling disease risk.

Because environmental conditions within a stream are often manifestations of activities within the watershed, anthropogenic alterations of the landscape (e.g., agricultural practices, mining and construction) can change the natural stream habitat and modify the physical, chemical, and biological processes within a stream, thus altering biological integrity (Karr 1991). Karr and Dudley (1981, pg 56) define biological integrity as “the capability of supporting and maintaining a balanced, integrated, adaptive community of organisms having a species composition, diversity, and functional organization comparable to that of the natural habitat of the region.” The primary mechanisms by which anthropogenic disturbances threaten the biological integrity of stream ecosystems is through sedimentation, nutrient enrichment, contaminant pollution, hydrologic alteration, loss of large woody debris, riparian clearing, and canopy opening (Allan 2004). For example, streams that drain deforested watersheds are often characterized by higher water temperatures and elevated levels of nutrients and sediments as compared to those draining forested catchments. These streams also have decreased energy input in the form of allochthonous material and woody debris that influences channel morphology, flow regimes, and provides cover for aquatic life (Webster et al. 1992; Bourque and Pomeroy 2001; Scott et al. 2002). Additionally, numerous watershed studies have documented relationships between an increase in agricultural practices and a decline in water quality, habitat, and biological assemblages (Lenat 1984; Sponseller et al. 2001), which support fewer sensitive fish and insect taxa than streams within forested watersheds (Genito et al. 2002). Fish and invertebrate assemblages often respond predictably to anthropogenic disturbances, making them a commonly used bioassessment tool to indicate the biological integrity of a stream.

Aquatic invertebrates are the most commonly used indicators of biological stream integrity (Rosenberg and Resh 1993). Invertebrates are commonly used because they have relatively short life spans, cannot easily avoid pollution or other disturbances, and are continuously responding to the cumulative effects of multiple stressors within their environment. Thus, they provide an indication of water quality, habitat conditions, and the potential causes and/or sources of impairment within the system. Streams lacking invertebrate diversity and sensitive taxa, those intolerant of pollution and other disturbances, indicate low biological integrity. For example, high abundances of *T. tubifex* are often an indicator of degraded habitat (Chapman and Brinkhurst 1984), associated with areas of increased sediment deposition (Zendt and Bergersen 2000) and organic matter, low dissolved oxygen concentrations, and slow water velocities (Brinkhurst 1996). Often times however, matching a biological indicator's response to a specific stressor is a complex task because streams are frequently influenced by multiple stressors that may interact (Allan 2004). Therefore, the primary purpose is often not to link a specific response to a specific anthropogenic stressor, but rather to determine the response of the system to the multitude of possible anthropogenic stressors within the environment.

It has been suggested that many of the streams where *M. cerebralis* is present are those with altered or degraded habitat (Allendorf et al. 2001). The water quality in 9,826 miles of Montana's streams is listed as impaired, unable to fully or partially support at least one designated beneficial use as defined by the state's Water-Use Classification System due to anthropogenic activities (MTDEQ 2004). Designated uses are categorized as water supplies supporting agricultural, industrial, recreation, drinking water, aquatic life, and fisheries (cold and warm) purposes. Water quality impairments are primarily due to increased nutrients and siltation, thermal modifications, and alteration of flow regimes as a result of agricultural and silvicultural practices, construction, mining and milling processes, hydromodification, and habitat modification such as removal of riparian vegetation and/or streambank alteration (MTDEQ 2004). Possibly, these water quality impairments also result in increased whirling disease risk.

A clearer understanding of the relationships between whirling disease risk and land use practices, measures of biological stream integrity, and the physicochemical features of streams are needed to help explain the broad scale variation in whirling disease risk. By identifying possible contributors to, and indicators of, stream degradation and their relationship to whirling disease risk, streams can be identified that are at the greatest risk of parasite establishment and proliferation. In addition, streams that would benefit from remediation can be identified and attempts to mitigate the effect of this disease on wild rainbow trout populations can be undertaken.

STUDY AREAS

The study area encompassed the mainstem and selected tributaries of four major drainages (Missouri, Madison, Blackfoot and Rock Creek) within western Montana (Figure 1). This study analyzed data collected in a much larger field monitoring project that was initiated in response to the detection of whirling disease within the state in 1994. Two of the drainages (Blackfoot and Rock Creek) are west of the continental divide and two (Madison and Missouri) are east of the divide, thus providing a broad range of environmental conditions. Physical, chemical, and invertebrate data were collected from 1997 through 2000 and whirling disease risk (i.e., infection severity and prevalence in caged sentinel trout, see Whirling Disease Risk) was assessed from 1997 through 2004, although the number of times sites within drainages were sampled varied (see METHODS; Appendix A). Selection of sampling sites was based on accessibility, previous data, and opinions of Montana Department of Fish, Wildlife and Parks (MFWP) personnel. In most cases, the designated beneficial uses (support of agriculture, industrial, recreation, drinking water, aquatic life and fisheries (cold) water supply) of all streams were assessed by MTDEQ (2004). The beneficial use(s) of some streams were not assessed due to a lack of sufficient data or because a stream was not designated to support one or more specific uses. If any beneficial use for a stream was not assessed it is noted in that stream's respective site description.

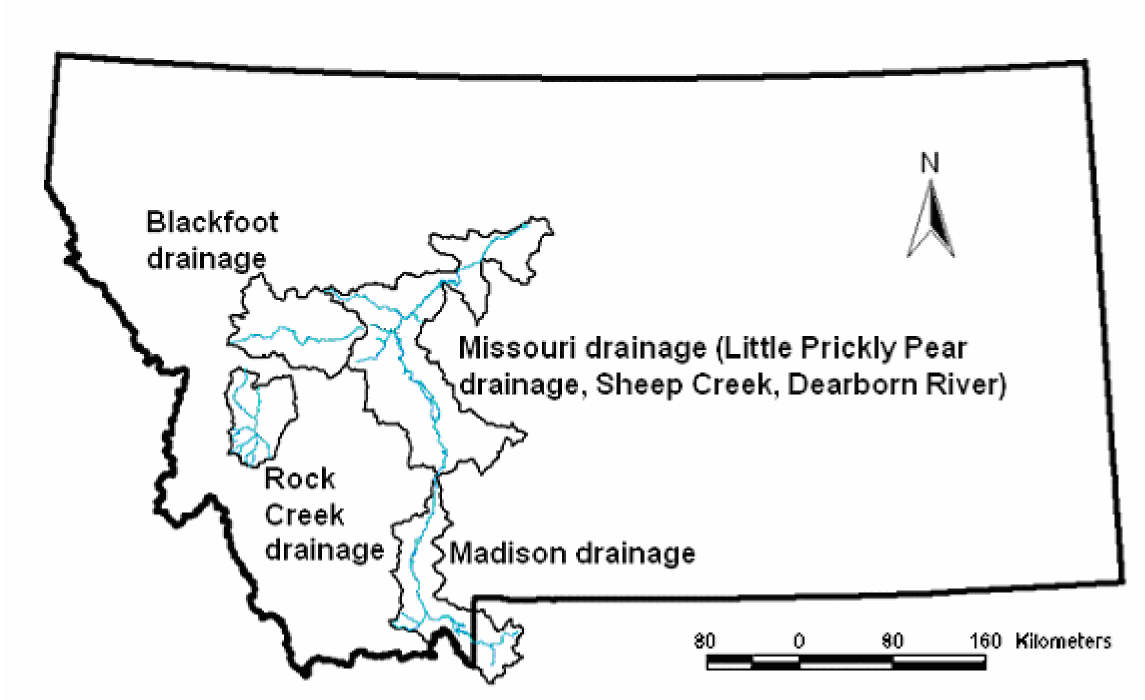

Figure 1. The mainstem and select tributaries of the Missouri River, Madison River, Blackfoot River and Rock Creek drainages within western Montana.

Missouri River Drainage

Tributaries of the Missouri River downstream of the Holter Dam that were selected for this study included Little Prickly Pear Creek, the Dearborn River, and Sheep Creek. Two tributaries of Little Prickly Pear Creek, Lyons Creek and Wolf Creek, and two tributaries of the Dearborn River, the South and Middle Fork were also included.

Little Prickly Pear Creek

The mainstem of Little Prickly Pear Creek originates at approximately 1480 m at the confluence of the North and South Forks of Little Prickly Pear Creek, draining an area of 1032 km^2 (USDA 1996). The creek is approximately 54 km long and enters the

Missouri River 4.4 km downstream of Holter Dam (iGage Mapping Corporation 2002). Six sites were located on Little Prickly Pear Creek at 7.5 (PR1), 18.3 (PR2), 28.3 (PR3), 34.3 (PR4), 37.7 (PR5), and 42.2 km (PR6) upstream from the mouth (Figure 2). Catchment elevations range between roughly 2500 m in the upper headwater reaches and 1050 m in the lowland plain areas. The predominant lithology of the watershed is meta-argillite, quartzite, carbonate, diorite, and volcanic rock with alluvial deposits along the creek (Stoeser et al. 2005). MTDEQ (2004) lists Little Prickly Pear Creek as having one or more beneficial uses impaired. Probable anthropogenic causes and sources listed are numerous (Appendix B).

Lyons Creek

Lyons Creek is approximately 7.5 km long, entering Little Prickly Pear Creek 16 km above its confluence with the Missouri River (iGage Mapping Corporation 2002). Lyons Creek originates at approximately 1260 m at the confluence of the North and South Forks of Lyons Creek and drains an area of 77 km^2. Elevations within the catchment range from 2300 m to 1150 m. One site on Lyons Creek (LY), approximately 1200 m upstream of the mouth, was selected for the study (Figure 2). Meta-argillite and carbonate are the dominant rock types (Stoeser et al. 2005). No assessment of the creek's beneficial uses has been conducted. Thus, this stream is not listed on the state's integrated water quality report.

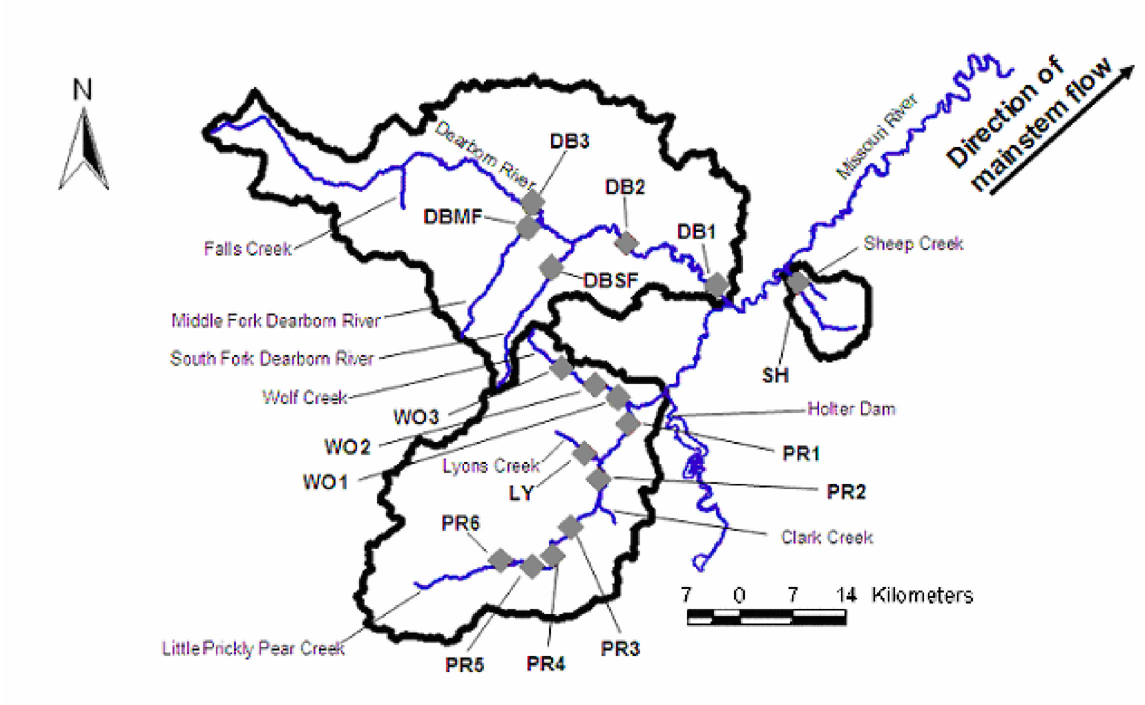

Figure 2. Site locations (indicated by diamonds) within the Missouri River drainage. Sites shown from downstream to upstream on Little Prickly Pear Creek are PR1, PR2, PR3, PR4, PR5, and PR6. Sites shown from downstream to upstream on Wolf Creek are WO1, WO2, and WO3. Sites shown from downstream to upstream on the Dearborn River are DB1, DB2, and DB3. Sites LY, SH, DBMF, and DBSF are located on Lyons Creek, Sheep Creek, Middle Fork Dearborn River, and the South Fork Dearborn River, respectively.

<u>Wolf Creek</u>

Wolf Creek originates at approximately 1700 m and is 19 km long (iGage Mapping Corporation 2002). The creek drains roughly 99 km^2 before entering Little Prickly Pear Creek 6.2 km above its confluence with the Missouri River. Elevations range from 2100 m in the upper sections of the catchment to 1100 m near its confluence with Little Prickly Pear Creek. Sites selected on Wolf Creek were 0.1 (WO1), 4.8 (WO2), and 10.9 km (WO3) upstream of the mouth (Figure 2). The lithology of the catchment is primarily meta-argillite, carbonate, diorite, and volcanic rock (Stoeser et al. 2005). No assessment of the creek's beneficial uses has been conducted. Thus, this stream is not listed on the state's integrated water quality report.

Dearborn River

The Dearborn River originates at 2260 m and is approximately 113 km long, draining an area of 1418 km^2 before entering the Missouri river 22 km below Holter Dam (iGage Mapping Corporation 2002; USDA 1996). Elevations range between 2700 m in the upper-most sections of the catchment and 1050 m near the Missouri River confluence. The predominant rock type is carbonate, clastic, volcanic, and meta-argillite, with pockets of alluvium and quartzite in the headwater reaches (Stoeser et al. 2005). Sites were located 0.6 (DB1), 30.9 (DB2), and 47.3 km (DB3) upstream of the mouth (Figure 2). The Dearborn River (Falls Creek to confluence Missouri River) is listed as having one or more beneficial uses impaired (MTDEQ 2004). Probable causes are related to agricultural practices and hydromodification (Appendix B).

Middle Fork Dearborn River

The Middle Fork of the Dearborn River is 23 km long (iGage Mapping Corporation 2002) and drains an area of approximately 175 km^2 (USDA 1996). The Middle Fork originates at approximately 1675 m with catchment elevations ranging between 2200 m and 1250 m. One site on the Middle Fork (DBMF) was selected approximately 123 m upstream of the confluence with the Dearborn River (Figure 2). The predominant lithology is meta-argillite, clastic, carbonate, and volcanic rock (Stoeser et al. 2005). The Middle Fork of the Dearborn River is listed as fully supporting all uses (MTDEQ 2004, Appendix B).

South Fork Dearborn River

The South Fork of the Dearborn River is approximately 26 km long (iGage Mapping Corporation 2002) and drains an area of 108 km^2. Catchment elevations range from 2150 m to 1210 m. The creek originates at approximately 1880 m. The predominant lithology is meta-argillite, clastic, carbonate, and volcanic rock (Stoeser et al. 2005). One site was selected on the South Fork (DBSF), approximately 5.6 km upstream of the confluence with the Dearborn River (Figure 2). The South Fork of the Dearborn River is listed as only partially supporting two beneficial uses (MTDEQ 2004). Probable causes are related to agricultural practices (Appendix B).

Sheep Creek

The mainstem of Sheep Creek enters the Missouri River 38 km below Holter Dam, originating at the confluence of the North and South Forks of Sheep Creek at an elevation of 1060 m (iGage Mapping Corporation 2002). The creek drains an area of roughly 93 km^2 and is approximately 3.3 km long. Elevations within the catchment range from 2150 m to 1030 m and is comprised primarily of volcanic rock with a few pockets of alkalic intrusive rock and some sandstone on the northern boundary of the catchment (Stoeser et al. 2005). The Sheep Creek site (SH) was located 926 m above the mouth of the creek (Figure 2). No assessment of the creek's beneficial uses has been conducted. Thus, this stream is not listed on the state's integrated water quality report.

Madison River Drainage

The mainstem of the Madison River between Hebgen Lake and Ennis Lake was included in this study as well as two tributaries of the Madison River, the West Fork of the Madison River and Squaw Creek.

Madison River

The mainstem of the Madison River drains an area of 6,606 km^2 (USDA 1996) and is approximately 193 km long. The river originates at 2073 m at the confluence of the Gibbon and Firehole River in Yellowstone National Park, Wyoming and terminates at its confluence with the Jefferson and Gallatin Rivers to form the Missouri River. Catchment elevations range roughly between 3436 m in portions of the Beaverhead National Forest near the headwaters of Squaw Creek to 1230 m near the confluence with the Jefferson and Gallatin Rivers. The dominant rock type is clastic, metamorphic, volcanic, and carbonate with pockets of glacial drift and sandstone (Stoeser et al. 2005). A large portion of the eastern section of the drainage near Ennis is comprised of diorite and there are many alluvial deposits along the mainstem of the Madison River. Eight sites (MA1-MA8) were located between Hebgen Lake and Ennis Lake, Montana (Figure 3). Hebgen Dam controls the flow of the Madison River, with a required minimum discharge of 17m^3/s (Vincent 1996). The Madison River between Hebgen Dam and Ennis Lake is listed as fully supporting all beneficial uses (MTDEQ 2004, Appendix B).

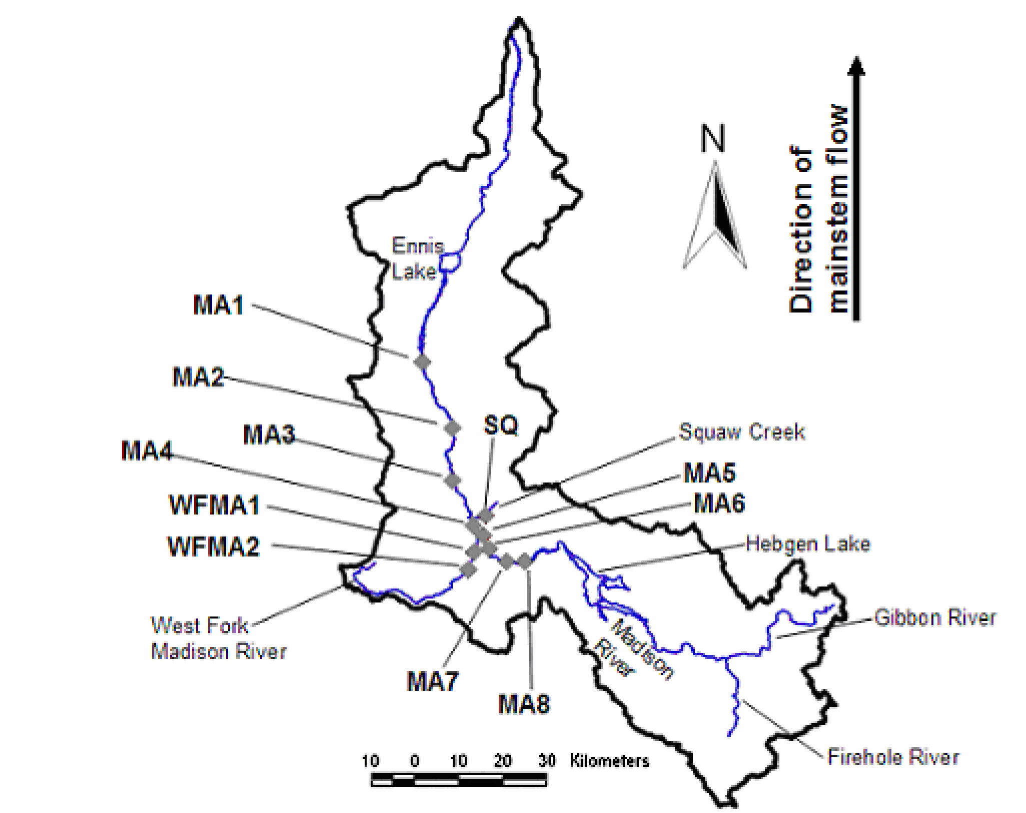

Figure 3. Site locations (indicated by diamonds) within the Madison River drainage. Sites shown from downstream to upstream on the Madison River mainstem are MA1, MA2, MA3, MA4, MA5, MA6, MA7, and MA8. Sites shown from downstream to upstream on the West Fork of the Madison River are WFMA1 and WFMA2. Site SQ is located on Squaw Creek.

West Fork Madison River

The West Fork is approximately 61 km long and originates at roughly 2800 m, entering the mainstem of the Madison River 30 km below Hebgen Dam (iGage Mapping Corporation 2002). The river drains an area of 574 km^2 (USDA 1996). The dominant lithology is metamorphic and volcanic rock with pockets of clastic, carbonate, and sandstone near the headwaters (Stoeser et al. 2005). Elevations range from 3000 m in the upper sections of the catchment to 1800 m near the confluence. Two sites were located on the West Fork, sites WFMA1 and WFMA2, approximately 0.3 and 4.3 km above the mouth, respectively (Figure 3). MTDEQ (2004) lists the West Fork of the Madison River as not supporting one or more beneficial uses. Probable causes and sources reported are numerous (Appendix B).

Squaw Creek

Squaw Creek is 12.5 km long and originates at 2800 m, entering the Madison River 34 km below Hebgen Dam (iGage Mapping Corporation 2002). The creek drains an area of 47 km^2. Elevations range between 3436 m in the upper sections of catchment and 1780 m near the confluence. The Squaw Creek site (SQ) was located roughly 80 m upstream of the creek mouth (Figure 3). The predominant lithology is metamorphic and clastic rock with a small pocket of volcanic rock near the confluence (Stoeser et al. 2005). No assessment of the creek’s beneficial uses has been conducted. Thus, this stream is not listed on the state’s integrated water quality report.

Rock Creek Drainage

Within the Rock Creek drainage, the mainstem along with select tributaries, the East Fork, West Fork, Middle Fork, Ross Fork, Willow Creek, Stony Creek, and Hogback Creek, were included in this study.

Rock Creek

Rock Creek originates at the confluence of the West and Middle Fork of Rock Creek at approximately 1600 m. The Rock Creek mainstem is 84 km long (iGage Mapping Corporation 2002) and drains an area of roughly 2304 km^2 (USDA 1996). Catchment elevations range from 3183 m in the upper sections of the Middle Fork drainage and 1072 m near its confluence with the Clark Fork River. Five sites in Rock Creek (RO1-RO5) were selected for this study (Figure 4). The predominant lithology is quartzite and meta-argillite (Stoeser et al. 2005). Large portions of carbonate, glacial drift, and granitoid are also present, with smaller pockets of clastic and volcanic rock. Alluvial deposits are found along the majority of the mainstem and some tributaries. The Rock Creek mainstem is listed as fully supporting all beneficial uses (MTDEQ 2004, Appendix B).

East Fork Rock Creek

The East Fork of Rock Creek originates at 2150 m. The creek drains an area of 207 km^2 before terminating at the confluence with the Middle Fork Rock Creek (USDA 1996). The creek is approximately 23 km long with a dam located 13.5 km upstream of the mouth (iGage Mapping Corporation 2002). Catchment elevations range from 3050 m to 1644 m. The East Fork site (EFRO) was located approximately 1400 m above the confluence with the Middle Fork Rock Creek (Figure 4). The dominant rock type within the catchment is carbonate and quartzite (Stoeser et al. 2005). There are small pockets of glacial drift, meta-argillite, and volcanic rock, and alluvial deposits are present along the stream. The East Fork (mouth to 14 km upstream) is listed as fully supporting all evaluated beneficial uses (MTDEQ 2004, Appendix B). The creek's support of aquatic life and cold water fisheries was not assessed.

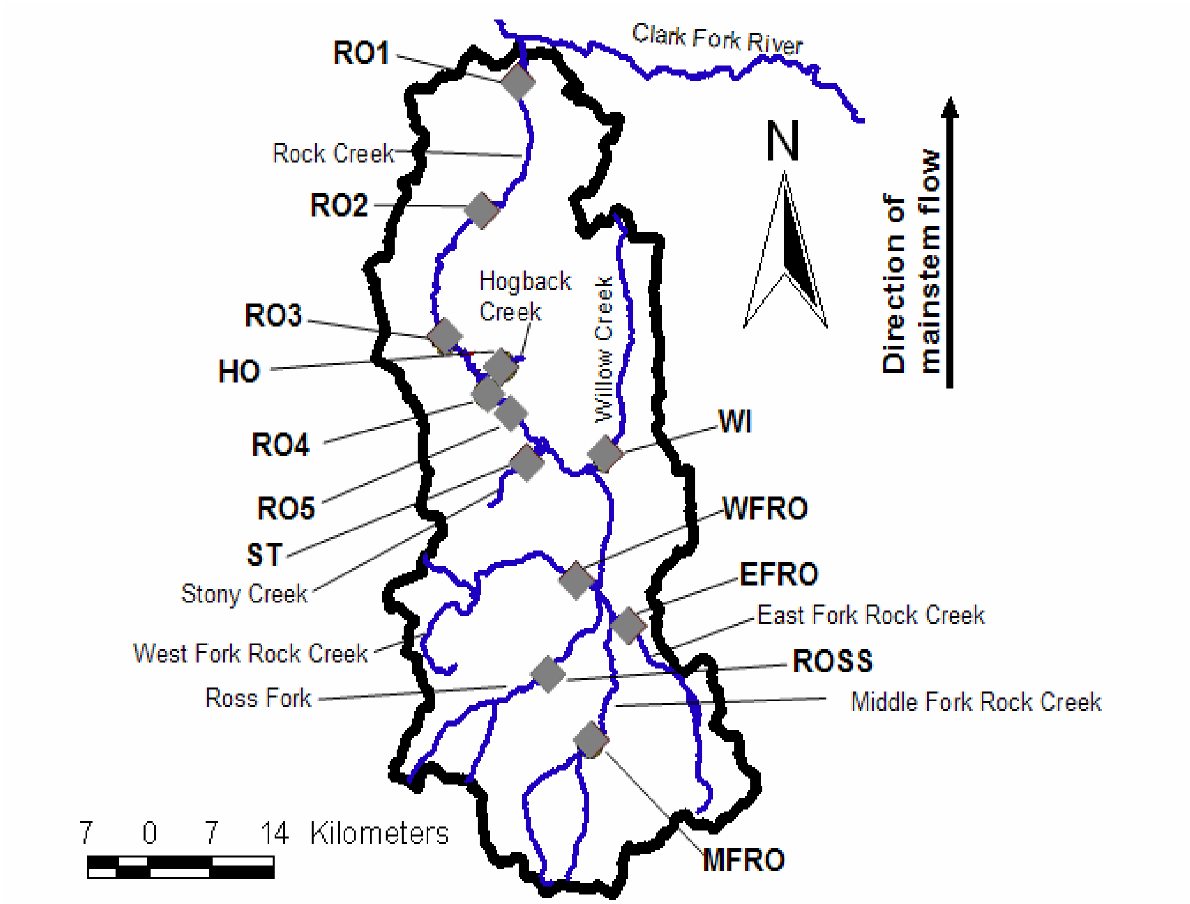

Figure 4. Site locations (indicated by diamonds) within the Rock Creek drainage. Sites shown from downstream to upstream on the Rock Creek mainstem are RO1, RO2, RO3, RO4, and RO5. Sites EFRO, MFRO, WFRO, ROSS, WI, ST, and HO are located on East Fork Rock Creek, Middle Fork Rock Creek, West Fork Rock Creek, Ross Fork, Willow Creek, Stony Creek, and Hogback Creek, respectively.

Middle Fork Rock Creek

The Middle Fork of Rock Creek originates at 2445 m. The Middle Fork is 34 km long (iGage Mapping Corporation 2002) and drains an area of 316 km^2 (USDA 1996). Elevations range from 3183 m to 1600 m. One site on the Middle Fork (MFRO) was selected approximately 23 km upstream from the mouth (Figure 4). The predominant lithology is glacial drift and quartzite (Stoeser et al. 2005). There are small pockets of carbonate and granitoid in the upper reaches of the catchment and alluvial deposits are present along the majority of the creek. No assessment of the creek's beneficial uses has been conducted. Thus, this stream is not listed on the state's integrated water quality report.

West Fork Rock Creek

The West Fork of Rock Creek originates at 2200 m. The creek is 39 km long (iGage Mapping Corporation 2002), draining an area of approximately 239 km^2 (USDA 1996). Elevations range from 2630 m near Mount Emerine to 1600 m near its confluence with Rock Creek. The West Fork site (WFRO) was located approximately 300 m from the creek's mouth (Figure 4). The dominant rock types are glacial drift, granitoid, and quartzite with pockets of carbonate, clastic, and volcanic rock (Stoeser et al. 2005). Alluvial deposits are present near the lower reaches of the creek. MTDEQ (2004) lists the West Fork as not supporting its beneficial use as a drinking water supply. Probable causes have been identified, yet the probable source is unknown (Appendix B). Its support of aquatic life and cold water fisheries was not assessed.

Ross Fork

Ross Fork is 37 km long and originates at 2310 m (iGage Mapping Corporation 2002). Catchment elevations range between 2710 and 1600 m. The creek drains an area of approximately 221 km^2 (USDA 1996). The predominant lithology is glacial drift, granitoid, and quartzite with pockets of carbonate and clastic rock (Stoeser et al. 2005). The Ross Fork site (ROSS) was located 13 km upstream from the mouth (Figure 4). No assessment of the creek's beneficial uses has been conducted. Thus, this stream is not listed on the state's integrated water quality report.

Willow Creek

Willow Creek is 32 km long and originates at 2115 m, entering the mainstem 14 km below its origination (iGage Mapping Corporation 2002). The creek drains an area of 247 km^2 (USDA 1996). Elevations range between 2525 m and 1500 m within the catchment. The Willow Creek site (WI) was located 30 m upstream from the confluence with the Rock Creek mainstem (Figure 4). Quartzite is the dominant rock type with small pockets of carbonate, clastic, meta-argillite, and volcanic rock (Stoeser et al. 2005). Alluvial deposits are present along the stream. MTDEQ (2004) lists Willow Creek as having one or more beneficial uses impaired (Appendix B). Its use as a drinking water supply was not assessed.

Stony Creek

Stony Creek is 16 km long, originating at 1705 m and enters the Rock Creek mainstem 20 km below its origination (iGage Mapping Corporation 2002). The site on

Stony Creek (ST) was located 1050 m upstream from the mouth (Figure 4). Elevations range between 2560 m at the catchment boundary and 1450 m near the confluence with Rock Creek. The creek drains an area of roughly 74 km^2. The predominant rock type is meta-argillite, with pockets of carbonate and volcanic rock (Stoeser et al. 2005). Glacial drift runs along upper reaches of the stream and alluvial deposits are found near the confluence. No assessment of the creek's beneficial uses has been conducted. Thus, this stream is not listed on the state's integrated water quality report.

Hogback Creek

Hogback Creek is 6.5 km long, entering the mainstem 42 km below its origination (iGage Mapping Corporation 2002). The creek originates at 1650 m and drains an area of 42 km^2. Elevations within the catchment range between 2500 m and 1370 m. The Hogback Creek site (HO) was located approximately 100 m upstream of the mouth (Figure 4). Meta-argillite is the dominant rock type (Stoeser et al. 2005). Quartzite is found in the upper portions of the catchment and alluvial deposits are found along the lower reaches of the stream. No assessment of the creek's beneficial uses has been conducted. Thus, this stream is not listed on the state's integrated water quality report.

Blackfoot Drainage

The mainstem of the Blackfoot River and several of its tributaries, Cottonwood Creek, Chamberlain Creek, Warren Creek, and the North Fork of the Blackfoot River along with one of its tributaries, Kleinschmidt Creek, were selected for the study.

Blackfoot River

The Blackfoot River originates at approximately 1650 m and is 200 km long (iGage Mapping Corporation 2002). The river drains an area of roughly 5990 km^2 (USDA 1996). Catchment elevations range from 2860 m to 1000 m near its confluence with the Clark Fork River. Three sites in the Blackfoot River were selected for this study approximately 0.5 (BL1), 55 (BL2), and 150 km (BL3) upstream of the mouth (Figure 5). The predominant lithology is carbonate, glacial drift, clastic, meta-argillite, quartzite, and volcanic rock (Stoeser et al. 2005). MTDEQ (2004) lists the Blackfoot River as having one or more beneficial uses impaired. Probable anthropogenic causes and sources listed are numerous (Appendix B).

Cottonwood Creek

Cottonwood Creek drains an area of approximately 180 km^2 (USDA 1996). The creek originates at 2190 m and is 28 km long (iGage Mapping Corporation 2002). Catchment elevations range from 2450 m to 1180 m near its confluence with the Blackfoot River mainstem. The Cottonwood Creek site (CO) was located roughly 1.3 km upstream of the mouth (Figure 5). The dominant rock type is carbonate and glacial drift, with alluvial deposits along the lower reaches of the stream (Stoeser et al. 2005). Cottonwood Creek is listed as fully supporting all beneficial uses (MTDEQ 2004, Appendix B).

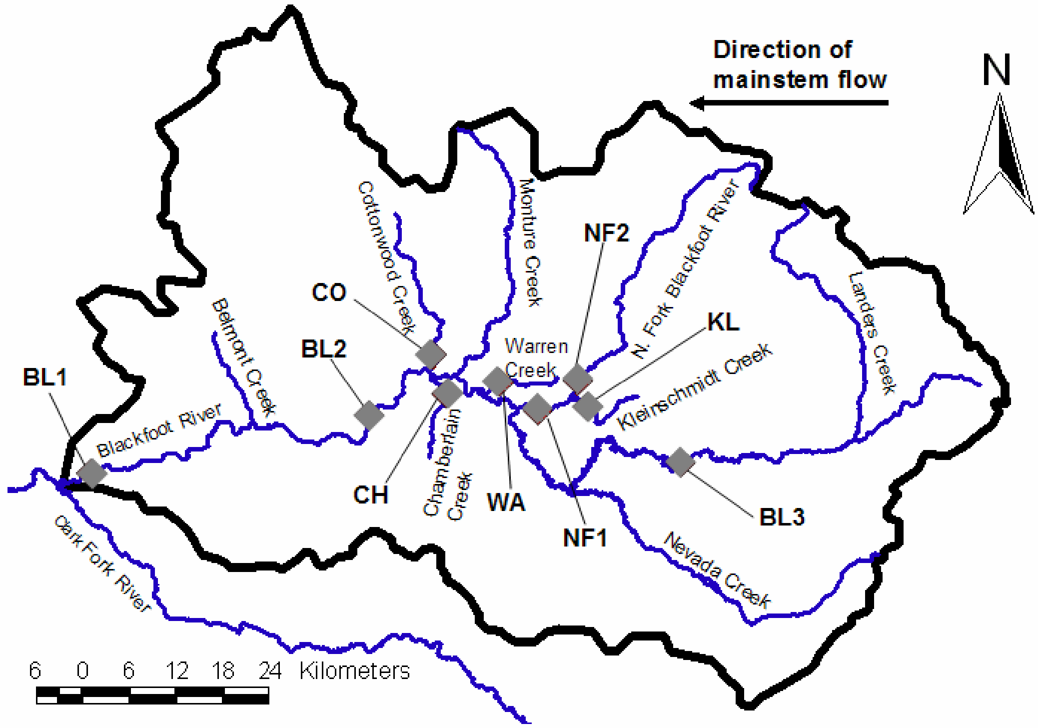

Figure 5. Site locations (indicated by diamonds) within the Blackfoot River drainage. Sites shown from downstream to upstream on the Blackfoot River mainstem are BL1, BL2, and BL3. Sites shown from downstream to upstream on the North Fork of the Blackfoot River are NF1 and NF2. Sites KL, WA, CH, and CO are located on Kleinschmidt Creek, Warren Creek, Chamberlain Creek, and Cottonwood Creek, respectively.

Chamberlain Creek

Chamberlain Creek originates at 1850 m and is roughly 18 km long (iGage Mapping Corporation 2002), draining an area of 57 km^2. Catchment elevations range from 2070 m to 1180 m near its confluence with the Blackfoot River mainstem. The Chamberlain Creek site (CH) was located 200 m upstream of the mouth (Figure 5). The catchment lithology is predominantly quartzite (Stoeser et al. 2005). MTDEQ (2004) assessed the lower section of Chamberlain Creek (mouth to 4.3 km upstream) and lists it as fully supporting all beneficial uses (Appendix B).

Warren Creek

Warren Creek originates at 2160 m near Ovando Mountain. The creek is approximately 26 km long (iGage Mapping Corporation 2002) and drains an area of 60 km^2. Catchment elevations range from 2340 m to 1210 m near the confluence with the Blackfoot River mainstem. The Warren Creek site (WA) was located approximately 80 m upstream of the mouth (Figure 5). The predominant lithology is carbonate, glacial drift, and alluvial deposits (Stoeser et al. 2005). Warren creek is listed as having one or more beneficial uses impaired (MTDEQ 2004). Probable causes are due to flow and other habitat alterations (Appendix B).

North Fork Blackfoot River

The North Fork of the Blackfoot River is 62 km long, originating at approximately 2430 m (iGage Mapping Corporation 2002). The river drains an area of 812 km^2 (USDA 1996). Catchment elevations range from 2800 m to 1230 m near its confluence with the mainstem of the Blackfoot River. Two sites were located on the North Fork of the Blackfoot River, sites NF1 and NF2, approximately 4.0 and 9.5 km above the mouth, respectively (Figure 5). The lithology of the catchment is predominantly alluvium, carbonate, meta-argillite, quartzite, and glacial drift (Stoeser et al. 2005). The North Fork of the Blackfoot River (mouth to 29 km upstream) is listed as fully supporting all beneficial uses (MTDEQ 2004, Appendix B).

Kleinschmidt Creek

Kleinschmidt Creek is approximately 7 km long (iGage Mapping Corporation 2002) and drains an area of 42 km^2. The creek originates at 1320 m and catchment elevations range between 2200 m and 1260 m. The Kleinschmidt Creek site (KL) was located 30 m upstream from its mouth (Figure 5). The dominant rock type is glacial drift, carbonate, meta-argillite, and alluvium (Stoeser et al. 2005). MTDEQ (2004) lists Kleinschmidt Creek (from mouth to 2.4 km upstream) as not supporting one or more assessed beneficial uses. Probable causes and sources reported are numerous (Appendix B).

METHODS

Whirling disease risk was assessed at two spatial scales within each drainage: a broad landscape scale (catchment) and a more localized site scale (reach). At the catchment scale, land use was quantified within the entire subbasin and riparian area upstream of the location where whirling disease risk (i.e., sentinel cages, see Whirling Disease Risk below) was measured. At the reach scale, land use within the subbasin and riparian zone, measures of biological stream integrity, and physicochemical parameters were quantified between the upper and lower sites in a reach (i.e., sentinel cages, see Whirling Disease Risk below). Land use was quantified at both the catchment and reach scale to assess whether relationships to risk differed at varying spatial extents. Biological stream integrity and physicochemical parameters were assessed at the reach scale only as they are often more indicative of localized site conditions than of broad landscape scale conditions.

Whirling Disease Risk

Whirling disease risk was quantified at each site as the infection severity and disease prevalence in sentinel rainbow trout. Sentinel fish are commonly employed for this task (e.g., Hiner and Moffitt 2001; Downing et al. 2002), providing an index of disease severity and an indirect measure of TAM abundance (Kerans and Zale 2002). Data from field exposures of sentinel rainbow trout with relatively equal susceptibility to whirling disease (Eagle Lake, Erwin, and Arlee strains) (Vincent 2002) were provided by

MFWP. Fifty rainbow trout fry between 40 and 60 mm in length and 60 to 80 days old were placed in each sentinel cage. Field exposures lasted ten days and generally occurred from mid-May through June from 1997 through 2004. After exposure, sentinel trout were returned to either the Wild Trout Laboratory in Bozeman, MT or the MFWP Laboratory in Pony, MT where surviving fish were sacrificed by exposure to 5% tricaine methanesulfonate after approximately 90 days to allow sufficient time for myxospores to develop. Gill arches and heads were removed and preserved and sent to the Washington Animal Disease Diagnostic Laboratory for histological examination. Each fish was scored on the MacConnell-Baldwin scale of zero to five based on the amount and severity of cartilage degeneration and infiltration of leukocytes (Baldwin et al. 2000). A score of zero indicates no abnormalities and five, the highest score, indicates severe infection including bone displacement into the brain or spinal cord.

The sentinel cage with the highest mean histological score among fish was used in these analyses to represent the maximum “risk” at each site. Because not all sites had sentinel cages after the years of invertebrate sampling, risk was quantified for two temporal scales: 1) when invertebrates were sampled in that particular drainage and 2) throughout the entire field exposure period (1997 – 2004). Both infection severity and disease prevalence were quantified at each temporal scale. Infection severity was calculated as the proportion of infected fish with a histological score greater than or equal to four during invertebrate sample years (SEVSAMP) and over all years (SEVALL). Disease prevalence was calculated as the proportion of infected fish during invertebrate sample years (PREVSAMP) and over all years (PREVALL). Disease risk was measured as both infection severity and disease prevalence to represent the two “extremes” of whirling disease within a system and assess whether features of the environment that permit establishment of the parasite within a system differ from those that promote severe infection within the fish host.

It is likely that infection severity and disease prevalence at sites within streams are spatially autocorrelated; especially given that TAMs can potentially drift in the water column farther than the distance between sentinel cages. To help minimize the effects of some possible spatial autocorrelation for statistical analyses, a decline in SEVSAMP values was used to classify sites into stream reaches. A decrease in the SEVSAMP value of 0.15 was chosen as a decline in infection severity great enough to indicate a point where sentinel fish were suspected to be unaffected by upstream sources of TAMs based on the difference in average infection severity in replicate sentinel rainbow trout cages (Erwin strain, mean = 0.077, standard deviation=0.107, n=3) (calculated using data from Anderson (2004)). SEVSAMP values were used to define reaches for two reasons: 1) severe infection represents the ultimate risk to fish, death, and therefore should relate more closely to fish population dynamics (E. R. Vincent, Montana Department of Fish, Wildlife and Parks, Bozeman, MT, personal communication) and 2) not all sites had sentinel cages after the years of invertebrate sampling; therefore, the period of invertebrate sampling represented sites when all had sentinel cages. An exception to this was the Missouri River drainage where many new locations were added (i.e., sites DBSF, DBMF, DB1, and DB3) after the period of invertebrate sampling. Therefore, stream reaches were defined separately for the period of invertebrate sampling using SEVSAMP values and throughout the entire field exposure period using SEVALL values.

Stream reaches within each drainage were defined by classifying the most upstream sites in headwater reaches and tributaries first, and then sites in a downstream direction, the last site defined being the one closest to the mouth of the drainage on the mainstem (Figures 6-9). The most upstream site in a headwater reach or tributary was defined as the upper site in that reach. Subsequent sites located downstream were considered to be part of the same reach as long as the SEVSAMP values increased or remained the same. Any decrease in the SEVSAMP value ≥ 0.15 (mean decrease = 0.36) terminated that reach. The site where the SEVSAMP decrease ≥ 0.15 occurred became the beginning of the new reach, and the site above was defined as the lowermost cage in the terminated reach. Reaches in mainstems terminated at confluences, unless the adjoining tributary was negative or its infection status was unknown, and the next downstream cage marked the beginning of a new reach. In situations where only one site was located on a tributary or headwater reach, it was considered to be the lowermost site in the reach under the assumption that at some point upstream SEVSAMP values must equal zero.

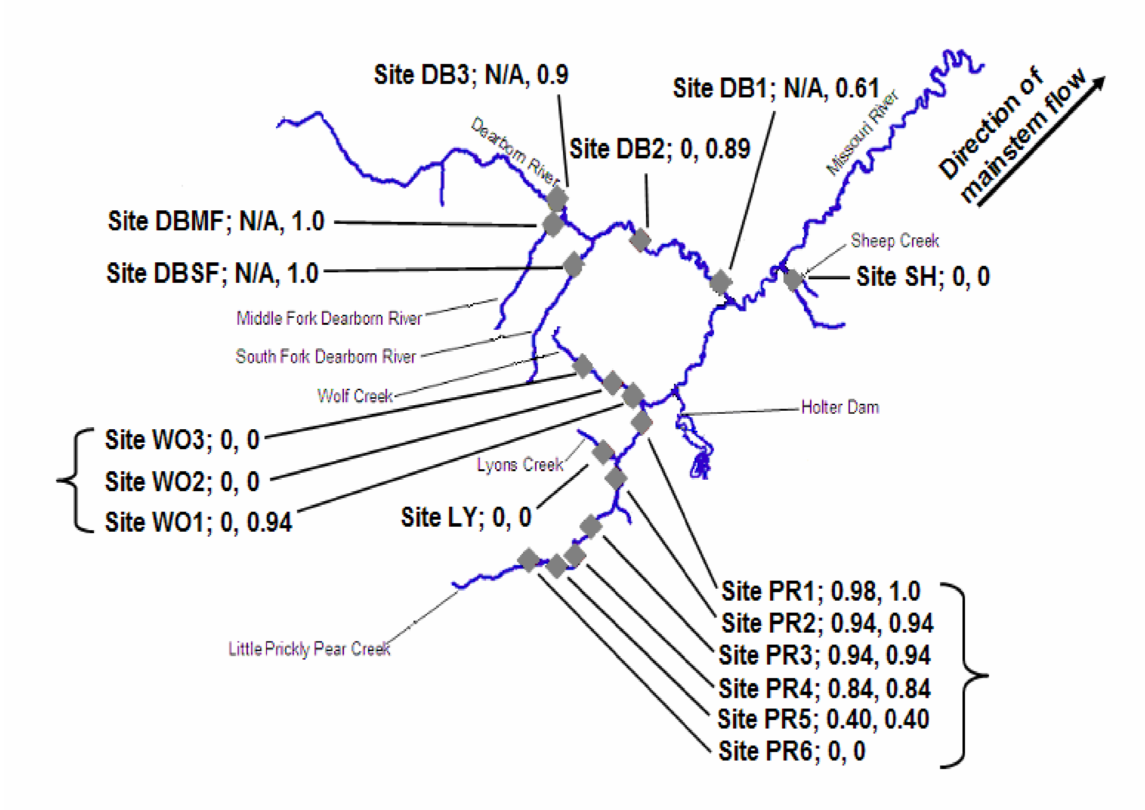

Figure 6. Sites within the Missouri River drainage and their respective SEVSAMP values (proportions of infected fish with a histological score greater than or equal to four during invertebrate sample years) and SEVALL values (proportions of infected fish with a histological score greater than or equal to four over all years) used to define reaches for each temporal scale; 1) when invertebrates were sampled within the Missouri River drainage and 2) throughout the entire field exposure period (1997 – 2004). Stream reaches were defined separately for the period of invertebrate sampling (SEVSAMP) and throughout the entire field exposure period (SEVALL) because many new locations were added (i.e., sites DBSF, DBMF, DB3, and DB1) after the period of invertebrate sampling. Thus, a SEVSAMP value could not be quantified at all sites, indicated by N/A. Reaches comprised of multiple sites are designated by brackets.

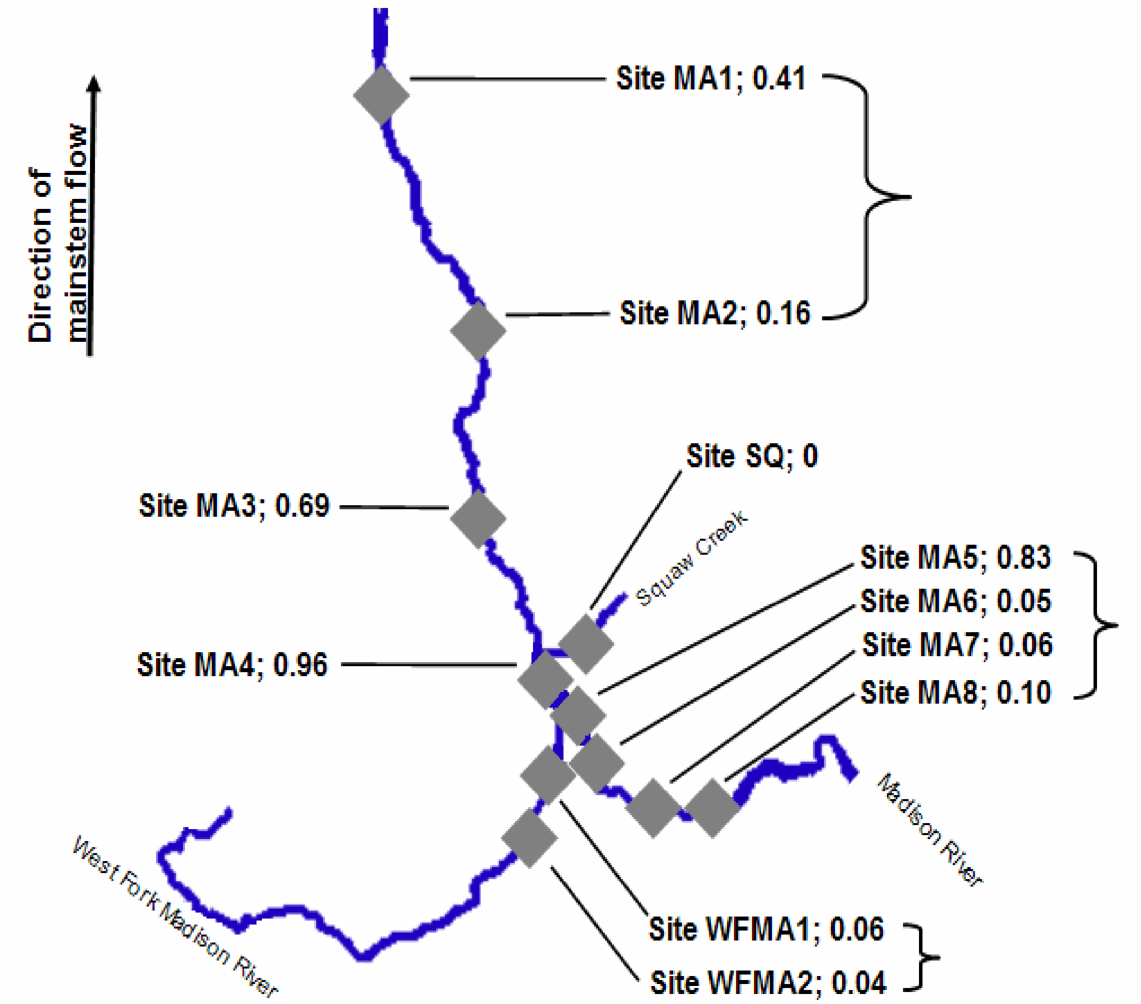

Figure 7. Sites within the Madison River drainage and their respective SEVSAMP values (proportions of infected fish with a histological score greater than or equal to four during invertebrate sample years) used to define reaches. Reaches comprised of multiple sites are designated by brackets.

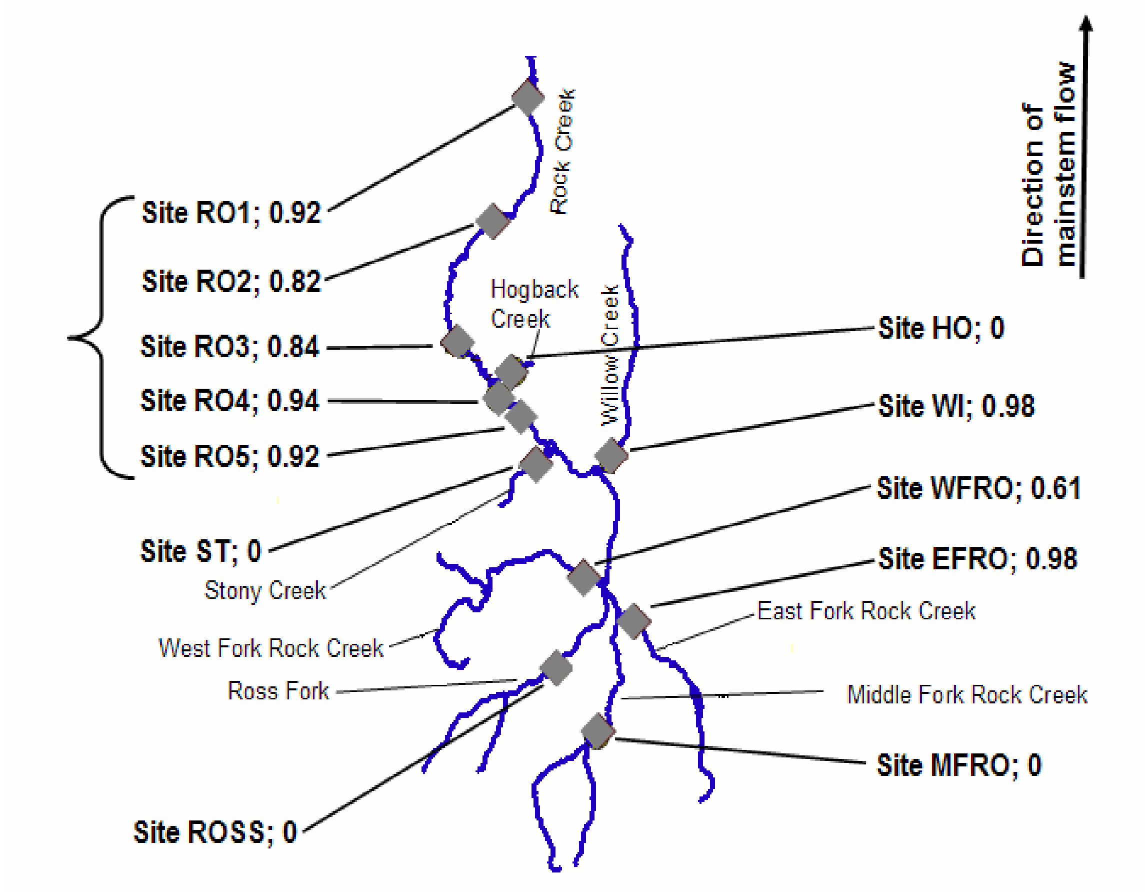

Figure 8. Sites within the Rock Creek drainage and their respective SEVSAMP values (proportions of infected fish with a histological score greater than or equal to four during invertebrate sample years) used to define reaches. Reaches comprised of multiple sites are designated by brackets.

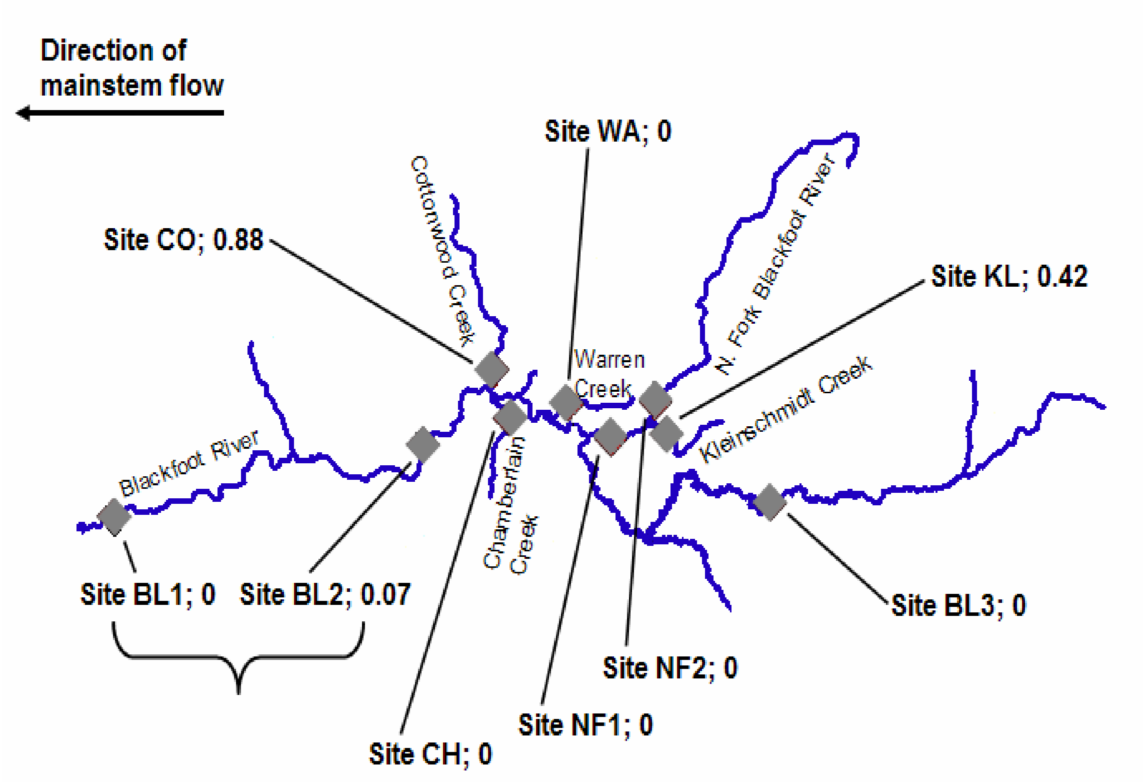

Figure 9. Sites within the Blackfoot River drainage and their respective SEVSAMP values (proportions of infected fish with a histological score greater than or equal to four during invertebrate sample years) used to define reaches. Reaches comprised of multiple sites are designated by brackets.

Whirling Disease Risk at the
Catchment Scale

Whirling disease risk was characterized by the values associated with the lowermost site in a reach. The whirling disease risk of sites isolated by the reach definition process (i.e., a site in between reaches, see site MA3 in Figure 7 for example) were characterized by the risk values associated with that site.

Whirling Disease Risk at the Reach Scale

Whirling disease risk was characterized as the change in risk values between the upper and lower site in a reach (hereafter, values associated with the change in risk are prefaced with Δ). Sites isolated by the reach definition process (i.e., a site in between reaches, see site MA3 in Figure 7 for example) were not used in the reach scale analyses because an upstream site for that reach was not defined.

Land Use

Land use and land cover data were assessed using the Geographic Information System (GIS) program ArcView 3.2 (ESRI 1996-1999). Geospatial data, compiled by state and federal agencies, were used to quantify proportions of land use in each drainage identified as conifer forest, agriculture, disturbance and residential areas, as well as road density and mine density (Table 1). The proportions of conifer forest, agriculture, disturbance, and residential area were derived from the 1992 National Land Cover Database; a dataset based on 30 m resolution Landsat thematic mapper satellite data resulting in 21 land type classes, nine of which were used in this study (Appendix C) (USGS 2000a, 2000b). A detailed description of the classification methods for each land type class for this data set is available online (http://landcover.usgs.gov/classes.php/) and Wickham et al. (2004) gives a detailed accuracy assessment of each land type class. Road density was quantified from 1:100,000 scale TIGER file and digital line graph data as the total road length (km) divided by the total area (km^2) (U.S. Census Bureau 2001; Geographic Information and Analysis Center and Spatial Analysis Center, Yellowstone

National Park, unpublished). Mine density was quantified as the total number of active, inactive, and abandoned mines divided by the total area (km^2) (MTDEQ 2005).

These land use types were chosen to represent undisturbed or "natural" (i.e., conifer forest) and anthropogenic (i.e., agriculture, disturbance, residential, roads and mines) land uses for numerous reasons. Only one measure of natural land use was chosen because proportions of land use types are not independent of each other; if the proportion of one land use type is increased it inevitably results in a decrease in proportion of one or more other land use types (Van Sickle 2003). Rather than include all proportions of all natural vegetation in analyses, which often summed to over 90% of the land use within drainages, one type was selected as the best representative of natural land use. Conifer forest was chosen as the better measure of natural land use over grasslands, the other dominant land cover type within drainages, because grasses are more easily misclassified as anthropogenic land use (e.g., agriculture) (Wickham et al. 2004). The land use types chosen to represent anthropogenic land use have been linked to declines in biological stream integrity and adverse effects in fish populations in many previous watershed assessments (Barton et al. 1985; Eaglin and Hubert 1993; Harding et al. 1998; Morley and Karr 2002; Bruns 2005). Furthermore, these data provided consistent measures of the land use types across the entire study area. Although other measures of anthropogenic land use (e.g., proportion of logged forest) and more detailed land use data were available, methods of classification and quantification of local data by various state and private agencies varied too much to provide an equal comparison of land use across drainages.

Table 1. Land use predictor variables used in analyses.

Land Use Type	**Description**	**Source**
Conifer Forest	Areas dominated by tree species that keep their leaves year round	USGS (2000a); USGS (2000b)
Agriculture (pasture/hay, row crops, small grains, fallow, and urban/recreational grasses)	Areas used for the production of crops or for livestock grazing or developed for recreation, erosion control, or aesthetic purposes	USGS (2000a); USGS (2000b)
Disturbance	Areas that are sparsely vegetated (<25% cover) and dynamically changing. Change can be a result of natural or anthropogenic activities (e.g., forest clearcut, fire)	USGS (2000a); USGS (2000b)
Residential (Low- and high-intensity residential)	Low-intensity residential includes areas where constructed materials account for 30-80% of the cover, representative of single-family housing units. High-intensity residential include areas where constructed materials account for 80-100% of the cover, representative of row houses and apartment complexes	USGS (2000a); USGS (2000b)
Roads	Hard-surface highways, local, city, frontage and service roads, alleys, driveways, and unimproved or dirt four-wheel drive roads and pedestrian trails	U.S. Census Bureau (2001); Geographic Information and Analysis Center and Spatial Analysis Center, Yellowstone National Park, unpublished
Mines	Active, inactive, and abandoned surface, underground and processing mines	MTDEQ (2005)

Land Use at the Catchment Scale

Land use types at the catchment scale were quantified by first calculating flow direction and flow accumulation from a 30 m resolution digital elevation model (USGS 2002) using the Geospatial Hydrologic Modeling Extension (HEC-GeoHMS) (U. S. Army Corps of Engineers 2003) to delineate the upstream subbasin area. The upstream subbasin area was defined as the entire surface drainage area within the catchment upstream of a sample site (i.e., sentinel cage) (Figure 10 A). A 400 m buffer of the delineated stream extending upstream from each site was also converted to a polygon to represent the upstream riparian zone (Figure 10 B). This riparian corridor width was selected to prevent exclusion of any important processes associated with the riparian area due to inaccuracies associated with the catchment delineation and stream buffering process (Morley and Karr 2002). The delineated subbasin and riparian zone for each site were then converted to polygons. Polygons were superimposed over the land coverage data to determine land use proportions and densities within these areas.

Land Use at the Reach Scale

To identify the area contributing to a reach, the delineated subbasin and riparian area between the upper and lower sites in a reach were converted to polygons (Figure 10 C and D, respectively). Polygons were superimposed over the land coverage data to determine land use proportions and densities within these areas.

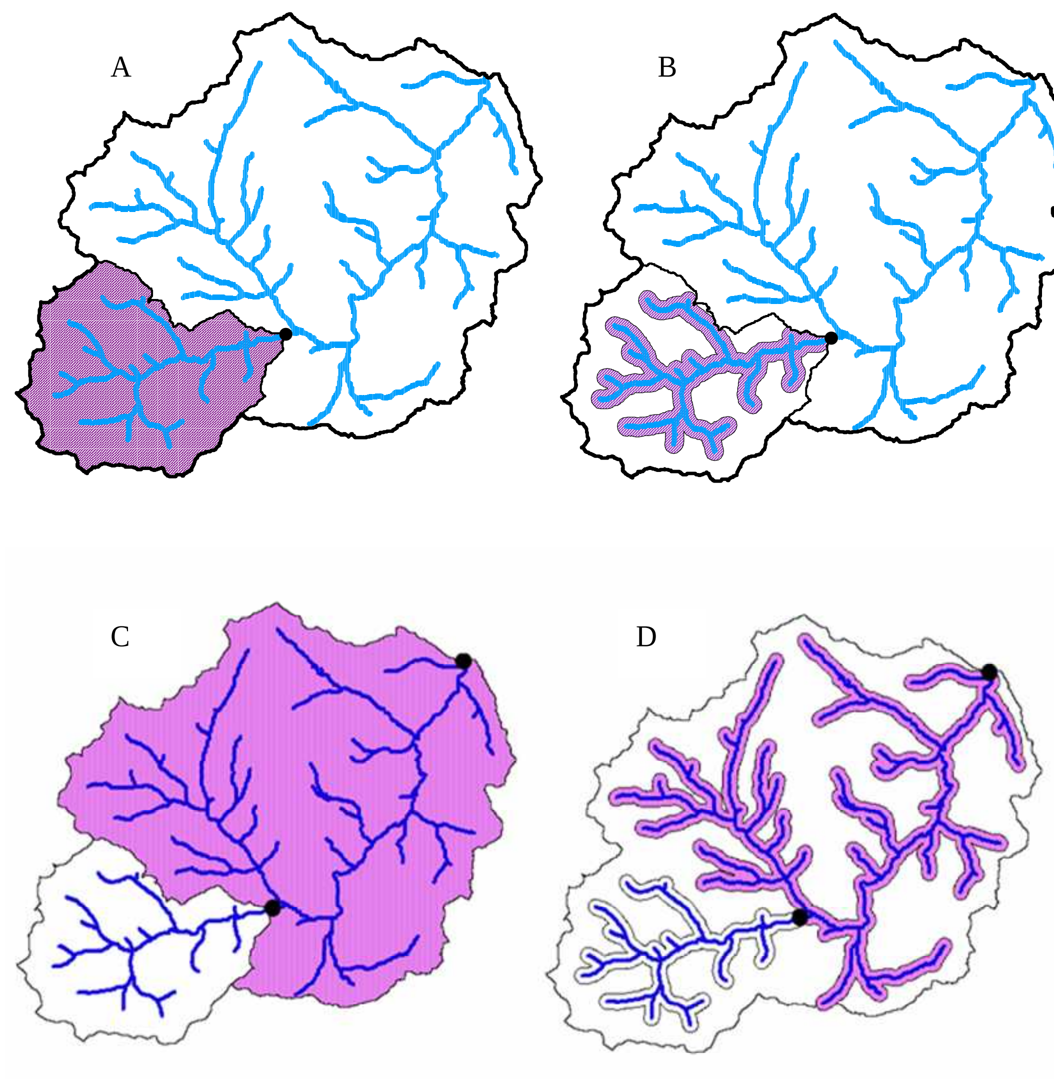

Figure 10. Diagram of spatial scales used in study. The shaded area in A represents the upstream subbasin area defined as the entire surface drainage area within a catchment upstream of a sample site. The shaded area in B represents the entire riparian zone (400 m) within a catchment upstream of a sample site. The shaded area in C represents the reach subbasin, defined as the contributing subbasin area between the upper and lower sample sites in a reach. The shaded area in D represents the reach riparian zone, defined as the contributing riparian area within the subbasin between the upper and lower sample sites in a reach.

Biological Stream Integrity

Within all drainages, a 42 mm diameter core sampler (Brinkhurst and Kennedy 1965) was used to collect six replicate samples of oligochaetes living within the upper 5 to 30 cm of soft sediment in depositional areas. Samples were collected from 1997 through 2000, although the number of time samples were collected varied (Appendix A). Samples were usually collected between 0-150 m upstream of the sentinel cage (<5% of samples were collected at >150 m upstream of sentinel trout cage) and preserved in Kahle’s solution (Pennak 1978). Samples were returned to the laboratory where they were washed through a 125 µm sieve, rinsed, and stored in 70% ethanol until sorted. All oligochaetes were typically slide-mounted (in <5% only a subsample was mounted) and identified using a Nikon Optiphot-2 compound microscope with phase contrast. Oligochaetes were identified to the family level and mature Tubificidae were identified to the species level when possible (Kathman and Brinkhurst 1998).

Within the Missouri and Rock Creek drainage four replicate samples of the macroinvertebrate assemblage were typically collected at each site and two replicate samples were collected at each site within the Madison River drainage at 0, 50, 100, and 150-m upstream from the sentinel trout cage (<5% of samples were collected at >150 m upstream of sentinel trout cage) from 1997 through 2000 (Appendix A). No macroinvertebrate samples were collected within the Blackfoot River drainage. Samples were collected along traveling D-frame kick net transects (mesh size of 500 µm, time = 2 minutes), targeting macroinvertebrates living at the sediment surface and within interstitial spaces of riffle areas. Samples were preserved in Kahle's solution (Pennak 1978) in the field and returned to the laboratory. When necessary, a plankton splitter was used to split the sample into fourths or eighths to reduce sample size (~ 50% of all samples were split). The sample or subsample was then stored in 70% ethanol until sorted and identified to the family level using standard taxonomic keys (Merritt and Cummins 1996).

The biological integrity of each site was determined by quantifying characteristics of the invertebrate community structure and composition (Table 2). Metrics quantified from the oligochaete samples (i.e., density oligochaetes, density tubificids, density *T. tubifex*, and density *L. hoffmeisteri*) were selected because of their apparent relationship to *T. tubifex* and whirling disease and because of their use as indicators of anoxic or eutrophic conditions (Milbrink 1994; Verdonschot 1996). The macroinvertebrate bioassessment metrics quantified from the kick samples that were selected are commonly used to determine the biological condition of a stream (Kerans et al. 1992; Kerans and Karr 1994) and in western Montana streams, they have been useful in the detection of impairment (Bollman 1998; R. Bukantis, Water Quality Division, Department of Environmental Quality, Helena, MT, personal communication).

Biological Stream Integrity at the
Reach Scale

Mean bioassessment metrics were quantified from nontransformed data for each reach from sites within reaches.

Table 2. Bioassessment metrics used in analyses to evaluate the biological condition of a site and their hypothesized direction of response to increased whirling disease risk.

Metric	**Description**	**Hypothesized Direction of Metric Change in Response to Increasing Whirling Disease Risk**
Density Oligochaetes	The total number of oligochaetes divided by the surface area of the core sample	Increase
Density Tubificids	The total number of tubificids divided by the surface area of the core sample	Increase
Density *T. tubifex*	The total number of *Tubifex tubifex* divided by the surface area of the core sample	Increase
Density *L. hoffmeisteri*	The total number of *Limnodrilus hoffmeisteri* divided by the surface area of the core sample	Increase
Total Taxa Richness	The total number of taxa identified at the family level	Decrease
Ephemeroptera Taxa Richness	The number of Ephemeroptera (mayfly) taxa	Decrease
Plecoptera Taxa Richness	The number of Plecoptera (stonefly) taxa	Decrease
Trichoptera Taxa Richness	The number of Trichoptera (caddisfly) taxa	Decrease
EPT Taxa Richness	The number of taxa belonging to the orders Ephemeroptera, Plecoptera, and Trichoptera	Decrease
Proportion Ephemeroptera	The proportion of the sample comprised of individuals from the order Ephemeroptera	Decrease
Proportion Plecoptera	The proportion of the sample comprised of individuals from the order Plecoptera	Decrease
Proportion EPT	The proportion of the sample comprised of individuals from the order Ephemeroptera, Plecoptera, and Trichoptera	Decrease
Proportion Dominant	The proportion of the sample comprised of the single most abundant taxon	Increase
Shannon Diversity Index	This index characterizes diversity by accounting for the abundance and evenness of taxa, quantified at the family level. This index is calculated as: $H = -\sum p_i \ln(p_i)$ where p_i is the proportion of the total number of taxa made up of the *i*th taxon	Decrease

Physical and Chemical Environment

At most sites where invertebrates were collected within the Madison, Missouri, and Rock Creek drainages the depth, width, velocity, proportion of organic matter in sediment, temperature, conductivity, pH, and concentration of dissolved oxygen were recorded (Appendix A). Elevation and slope were quantified for all reaches within all drainages.

Typically at each site, three measurements of depth and velocity at 0.6 of water depth were calculated using a Model 3000 Swoffer Instrument flow meter (Swoffer Instruments, Inc., Seattle, Washington USA) at three equidistant points along four transects at 0, 50, 100, and 150-m upstream of each sentinel cage. The width of each transect was also recorded. The mean depth and velocity for each transect were quantified and an overall mean depth, velocity and width were quantified for each site. A mean discharge value in cubic meters per second (cms) was calculated for each site as the mean depth multiplied by the mean width multiplied by the mean velocity for that site.

The proportion of organic matter in sediment was assessed by the dry weight analysis of benthic sediments. Usually at each site, one sediment sample was collected at each transect in soft sediment areas using a trowel for a total of four samples. Sediment samples were returned to the laboratory and frozen until processed. The proportion of organic material in each sample (30 g wet sediment) was determined as the difference between the dry weight (105 °C for 24 hr) and the weight after the sample was burned in a muffle furnace (550 °C for 2 hr), divided by the original dry weight (Wetzel and Likens 1991). A mean proportion of organic material in sediment was quantified for each site.

At most sites, temperature (°C), conductivity (mS/cm), pH, and the concentration of dissolved oxygen (mg/l) were recorded at two randomly selected points within each reach using a Yellow Springs Instrument meter (Yellow Springs Instrument Co., Inc., Yellow Springs, Ohio USA). A mean of each parameter was then quantified for each site. Furthermore, the elevation (m) at each site was identified from a 30 m resolution digital elevation model (USGS 2002) in the GIS program Arcview 3.2 (ESRI 1996-1999).

Physical and Chemical Environment at the Reach Scale

Mean discharge (cms), proportion of organic material in sediment, temperature (°C), conductivity (mS/cm), pH, and concentration of dissolved oxygen (mg/l) were quantified from untransformed data for each reach from sites within reaches where data were collected. Reaches within all drainages were characterized by the elevation of the lowermost site in the reach. To quantify the slope of each reach, an elevation profile for each stream was created using HEC-GeoHMS software (U. S. Army Corps of Engineers 2003). Slope changes within the elevation profile of each stream were visually identified to assess the slope at each site and the reach slope was quantified as the difference in elevation (m) between the upper and lower site in the reach divided by the length (m) of the reach.

Statistical Analysis

Prior to analyses of bioassessment metrics and physicochemical parameters, differences among sampling dates were examined using analysis of variance (PROC GLM, SAS Institute 2004). Seasonal differences were detected for most bioassessment metrics, temperature, pH, concentration of dissolved oxygen, and discharge. Because seasonal variances were detected, seasons during which the largest proportion of sites were sampled were used in data analyses. Thus, bioassessment metrics quantified from fall 1998 data within the Missouri and Madison drainages, and from summer 1998 data within the Rock Creek and Blackfoot drainages, were used to assess the relationship between risk and biological stream integrity. Temperature, pH, concentration of dissolved oxygen, and discharge data collected in the summer of 1997 and 1998 within the Missouri and Madison drainage and the summer of 1998 within the Rock Creek drainage were used to assess the relationship between risk and these physicochemical parameters. No seasonal differences were detected for conductivity and proportion of organic matter in sediment; therefore, all available data within each drainage were used in analyses of these physicochemical parameters in relation to risk.

Relationships between whirling disease risk and predictor variables were assessed at the catchment and reach scale within and among drainages (Figure 11). To assess risk at the catchment scale, whirling disease risk values associated with the lowermost site in a reach were correlated with land use within the upstream subbasin and riparian zone. To assess risk at the reach scale, Δwhirling disease risk values were paired with land use within the contributing subbasin and riparian zone, reach mean bioassessment metrics quantified from the oligochaete community, reach mean bioassessment metrics quantified from the macroinvertebrate community, reach mean physicochemical parameters (i.e., discharge, proportion of organic matter in sediment, temperature, conductivity, pH, and concentration of dissolved oxygen), elevation, and slope.

All within-drainage analyses, as well as the among-drainage analysis of physicochemical parameters, were conducted using Spearman's rank correlation (PROC CORR, SAS Institute 2004). Spearman's rank correlation analyses were employed because data did not exhibit a bivariate normal distribution and no appropriate transformations of the predictor and/or response variables could be identified to permit the use of a parametric correlation analysis.

For each set of correlations between risk and predictor variables within each drainage, the alpha level of significance was adjusted to account for multiple response variables from a single sample using the Holm's procedure. The Holm's procedure is a modification of the Bonferroni procedure that is less conservative yet still retains the experimentwise error rate (alpha 0.05 level of significance) (Wright 1992). In accordance with the Holm's procedure, unadjusted p-values were ordered from smallest to largest; the smallest p-value was compared to alpha (i.e., 0.05) divided by the number of correlations, then the next smallest p-value was compared to alpha divided by the number of correlations minus one and so on, until a non-significant test statistic result (a p-value greater than the adjusted p-value) was obtained. The last comparison resulting in a significant test statistic represented the adjusted p-value for that set of correlations. P-values less than or equal to the adjusted p-value provide strong evidence against the null hypothesis under traditional hypothesis testing. Nevertheless, p-values close to this value still provide supporting evidence that a relationship may exist. Considering the latter and because this was an exploratory study, relationships exhibiting p-values between the adjusted alpha level of significance and the alpha 0.05 level were considered to be marginally significant.

Regression tree analyses were used to model relationships between the response variable, whirling disease risk, and the predictor variables, land use and biological stream integrity, among drainages (package: tree, R Development Core Team 2005; Ripley 2005). Regression tree analyses identify important predictor variables by splitting the data based on minimizing the squared residual deviance, a value analogous with variance (Breiman et al. 1984). Regression tree models often over fit the data so a 10-fold cross-validation method was used to prune trees. The cross-validation is performed by randomly selecting and holding out 10% of the data. The tree is fit to the remaining 90% of the data, and the data that were held out are then "dropped" through the tree. This process is repeated 10 times, holding out a different randomly-selected 10% of the data each time, to identify the most parsimonious tree. Land use at both the catchment and riparian scale were included as predictor variables to assess the importance of land use at both scales in explaining whirling disease risk. A regression tree analysis of physicochemical variables was not conducted because of an excess of missing values, which could have biased the model's results.

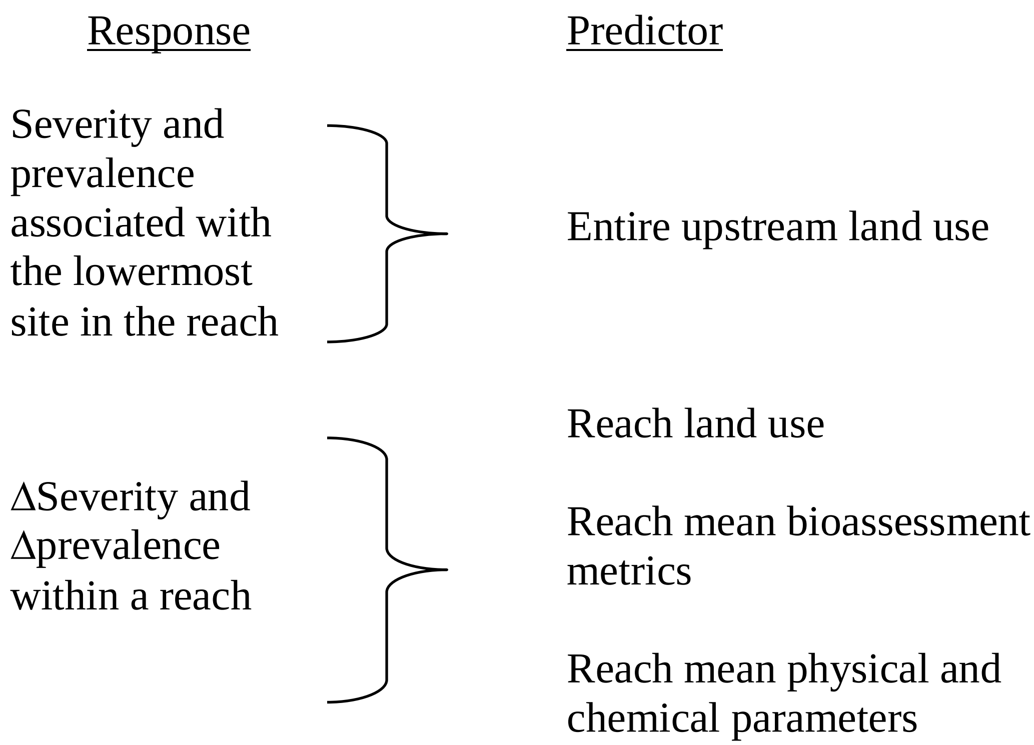

Figure 11. Relationships between whirling disease risk and predictor variables assessed at the catchment and reach scale within and among drainages. Relationships between the prevalence and severity values at the lowermost site in a reach and the land use within the entire upstream from that point were identified as well as relationships between the change in the prevalence and severity values within a reach and the land use that contributes to that reach, reach mean bioassessment metrics, and the mean physical and chemical parameters of the reach. The relationship between risk and land use was assessed within the entire upstream area and also the contributing reach area to determine whether relationships differed at varying spatial extents. Relationships between risk and the bioassessment metrics and the physical and chemical parameters were assessed at the reach scale only because these measures are often more indicative of localized site conditions than of broad landscape scale conditions.

RESULTS

Missouri River Drainage

Whirling Disease Risk

Myxobolus cerebralis was never detected in sentinel rainbow trout at the uppermost site on Little Prickly Pear Creek (PR6), the uppermost site on Wolf Creek (WO3), or the Lyons Creek site (LY) (Table 3). In 1998 low levels of infection were detected at site WO2 on Wolf Creek and the Sheep Creek site (SH), but since then sentinel fish have remained parasite free. PREVALL values were high (≥ 0.80) at all other locations as were SEVALL values, except at an upper site on Little Prickly Pear (PR5) and the lowermost site on the Dearborn River (DB1), where SEVALL values were 0.4 and 0.61, respectively.

Correlations at the Catchment Scale

Land Use. The proportion of conifer forest was by far the dominant land cover, accounting for roughly 0.30 to 0.70 of the total area within the entire upstream subbasin and riparian zone (Appendix D). The proportion of land classified as agriculture within subbasins ranged between 0 and 0.04 and between 0 and 0.11 within the riparian area. Land classified as disturbance comprised a small proportion of the total subbasin area (between 1.71E-03 and 3.77E-03) and land identified as disturbance was not present within the entire upstream riparian zone. The proportion of residential area and mine density were also minor components of the landscape (<0.001) within the entire upstream subbasin and riparian zone. The density of roads within the riparian zone (0.18-2.82 km/km^2) was often much higher than road density within the subbasin (0.07-0.80 km/km^2) upstream of each site. Risk was positively associated with the proportion of disturbed area within the subbasin, and the proportion of residential area and road density within the riparian zone (Table 4).

Correlations at the Reach Scale

Land Use. Once again, the proportion of forest was by far the dominant land cover, accounting for roughly 0.35 to 0.70 of the total area within the subbasin and riparian zone at the reach scale (Appendix E). At the reach scale, proportions of agriculture, disturbance, and residential, as well as the density of roads and mines, were similar to those values identified at the catchment scale. Significant positive relationships were identified between Δrisk and the proportion of disturbed area within the contributing subbasin, the proportion of residential and agricultural area within the contributing riparian zone, and road density within the contributing subbasin area (Table 4). Significant negative correlations were identified between Δrisk and the proportion of forest within the contributing subbasin and riparian zone.

Biological Stream Integrity. The highest means for eight of the nine metrics that indicate high biological stream integrity as their values increase (e.g., total taxa richness, EPT taxa richness) were recorded at sites and reaches where the parasite has not been detected or disease prevalence and severity has remained low (i.e., sites PR6, LY, WO2, SH) (Appendix F; Appendix G). Furthermore, *T. tubifex* was not detected at these sites.

Significant positive relationships were identified between Δwhirling disease risk and the mean density of *T. tubifex* and *L. hoffmeisteri* (Table 5). Mean density values for these species ranged between roughly 0 and 200 worms per square meter among reaches (Appendix G).

Physical and Chemical Environment. Physical and chemical data for sites and reaches are summarized in Appendix H and Appendix I, respectively. No statistically significant relationships were detected between Δwhirling disease risk and any physicochemical parameters (Table 6).

Table 3. Whirling disease risk within the Missouri River drainage. Whirling disease risk values recorded during the invertebrate collection period (1997 – 1999) are reported as the proportions of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportions of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period (1997 – 2004) are reported as the proportions infected with a histological score of grade four or greater (SEVALL) and the proportions of infected rainbow trout (PREVALL). Values prefaced with a Δ indicate the change in that respective value between the upper and lower sites in that reach. Multiple sites within a reach are indicated by brackets and are identified by the lowermost site in that reach, signified by boldface type. N/A for PREVSAMP, SEVSAMP, PREVALL and SEVALL values indicates that no samples were collected at that site during that time period. N/A for ΔPREVSAMP, ΔSEVSAMP, ΔPREVALL and ΔSEVALL values signifies that that reach was not defined during that time period.

	Catchment				**Reach**			
Site	**SEVSAMP**	**SEVALL**	**PREVSAMP**	**PREVALL**	**ΔSEVSAMP**	**ΔSEVALL**	**ΔPREVSAMP**	**ΔPREVALL**
LY	0	0	0	0	0	0	0	0
WO3	0	0	0	0				
WO2	0	0	0.09	0.09				
WO1	0	0.94	0.13	1	0	0.94	0.13	1
PR6	0	0	0	0				
PR5	0.4	0.4	0.8	0.8				
PR4	0.84	0.84	0.91	0.91				
PR3	0.94	0.94	1	1				
PR2	0.94	0.94	0.98	0.98				
PR1	0.98	1	1	1	0.98	1	1	1
DBMF	N/A	1	N/A	1	N/A	1	N/A	1
DBSF	N/A	1	N/A	1	N/A	1	N/A	1
DB3	N/A	0.9	N/A	1	N/A	0.9	N/A	1
DB2	0	0.89	0	1	0	N/A	0	N/A
DB1	N/A	0.61	N/A	0.92	N/A	N/A	N/A	N/A
SH	0	0	0.09	0.09	0	0	0.09	0.09

Table 4. Spearman's rank correlation coefficients (r) and p-values relating upstream land use (predictor variable) to whirling disease risk and Δwhirling disease risk (response variable) within the Missouri River drainage. Whirling disease risk values recorded during the invertebrate collection period are reported as the proportions of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportions of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period are reported as the proportions infected with a histological score of grade four or greater (SEVALL) and the proportions of infected rainbow trout (PREVALL). Values prefaced with a Δ indicate the change in the respective value between the upper and lower site in that reach. n = 5 for PREVSAMP, SEVSAMP, ΔPREVSAMP and ΔSEVSAMP correlations. n = 9 for PREVALL and SEVALL correlations. n = 7 for ΔPREVALL and ΔSEVALL. Statistically significant relationships at the adjusted alpha level ($p \leq 0.0006$) are indicated by bold face type and an asterisk. Relationships considered marginally significant ($0.0006 < p \leq 0.05$) are in bold face type only.

	Catchment Correlations				**Reach Correlations**			
Land Use	**SEVSAMP**	**SEVALL**	**PREVSAMP**	**PREVALL**	**ΔSEVSAMP**	**ΔSEVALL**	**ΔPREVSAMP**	**ΔPREVALL**
<u>Subbasin</u>								
Proportion Conifer Forest	-0.71 p = 0.182	-0.42 p = 0.264	-0.41 p = 0.493	-0.41 p = 0.278	-0.71 p = 0.180	**-0.77** **p = 0.044**	-0.41 p = 0.4932	**-0.76** **p = 0.049**
Proportion Agriculture	0.73 p = 0.165	0.23 p = 0.560	0.34 p = 0.573	0.14 p = 0.718	0.73 p = 0.165	0.74 p = 0.058	0.34 p = 0.573	0.37 p = 0.414
Proportion Disturbance	**1.00** **p = <0.0001***	0.42 p = 0.261	0.73 p = 0.165	0.24 p = 0.527	**1.00** **p = <0.0001***	0.43 p = 0.338	0.73 p = 0.165	0.25 p = 0.582
Proportion Residential	0.79 p = 0.111	-0.13 p = 0.746	0.69 p = 0.199	-0.29 p = 0.452	0.79 p = 0.111	0.02 p = 0.960	0.69 p = 0.199	-0.11 p = 0.813
Road Density	0.71 p = 0.182	0.66 p = 0.051	0.67 p = 0.219	0.50 p = 0.166	0.71 p = 0.182	**0.82** **p = 0.023**	0.67 p = 0.219	0.58 p = 0.173
Mine Density	0.73 p = 0.165	0.24 p = 0.530	0.76 p = 0.133	0.36 p = 0.344	0.73 p = 0.165	0.37 p = 0.410	0.76 p = 0.133	0.39 p = 0.389
<u>Riparian</u>								
Proportion Conifer Forest	-0.35 p = 0.559	-0.34 p = 0.370	0.21 p = 0.741	-0.30 p = 0.438	-0.35 p = 0.559	**-0.92** **p = 0.004**	-0.05 p = 0.935	**-0.76** **p = 0.049**
Proportion Agriculture	0.79 p = 0.111	0.37 p = 0.322	0.34 p = 0.571	0.23 p = 0.556	0.79 p = 0.111	**0.87** **p = 0.012**	0.34 p = 0.571	0.52 p = 0.236

Table 4 continued.

Proportion Disturbance	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A
Proportion Residential	**1.00** **p = <0.0001***	0.16 p = 0.675	0.73 p = 0.165	-0.01 p = 0.972	**1.00** **p = <0.0001***	0.43 p = 0.338	0.73 p = 0.165	0.25 p = 0.582
Road Density	0.35 p = 0.559	**0.77** **p = 0.016**	0.56 p = 0.322	0.64 p = 0.061	0.35 p = 0.559	0.67 p = 0.097	0.56 p = 0.322	0.58 p = 0.173
Mine Density	0.36 p = 0.549	0.19 p = 0.631	0.66 p = 0.228	0.28 p = 0.462	0.00 p = 1.00	0.28 p = 0.543	0.29 p = 0.637	0.39 p = 0.389

Table 5. Spearman's rank correlation coefficients (r) and p-values relating mean reach biological stream integrity metrics (predictor variable) and Δwhirling disease risk values (response variable) within the Missouri River drainage. Whirling disease risk values recorded during the invertebrate collection period are reported as the proportion of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportion of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period are reported as the proportion infected with a histological score of grade four or greater (SEVALL) and the proportion of infected rainbow trout (PREVALL). The Δ indicates the change in the respective risk value between the upper and lower sites in the reach. n = 5 for ΔPREVSAMP and ΔSEVSAMP correlations and n = 4 for ΔPREVALL and ΔSEVALL correlations. Statistically significant relationships at the adjusted alpha level ($p \leq 0.0009$) are indicated by bold face type and an asterisk. Relationships considered marginally significant ($0.0009 < p \leq 0.05$) are in bold face type only.

	Reach Correlations			
Metric	**ΔSEVSAMP**	**ΔSEVALL**	**ΔPREVSAMP**	**ΔPREVALL**
Density Oligochaetes	0.71 p = 0.182	0.95 p = 0.051	0.82 p = 0.089	0.95 p = 0.051
Density Tubificids	0.71 p = 0.182	0.63 p = 0.368	0.87 p = 0.054	0.63 p = 0.368
Density *T. tubifex*	**1.0** **p = <0.0001***	0.82 p = 0.184	0.72 p = 0.165	0.54 p = 0.456
Density *L. hoffmeisteri*	0.73 p = 0.165	0.95 p = 0.051	**1.0** **p = <0.0001***	0.95 p = 0.051
Total Taxa Richness	0.00 p = 1.00	0.21 p = 0.789	0.67 p = 0.219	0.63 p = 0.378
Ephemeroptera Taxa Richness	-0.79 p = 0.111	-0.83 p = 0.167	-0.46 p = 0.437	-0.50 p = 0.500
Plecoptera Taxa Richness	-0.73 p = 0.165	-0.33 p = 0.667	-0.13 p = 0.833	0.00 p = 1.00
Trichoptera Taxa Richness	0.35 p = 0.559	0.74 p = 0.262	0.56 p = 0.322	0.74 p = 0.262
EPT Taxa Richness	-0.73 p = 0.165	-0.32 p = 0.684	-0.29 p = 0.636	-0.11 p = 0.895
Proportion Ephemeroptera	0.35 p = 0.559	-0.21 p = 0.789	-0.15 p = 0.805	-0.63 p = 0.368
Proportion Plecoptera	0.00 p = 1.00	0.11 p = 0.895	-0.56 p = 0.322	-0.32 p = 0.684
Proportion EPT	0.35 p = 0.559	-0.21 p = 0.789	-0.15 p = 0.805	-0.63 p = 0.368
Proportion Dominant	0.00 p = 1.00	0.21 p = 0.789	0.67 p = 0.219	0.63 p = 0.368
Shannon Diversity	0.00 p = 1.00	-0.21 p = 0.789	-0.67 p = 0.219	-0.63 p = 0.368

Table 6. Spearman's rank correlation coefficients (r) and p-values relating physicochemical parameters (predictor variable) to Δwhirling disease risk (response variable) within the Missouri River drainage. Whirling disease risk values recorded during the invertebrate collection period are reported as the proportion of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportion of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period are reported as the proportion infected with a histological score of grade four or greater (SEVALL) and the proportion of infected rainbow trout (PREVALL). The Δ indicates the change in the respective risk value between the upper and lower site in the reach. n = 7 for correlation of ΔPREVALL and ΔSEVALL with elevation and slope. n = 5 for all other correlations. Statistically significant relationships at the adjusted alpha level ($p \leq 0.0016$) are indicated by bold face type and an asterisk. Relationships considered marginally significant ($0.0016 < p \leq 0.05$) are in bold face type only.

Reach Correlations

Physicochemical Variable	**ΔSEVSAMP**	**ΔSEVALL**	**ΔPREVSAMP**	**ΔPREVALL**
Discharge	0.35 p = 0.560	0.31 p = 0.684	-0.15 p = 0.805	0.11 p = 0.895
Proportion Organic Matter	0.71 p = 0.182	0.63 p = 0.368	0.46 p = 0.434	0.63 p = 0.368
Temperature	0.35 p = 0.560	0.11 p = 0.895	0.41 p = 0.493	0.32 p = 0.684
Conductivity	0.71 p = 0.182	0.95 p = 0.051	0.67 p = 0.219	0.74 p = 0.262
pH	0.35 p = 0.560	0.63 p = 0.368	-0.21 p = 0.741	0.21 p = 0.789
Dissolved Oxygen	0.00 p = 1.00	-0.11 p = 0.894	-0.05 p = 0.935	-0.32 p = 0.684
Elevation	0.00 p = 1.00	0.54 p = 0.208	-0.56 p = 0.322	0.40 p = 0.373
Slope	-0.35 p = 0.560	-0.60 p = 0.155	-0.10 p = 0.870	-0.67 p = 0.101

Madison River Drainage

Whirling Disease Risk

Within the Madison River drainage, *Myxobolus cerebralis* was never detected in sentinel rainbow trout at the Squaw Creek site (SQ) and whirling disease risk remained low (≤ 0.10) at the uppermost site on the West Fork Madison River (WFMA2) (Table 7). All other sites exhibited high PREVALL values (≥ 0.86) and SEVALL values were variable, ranging from 0.16 – 1.0.

Correlations at the Catchment Scale

Land Use. The proportions of forested and disturbed area were the predominant land uses within the upstream subbasin and riparian zone, accounting for roughly 0.20 to 0.50 and 0 to 0.33 of the total area within the entire upstream subbasin and riparian zone, respectively (Appendix J). The proportion of agricultural area ranged between 0 and 0.05 within the subbasin and riparian zone. The proportion of residential area and density of mines were small components of the landscape (<0.001). Road density was much higher within the riparian zone (0.87-1.30 km/km^2) than within the subbasin area (0.13-0.31 km/km^2) upstream of each site. Significant positive relationships were identified between whirling disease risk and the proportion of disturbance and residential area within the subbasin and riparian area as well as the density of roads within the riparian zone (Table 8). It is noteworthy here that these land use types were all significantly correlated regardless of the value used to represent risk, except one relationship between risk and the proportion of residential area, which was marginally significant.

Correlations at the Reach Scale

Land Use. The proportion of forest was the predominant land cover, ranging between 0.04 and 0.82 within the contributing riparian zone and 0.28 and 0.72 within the contributing subbasin (Appendix K). The proportion of land identified as disturbed was far less at the reach scale, ranging between 8.0E-05 and 0.03. As at the catchment scale, road density was higher within the contributing riparian zone than within the contributing subbasin area. The proportion of agricultural area within the contributing subbasin area comprised ≤ 0.05 of the total reach area and the proportion of residential area and mine density comprised a small portion of the contributing subbasin area (<0.001). Within the contributing riparian area no land was classified as disturbance or residential and no mines were present. No significant relationships were identified between Δrisk and land use at the reach scale (Table 8).

Biological Stream Integrity. The highest means for six of the nine metrics that indicate high biological stream integrity as their values increase (e.g., Ephemeroptera taxa richness, proportion Plecoptera) were recorded at sites where the parasite has not been detected or disease prevalence and severity has remained low (i.e., sites WFMA2 and SQ) (Appendix L). The lowest densities of tubificids, an indicator of high biological stream integrity as values decrease, were detected at these sites and reaches as well (Appendix L; Appendix M). However, site SQ was one of the few sites where *T. tubifex* was collected. Two significant negative relationships were identified between Δwhirling disease risk and mean Plecoptera taxa richness and the mean proportion of Plecoptera taxa (Table 9). Mean Plecoptera taxa richness ranged between 2.00 and 4.50 and the mean proportion of Plecoptera ranged between 0.01 and 0.23 among reaches (Appendix M). A significant positive relationship was identified between Δwhirling disease risk and the mean proportion of dominant taxa and two significant relationships, contrary to their predicted direction, were also detected between Δwhirling disease risk and mean total taxa richness and mean EPT taxa richness (Table 9). The mean proportion of dominant taxa ranged between 0.29 and 0.34 among reaches (Appendix M). Mean total taxa richness ranged between 18.50 and 25.00 and mean EPT taxa richness ranged between 11.00 and 12.00 among reaches.

Physical and Chemical Environment. Physical and chemical data for sites and reaches are summarized in Appendix N and Appendix O, respectively. Significant positive relationships were detected between Δwhirling disease risk and temperature and pH; significant negative correlations were detected between Δwhirling disease risk and dissolved oxygen (Table 10). Temperature reach means ranged between 9.26 and 17.34°C; pH reach means ranged between 8.11 and 8.77; the reach mean of dissolved oxygen concentrations ranged between 8.14 and 10.26 (mg/l) (Appendix O).

Table 7. Whirling disease risk within the Madison River drainage. Whirling disease risk values recorded during the invertebrate collection period (1997 - 1999) are reported as the proportions of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportions of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period (1997 – 2004) are reported as the proportions infected with a histological score of grade four or greater (SEVALL) and the proportions of infected rainbow trout (PREVALL). Values prefaced with a Δ indicate the change in that respective value between the upper and lower sites in that reach. Multiple sites within a reach are indicated by brackets and are identified by the lowermost site in the reach, signified by boldface type.

	Catchment				Reach			
Site	**SEVSAMP**	**SEVALL**	**PREVSAMP**	**PREVALL**	**ΔSEVSAMP**	**ΔSEVALL**	**ΔPREVSAMP**	**ΔPREVALL**
WFMA2	0.04	0.04	0.10	0.10				
WFMA1	0.06	0.26	0.54	0.90	0.02	0.22	0.44	0.80
SQ	0	0	0	0	0	0	0	0
MA8	0.10	0.98	0.68	1.00				
MA7	0.06	0.71	0.56	1.00				
MA6	0.05	1.00	0.55	1.00				
MA5	0.83	1.00	0.94	1.00	0.73	0.02	0.26	0
MA4	0.96	1.00	0.98	1.00				
MA3	0.69	0.69	0.94	0.94				
MA2	0.16	0.16	0.86	0.86				
MA1	0.41	0.41	0.93	0.93	0.25	0.25	0.07	0.07

Table 8. Spearman's rank correlation coefficients (r) and p-values relating upstream land use (predictor variable) to whirling disease risk and Δwhirling disease risk (response variable) within the Madison River drainage. Whirling disease risk values recorded during the invertebrate collection period are reported as the proportions of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportions of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period are reported as the proportions infected with a histological score of grade four or greater (SEVALL) and the proportions of infected rainbow trout (PREVALL). Values prefaced with a Δ indicate the change in that respective value between the upper and lower sites in that reach. n = 6 for PREVSAMP, SEVSAMP, PREVALL and SEVALL correlations. n = 4 for ΔPREVSAMP, ΔSEVSAMP, ΔPREVALL and ΔSEVALL correlations. N/A indicates correlations not conducted because land use at that scale was not present. Statistically significant relationships at the adjusted alpha level ($p \leq 0.0006$) are indicated by bold face type and an asterisk. Relationships considered marginally significant ($0.0006 < p \leq 0.05$) are in bold face type only.

	Catchment Correlations				**Reach Correlations**			
Land Use	**SEVSAMP**	**SEVALL**	**PREVSAMP**	**PREVALL**	**ΔSEVSAMP**	**ΔSEVALL**	**ΔPREVSAMP**	**ΔPREVALL**
Subbasin								
Proportion Conifer Forest	-0.14 p = 0.787	-0.12 p = 0.827	-0.09 p = 0.872	-0.12 p = 0.827	-0.80 p = 0.200	0.00 p = 1.00	0.20 p = 0.800	0.63 p = 0.368
Proportion Agriculture	0.43 p = 0.397	0.49 p = 0.321	0.37 p = 0.469	0.49 p = 0.321	0.60 p = 0.400	0.40 p = 0.600	-0.40 p = 0.600	-0.32 p = 0.684
Proportion Disturbance	**0.94** **p = 0.005**	**0.99** **p = 0.0003***	**0.83** **p = 0.042**	**0.99** **p = 0.0003***	-0.80 p = 0.20	0.00 p = 1.00	0.20 p = 0.800	0.63 p = 0.368
Proportion Residential	**0.89** **p = 0.019**	**0.93** **p = 0.008**	0.77 p = 0.072	**0.93** **p = 0.008**	-0.63 p = 0.368	-0.32 p = 0.684	-0.95 p = 0.051	-0.39 p = 0.611
Road Density	0.49 p = 0.329	0.52 p = 0.289	0.31 p = 0.544	0.52 p = 0.289	0.80 p = 0.20	0.20 p = 0.800	0.80 p = 0.20	0.11 p = 0.895
Mine Density	0.58 p = 0.228	0.62 p = 0.191	0.46 p = 0.354	0.62 p = 0.191	0.26 p = 0.742	0.77 p = 0.225	-0.26 p = 0.742	0.27 p = 0.728
Riparian								
Proportion Conifer Forest	0.14 p = 0.787	0.09 p = 0.870	0.31 p = 0.544	0.09 p = 0.870	-0.40 p = 0.600	0.60 p = 0.400	0.40 p = 0.600	0.94 p = 0.051
Proportion Agriculture	-0.26 p = 0.623	-0.23 p = 0.658	-0.37 p = 0.469	-0.23 p = 0.658	0.00 p = 1.00	0.20 p = 0.800	-0.80 p = 0.200	-0.32 p = 0.684

Table 8 continued.

Proportion Disturbance	**0.93** **p = 0.008**	**0.97** **p = 0.001**	**0.81** **p = 0.050**	**0.97** **p = 0.001**	N/A N/A	N/A N/A	N/A N/A	N/A N/A
Proportion Residential	**0.93** **p = 0.008**	**0.97** **p = 0.001**	**0.81** **p = 0.050**	**0.97** **p = 0.001**	N/A N/A	N/A N/A	N/A N/A	N/A N/A
Road Density	**0.94** **p = 0.005**	**0.99** **p = 0.0003***	**0.83** **p = 0.042**	**0.99** **p = 0.0003***	0.40 p = 0.60	-0.40 p = 0.600	0.60 p = 0.400	-0.21 p = 0.789
Mine Density	-0.13 p = 0.805	-0.13 p = 0.802	-0.13 p = 0.805	-0.13 p = 0.802	N/A N/A	N/A N/A	N/A N/A	N/A N/A

Table 9. Spearman's rank correlation coefficients (r) and p-values relating mean reach biological stream integrity metrics (predictor variable) and Δwhirling disease risk values (response variable) within the Madison River drainage. Whirling disease risk values recorded during the invertebrate collection period are reported as the proportions of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportions of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period are reported as the proportions infected with a histological score of grade four or greater (SEVALL) and the proportions of infected rainbow trout (PREVALL). The Δ indicates the change in the respective risk value between the upper and lower sites in the reach; n = 4. Statistically significant relationships at the adjusted alpha level ($p \leq 0.0009$) are indicated by bold face type and an asterisk. Relationships considered marginally significant ($0.0009 < p \leq 0.05$) are in bold face type only.

	Reach Correlations			
Metric	**ΔSEVSAMP**	**ΔSEVALL**	**ΔPREVSAMP**	**ΔPREVALL**
Density Oligochaetes	0.80 p = 0.200	0.20 p = 0.800	0.80 p = 0.200	0.11 p = 0.895
Density Tubificids	0.80 p = 0.200	0.80 p = 0.200	0.20 p = 0.800	0.21 p = 0.789
Density *T. tubifex*	-0.21 p = 0.789	0.32 p = 0.684	-0.73 p = 0.262	-0.06 p = 0.944
Density *L. hoffmeisteri*	0.63 p = 0.368	0.95 p = 0.051	0.32 p = 0.684	0.50 p = 0.500
Total Taxa Richness	**1.00** **p = <0.0001***	0.40 p = 0.600	0.40 p = 0.600	-0.11 p = 0.895
Ephemeroptera Taxa Richness	-0.74 p = 0.262	-0.21 p = 0.789	0.32 p = 0.684	0.50 p = 0.500
Plecoptera Taxa Richness	-0.40 p = 0.600	-0.40 p = 0.600	**-1.00** **p = <0.0001***	-0.63 p = 0.368
Trichoptera Taxa Richness	0.20 p = 0.800	0.80 p = 0.200	0.80 p = 0.200	0.95 p = 0.051
EPT Taxa Richness	0.40 p = 0.600	**1.00** **p = <0.0001***	0.40 p = 0.600	0.74 p = 0.262
Proportion Ephemeroptera	-0.80 p = 0.200	-0.80 p = 0.200	-0.20 p = 0.800	-0.21 p = 0.790
Proportion Plecoptera	-0.40 p = 0.600	-0.40 p = 0.600	**-1.00** **p = <0.0001***	-0.63 p = 0.368
Proportion EPT	0.20 p = 0.800	0.80 p = 0.200	0.80 p = 0.200	0.95 p = 0.051
Proportion Dominant	0.40 p = 0.600	0.40 p = 0.600	**1.00** **p = <0.0001***	0.63 p = 0.368
Shannon Diversity	-0.20 p = 0.800	-0.80 p = 0.200	-0.80 p = 0.200	-0.95 p = 0.051

Table 10. Spearman's rank correlation coefficients (r) and p-values relating physicochemical parameters (predictor variable) to Δwhirling disease risk (response variable) within the Madison River drainage. Whirling disease risk values recorded during the invertebrate collection period are reported as the proportions of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportions of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period are reported as the proportions infected with a histological score of grade four or greater (SEVALL) and the proportions of infected rainbow trout (PREVALL). The Δ indicates the change in the respective risk value between the upper and lower sites in the reach. n = 4 for all correlations except temperature, pH, and dissolved oxygen (n = 3). Statistically significant relationships at the adjusted alpha level ($p \leq 0.0016$) are indicated by bold face type and an asterisk. Relationships considered marginally significant ($0.0016 < p \leq 0.05$) are in bold face type only.

	Reach Correlations			
Physicochemical Variable	**ΔSEVSAMP**	**ΔSEVALL**	**ΔPREVSAMP**	**ΔPREVALL**
Discharge	0.80 p = 0.200	0.80 p = 0.200	0.20 p = 0.800	0.21 p = 0.789
Proportion Organic Matter	-0.80 p = 0.200	0.00 p = 1.00	0.20 p = 0.800	0.63 p = 0.368
Temperature	**1.00** **p = <0.0001***	0.50 0.667	0.50 p = 0.667	0.00 1.00
Conductivity	0.40 p = 0.600	-0.60 p = 0.400	-0.40 p = 0.600	-0.95 p = 0.051
pH	0.50 p = 0.667	**1.00** **p = <0.0001***	**1.00** **p = <0.0001***	0.87 p = 0.333
Dissolved Oxygen	-0.50 p = 0.667	**-1.00** **p = <0.0001***	**-1.00** **p = <0.0001***	-0.87 p = 0.333
Elevation	0.00 1.00	-0.20 p = 0.800	0.80 p = 0.200	0.32 0.684
Slope	-0.80 p = 0.200	-0.80 p = 0.200	-0.20 p = 0.800	-0.21 p = 0.789

Rock Creek Drainage

Whirling Disease Risk

Myxobolus cerebralis was never detected in sentinel rainbow trout at the Middle Fork Rock Creek site (MFRO), the Hogback Creek site (HO), or the Stony Creek site (ST) (Table 11). Low levels of infection were detected at the Ross Fork site (ROSS). PREVSAMP and PREVALL values were high (≥ 0.97) at all other locations. SEVSAMP and SEVALL values were also high at these sites (≥ 0.82); except for the West Fork Rock Creek site (WFRO), where values were 0.61.

Correlations at the Catchment Scale

Land Use. The dominant land use was the proportion of forest, accounting for roughly 0.25 to 0.95 of the total area within the entire upstream subbasin and riparian zone (Appendix P). No land was identified as residential within the entire drainage and disturbance was not present within the riparian zone. The proportion of agricultural area ranged between 0 and 0.01 within the entire upstream subbasin and between 0 and 0.08 within the entire upstream riparian area. The proportion of disturbance within the subbasin, as well as mine density within the subbasin and riparian zone made up a small component of the total respective areas (>0.001). Road densities ranged between 0.01 and 0.62 km/km^2 within the entire upstream subbasin and between 0.15 and 2.58 km/km^2 within the entire upstream riparian zone. Significant positive correlations were identified between risk and the proportions of agriculture and the densities of roads within the subbasin (Table 12). Significant negative correlations were identified between whirling disease risk and the proportions of forest within the subbasin and riparian zone. Similar to the Madison River drainage, relationships were significant regardless of the value used to represent risk.

Correlations at the Reach Scale

Land Use. Land use values identified at the reach scale were similar to those described at the catchment scale (Appendix Q). As at the entire catchment scale, positive correlations were identified between Δwhirling disease risk and the proportion of agricultural area and road density within the contributing subbasin. Furthermore, negative correlations between Δwhirling disease risk and the proportion of forested area were identified within the contributing subbasin (Table 12).

Biological Stream Integrity. The highest means for four of the nine metrics that indicate high biological stream integrity as their values increase (i.e., Plecoptera taxa richness, proportion Ephemeroptera, proportion Plecoptera, proportion EPT) were recorded at the Stony Creek site (ST) where the parasite was never detected (Appendix R). However, this site did have the highest proportion of dominant taxa (0.48), which is an indicator of low biological integrity as its value increases. These trends also held when mean values were compared among reaches (Appendix S). Furthermore, the East Fork Rock Creek site (EFRO), a highly infected site, had the highest means for four of the five metrics that indicate low biological integrity as their values increase (i.e., density of oligochaetes, tubificids, *T. tubifex*, and *L. hoffmeisteri*). Significant positive relationships between Δrisk and the density of oligochaetes were identified (Table 13).

Furthermore, positive relationships between Δrisk and Shannon diversity were identified, which was contrary to the initial predictions. Mean oligochaete densities ranged between 480.58 and 7.54E+04 and mean Shannon diversity values ranged between 1.62 and 2.08 among reaches.

Physical and Chemical Environment. The physical and chemical data for sites and reaches are summarized in Appendix T and Appendix U, respectively. No statistically significant relationships were detected between Δwhirling disease risk and any of the physicochemical parameters (Table 14).

Table 11. Whirling disease risk within the Rock Creek drainage. Whirling disease risk values recorded during the invertebrate collection period (1998 - 2000) are reported as the proportions of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportions of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period (1997 - 2004) are reported as the proportions infected with a histological score of grade four or greater (SEVALL) and the proportions of infected rainbow trout (PREVALL). Values prefaced with a Δ indicate the change in that respective value between the upper and lower sites in that reach. Multiple sites within a reach are indicated by brackets and are identified by the lowermost site in a reach, signified by boldface type.

	Catchment				**Reach**			
Site	**SEVSAMP**	**SEVALL**	**PREVSAMP**	**PREVALL**	**ΔSEVSAMP**	**ΔSEVALL**	**ΔPREVSAMP**	**ΔPREVALL**
EFRO	0.98	1	1	1	0.98	1	1	1
MFRO	0	0	0	0	0	0	0	0
WFRO	0.61	0.61	0.97	0.97	0.61	0.61	0.97	0.97
ROSS	0	0	0.04	0.04	0	0	0.04	0.04
WI	0.98	0.98	1	1	0.98	0.98	1	1
ST	0	0	0	0	0	0	0	0
HO	0	0	0	0	0	0	0	0
RO5	0.92	0.92	1	1				
RO4	0.94	0.94	1	1				
RO3	0.84	0.98	1	1				
RO2	0.82	0.82	1	1				
RO1	0.92	0.92	0.98	0.98	0	0	-0.02	-0.02

Table 12. Spearman's rank correlation coefficients (r) and p-values relating upstream land use (predictor variable) to whirling disease risk and Δwhirling disease risk within the Rock Creek drainage. Whirling disease risk values recorded during the invertebrate collection period are reported as the proportions of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportions of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period are reported as the proportions infected with a histological score of grade four or greater (SEVALL) and the proportions of infected rainbow trout (PREVALL). Values prefaced with a Δ indicate the change in that respective value between the upper and lower sites in that reach. n = 8 for all correlations. N/A indicates correlations not conducted because land use at that scale was not present. Statistically significant relationships at the adjusted alpha level ($p \leq 0.0007$) are indicated by bold face type and an asterisk. Relationships considered marginally significant ($0.0007 < p \leq 0.05$) are in bold face type only.

	Catchment Correlations				**Reach Correlations**			
Land Use	**SEVSAMP**	**SEVALL**	**PREVSAMP**	**PREVALL**	**ΔSEVSAMP**	**ΔSEVALL**	**ΔPREVSAMP**	**ΔPREVALL**
Subbasin								
Proportion	**-0.93**	**-0.94**	**-0.82**	**-0.82**	**-0.87**	**-0.87**	-0.58	-0.58
Conifer Forest	**p = 0.0007***	**p = 0.0006***	**p = 0.012**	**p = 0.012**	**p = 0.005**	**p = 0.005**	p = 0.134	p = 0.134
Proportion	**0.99**	**0.97**	**0.95**	**0.95**	**0.92**	**0.90**	0.69	0.69
Agriculture	**p = <0.0001***	**p = <0.0001***	**p = 0.0002***	**p = 0.0002***	**p = 0.001**	**p = 0.002**	p = 0.057	p = 0.057
Proportion	0.63	0.70	0.60	0.60	0.13	0.20	-0.17	-0.17
Disturbance	p= 0.096	p= 0.055	p = 0.114	p= 0.114	p= 0.750	p= 0.641	p= 0.690	p= 0.690
Proportion	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Residential	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A
Road	**0.91**	**0.89**	**0.85**	**0.85**	**0.87**	**0.85**	0.68	0.68
Density	**p= 0.002**	**p= 0.003**	**p= 0.008**	**p= 0.008**	**p= 0.005**	**p= 0.008**	p= 0.066	p= 0.066
Mine	0.01	-0.03	-0.14	-0.14	0.04	0.00	-0.09	-0.09
Density	p= 0.976	p= 0.95	p= 0.748	p= 0.748	p= 0.922	p= 1.00	p= 0.839	p= 0.839
Riparian								
Proportion	**-0.87**	**-0.85**	**-0.75**	**-0.75**	-0.70	-0.68	-0.43	-0.43
Conifer Forest	**p= 0.005**	**p= 0.008**	**p= 0.033**	**p= 0.033**	p= 0.053	p= 0.063	p= 0.288	p= 0.288
Proportion	0.66	0.58	0.63	0.63	0.67	0.59	0.60	0.60
Agriculture	p= 0.078	p= 0.131	p= 0.092	p= 0.092	p= 0.066	p= 0.124	p= 0.114	p= 0.114

Table 12 continued.

Proportion Disturbance	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A
Proportion Residential	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A	N/A N/A
Road Density	0.05 p= 0.904	0.03 p= 0.953	-0.05 p= 0.910	0.05 p= 0.910	0.16 p= 0.700	0.13 p= 0.747	0.05 p= 0.908	0.05 p= 0.908
Mine Density	-0.21 p= 0.619	-0.25 p= 0.556	-0.34 p= 0.411	-0.34 p= 0.411	-0.07 p= 0.877	-0.10 p= 0.811	-0.06 p= 0.890	0.06 p= 0.890

Table 13. Spearman's rank correlation coefficients (r) and p-values relating mean reach biological stream integrity metrics (predictor variable) and Δwhirling disease risk values (response variable) within the Rock Creek drainage. Whirling disease risk values recorded during the invertebrate collection period are reported as the proportions of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportions of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period are reported as the proportions infected with a histological score of grade four or greater (SEVALL) and the proportions of infected rainbow trout (PREVALL). The Δ indicates the change in the respective risk value between the upper and lower sites in the reach ; n = 5. Statistically significant relationships at the adjusted alpha level ($p \leq 0.0009$) are indicated by bold face type and an asterisk. Relationships considered marginally significant ($0.0009 < p \leq 0.05$) are in bold face type only.

	Reach Correlations			
Metric	**ΔSEVSAMP**	**ΔSEVALL**	**ΔPREVSAMP**	**ΔPREVALL**
Density Oligochaetes	**0.89 p = 0.041**	**0.89 p = 0.041**	0.70 p = 0.188	0.70 p = 0.188
Density Tubificids	0.75 p = 0.144	0.75 p = 0.144	0.67 p = 0.215	0.67 p = 0.215
Density *T. tubifex*	0.66 p = 0.150	0.66 p = 0.150	0.66 p = 0.150	0.66 p = 0.150
Density *L. hoffmeisteri*	0.69 p = 0.132	0.17 p = 0.745	0.17 p = 0.745	0.17 p = 0.745
Total Taxa Richness	0.67 p = 0.215	0.67 p = 0.215	0.40 p = 0.505	0.40 p = 0.505
Ephemeroptera Taxa Richness	-0.13 p = 0.841	-0.13 p = 0.841	0.11 p = 0.858	0.11 p = 0.858
Plecoptera Taxa Richness	-0.67 p = 0.215	-0.67 p = 0.215	-0.80 p = 0.104	-0.80 p = 0.104
Trichoptera Taxa Richness	0.29 p = 0.640	0.29 p = 0.640	0.56 p = 0.322	0.56 p = 0.322
EPT Taxa Richness	-0.67 p = 0.215	-0.67 p = 0.215	-0.50 p = 0.391	-0.50 p = 0.391
Proportion Ephemeroptera	-0.11 p = 0.858	-0.11 p = 0.858	-0.30 p = 0.624	-0.30 p = 0.624
Proportion Plecoptera	-0.67 p = 0.215	-0.67 p = 0.215	-0.80 p = 0.104	-0.80 p = 0.104
Proportion EPT	0.34 p = 0.581	0.34 p = 0.581	0.40 p = 0.505	0.40 p = 0.505
Proportion Dominant	-0.78 p = 0.118	-0.78 p = 0.118	-0.60 p = 0.285	-0.60 p = 0.285
Shannon Diversity	**0.89 p = 0.041**	**0.89 p = 0.041**	**0.90 p = 0.037**	**0.90 p = 0.037**

Table 14. Spearman's rank correlation coefficients (r) and p-values relating physicochemical parameters (predictor variable) to Δwhirling disease risk (response variable) within the Rock Creek drainage. Whirling disease risk values recorded during the invertebrate collection period are reported as the proportions of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportions of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period are reported as the proportions infected with a histological score of grade four or greater (SEVALL) and the proportions of infected rainbow trout (PREVALL). The Δ indicates the change in the respective risk value between the upper and lower sites in the reach. n = 4 for temperature, dissolved oxygen, and pH correlations. n = 5 for discharge and proportion organic matter correlations. n = 7 for conductivity correlations. n = 8 for elevation and slope correlations. Statistically significant relationships at the adjusted alpha level ($p \leq 0.0016$) are indicated by bold face type and an asterisk. Relationships considered marginally significant ($0.0016 < p \leq 0.05$) are in bold face type only.

	Reach Correlations			
Physicochemical Variable	**ΔSEVSAMP**	**ΔSEVALL**	**ΔPREVSAMP**	**ΔPREVALL**
Discharge	-0.11 p = 0.858	-0.11 p = 0.858	-0.30 p = 0.624	-0.30 p = 0.624
Proportion Organic Matter	0.34 p = 0.581	0.34 p = 0.581	0.20 p = 0.747	0.20 p = 0.747
Temperature	0.26 p = 0.742	0.26 p = 0.742	0.60 p = 0.400	0.60 p = 0.400
Conductivity	0.02 p = 0.966	0.10 p = 0.834	-0.20 p = 0.667	-0.20 p = 0.667
pH	-0.77 p = 0.225	-0.77 p = 0.225	-0.80 p = 0.200	-0.80 p = 0.200
Dissolved Oxygen	-0.77 p = 0.225	-0.77 p = 0.225	-0.80 p = 0.200	-0.80 p = 0.200
Elevation	0.16 p = 0.696	0.19 p = 0.651	0.44 p = 0.273	0.44 p = 0.273
Slope	-0.21 p = 0.624	-0.16 p = 0.699	0.01 p =0.977	0.01 p = 0.977

Blackfoot River Drainage

Whirling Disease Risk

Myxobolus cerebralis was detected at all sites within the Blackfoot River drainage (Table 15). Whirling disease risk increased beyond the period of invertebrate sampling at all sites sampled.

Correlations at the Catchment Scale

Land Use. The proportion of forest was identified as the dominant land cover, accounting for roughly 0.20 to 0.85 of the total area within the entire upstream subbasin and riparian zone (Appendix V). The proportion of land classified as disturbance ranged between 0 and 0.01 within the subbasin and riparian zone, while the proportion of agricultural area accounted for as much as 0.17 of the total riparian area. The proportion of residential area and density of mines comprised small portions of the total landscape (>0.001). Road densities within the entire upstream subbasin and riparian zone ranged between 0.10 and 1.83 km/km^2. Significant positive relationships were identified between whirling disease risk and the proportion of agricultural area, disturbed area, and road density within the subbasin (Table 16). Significant negative correlations were identified between risk and the proportion of forest within the subbasin and riparian area.

Correlations at the Reach Scale

Land Use. Land use proportions and densities identified at the reach scale were comparable to those described at the catchment scale (Appendix W). No significant relationships were identified between Δwhirling disease risk and land use within the contributing subbasin or riparian zone (Table 16).

Biological Stream Integrity. The lowermost site on the North Fork Blackfoot River (NF1) had the highest mean density of oligochaetes at the various taxonomic levels (Appendix X). No oligochaetes were detected at the uppermost site on the Blackfoot River mainstem (BL3) (Appendix X; Appendix Y). No significant relationships were identified between Δwhirling disease risk and any of the bioassessment metrics (Table 17).

Physical and Chemical Environment. Site elevation and reach slope data are summarized for sites and reaches in Appendix Z and Appendix AA, respectively. No statistically significant relationships were detected between Δwhirling disease risk and elevation or reach slope (Table 18).

Table 15. Whirling disease risk values within the Blackfoot River drainage. Whirling disease risk values recorded during the invertebrate collection period (1998) are reported as the proportions of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportions of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period (1997 – 2004) are reported as the proportions infected with a histological score of grade four or greater (SEVALL) and the proportions of infected rainbow trout (PREVALL). Values prefaced with a Δ indicate the change in that respective value between the upper and lower sites in that reach. Multiple sites within a reach are indicated by brackets and are identified by the lowermost site in a reach, signified by boldface type.

	Catchment				**Reach**			
Site	**SEVSAMP**	**SEVALL**	**PREVSAMP**	**PREVALL**	**ΔSEVSAMP**	**ΔSEVALL**	**ΔPREVSAMP**	**ΔPREVALL**
KL	0.42	1	0.90	1	0.42	1	0.90	1
NF2	0	0.03	0	0.64	0	0.03	0	0.64
NF1	0	0	0.12	0.12	0	0	0.12	0.12
WA	0	0.2	0.19	0.89	0	0.2	0.19	0.89
CH	0	0.86	0.08	0.98	0	0.86	0.08	0.98
CO	0.88	0.96	0.94	1	0.88	0.96	0.94	1
BL3	0	0.25	0	0.94	0	0.25	0	0.94
{ BL2	0.07	0.59	0.52	0.97				
BL1	0	0	0.14	0.14	-0.07	-0.59	-0.38	-0.83

Table 16. Spearman's rank correlation coefficients (r) and p-values relating upstream land use (predictor variable) to whirling disease risk and Δwhirling disease risk (response variable) within the Blackfoot River drainage. Whirling disease risk values recorded during the invertebrate collection period are reported as the proportions of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportions of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period are reported as the proportions infected with a histological score of grade four or greater (SEVALL) and the proportions of infected rainbow trout (PREVALL). Values prefaced with a Δ indicate the change in that respective value between the upper and lower sites in that reach. n = 8 for PREVSAMP, SEVSAMP, PREVALL and SEVALL correlations. n = 7 for ΔPREVSAMP, ΔSEVSAMP, ΔPREVALL and ΔSEVALL correlations. N/A indicates correlations not conducted because land use at that scale was not present. Statistically significant relationships at the adjusted alpha level ($p \leq 0.0005$) are indicated by bold face type and an asterisk. Relationships considered marginally significant ($0.0005 < p \leq 0.05$) are in bold face type only.

	Catchment Correlations				**Reach Correlations**			
Land Use	**SEVSAMP**	**SEVALL**	**PREVSAMP**	**PREVALL**	**ΔSEVSAMP**	**ΔSEVALL**	**ΔPREVSAMP**	**ΔPREVALL**
Subbasin								
Proportion Conifer Forest	-0.34 p = 0.406	-0.28 p = 0.509	**-0.80** **p = 0.017**	-0.29 p = 0.490	-0.43 p = 0.331	-0.50 p = 0.253	-0.74 p = 0.058	-0.49 p = 0.268
Proportion Agriculture	0.34 p = 0.406	0.07 p = 0.866	**0.71** **p = 0.050**	0.02 p = 0.955	0.43 p = 0.331	0.32 p = 0.482	0.58 p = 0.175	0.31 p = 0.504
Proportion Disturbance	**0.77** **p = 0.026**	0.58 p = 0.133	0.61 p = 0.110	0.63 p = 0.100	0.32 p = 0.487	0.38 p = 0.403	0.28 p = 0.540	0.42 p = 0.351
Proportion Residential	-0.43 p = 0.290	-0.26 p = 0.533	-0.08 p = 0.846	-0.22 p = 0.601	-0.61 p = 0.147	-0.59 p = 0.162	-0.39 p = 0.390	-0.60 p = 0.158
Road Density	0.34 p = 0.406	0.34 p = 0.417	**0.71** **p = 0.050**	0.36 p = 0.382	-0.10 p = 0.834	-0.07 p = 0.879	0.18 p = 0.699	-0.09 p = 0.848
Mine Density	-0.43 p = 0.289	-0.51 p = 0.199	-0.40 p = 0.329	-0.49 p = 0.214	-0.54 p = 0.210	-0.40 p = 0.373	-0.67 p = 0.100	-0.40 p = 0.368
Riparian								
Proportion Conifer Forest	-0.48 p = 0.225	-0.36 p = 0.382	**-0.80** **p = 0.017**	-0.37 p = 0.365	-0.33 p = 0.463	-0.32 p = 0.482	-0.58 p = 0.175	-0.31 p = 0.504
Proportion Agriculture	0.17 p = 0.685	0.01 p = 0.978	0.53 p = 0.180	-0.06 p = 0.888	0.06 p = 0.900	0.11 p = 0.819	0.18 p = 0.699	0.04 p = 0.939

Table 16 continued.

Proportion Disturbance	0.27 p = 0.523	0.14 p = 0.733	0.16 p = 0.700	0.22 p = 0.606	-0.16 p = 0.733	-0.02 p = 0.970	-0.28 p = 0.540	0.00 p = 1.00
Proportion Residential	-0.33 p = 0.430	-0.37 p = 0.370	-0.26 p = 0.536	-0.30 p = 0.474	-0.66 p = 0.104	-0.53 p = 0.216	-0.74 p = 0.056	-0.54 p = 0.212
Road Density	-0.02 p = 0.971	0.11 p = 0.800	0.16 p = 0.713	0.16 p = 0.713	-0.43 p = 0.331	-0.28 p = 0.535	-0.34 p = 0.452	-0.31 p = 0.504
Mine Density	-0.33 p = 0.430	-0.26 p = 0.536	-0.37 p = 0.370	-0.20 p = 0.628	-0.54 p = 0.210	-0.40 p = 0.372	-0.67 p = 0.100	-0.40 p = 0.368

Table 17. Spearman's rank correlation coefficients (r) and p-values relating mean reach biological stream integrity metrics (predictor variable) and Δwhirling disease risk values (response variable) within the Blackfoot River drainage. Whirling disease risk values recorded during the invertebrate collection period are reported as the proportions of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportions of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period are reported as the proportions infected with a histological score of grade four or greater (SEVALL) and the proportions of infected rainbow trout (PREVALL). The Δ indicates the change in the respective risk value between the upper and lower sites in the reach; n = 7. Statistically significant relationships at the adjusted alpha level ($p \leq 0.003$) are indicated by bold face type and an asterisk. Relationships considered marginally significant ($0.003 < p \leq 0.05$) are in bold face type only.

	Reach Correlations			
Metric	**ΔSEVSAMP**	**ΔSEVALL**	**ΔPREVSAMP**	**ΔPREVALL**
Density Oligochaetes	0.06 p = 0.900	0.21 p = 0.645	0.34 p = 0.452	0.14 p = 0.758
Density Tubificids	0.06 p = 0.900	0.25 p = 0.585	0.34 p = 0.448	0.18 p = 0.700
Density *T. tubifex*	-0.54 p = 0.210	-0.22 p = 0.631	-0.40 p = 0.368	-0.22 p = 0.628
Density *L. hoffmeisteri*	0.03 p = 0.945	0.16 p = 0.736	0.20 p = 0.799	0.23 p = 0.622

Table 18. Spearman's rank correlation coefficients (r) and p-values relating physicochemical parameters (predictor variable) to Δwhirling disease risk (response variable) within the Blackfoot River drainage. Whirling disease risk values recorded during the invertebrate collection period are reported as the proportions of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportions of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period are reported as the proportions infected with a histological score of grade four or greater (SEVALL) and the proportions of infected rainbow trout (PREVALL). The Δ indicates the change in the respective risk value between the upper and lower sites in the reach. n = 7 for all correlations. Statistically significant relationships at the adjusted alpha level ($p \leq 0.006$) are indicated by bold face type and an asterisk. Relationships considered marginally significant ($0.006 < p \leq 0.05$) are in bold face type only.

Physicochemical Variable	**Reach Correlations**			
	ΔPREVSAMP	**ΔSEVSAMP**	**ΔPREVALL**	**ΔSEVALL**
Elevation	0.14 p = 0.758	0.35 p = 0.435	0.27 p = 0.558	0.32 p = 0.482
Slope	0.52 p = 0.229	0.24 p = 0.610	0.18 p = 0.700	0.14 p = 0.760

Among Drainages

Correlations at the Catchment Scale

Land Use. Final regression trees constructed with land use as the explanatory variables accounted for 42 - 51% of the variation in whirling disease risk (Table 19). The land use variables identified as most important (the variable that appears first in the regression tree model) in predicting whirling disease risk were the proportion of forest within the riparian zone and road density within the subbasin (Figure 12). Whirling disease risk was predicted to be lower in areas with a proportion of upstream riparian forest greater than 0.59 by the regression tree models with SEVSAMP, SEVALL, and PREVSAMP as the dependent variables. Whirling disease risk (PREVALL) was predicted to be lower in subbasins with lower road densities (<0.29 km/km^2), and a small reduction in deviance was further accounted for by the proportion of forested area within the riparian zone.

Correlations at the Reach Scale

Land Use. Final regression trees constructed with land use as the explanatory variables accounted for 25 - 56% of the variation in Δwhirling disease risk (Table 19). Once again, the land use variables identified as most important in predicting risk were the proportion of forest within the riparian zone and road density within the subbasin (Figure 13). ΔWhirling disease severity was predicted to be greater in less forested riparian areas (<~0.60). ΔWhirling disease prevalence was predicted to be greater in subbasins with increased road densities (>~0.40km/ km^2). The proportion of forested riparian area accounted for a further reduction in deviance after subbasin road density in the ΔPREVSAMP regression tree model.

Biological Stream Integrity. Final regression trees with metrics quantified from the oligochaete community as explanatory variables accounted for 14 - 36% of the variation in Δwhirling disease risk (Table 20). The density of oligochaetes and *L. hoffmeisteri* were identified as the most important explanatory variables (Figure 14). ΔWhirling disease severity was predicted to be higher in reaches with densities of *L. hoffmeisteri* greater than 168.21/m^2. ΔWhirling disease prevalence was predicted to be higher in reaches with greater oligochaete densities (>~30,000 oligochaetes/ m^2).

Final regression trees with metrics quantified from the macroinvertebrate assemblage as explanatory variables accounted for 27 – 61% of the variation in Δwhirling disease risk (Table 21). The most important predictor variable identified by regression tree models to account for variation in Δwhirling disease risk during the period of invertebrate sampling was Plecoptera taxa richness (Figure 15). ΔWhirling disease risk was predicted to be higher in areas with less than 2.48 Plecoptera taxa present. Over the entire field exposure period, the proportion of Plecoptera taxa present accounted for the largest reduction in deviance in Δwhirling disease risk. ΔWhirling disease risk was predicted to be higher in areas with a proportion of Plecoptera taxa less than 0.01.

Physical and Chemical Environment. No significant relationships were detected between Δwhirling disease risk and any physicochemical parameters (Table 22).

Table 19. Predictor land use variables included in (indicated by X) and the amount of variability explained (D^2) by each regression tree. Whirling disease risk values recorded during the invertebrate collection period are reported as the proportions of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportions of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period are reported as the proportions infected with a histological score of grade four or greater (SEVALL) and the proportions of infected rainbow trout (PREVALL). Values prefaced with a Δ indicate the change in the respective value between the upper and lower sites in that reach.

	Regression Tree							
	Catchment				**Reach**			
Predictor Variables	**SEVSAMP**	**SEVALL**	**PREVSAMP**	**PREVALL**	**ΔSEVSAMP**	**ΔSEVALL**	**ΔPREVSAMP**	**ΔPREVALL**
Subbasin								
Road Density				X			X	X
Proportion Residential								
Proportion Agriculture								
Proportion Disturbance								
Proportion Conifer Forest							X	
Mine Density								
Riparian								
Road Density								
Proportion Residential								
Proportion Agriculture								
Proportion Disturbance								
Proportion Conifer Forest	X	X	X	X	X	X		
Mine Density								
D^2	0.43	0.42	0.48	0.51	0.40	0.40	0.56	0.25

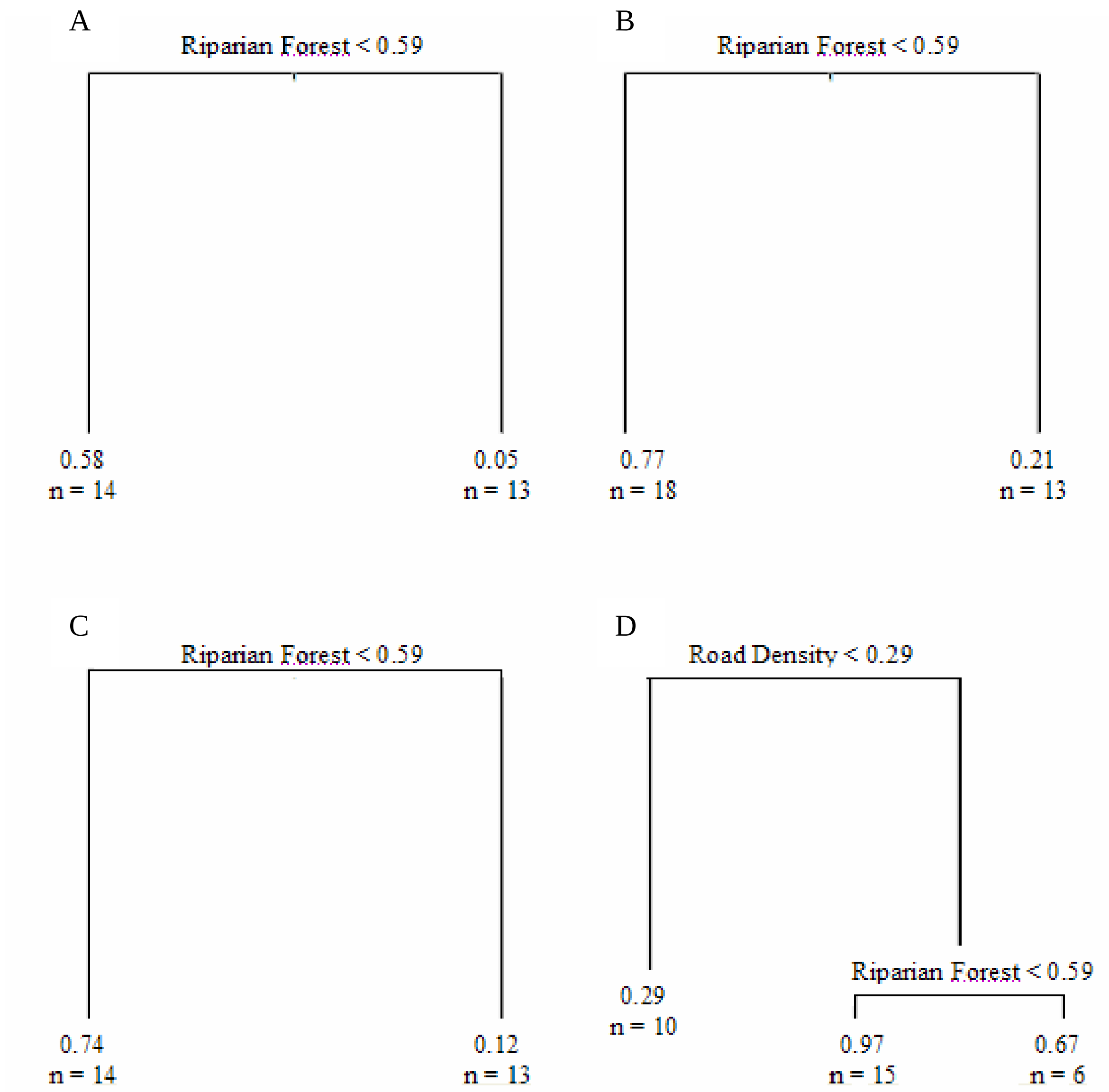

Figure 12. Final regression trees for whirling disease risk with land use within the entire upstream subbasin and riparian zone as explanatory variables. Whirling disease risk values recorded during the invertebrate collection period are reported as the proportions of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportions of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period are reported as the proportions infected with a histological score of grade four or greater (SEVALL) and the proportions of infected rainbow trout (PREVALL). The response variable for each regression tree model is as follows: A) SEVSAMP, B) SEVALL, C) PREVSAMP, and D) PREVALL. Text and value at split indicates criteria for branching. Numbers at terminal nodes indicate the estimated value of whirling disease risk and the number of observations in that group.

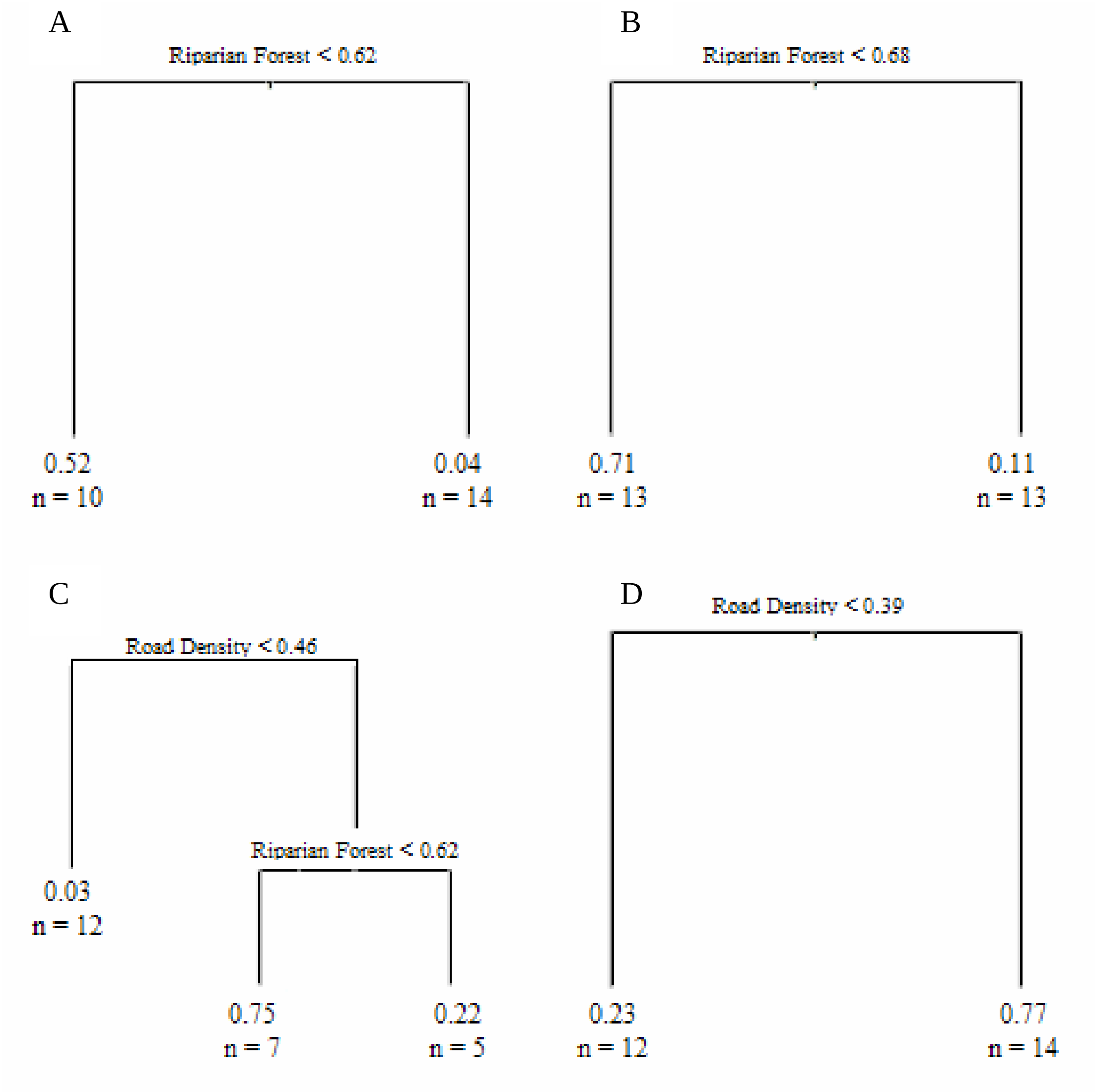

Figure 13. Final regression trees for Δwhirling disease risk with land use contributing to the reach subbasin and riparian zone as explanatory variables. Whirling disease risk values recorded during the invertebrate collection period are reported as the proportions of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportions of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period are reported as the proportions infected with a histological score of grade four or greater (SEVALL) and the proportions of infected rainbow trout (PREVALL). Δ indicates the change in that respective value between the upper and lower sites in that reach. The response variable for each regression tree model is as follows: A) ΔSEVSAMP, B) ΔSEVALL, C) ΔPREVSAMP, and D) ΔPREVALL. Text and value at split indicates criteria for branching. Numbers at terminal nodes indicate the estimated value of Δwhirling disease risk and the number of observations in that group.

Table 20. Predictor oligochaete bioassessment metric variables included in (indicated by X) and the amount of variability explained (D^2) by each regression tree. Whirling disease risk values recorded during the invertebrate collection period are reported as the proportions of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportions of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period are reported as the proportions infected with a histological score of grade four or greater (SEVALL) and the proportions of infected rainbow trout (PREVALL). The Δ indicates the change in the respective risk value between the upper and lower sites in the reach.

	Regression Tree			
	Reach			
Predictor Variables	**ΔSEVSAMP**	**ΔSEVALL**	**ΔPREVSAMP**	**ΔPREVALL**
Density Oligochaetes			X	X
Density Tubificids				
Density *T. tubifex*				
Density *L. hoffmeisteri*	X	X		
D^2	0.36	0.36	0.27	0.14

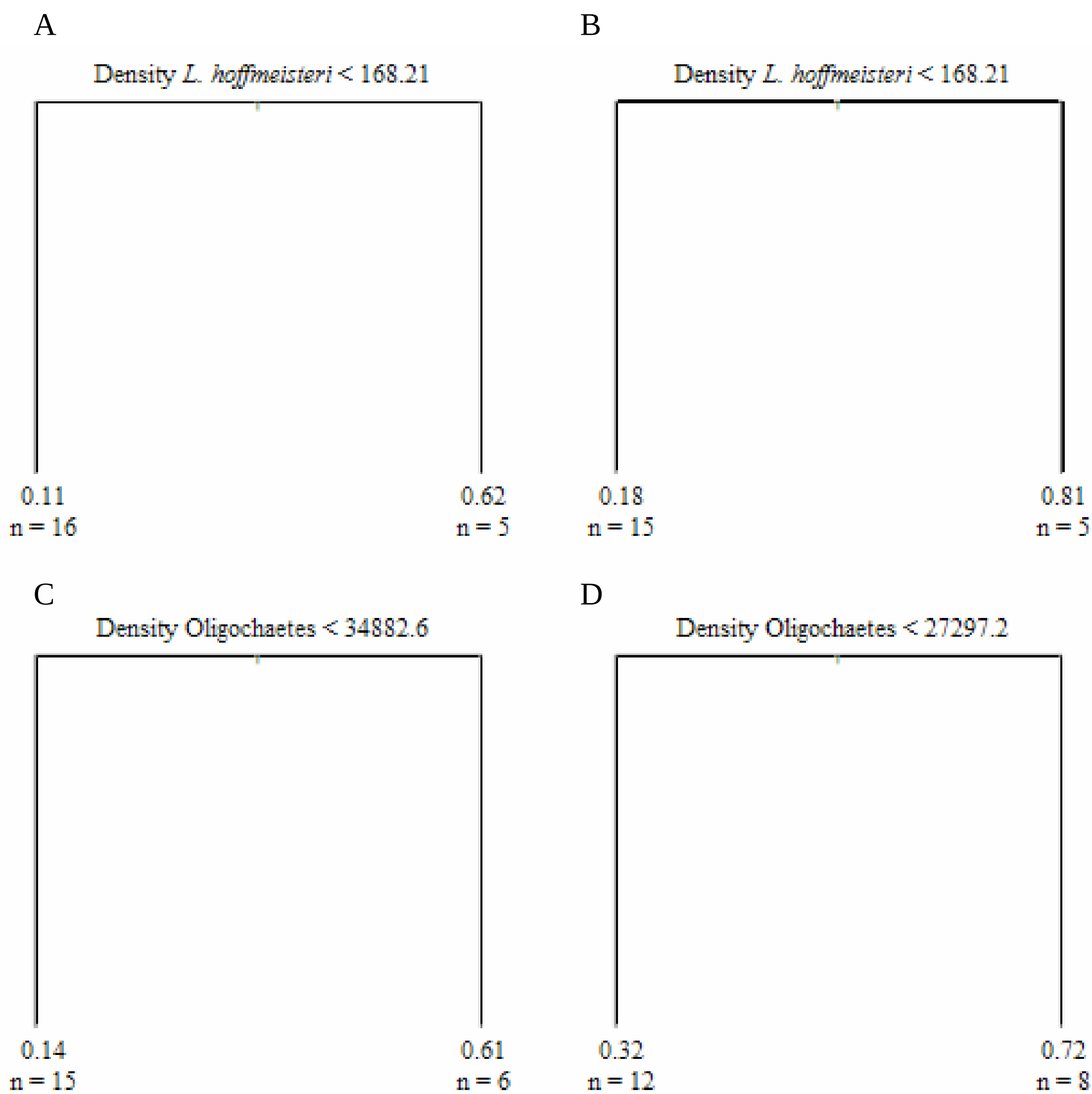

Figure 14. Final regression trees for Δwhirling disease risk with reach mean bioassessment metrics quantified from the oligochaete community as explanatory variables. Whirling disease risk values recorded during the invertebrate collection period are reported as the proportions of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportions of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period are reported as the proportions infected with a histological score of grade four or greater (SEVALL) and the proportions of infected rainbow trout (PREVALL). Δ indicates the change in that respective value between the upper and lower sites in that reach. The response variable for each regression tree model is as follows: A) ΔSEVSAMP, B) ΔSEVALL, C) ΔPREVSAMP, and D) ΔPREVALL. Text and value at split indicates criteria for branching. Numbers at terminal nodes indicate the estimated value of Δwhirling disease risk and the number of observations in that group.

Table 21. Predictor macroinvertebrate bioassessment metric variables included in (indicated by X) and the amount of variability explained (D^2) by each regression tree. Whirling disease risk values recorded during the invertebrate collection period are reported as the proportions of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportions of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period are reported as the proportions infected with a histological score of grade four or greater (SEVALL) and the proportions of infected rainbow trout (PREVALL). The Δ indicates the change in the respective risk value between the upper and lower sites in the reach.

	Regression Tree			
	Reach			
Predictor Variables	**ΔSEVSAMP**	**ΔSEVALL**	**ΔPREVSAMP**	**ΔPREVALL**
Total Taxa Richness				
Ephemeroptera Taxa Richness				
Plecoptera Taxa Richness	X		X	
Trichoptera Taxa Richness				
EPT Taxa Richness				
Proportion Ephemeroptera				
Proportion Plecoptera		X		X
Proportion EPT				
Proportion Dominant				
Shannon Diversity Index				
D^2	0.27	0.40	0.61	0.47

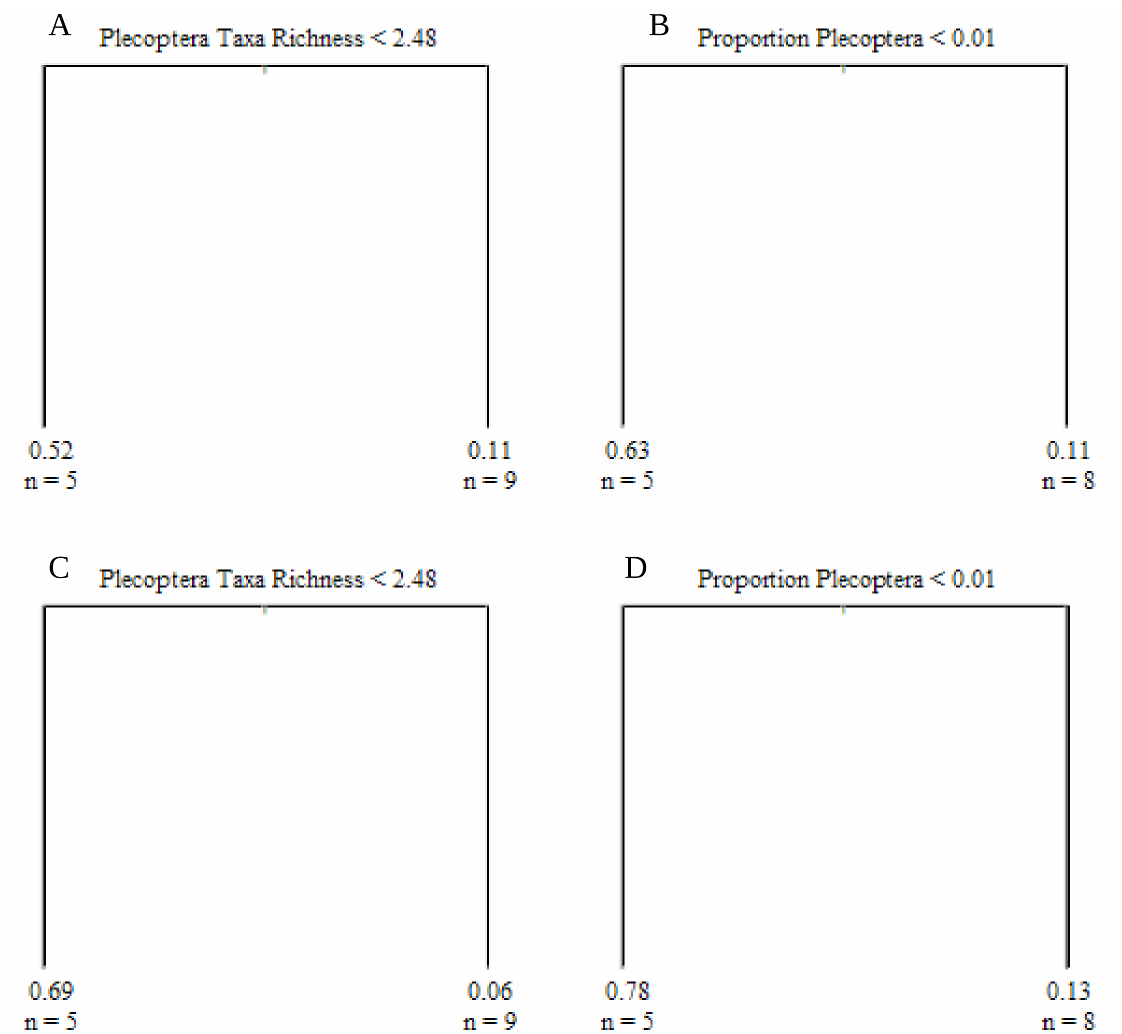

Figure 15. Final regression trees for Δwhirling disease risk with reach mean bioassessment metrics quantified from the macroinvertebrate community as explanatory variables. Whirling disease risk values recorded during the invertebrate collection period are reported as the proportions of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportions of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period are reported as the proportions infected with a histological score of grade four or greater (SEVALL) and the proportions of infected rainbow trout (PREVALL). Δ indicates the change in that respective value between the upper and lower sites in that reach. The response variable for each regression tree model is as follows: A) ΔSEVSAMP, B) ΔSEVALL, C) ΔPREVSAMP, and D) ΔPREVALL. Text and value at split indicates criteria for branching. Numbers at terminal nodes indicate the estimated value of Δwhirling disease risk and the number of observations in that group.

Table 22. Spearman's rank correlation coefficients (r) and p-values relating physicochemical parameters (predictor variable) to Δwhirling disease risk (response variable) among drainages. Whirling disease risk values recorded during the invertebrate collection period are reported as the proportions of infected rainbow trout with a histological score of grade four or greater (SEVSAMP) and the proportions of infected rainbow trout (PREVSAMP). Whirling disease risk values recorded throughout the study period are reported as the proportions infected with a histological score of grade four or greater (SEVALL) and the proportions of infected rainbow trout (PREVALL). The Δ indicates the change in the respective risk value between the upper and lower sites in the reach. n = 12 for temperature, dissolved oxygen, and pH correlations during the period of invertebrate sampling; n = 11 for correlations over the entire study period. n = 14 for discharge and proportion of organic material in sediment for sampling period correlations; n = 13 for correlations over all years. n = 16 for conductivity correlations during the sampling period and n = 15 for correlations over the entire study period. n = 24 for elevation and slope correlations during the period of invertebrate sampling and n = 26 for elevation and slope over the entire study period. Statistically significant relationships at the adjusted alpha level ($p \leq 0.0016$) are indicated by bold face type and an asterisk. Relationships considered marginally significant ($0.0016 < p \leq 0.05$) are in bold face type only.

	Reach Correlations			
Physicochemical Variable	**ΔSEVSAMP**	**ΔSEVALL**	**ΔPREVSAMP**	**ΔPREVALL**
Discharge	0.26 p = 0.364	0.07 p = 0.814	-0.09 p = 0.768	-0.22 p = 0.462
Proportion Organic Matter	0.23 p = 0.421	0.25 p = 0.407	0.24 p = 0.417	0.32 p = 0.283
Temperature	0.49 p = 0.109	0.26 p = 0.444	0.48 p = 0.117	0.18 p = 0.602
Conductivity	0.18 p = 0.497	0.36 p = 0.185	0.15 p = 0.580	0.13 p = 0.648
pH	0.17 p = 0.605	-0.10 p = 0.761	-0.14 p = 0.675	-0.40 p = 0.227
Dissolved Oxygen	-0.45 p = 0.147	-0.14 p = 0.684	-0.37 p = 0.241	-0.05 p = 0.892
Elevation	0.36 p = 0.087	-0.05 p = 0.818	0.20 p = 0.337	-0.14 p = 0.505
Slope	-0.16 p = 0.452	-0.31 p = 0.123	0.02 p = 0.914	-0.20 p = 0.326

DISCUSSION

The goal of this study was to examine the relationships between whirling disease risk and possible contributors to, and indicators of, stream degradation to help explain the broad scale variation of disease prevalence and severity within Montana watersheds. Land use such as roads and agriculture can degrade streams by altering the physical and chemical regimes of streams (e.g., increase sedimentation and nutrient input, decrease flow), which may increase whirling disease risk by creating favorable habitat for *T. tubifex*, and therefore, favorable habitat for *M. cerebralis*. Although high proportions of rainbow trout infected with *M. cerebralis* have been documented in stream reaches considered to be less than optimal *T. tubifex* habitat (i.e., cold, oligotrophic, sediment-poor) (Allen and Bergersen 2002); many studies have suggested that aquatic systems influenced by anthropogenic activities may exacerbate whirling disease (Zendt and Bergersen 2000; Nehring et al. 2003; Beauchamp et al. 2005; Kaeser and Sharpe 2006). For example, Kaeser and Sharpe (2006) found that high oligochaete and *T. tubifex* density were associated with proximity to cow pastures and other sources of organic enrichment. Similarly, Zendt and Bergersen (2000) reported that *T. tubifex* abundance, relative to other species, was higher in areas influenced by increased sediment input due to anthropogenic land use and water management practices than those areas unaffected by anthropogenic activities. Reducing erosion, the input of organic material, and mitigation of other contributors to stream degradation may reduce habitat conditions that promote high densities of *T. tubifex*, resulting in a decrease in whirling disease (Brinkhurst 1996).

The hypothesis in this study was that whirling disease risk is influenced by anthropogenic land use practices that create favorable habitat for *T. tubifex* which is reflected in the biological integrity and physicochemical features of the stream. Streams that drain catchments with a greater amount of anthropogenic land use (i.e., agriculture, residential, disturbance, mines and roads), low proportions of forest, low biological integrity, and physicochemical features that provide suitable habitat for *T. tubifex* will have a higher risk of whirling disease than those with less anthropogenic land use, high proportions of forest, high biological stream integrity, and physicochemical features that provide less suitable *T. tubifex* habitat. Within drainages, all land use types except mine density exhibited numerous significant relationships with whirling disease risk and Δwhirling disease risk. Several of the bioassessment metrics also exhibited statistically significant relationships with Δwhirling disease risk within drainages, although some were significant in the direction not predicted. Additionally, significant relationships were detected between Δwhirling disease risk and physicochemical features (i.e., temperature, pH, and dissolved oxygen) within the Madison River drainage.

Although correlations were identified within each drainage, the number, type and strength of the correlation varied significantly among the drainages. The number of relationships identified between measures of risk and all predictor variables was greatest within the Madison River and Rock Creek drainages. Close to 30 were detected within each drainage; approximately 20 of those relationships were detected between risk and land use. Within the Madison River drainage the strongest correlations, those ≤ the adjusted p-value, were identified between risk and the proportion of disturbance within the subbasin and roads within the riparian zone, while in the Rock Creek drainage, the proportion of agriculture and conifer forest provided the strongest correlations. The greatest number of significant correlations between risk and bioassessment metrics were also detected within these two drainages. Roughly five were detected within each drainage, although over half were significant in the direction not predicted and those within the Rock Creek drainage were only marginally significant. Furthermore, significant relationships between risk and physicochemical parameters were only detected within the Madison River drainage.

Far fewer correlations were identified within the Missouri River and Blackfoot River drainages. Fewer than 10 relationships between risk and land use were detected within each and within the Blackfoot all relationships identified were only of marginal significance. Only two relationships were detected between risk and bioassessment metrics within the Missouri River drainage, though highly significant in the direction predicted, and none were identified within the Blackfoot River drainage.

Although numerous relationships were detected between whirling disease risk and land use (except mine density) at both the catchment and reach scale, the most important predictors of risk among drainages were the proportion of forested riparian area and road density within the subbasin. The riparian zone, although only a minor proportion of the total catchment area, plays a substantial role in regulating many stream processes. Forested riparian areas act as buffers, moderating temperature, flow regimes, and the amount of nutrients and sediment transported to the stream from the surrounding landscape (Gregory et al. 1991). Therefore, intact forested riparian areas may help reduce disease risk by influencing parameters of the stream that limit the amount and suitability of available habitat for increased abundances of *T. tubifex* and success of the parasite at various stages within its lifecycle.

Riparian forest influences water temperature, which affects the availability of oxygen and nutrients, and also the variability of stream discharge (Barton et al. 1985). The riparian canopy reduces the amount of solar radiation reaching the water's surface, minimizing temperature fluctuations and extremes (Malcolm et al. 2004). Increased temperatures decrease the oxygen-carrying capacity of streams and increase the rate at which nutrients become available to biota (USEPA 2006). The availability of nutrients is also determined by the amount transported to the stream from the surrounding landscape. Streams draining deforested riparian areas often have highly variable discharge (Barton et al. 1985), characterized by low baseflow levels and high peak discharges during storm events. During peak discharge events, increased loads of nutrients, and the sediment to which they are attached, may be delivered to the stream (USEPA 2006).

The riparian forest essentially acts as a filter or sink, reducing the input of nutrients and sediment to the stream from the surrounding catchment. Forested streambanks physically trap sediment as it is transported via overland flow and also stabilize the bank, reducing the process of erosion (Naiman and Decamps 1997). Nutrient input to the stream is further reduced by the uptake of dissolved nutrients by plant roots in the shallow subsurface zone. Lowrance et al. (1984) found that riparian forests retained more than 65% of the nitrogen and 30% of the phosphorus contributed through overland and subsurface flow from surrounding agricultural lands. Therefore, the riparian forest may indirectly affect worm populations by reducing the amount of sediment and nutrients that reach the stream.

Forested riparian areas do not provide favorable habitat conditions for the worm host or the parasite. Increased abundances of *T. tubifex* are often found in areas of reduced oxygen conditions, increased siltation, and organic pollution, due to a tolerance to anoxia, lack of competition, and an increased food supply (Brinkhurst and Jamison 1971; Brinkhurst et al. 1983; Juget and LaFont 1994; Brinkhurst 1996). Furthermore, increased temperatures (Halliday 1973; El-Matbouli et al. 1999; Schisler et al. 2000; Blazer et al. 2003; Kerans et al. 2005) and low flow (Kerans and Zale 2002; Krueger et al. 2006) may contribute to the success of the parasite. Kaeser and Sharpe (2006) found that sites occupied by *T. tubifex* were associated with a lower proportion of upstream riparian forest. Therefore, forested riparian areas may significantly reduce the proliferation of *T. tubifex* and *M. cerebralis*, possibly reducing the risk of disease to rainbow trout populations.

The presence of roads within catchments has also been correlated to changes in the chemical, physical, and biological processes of stream systems. Road maintenance procedures and automobile emissions contribute sediment, heavy metals, salts, and nutrients to the surrounding land and water (Gjessing et al. 1984; Grayson et al. 1993). Additionally, roads can alter the direction of runoff, the flow of surface and subsurface water (Wemple et al. 1996), and intensify erosion (Richardson et al. 2001). Roads have also been identified as vectors for the spread of introduced species (Schedl 1991). For whirling disease, the increased stream accessibility that roads provide could facilitate the transport of the parasite on angler gear. The number of anglers, the extent and frequency of their movement among waters, the amount of sediment that is typically transported on gear, and the myxospore's propensity to readily adhere to gear such as felt-soled boots suggests that anglers could be a vector of transport (K. Gates, Montana State University, Bozeman, MT, personal communication). Unfortunately, Gates was unable to verify this hypothesis because current methods are too insensitive to detect myxospores in sediment at levels thought to occur "naturally" within systems.

Although this study does not identify the exact mechanism by which roads affect whirling disease risk, numerous other studies have linked the adverse effects of roads to fish populations. The standing stock of trout (fish $\geq$ 100 mm total length) was negatively correlated to the density of stream-road crossings within a portion of the Medicine Bow National Forest, Wyoming (Eaglin and Hubert 1993). Within the Columbia River Basin, bull trout populations were predicted to be lower in areas with increased road densities (Reiman et al. 1997) and the number of bull trout redds in the Swan Basin, Montana was negatively correlated to the density of logging roads in spawning tributaries (Baxter et al. 1999).

Even at the reach scale, subbasin road density was significantly correlated to Δwhirling disease risk, suggesting that risk may be influenced by the deleterious effects of roads within even a relatively small portion of the surrounding catchment. Possibly, localized sources of degradation are enough to sustain parasite populations by providing favorable habitat for *T. tubifex*. Lenat et al. (1981) documented shifts in the invertebrate community to sediment-tolerant taxa in response to increased sediment input from road construction. In the Madison River, Montana, Krueger et al. (2006) found that fine sediment substrate in side channels was a strong predictor of infected *T. tubifex* density and, in turn, the density of infected *T. tubifex* was the best predictor of the site-specific contribution to whirling disease risk. Furthermore, risk throughout the entire system was found to be greatly influenced by these highly infected side channels.

At the reach scale, regression tree models identified various bioassessment metrics quantified from the oligochaete and macroinvertebrate community as the best predictors of Δwhirling disease risk among drainages in their respective analysis. Of the bioassessment metrics quantified from the oligochaete community, oligochaetes at the subclass (density oligochaetes) and species (density *L. hoffmeisteri*) level were identified as important predictors of Δwhirling disease risk. Oligochaetes at various taxonomic levels are often used as indicators of stressors such as anoxia or organic pollution in ecosystem assessments (Boreham et al. 1989; Milbrink 1994; Verdonschot 1996; Weerasundara 2000) and *L. hoffmeisteri*, in particular, often co-occurs with *T. tubifex* in natural settings (Brinkhurst et al. 1972). Interestingly, only one significant correlation was identified between Δwhirling disease risk and *T. tubifex* density in the within-drainage analyses and *T. tubifex* density was not included in any of the regression tree models which was surprising and warrants further investigation.

Insignificant or weak correlations between whirling disease risk and the density of *T. tubifex* have been demonstrated in previous studies, suggesting that measures of *T. tubifex* density alone may not provide a clear picture of whirling disease risk (e.g., Hiner and Moffitt 2001). Krueger et al. (2006) point out that a low density of highly infected *T.*

tubifex can still result in a high contribution to whirling disease risk. Limitations of tubificid identification may have also influenced the exclusion of *T. tubifex* density as a predictor of risk. The density of *T. tubifex* may have been inaccurately portrayed because fall data were used to conduct analyses. If the sampling effort occurred when most *T. tubifex* were not sexually mature, and therefore morphologically unidentifiable, the density of *T. tubifex* would have been underrepresented. This caveat should also be heeded when considering *L. hoffmeisteri* as an indicator of risk, as their density estimate may have also been an affected by when samples were collected and samples taken at other times may show no correlation. As a result, estimates at a higher taxonomic level may be a more reliable predictor of risk. Krueger (2002) suggested that if risk was to be estimated based on worm density alone, density measures at the family level provided a more accurate estimate of risk than density measures at the species level (i.e., *T. tubifex*) because of limitations of identification.

Of the bioassessment metrics quantified from the macroinvertebrate assemblages, regression trees identified the richness and proportion of Plecoptera as important predictors of Δwhirling disease risk. As predicted, areas with higher values of Plecoptera taxa richness and proportions of Plecoptera were estimated to have lower Δwhirling disease risk. Plecopterans (stoneflies) are particularly sensitive to impairments at the reach scale such as loss of riparian canopy, streambank instability, and channelization (Bollman 2004). Interestingly, metrics associated with plecopterans were only significant twice in the within-drainage analyses and both occurred within the Madison River drainage. The surprising lack of significant correlations between Δwhirling disease risk and bioassessment metrics, along with the number in the direction not predicted, may be attributable to taxonomic resolution, issues of scale, levels of disturbance, and comparisons made among streams of varying order.

The taxonomic resolution at which bioassessment metrics were quantified from the macroinvertebrate community may have been too coarse to account for the wide range of within-family tolerance levels to environmental stressors. For example, Hilsenhoff (1988) found that organic pollution in a stream was greatly underestimated when a macroinvertebrate biotic index was quantified at the family versus species level. This overestimation of the stream's biological integrity occurred because tolerant genera and species within relatively intolerant families were the dominant organisms. By the same rationale, the biological integrity of an unpolluted stream may be underestimated.

The lack of significant results may have also been an issue of scale. The spatial scale at which bioassessment metrics detect impairment may not be the scale at which the parasite influences disease risk. The invertebrate community can respond fairly quickly to some point sources of degradation. Storey and Cowley (1997) found that macroinvertebrate assemblage structure within a forested riparian area 600 m downstream of a pasture was more similar to those found in forested control streams than those in the upstream agriculture site. In contrast, it is unknown how far downstream the parasite can influence disease risk after release from its worm host. TAMs produced within a relatively small portion of the stream may contribute significantly to the overall risk in a system (Krueger et al. 2006). It is possible that TAMs produced at localized points of degradation can travel past the point where the stream's biological integrity recovers. Therefore, the scale at which bioassessment metrics detect impairment may be smaller than the scale on which the parasite influences disease risk.

The unexpected correlations between ∆whirling disease risk and bioassessment metrics may have been influenced by disturbance levels and stream order. The frequency and intensity of disturbance can affect diversity; at low levels, competition reduces diversity whereas at high levels, species are actively excluded because few can cope with constant or frequent disturbances (Ricklefs and Miller 2000). However, intermediate levels of disturbance within a system may actually increase diversity by creating a mosaic of habitat types, or niche spaces. This increased heterogeneity of niche space at moderate disturbance levels within a stream could result in an overlap of species both tolerant and intolerant of pollution and habitat degradation. Many streams in the West are characterized by low species richness; disturbances that change the community composition may increase diversity and is often an indication that tolerant or exotic species are present (Karr et al. 1986). Alternatively, increased measures of diversity and richness in areas of high risk may have simply been a function of stream order, which was not controlled in this study. Whirling disease risk often was lower in the tributaries than in the mainstems. Moving downstream from small tributaries to larger, cobble-bottomed mainstem rivers, more niche space theoretically becomes available, simply due to an increase in physical space, which may allow for more species to coexist.

The small number of significant relationships identified between whirling disease risk and characteristics of the physical and chemical environment within drainages, and lack of among drainages, was not overly surprising. Relationships among risk and physicochemical factors reported in other studies differ in their assessment of significance and effect. For example, many inconsistent findings have been documented between whirling disease and temperature. In this study a positive relationship between risk and mean reach temperature (8.5-17.3°C) was detected within the Madison River drainage, whereas whirling disease risk was found to be negatively correlated to mean water temperatures (10.2–13.6°C) within the side channels of the Madison River, Montana (Krueger et al. 2006), positively correlated to water temperatures (6.2-11.2°C) within Willow Creek, Montana (Baldwin et al. 2000), and not correlated to mean water temperatures (7.8-10.8°C) in Idaho (Hiner and Moffitt 2001). Furthermore, Downing et al. (2002) found that peak infection in fish occurred at temperatures near 12°C. Although these contrasting results may have been influenced by the varying temperature regimes within each system, the findings presented here and those of others suggest that a single abiotic factor is most likely not the sole determinant of whirling disease risk.

The results of this study should prove useful in the management of whirling disease by helping to guide identification of those streams most at risk of parasite establishment and proliferation and bolstering arguments for mitigation of areas that would benefit from remediation efforts. The results suggest that at the catchment and reach scale, riparian forest and subbasin road density may be a reliable predictor of whirling disease risk. If land use data are not readily available, characteristics of the invertebrate community such as the density of oligochaetes or the proportion of plecopterans may be useful predictors of risk.

Some potential limitations to this study do exist. Not all sites were sampled consistently and the location of sentinel cages was not random, which may have biased results. Land use data were collected primarily in the year 1992 but land use activity within watersheds several years, even decades prior, may influence fish and invertebrate assemblages (Harding et al. 1998) and possibly whirling disease risk. Additionally, map classification errors may have influenced results. A case in point is conifer forest within the Rocky Mountain region, which encompasses the extent of this study. A comparison of the map pixel data to a set of reference data revealed that conifer forest within the Rocky Mountain region is underrepresented by the GIS data layer (Wickham et al. 2004). Furthermore, although a significant relationship between risk and elevation was not detected in this study, the natural physical gradients within watersheds that possibly affect whirling disease risk (e.g., temperature, substrate) may be associated with land use practices that often are not randomly distributed along those gradients. For example, agriculture is often concentrated in the lower elevation of catchments which may be naturally associated with higher water temperatures, decreased velocities, and fine sediments.

Characteristics of the invertebrate community may also be associated with natural physical gradients within watersheds. Stoneflies are often associated with cooler water temperatures and many identified in this study were shredders (Merritt and Cummins 1996). Therefore, one would expect to find them in the upper headwater reaches where water temperatures are cooler and course particulate organic matter is abundant. The upper headwater reaches are also where whirling disease risk values tended to be low.

Another possible caveat of this study is that invertebrate data were generalized beyond the stream reaches in which they were sampled to represent the biological integrity of the stream. However, variability of bioassessment metrics (e.g., total taxa richness, EPT taxa richness, and Shannon diversity) among reaches is negligible within a stream segment (8-16 km) and the additional sampling of one or two stream reaches results in a minimal increase (<10%) in the ability to detect impairment (Rabeni et al. 1999). Relationships found here should be assessed in other systems to evaluate whether or not they consistently hold across multiple systems.

Managing the parasite by eliminating one part of the life cycle is not realistic, but efforts to limit the effects of land use practices that create favorable habitat for *T. tubifex*, and therefore the parasite, could prove useful. The ability to predict those areas at risk and those areas that would benefit from remediation efforts is a crucial component to alleviating the effects of this disease on wild rainbow trout populations. The conservation and preservation of streams and rivers should be a priority to support wild salmonid populations within Montana and elsewhere.

REFERENCES CITED

Allan, J. D. 2004. Landscapes and riverscapes: the influence of land use on stream ecosystems. Annual Review of Ecology, Evolution, and Systematics 35:257-284.

Allen, M. B., and E. P. Bergersen. 2002. Factors influencing the distribution of *Myxobolus cerebralis*, the causative agent of whirling disease, in the Cache la Poudre River, Colorado. Diseases of Aquatic Organisms 49:51-60.

Allendorf, F. W., P. O. Spruell, and F. M. Utter. 2001. Whirling disease and wild trout: darwinian fisheries management. Fisheries 26(5):27-29.

Anderson, R. A. 2004. Occurrence and seasonal dynamics of the whirling disease parasite, *Myxobolus cerebralis*, in Montana spring creeks. Master's thesis. Montana State University, Bozeman, Montana.

Anlauf, A., and D. Neumann. 1997. The genetic variability of *Tubifex tubifex* (Muller) in 20 populations and its relation to habitat type. Archiv fur Hydrobiologie 139:145-162.

Baldwin, T. J., J. E. Peterson, G. C. McGhee, K. D. Staigmiller, E. S. Motteram, C. C. Downs, and D. R. Stanek. 1998. Distribution of *Myxobolus cerebralis* in salmonid fishes in Montana. Journal of Aquatic Animal Health 10:361-371.

Baldwin, T. J., E. R. Vincent, R. M. Silflow, and D. Stanek. 2000. *Myxobolus cerebralis* infection in rainbow trout (*Oncorhynchus mykiss*) and brown trout (*Salmo trutta*) exposed under natural stream conditions. Journal of Veterinary Diagnostic Investigation 12:312-321.

Barouque, C. P. A., and J. H. Pomeroy. 2001. Effects of forest harvesting on summer stream temperatures in New Brunswick, Canada: an inter-catchment, multiple year comparison. Hydrology and Earth System Sciences 5:599-613.

Bartholomew, J., B. L. Kerans, R. P. Hedrick, S. C. Macdiarmid, and J. R. Winton. 2005. A risk assessment based approach for the management of whirling disease. Reviews in Fisheries Science 13:205-230.

Bartholomew, J., and P. W. Reno. 2002. The history and dissemination of whirling disease. Pages 3-24 *in* J. L. Bartholomew and J. C. Wilson, editors. Whirling disease: reviews and current topics. American Fisheries Society, Symposium 29, Bethesda, Maryland.

Barton, D.R., W. D. Taylor, and R.M. Biette. 1985. Dimensions of riparian buffer strips required to maintain trout habitat in southern Ontario streams. North American Journal of Fisheries Management 5:364-378.

Baxter, C. V., C. A. Frissell, and F. R. Hauer. 1999. Geomorphology, logging roads, and the distribution of bull trout (*Salvelinus confluentus*) spawning in a forested river basin: implications for management and conservation. Transactions of the American Fisheries Society 128:854-867.

Beauchamp, K. A., M. El-Matbouli, M. Gay, M. P. Georgiadis, R. B. Nehring, and R. P. Hedrick. 2006. The effect of cohabitation of *Tubifex tubifex* (Oligochaeta: Tubificidae) populations on infections to *Myxobolus cerebralis* (Myxozoa: Myxobolidae). Journal of Invertebrate Pathology 91:1-8.

Beauchamp, K. A., M. Gay, G. O. Kelley, M. El-Matbouli, R. D. Kathman, R. B. Nehring, and R. P. Hedrick. 2002. Prevalence and susceptibility of infection to *Myxobolus cerebralis* and genetic differences among populations of *Tubifex tubifex*. Diseases of Aquatic Organisms 51:113-121.

Beauchamp, K. A., G. O. Kelley, R. B. Nehring, and R. P. Hedrick. 2005. The severity of whirling disease among wild trout corresponds to the differences in genetic composition of *Tubifex tubifex* populations in Colorado. Journal of Parasitology 91:53-60.

Blazer, V. S., T. B. Waldrop, W. B. Schill, C. L. Densmore, and D. Smith. 2003. Effects of water temperature and substrate type on spore production and release in eastern *Tubifex tubifex* worms infected with *Myxobolus cerebralis*. Journal of Parasitology 89:21-26.

Bollman, W. 1998. Improving stream bioassessment methods for the Montana valleys and foothill prairies ecoregion. Master's thesis. University of Montana, Missoula, Montana.

Bollman, W. 2004. A biological assessment of sites in the Madison River drainage, Madison and Beaverhead counties, Montana TMDL-M06 August-September, 2003. Report prepared for the Montana Department of Environmental Quality Planning, Prevention and Assistance Division.

Boreham, S., A. R. Hough, and P. Birch. 1989. The effects of livestock slurry pollution on the benthic macro-invertebrate fauna of a clay stream. The London Naturalist 68:77-80.

Breiman, L., J. H. Friedman, R. A. Olsen, and C. J. Stone. 1984. Classification and regression trees. Wadsworth International Group, Belmont, California, USA.

Brinkhurst, R. O. 1996. On the role of tubificid oligochaetes in relation to fish disease with special reference to the myxozoa. Annual Review of Fish Diseases 6:29-40.

Brinkhurst, R. O., P. M. Chapman, and M. A. Farrell. 1983. A comparative study of respiration rates of some aquatic oligochaetes in relation to sublethal stress. Internationale Revue Der Gesamten Hydrobiologie 68:683-699.

Brinkhurst, R. O., K. E. Chua, and N. K. Kaushick. 1972. Interspecific interactions and selective feeding of tubificid oligochaetes. Limnology and Oceanography 17:122-133.

Brinkhurst, R. O., and B. G. M. Jamieson. 1971. Aquatic Oligochaeta of the world. Oliver and Boyd, Edinburgh.

Brinkhurst, R. O., and C. R. Kennedy. 1965. Studies on the biology of the Tubificidae (Annelida, Oligochaeta) in a polluted stream. Journal of Animal Ecology 34:429-443.

Bruns, D. A. 2005. Macroinvertebrate response to land cover, habitat, and water chemistry in a mining-impacted river ecosystem: a GIS watershed analysis. Aquatic Sciences 67:403-423.

Chapman, P. M., and R. O. Brinkhurst. 1984. Lethal and sublethal tolerances of aquatic oligochaetes with reference to their use as a biotic index of pollution. Hydrobiologia 115:139-144.

Downing, D. C., T. E. McMahon, B. L. Kerans, and E. R. Vincent. 2002. Relation of spawning and rearing life history of rainbow trout and susceptibility to *Myxobolus cerebralis* infection in the Madison River, Montana. Journal of Aquatic Animal Health 14:191-203.

DuBey, R., and C. Caldwell. 2004. Distribution of *Tubifex tubifex* lineages and *Myxobolus cerebralis* infection in the tailwater of the San Juan River, New Mexico. Journal of Aquatic Animal Health 16:179-185.

Eaglin, G. S., and W. A. Hubert. 1993. Effects of logging roads on substrate and trout in streams of the Medicine Bow National Forest, Wyoming. North American Journal of Fisheries Management 13:844-846.

El-Matbouli, M., T. Fischer-Scherl, and R. W. Hoffmann. 1992. Present knowledge on the life cycle, taxonomy, pathology and therapy of some Myxosporea spp. important for freshwater fish. Annual Review of Fish Diseases 2:367-402.

El-Matbouli, M., and R. W. Hoffmann. 1989. Experimental transmission of two *Myxobolus* spp. developing bisporogeny via tubificid worms. Parasitology Research 75:461-464.

El-Matbouli, M., and R. W. Hoffmann. 1991. Effects of freezing, aging, and passage through the alimentary canal of predatory animals on the viability of *Myxobolus cerebralis* spores. Journal of Aquatic Animal Health 3:260-262.

El-Matbouli, M., R. W. Hoffmann, and C. Mandok. 1995. Light and electron microscopic observations on the route of the triactinomyxon-sporoplasm of *Myxobolus cerebralis* from epidermis to rainbow trout cartilage. Journal of Fish Biology 46:919-935.

El-Matbouli, M., T. S. McDowell, D. B. Antonio, K. B. Andree, and R. P. Hedrick. 1999. Effect of water temperature on the development, release and survival of the triactinomyxon stage of *Myxobolus cerebralis* in its oligochaete host. International Journal for Parasitology 29:627-641.

ESRI (Environmental Systems Research Institute). 1996-1999. ArcView GIS 3.2. Redlands, California.

Genito, D., W. J. Gburek, and A. N. Sharpley. 2002. Response of stream macro invertebrates to agricultural land cover in a small watershed. Journal of Freshwater Ecology 17:109-119.

Gilbert, M. A., and W. O. Granath. 2001. Persistent infection of *Myxobolus cerebralis*, the causitive agent of salmonid whirling disease, in *Tubifex tubifex*. Journal of Parasitology 87:101-107.

Gjessing, E., E. Lygren, L. Berglind, T. Gulbrandsen, and R. Skanne. 1984. Effect of highway runoff on lake water quality. Science of the Total Environment 33:247-257.

Grayson, R. B., S. R. Haydon, M. D. A. Jayasuriya, and B. C. Enlayson. 1993. Water quality in mountain ash forests - separating the impacts of roads from those of logging operations. Journal of Hydrology 150:459-480.

Gregory, S.V., F. J. Swanson, W. A. McKee, and K. W. Cummins. 1991. An ecosystem perspective of riparian zones. BioScience 41:540-551.

Halliday, M. M. 1973. Studies of *Myxosoma cerebralis*, a parasite of salmonids. II. The development and pathology of *Myxosoma cerebralis*, in experimentally infected rainbow trout (*Salmo gairdneri*) fry reared at different water temperatures. Nordisk Veterinarmedicin 25:349-358.

Harding, J. S., E. F. Benfield, P. V. Bolstad, G. S. Helfman, and E. B. D. Jones III. 1998. Stream biodiversity: the ghost of land use past. Proceedings of the National Academy of Sciences 95:14843-14847.

Hedrick, R. P., M. El-Matbouli, M. A. Adkison, and E. MacConnell. 1998. Whirling disease: re-emergence among wild trout. Immunological Reviews 166:365-376.

Hilsenhoff, W. L. 1988. Rapid field assessment of organic pollution with a family-level biotic index. The North American Benthological Society 7:65-68.

Hiner, M., and C. M. Moffitt. 2001. Variations in *Myxobolus cerebralis* infections in field exposed cutthroat, and rainbow trout in Idaho. Journal of Aquatic Animal Health 13:124-132.

Hiner, M., and C. M. Moffitt. 2002. Modeling *Myxobolus cerebralis* infections in trout: associations with habitat variables. Pages 167-179 *in* J. L. Bartholomew and J. C. Wilson, editors. Whirling disease: reviews and current topics. American Fisheries Society, Symposium 29, Bethesda, Maryland.

Hőfer, B. 1903. Uber die drehkrankheit der regenbogenforelle. Allgemeine Fischerei Zeitschrift 28:7-8.

Hoffman, G. L. 1970. Intercontinental and transcontinental dissemination and transfaunation of fish parasites with emphasis on whirling disease (*Myxosoma cerebralis*). Pages 69-81 *in* S. F. Snieszko, editor. Symposium on diseases of fishes and shellfishes.
American Fisheries Society, Special Publication 5, Bethesda, Maryland.

Hoffman, G. L. 1990. *Myxobolus cerebralis*, a worldwide cause of salmonid whirling disease. Journal of Aquatic Animal Health 2:30-37.

Hoffman, G. L., C. E. Dunbar, and A. Bradford. 1962. Whirling disease of trout caused by *Myxosoma cerebralis* in the United States. U.S. Fish and Wildlife Service, Special Scientific Report-Fisheries, Report 427 14pp.

iGage Mapping Corporation. 2002. All Topo Maps: Montana. Salt Lake City, Utah. (CD-ROM).

Juget, L., and M. Lafont. 1994. Theoretical habitat templets, species traits, and species richness: aquatic oligochaetes in the upper Rhone River and its floodplain. Freshwater Biology 31:327-340.

Kaeser, A. J., and W. E. Sharpe. 2006. Patterns of distribution and abundance of *Tubifex tubifex* and other aquatic oligochaetes in *Myxobolus cerebralis* enzootic areas in Pennsylvania. Journal of Aquatic Animal Health 18:64-78.

Kathman, R. D., and R. O. Brinkhurst. 1998. Guide to freshwater oligochaetes in North America. Aquatic Resources Center, College Grove, Tennessee.

Karr, J. R. 1991. Biological integrity: a long-neglected aspect of water resource management. Ecological Applications 1:66-84.

Karr, J. R. and E. W. Chu. 1999. Restoring life in running waters: better biological monitoring. Island Press, Washington, D.C.

Karr, J. R., and D. R. Dudley. 1981. Ecological perspective on water quality goals. Environmental Management 5:55-68.

Karr, J. R., K. D. Fausch, P. L. Angermeier, P. R. Yant, and I. J. Schlosser. 1986. Assessing biological integrity in running waters a method and its rationale. Illinois Natural History Survey, Special Publication 5.

Kent, K. L., K. B. Andree, J. L. Bartholomew, M. El-Matbouli, S. S. Desser, R. H. Delvin, S. W. Feist, R. P. Hedrick, R. W. Hoffman, J. Khattra, S. L. Hallett, R. J. G. Lester, M. Longshaw, O. Palenzeula, M. E. Siddall and C. Xiao. 2001. Recent advances in our knowledge of the Myxozoa. The Journal of Eukaryotic Microbiology 48:395-413.

Kerans, B. L., J. Karr, and S. A. Ahlstedt. 1992. Aquatic invertebrate assemblages: spatial and temporal differences among sampling protocols. The North American Benthological Society 11:377-390.

Kerans, B. L., and J. Karr. 1994. A benthic index of biotic integrity (B-IBI) for rivers of the Tennessee Valley. Ecological Applications 4:768-785.

Kerans, B. L., C. Rasmussen, R. Stevens, A. E. L. Colwell, and J. R. Winton. 2004. Differential propagation of the metazoan parasite *Myxobolus cerebralis* by *Limnodrilus hoffmeisteri*, *Ilyodrilus templetoni*, and genetically distinct strains of *Tubifex tubifex*. Journal of Parasitology 90:1366-1373.

Kerans, B. L., R. I. Stevens, and J. C. Lemmon. 2005. Water temperature affects a host-parasite interaction: *Tubifex tubifex* and *Myxobolus cerebralis*. Journal of Aquatic Animal Health 17:216-221.

Kerans, B. L., and A. V. Zale. 2002. The ecology of *Myxobolus cerebralis*. Pages 145-166 *in* J. L. Bartholomew and J. C. Wilson, editors. Whirling disease: reviews and current topics. American Fisheries Society, Symposium 29, Bethesda, Maryland.

Krueger, R. C. 2002. Correlations among environmental features, *Myxobolus cerebralis* infection prevalence in oligochaetes, and salmonid infection fish in the Madison River, Montana. Master's thesis. Montana State University, Bozeman, Montana.

Krueger, R. C., B. L. Kerans, E. R. Vincent and C. Rasmussen. 2006. Risk of *Myxobolus cerebralis* infection to rainbow trout in the Madison River, Montana. Ecological Applications 16:770-783.

Kwain, W. 1975. Embryonic development, early growth, and meristic variation in rainbow trout (*Salmo gairdneri*) exposed to combinations of light intensity and temperature. Journal of the Fisheries Research Board of Canada 32:397-402.

Lenat, D. R. 1984. Agriculture and stream water quality: a biological evaluation of erosion control procedures. Environmental Management 8:333-343.

Lenat, D. R, D. L. Penrose, and K. W. Eagleson. 1981. Variable effects of sediment addition on stream benthos. Hydrobiologia 79:187-194.

Lestochova, E. I. 1994. Influence of small-river conditions on the abundance of Tubificidae. Hydrobiologia 278:129-131.

Lowrance, R., R. Todd, J. Fail Jr., O. Hendrickson Jr., R. Leonard, and L. Asmussen. 1984. Riparian forests as nutrient filters in agricultural watersheds. BioScience 34:374-377.

Malcolm, I. A., D. M. Hannah, M. J. Donaghy, C. Soulsby, and A. F. Youngson. 2004. The influence of riparian woodland on the spatial and temporal variability of stream water temperatures in an upland salmon stream. Hydrology and Earth System Sciences 8:449-459.

Markiw, M. E., and K. Wolf. 1983. *Myxosoma cerebralis* (Myxozoa: Myxosporea) etilogic agent of salmonid whirling disease requires tubificid worm (Annelida: Oligochaeta) in its life cycle. Journal of Protozoology 30:561-564.

MacConnell, E., and E. R. Vincent. 2002. The effects of *Myxobolus cerebralis* on the salmonid host. Pages 95-100 *in* J. L. Bartholomew and J. C. Wilson, editors. Whirling disease: review and current topics. American Fisheries Society, Symposium 29, Bethesda, Maryland.

Merritt, R. W., and K. W. Cummins. 1996. An introduction to the aquatic insects of North America, third edition. Kendall/Hunt, Dubuque, Iowa.

Milbrink, G. 1994. Oligochaetes and water-pollution in 2 deep Norwegian lakes. Hydrobiologia 278:213-222.

Modin, J. 1998. Whirling disease in California: a review of its history, distribution, and impacts, 1965-1997. Journal of Aquatic Animal Health 10:132-142.

Morley, S. A., and J. R. Karr. 2002. Assessing and restoring the health of urban streams in the Puget Sound basin. Conservation Biology 16:1498-1509.

MTDEQ (Montana Department of Environmental Quality). 2004. Water quality integrated report for Montana. MTDEQ Water Quality Planning Bureau.

MTDEQ (Montana Department of Environmental Quality). 2005. Montana Abandoned Mines Inventory. MTDEQ. Helena, Montana. (http://nris.mt.gov/nsdi/nris/shape/deq_abandoned_mines.zip).

Naiman, R. J. and H. Decamps. 1997. The ecology of interfaces: riparian zones. Annual Review of Ecology and Systematics 28:621-658.

Nehring, R. B., K. G. Thompson, D. L. Shuler, and T. M. James. 2003. Using sediment core samples to examine spatial distribution of *Myxobolus cerebralis* actinospore production in Windy Gap Reservoir, Colorado. North American Journal of Fisheries Management 23:376-384.

Nehring, R. P., and P. G. Walker. 1996. Whirling disease in the wild: the new reality in the intermountain west. Fisheries 21(6):28-30.

Pennak, R. W. 1978. Fresh-water invertebrates of the United States, second edition. John Wiley and Sons.

R Development Core Team. 2005. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria.

Rabeni, C. F, N. Wang, and R. J. Sarver. 1999. Evaluating adequacy of the representative stream reach used in invertebrate monitoring programs. The North American Benthological Society 18:284-291.

Ricklefs, R. E., and G. L. Miller. 2000. Ecology, fourth edition. W. H. Freeman and Company, New York.

Rieman, B. E., D. C. Lee, and R. F. Thurow. 1997. Distribution, status, and likely future trends of bull trout within the Columbia River and Klamath River basins. North American Journal of Fisheries Management 17:1111-1125.

Richardson, E. V., D. B. Simons, and P. F. Lagasse. 2001. River engineering for highway encroachments: highways in the river environment. U. S. Department of Transportation, Federal Highway Administration, Report FHWA NHI 01-004 HDS 6, Washington D.C.

Ripley, B. 2005. tree: classification and regression trees. R package version 1.0-20.

Rosenberg, D. M. and V. H. Resh. 1993. Introduction to freshwater biomonitoring and benthic macroinvertebrates. Pages 1-9 *in* D. M. Rosenberg and V. H. Resh, editors. Freshwater Biomonitoring and Benthic Macroinvertebrates. Chapman and Hall, New York.

Ryce, E. K. N., A. V. Zale, and E. MacConnell. 2004. Effects of fish age and development of whirling parasite dose on the disease in rainbow trout. Diseases of Aquatic Organisms 59:225-233.

Ryce, E. K. N., A. V. Zale, E. MacConnell, and M. Nelson. 2005. Effects of fish age versus size on the development of whirling disease in rainbow trout. Diseases of Aquatic Organisms 63:69-76.

Sandell, T. A., H. V. Lorz, D. G. Stevens, and J. L. Bartholomew. 2001. Dynamics of *Myxobolus cerebralis* in the Lostine River, Oregon: implications for resident and anadromous salmonids. Journal of Aquatic Animal Health 13:142-150.

SAS Institute. 2004. SAS statistical software, release 9.1.2. SAS Institute Inc., Cary, North Carolina.

Schedl, W. 1991. Invasion of the American buffalo treehopper (*Stictocephala bisonia* Kopp and Yonke 1977) into Austria (Homoptera, Auchenorrhyncha, Membracidae). Anzeiger fuer Schaedlingskunde, Pflanzenschutz, Umweltschutz 64:9-13.

Schisler, G. L., E. P. Bergersen, and P. G. Walker. 2000. Effects of multiple stressors on morbidity and mortality of fingerling rainbow trout infected with Myxobolus cerebralis. Transactions of the American Fisheries Society 129:859-865.

Scott, M. C., G. S. Helfman, M. E. McTammany, E. F. Benfield, and P. V. Bolstad. 2002. Multiscale influences on physical and chemical stream conditions across Blue Ridge landscapes. Journal of the American Water Resources Association 38:1379-1392.

Sponseller, R. A., E. F. Benfield, and H. M. Valett. 2001. Relationships between land use, spatial scale and stream macroinvertebrate communities. Freshwater Biology 46:1409-1424.

Steinbach Elwell, L. C., B. L. Kerans, C. Rasmussen, and J. R. Winton. 2006. Interactions among two strains of *Tubifex tubifex* (Oligochaeta: Tubificidae) and *Myxobolus cerebralis* (Myxozoa). Diseases of Aquatic Organisms 68:131-139.

Steinbach Elwell L. C., B. L. Kerans, and J. Zicvovich in preparation. Host-parasite interactions mediated by a competitor.

Stoeser, D. B., G. N. Green, L. C. Morath, W. D. Heran, A. B. Wilson, D. W. Moore, and B. S. Van Gosen. 2005. Preliminary integrated geologic map databases for the United States central states: Montana, Wyoming, Colorado, New Mexico, Kansas, Oklahoma, Texas, Missouri, Arkansas, and Louisiana, - the state of Montana. Version 1.0, U.S. Geological Survey, Geological Survey Open-File Report 2005-1351, Denver, CO. (http://pubs.usgs.gov/of/2005/1351/).

Storey, R. G., and D. R. Cowley. 1997. Recovery of three New Zealand rural streams as they pass through native forest remnants. Hydrobiologia 353:63-76.

U.S. Army Corps of Engineers. 2003. Geospatial Hydrologic Modeling Extension (HEC-GeoHMS). Version 1.1, U.S. Army Corps of Engineers Hydrologic Engineering Center, Davis, California.

U.S. Census Bureau. 2001. Montana Roads from TIGER/Line Files. Edition: redistricting census 2000. U.S. Census Bureau, Helena, Montana. (http://nris.state.mt.us/nsdi/nris/road2000/road2000.html).

USDA (U.S. Department of Agriculture). 1996. Montana 5th-code watersheds. USDA Natural Resources Conservation Service, Helena, Montana. (http://nris.state.mt.us/nsdi/nris/e00/hd109.zip).

USEPA (U.S. Environmental Protection Agency). 2006. National management measures to control nonpoint source pollution from hydromodification. U.S. Environmental Protection Agency Office of Wetlands, Oceans and Watersheds, Washington, D.C. EPA 841-D-06-001.

USGS (U.S. Geological Survey). 2000a. Montana Land Cover Data Set. Edition: 1, USGS, Sioux Falls, South Dakota. (http://edcwww.cr.usgs.gov/programs/lccp/natllandcover.html)

USGS (U.S. Geological Survey). 2000b. Wyoming Land Cover Data Set. Edition: 1, USGS, Sioux Falls, South Dakota. (http://edcwww.cr.usgs.gov/programs/lccp/natllandcover.html)

USGS (U.S. Geological Survey). 2002. National elevation dataset for Montana. USGS, Helena, Montana. (http://nris.state.mt.us/nsdi/nris/el10/dems.html).

Van Sickle, J. 2003. Analyzing correlations between stream and watershed attributes. Journal of the American Water Resources Association 39:717-726.

Verdonschot, P. F. M. 1996. Oligochaetes and eutrophication; an experiment over four years in outdoor mesocosms. Hydrobiologia 334:169-183.

Vincent, E. R. 1996. Whirling disease and wild trout: the Montana experience. Fisheries 21(6):32-33.

Vincent, E. R. 2002. Relative susceptibility of various salmonids to whirling disease with emphasis on rainbow and cutthroat trout. Pages 109-115 *in* J. L. Bartholomew and J. C. Wilson, editors. Whirling disease: reviews and current topics. American Fisheries Society, Symposium 29, Bethesda, Maryland, USA.

Webster, J. R., S. W. Golladay, E. F. Benfield, J. L. Meyer, W. T. Swank, and J. B. Wallace. 1992. Catchment disturbance and stream response: an overview of stream research at Coweeta Hydrologic Laboratory. Pages 231-253 *in* P.J. Boon, P. Calow, and G. E. Petts, editors. River Conservation and Management. John Wiley and Sons, New York, New York, USA.

Weerasundara, A., A. Pathiratne, and H. Costa. 2000. Species composition and abundance of littoral oligochaete fauna in Lunuwila Reservoir, Sri Lanka. International Review of Hydrobiology 85:223-230.

Wemple, B. C., J. A. Jones, and G. E. Grant. 1996. Channel network extension by logging roads in two basins, western Cascades, Oregon. Water Resources Bulletin 32:1195-1207.

Wetzel, R. G., and G. E. Likens. 1991. Limnological analyses. Second edition, Springer -Verlag Press, New York.

Wickham, J. D., S. V. Stehman, J. H. Smith, and L. Yang. 2004. Thematic accuracy of the 1992 national land-cover data for the western United States. Remote Sensing Environment 91:452-468.

Wolf, K., and M. E. Markiw. 1984. Biology contravenes taxonomy in the Myxozoa: new discoveries show alteration of invertebrate and vertebrate hosts. Science 225:1449-1452.

Wolf, K., M. E. Markiw, and J. K. Hiltunen. 1986. Salmonid whirling disease: *Tubifex tubifex* (Müller) identified as the essential oligochaete in the protozoan life cycle. Journal of Fish Diseases 9:83-85.

Wright, P. S. 1992. Adjusted p-values for simultaneous inference. Biometrics 48:1005-1013.

Zendt, J. S. and E. P. Bergersen. 2000. Distribution and abundance of the aquatic oligochaete host *Tubifex tubifex* for the salmonids whirling disease parasite *Myxobolus cerebralis* in the Upper Colorado River Basin. North American Journal of Fisheries Management 20:502-512.

APPENDICES

APPENDIX A

YEARS OF SENTINEL FISH, INVERTEBRATE, AND PHYSICOCHEMICAL DATA COLLECTION

Years that sentinel cage, invertebrate, and physicochemical data were collected within each drainage at each site. Sites on each stream are listed from upstream to downstream. Sentinel cage field exposures lasted ten days and generally occurred from mid-May through June during the years indicated. Invertebrate data and some or all physicochemical data (excluding elevation and slope) were collected up to three times (spring, summer, and fall) during the year indicated. N/A for invertebrate data indicates that samples from that site were not processed. ES for physicochemical data indicates that only elevation and slope data were quantified for that site.

DRAINAGE	SITE	SENTINEL CAGE	INVERTEBRATE	PHYSICOCHEMICAL
Missouri				
	PR6	1998	1998 through 1999	1998 through 1999
	PR5	1998	1998 through 1999	1998 through 1999
	PR4	1998	1998 through 1999	1998 through 1999
	PR3	1997 through 1999	1997 through 1999	1997 through 1999
	PR2	1997 through 1998	1997 through 1999	1997 through 1999
	PR1	1997 through 2004	1997 through 1999	1997 through 1999
	LY	1998 through 2004	1998 through 1999	1998 through 1999
	WO3	1999	1999	1999
	WO2	1998	1998	1998
	WO1	1999 through 2004	1999	ES
	SH	1997 through 2003	1997 through 1999	1997 through 1999
	DBMF	2002 through 2004	N/A	ES
	DBSF	2002 through 2004	N/A	ES
	DB3	2003 through 2004	N/A	ES
	DB2	1997 through 2004	1997 through 1999	1997 through 1999
	DB1	2000, 2001, 2003, 2004	N/A	ES
Madison				
	MA8	1998, 1999, 2000, 2001, 2002, 2004	1997 through 1999	1997 through 1999
	MA7	1998, 1999, 2002, 2003	1998, 1999	1998, 1999
	MA6	1997 through 2004	1997 through 1999	1997 through 1999
	MA5	1997 through 2004	1997 through 1999	1997 through 1999
	MA4	1997, 1998, 1999, 2000, 2002, 2003, 2004	1997 through 1999	1997 through 1999
	MA3	1997 through 1999	1997 through 1999	1997 through 1999
	MA2	1998 through 1999	1998, 1999	1998, 1999
	MA1	1998 through 1999	1998, 1999	1998, 1999
	WFMA2	1998	1998	1998
	WFMA1	1997 through 2000	1997 through 1999	1997 through 1999
	SQ	1997 through 1998	1997 through 1999	1997 through 1999
Rock Creek				
	RO5	2000, 2002, 2003, 2004	2000	ES
	RO4	1998, 1999	1998, 1999	1998, 1999
	RO3	1999 through 2004	1999	1999
	RO2	1998, 1999, 2000, 2002, 2003, 2004	1998, 1999	1998, 1999
	RO1	1998, 1999, 2000, 2002, 2003, 2004	1998, 1999	1998, 1999
	EFRO	1998, 1999, 2000, 2002, 2003, 2004	1998 through 2000	1998, 1999
	MFRO	2000, 2002, 2003, 2004	2000	ES
	WFRO	1998, 1999, 2000, 2002, 2003, 2004	1998, 1999	1998, 1999
	HO	1999, 2000, 2003	1999	1999

	ROSS	1998	1998	1998
	ST	1998, 1999, 2000, 2003, 2004	1998, 1999	1998, 1999
	WI	1999, 2000, 2002, 2003, 2004	1999 through 2000	1999
Blackfoot				
	BL3	1998, 2000, 2002	1998	ES
	BL2	1998, 1999, 2000, 2002, 2003, 2004	1998	ES
	BL1	1998	1998	ES
	NF2	1998	1998	ES
	NF1	1998	1998	ES
	KL	1998, 1999, 2003	1998	ES
	WA	1998, 1999, 2000, 2003	1998	ES
	CH	1998, 1999, 2000, 2002, 2004	1998	ES
	CO	1998, 1999, 2002	1998	ES

APPENDIX B

STREAM IMPAIRMENT CAUSES AND SOURCES

Probable impairment causes and sources/sub-sources for Montana streams listed as having beneficial uses partially or fully impaired (MTDEQ 2004). The potential beneficial uses of a waterbody are the ability to support agricultural, industrial, recreational, drinking water, aquatic life and fisheries (cold water) but not all streams are expected to support all beneficial uses. Therefore, one or all beneficial uses may have been evaluated for a waterbody depending upon what the expected uses are for that stream; if a beneficial use was not assessed for a stream it is noted.

Drainage	Stream	Section	Impaired Use	Impairment Cause	**Impairment Source/**Sub-source
Missouri	Little Prickly Pear	Upper (North and South Forks to Clark Creek)	Aquatic life Cold water fisheries	Flow alteration Other habitat alterations Siltation Thermal modifications	**Agriculture** Grazing related sources **Silviculture** Logging road construction/maintenance **Hydromodification** **Habitat Modification** (other than Hydromodification) Removal of riparian vegetation
	Little Prickly Pear	Lower (Clark Creek to the Missouri River confluence)	Aquatic life Cold water fisheries	Flow alteration Other habitat alterations Thermal modifications	**Construction** Highway/Road/Bridge construction **Hydromodification** Channelization Bridge construction **Habitat Modification** (other than Hydromodification) Removal of riparian vegetation
	Dearborn River	Falls Creek to confluence Missouri River	Recreational Aquatic life Cold water fisheries	Flow alteration Siltation Thermal modifications	**Agriculture** Grazing related sources **Hydromodification** Flow regulation/modification
	Middle Fork Dearborn River	Headwaters to confluence Dearborn River	None	None	None
	South Fork Dearborn River	Headwaters to confluence Dearborn River	Aquatic life Cold water fisheries	Dewatering Flow alteration Siltation	**Agriculture** Grazing related sources

Madison	Madison River	Quake Lake to Ennis Lake	None	None	None
	West Fork Madison	Headwaters to confluence Madison River	Aquatic life Cold water fisheries	Arsenic Bank erosion Cadmium Dewatering Fish habitat degradation Flow alteration Lead Metals Other habitat alterations Riparian degradation Thermal modifications	**Agriculture** Crop-related Sources Grazing related Sources **Silviculture** Harvesting	Restoration	Residue Management Logging Road Construction/Maintenance **Hydromodification** Flow Regulation/Modification Highway Maintenance and Runoff
Rock Creek	Rock Creek	Mainstem headwaters to confluence Clark Fork	None	None	None
	East Fork Rock Creek	From mouth to 14 km above	None (support of aquatic life and cold water fisheries not assessed)	None	None
	West Fork Rock Creek	Headwaters to confluence Rock Creek	Drinking water (support of aquatic life and cold water fisheries not assessed)	Mercury Metals	Source Unknown
	Willow Creek	Headwaters to confluence Rock Creek	Recreation Aquatic life Cold water fisheries (use as a drinking water supply was not assessed)	Flow alteration Other habitat alterations	**Agriculture** Crop-related Sources Grazing related Sources

Blackfoot	Blackfoot River	Headwaters to Landers Fork	Agriculture Aquatic life Cold water fisheries Drinking water	Metals Other habitat alterations	**Silviculture** **Resource Extraction** Acid Mine Drainage Abandoned mining **Habitat Modification** (other than Hydromodification) Bank or Shoreline Modification/Destabilization
	Blackfoot River	Landers Fork to Nevada Creek	Aquatic life Cold water fisheries	Siltation Other habitat alterations	**Agriculture** **Silviculture**
	Blackfoot River	Nevada Creek to Monture Creek	Aquatic life Cold water fisheries	Nutrients Thermal modifications	**Agriculture** Crop-related Sources
	Blackfoot River	Monture Creek to Belmont Creek	Aquatic life Cold water fisheries	Nutrients Thermal modifications	**Agriculture** **Silviculture** **Hydromodification** Flow Regulation/Modification
	Blackfoot River	Belmont Creek to Clark Fork confluence	Aquatic life Cold water fisheries	Unionized Ammonia	Source Unknown
	Cottonwood Creek	From mouth to 16.1 km above	None	None	None
	Chamberlain Creek	From mouth to 4.3 km above	None	None	None
	Warren Creek	Headwaters to confluence Blackfoot River	Aquatic life Cold water fisheries Recreation	Flow alteration Other habitat alterations	**Agriculture** Crop-related Sources Grazing related Sources **Hydromodification** Channelization
	North Fork Blackfoot River	From mouth to 29 km upstream	None	None	None

Klienschmidt Creek	From mouth to 2.4 km above	Aquatic life Cold water fisheries	Copper Fish habitat degradation Metals Other habitat alterations Riparian degradation Thermal modifications	**Agriculture** Grazing related Sources **Hydromodification** Dam Construction **Habitat Modification** (other than Hydromodification) Bank or Shoreline Modification/Destabilization

APPENDIX C

LAND COVER CLASSIFICATION

1992 National Land Cover Database land use class description (USGS 2000a; USGS 2000b). Land use classification column indicates how land use types were classified within this study. N/A indicates that the land use class was not included in this study.

Land Use Class Number	Land Use Class Description	Land Use Classification
11	Open Water	N/A
12	Perennial Ice/Snow	N/A
21	Low Intensity Residential	Residential
22	High Intensity Residential	Residential
23	Commercial/Industrial/Transportation	N/A
31	Bare Rock/Sand/Clay	N/A
32	Quarries/Strip Mines/Gravel Pits	N/A
33	Transitional	Disturbance
41	Deciduous Forest	N/A
42	Evergreen Forest	Conifer Forest
43	Mixed Forest	N/A
51	Shrubland	N/A
61	Orchards/Vineyards/Other	N/A
71	Grasslands/Herbaceous	N/A
81	Pasture/Hay	Agriculture
82	Row Crops	Agriculture
83	Small Grains	Agriculture
84	Fallow	Agriculture
85	Urban/Recreational Grasses	Agriculture
91	Woody Wetlands	N/A
92	Emergent Herbaceous Wetlands	N/A

APPENDIX D

MISSOURI RIVER DRAINAGE – LAND USE BY SITE

Land use within the entire upstream subbasin and riparian zone at each site within the Missouri River drainage.

Land Use	PR6	PR5	PR4	PR3	PR2	PR1	LY	WO3	WO2	WO1	SH	DBMF
Subbasin												
Proportion Conifer Forest	0.61	0.54	0.46	0.44	0.40	0.44	0.67	0.67	0.63	0.58	0.68	0.42
Proportion Agriculture	0.02	0.03	0.04	0.04	0.04	0.03	0	0	0	0	1E-05	7.17E-03
Proportion Disturbance	3.77E-03	3.28E-03	2.76E-03	2.60E-03	2.14E-03	1.71E-03	0	0	0	0	0	0
Proportion Residential	0	0	0	0	4.3E-05	5.5E-05	0	0	0	0	1E-05	0
Road Density	0.72	0.75	0.80	0.80	0.80	0.72	0.15	0.70	0.56	0.56	0.07	0.60
Mine Density	1.8E-07	1E-07	8.7E-08	8.2E-08	6.7E-08	5.9E-08	0	0	1E-08	1E-08	0	0
Riparian												
Proportion Conifer Forest	0.51	0.51	0.45	0.44	0.40	0.44	0.68	0.79	0.79	0.72	0.73	0.35
Proportion Agriculture	0.10	0.07	0.08	0.08	0.10	0.08	0	0	0	0	0	0.07
Proportion Disturbance	0	0	0	0	0	0	0	0	0	0	0	0
Proportion Residential	0	0	0	0	1.74E-04	3.05E-04	0	0	0	0	0	0
Road Density	2.22	2.16	2.14	2.17	2.13	2.13	0.64	2.82	2.54	2.58	0.18	1.76
Mine Density	2.4E-07	1.2E-07	1E-07	9.6E-08	8E-08	6.4E-08	0	0	1E-07	1E-07	0	0

Land Use	DBSF	DB3	DB2	DB1
<u>Subbasin</u>				
Proportion Conifer Forest	0.62	0.61	0.45	0.32
Proportion Agriculture	0.01	0	0.01	0.04
Proportion Disturbance	0	0	0	0
Proportion Residential	0	0	0	2E-06
Road Density	0.17	0.13	0.30	0.37
Mine Density	0	0	2E-09	2E-09
<u>Riparian</u>				
Proportion Conifer Forest	0.53	0.55	0.38	0.30
Proportion Agriculture	4.31E-03	0	0.04	0.11
Proportion Disturbance	0	0	0	0
Proportion Residential	0	0	0	2E-05
Road Density	1.40	0.45	0.70	0.70
Mine Density	0	0	2E-08	2E-08

APPENDIX E

MISSOURI RIVER DRAINAGE – LAND USE BY REACH

Contributing land use between the upper and lower site in each reach at the subbasin and riparian scale within the Missouri River drainage. Reaches are identified by the lowermost site in the reach.

LAND USE	**PR1**	**LY**	**WO1**	**SH**	**DBMF**	**DBSF**	**DB3**	**DB2**
<u>Subbasin</u>								
Proportion Conifer Forest	0.36	0.67	0.51	0.68	0.42	0.62	0.61	0.45
Proportion Agriculture	0.03	0	0	1E-05	7.17E-03	0.01	0	0.01
Proportion Disturbance	1.25E-03	0	0	0	0	0	0	0
Proportion Residential	7.9E-05	0	0	1E-05	0	0	0	0
Road Density	0.79	0.15	0.45	0.07	0.60	0.17	0.13	0.30
Mine Density	2.9E-08	0	2E-08	0	0	0	0	2E-09
<u>Riparian</u>								
Proportion Conifer Forest	0.40	0.68	0.67	0.73	0.35	0.53	0.55	0.38
Proportion Agriculture	0.08	0	0	0	0.07	4.31E-03	0	0.04
Proportion Disturbance	0	0	0	0	0	0	0	0
Proportion Residential	4.36E-04	0	0	0	0	0	0	0
Road Density	2.25	0.64	2.44	0.18	1.76	1.40	0.45	0.70
Mine Density	1.3E-08	0	2E-07	0	0	0	0	2E-08

APPENDIX F

MISSOURI RIVER DRAINAGE – BIOASSESSMENT METRIC MEANS BY SITE

Bioassessment metric means (fall 1998 data) at sites where invertebrates were collected within the Missouri River drainage. Data are means ± 1 standard error (± SE). n = 6 for oligochaete, tubificid, *T. tubifex*, and *L. hoffmeisteri* metrics; n = 4 for all other metrics.

Metric	**PR6**	**PR5**	**PR4**	**PR3**	**PR2**	**PR1**	**LY**
Total Taxa Richness	24.00 (± 1.96)	18.25 (± 2.95)	16.25 (± 2.69)	21.75 (± 1.25)	22.25 (± 1.38)	23.50 (± 2.22)	20.25 (± 1.11)
Ephemeroptera Taxa Richness	4.00 (± 0.00)	2.75 (± 0.63)	3.50 (± 0.50)	4.25 (± 0.25)	4.25 (± 0.25)	4.25 (± 0.25)	4.25 (± 0.25)
Plecoptera Taxa Richness	3.75 (± 0.63)	1.75 (± 0.48)	1.50 (± 0.65)	2.25 (± 0.48)	2.50 (± 0.29)	3.50 (± 0.29)	3.50 (± 0.29)
Trichoptera Taxa Richness	5.00 (± 0.71)	3.75 (± 1.11)	4.00 (± 0.41)	4.25 (± 0.48)	4.00 (± 0.58)	3.50 (± 0.65)	4.00 (± 0.71)
EPT Taxa Richness	12.75 (± 1.25)	8.25 (± 1.80)	9.00 (± 1.41)	10.75 (± 0.63)	10.75 (± 0.75)	11.25 (± 0.95)	11.75 (± 0.48)
Proportion Ephemeroptera	0.35 (± 0.13)	0.17 (± 0.06)	0.28 (± 0.05)	0.32 (± 0.06)	0.17 (± 0.03)	0.33 (± 0.05)	0.32 (± 0.05)
Proportion Plecoptera	0.06 (± 0.01)	0.01 (± 0.01)	0.01 (± 0.003)	0.02 (± 0.004)	0.01 (± 0.003)	0.01 (± 0.003)	0.05 (± 0.02)
Proportion EPT	0.45 (± 0.13)	0.22 (± 0.07)	0.40 (± 0.06)	0.41 (± 0.07)	0.43 (± 0.01)	0.42 (± 0.07)	0.51 (± 0.02)
Proportion Dominant	0.55 (± 0.08)	0.70 (± 0.07)	0.52 (± 0.07)	0.36 (± 0.04)	0.24 (± 0.02)	0.33 (± 0.06)	0.30 (± 0.04)
Shannon Diversity	1.72 (± 0.20)	1.15 (± 0.21)	1.54 (± 0.13)	1.99 (± 0.08)	2.22 (± 0.06)	2.09 (± 0.10)	2.08 (± 0.10)
Density Oligochaetes	1.02E+04 (±3.05E+03)	1.30E+04 (±3.86E+03)	5.09E+04 (±2.81E+04)	1.57E+05 (±2.77E+04)	1.04E+05 (±1.35E+04)	1.33E+05 (±6.29E+04)	7.90E+03 (±1.97E+03)
Density Tubificids	6.74E+02 (±4.58E+02)	7.80E+03 (±3.11E+03)	4.74E+04 (±2.73E+04)	9.39E+04 (±3.21E+04)	6.78E+04 (±1.58E+04)	1.92E+03 (±1.11E+03)	1.54E+03 (±1.32E+03)
Density *T. tubifex*	0.00 (± 0.0)	0.00 (± 0.0)	289.02 (± 197.44)	0.00 (± 0.0)	480.58 (± 377.19)	384.47 (± 192.23)	0.00 (± 0.0)
Density *L. hoffmeisteri*	0.00 (± 0.0)	0.00 (± 0.0)	0.00 (± 0.0)	0.00 (± 0.0)	768.93 (± 580.60)	288.35 (±196.98)	0.00 (± 0.0)

Metric	WO2	SH	DB2
Total Taxa Richness	27.50 (± 1.26)	21.25 (± 1.80)	18.00 (± 1.73)
Ephemeroptera Taxa Richness	4.25 (± 0.25)	4.50 (± 0.65)	4.25 (± 0.48)
Plecoptera Taxa Richness	4.00 (± 0.71)	3.50 (± 0.29)	2.75 (± 0.63)
Trichoptera Taxa Richness	4.50 (± 0.50)	2.50 (± 0.65)	3.50 (± 0.50)
EPT Taxa Richness	12.75 (± 0.85)	10.50 (± 1.19)	10.50 (± 0.96)
Proportion Ephemeroptera	0.05 (± 0.02)	0.19 (± 0.05)	0.12 (± 0.02)
Proportion Plecoptera	0.01 (± 0.01)	0.01 (± 0.003)	0.02 (± 0.003)
Proportion EPT	0.09 (± 0.02)	0.28 (± 0.11)	0.24 (± 0.03)
Proportion Dominant	0.57 (± 0.04)	0.49 (± 0.12)	0.28 (± 0.02)
Shannon Diversity	1.49 (± 0.09)	1.63 (± 0.30)	2.03 (± 0.07)
Density Oligochaetes	3.49E+04 (±9.87E+03)	1.68E+04 (±7.95E+03)	2.04E+04 (±9.10E+03)
Density Tubificids	2.31E+03 (±7.90E+02)	1.02E+04 (±4.94E+03)	2.21E+03 (±1.00E+03)
Density *T. tubifex*	0.00 (± 0.0)	0.00 (± 0.0)	0.00 (± 0.0)
Density *L. hoffmeisteri*	96.34 (± 96.34)	96.12 (± 96.12)	0.00 (± 0.0)

APPENDIX G

MISSOURI RIVER DRAINAGE – BIOASSESSMENT METRIC MEANS BY REACH

Bioassessment metric means (fall 1998 data) for each reach where invertebrates were collected within the Missouri River drainage. Data are means ± 1 standard error (± SE). Reaches are identified by the lowermost site in the reach. n = 6 for oligochaete, tubificid, *T. tubifex,* and *L. hoffmeisteri* density metrics; n = 4 for all other metrics for all reaches except reach PR1 (n = 36 for oligochaete, tubificid, *T. tubifex*, and *L. hoffmeisteri* density metrics; n = 24 for all other metrics at this site).

Metric	**PR1**	**LY**	**WO2**	**SH**	**DB2**
Total Taxa Richness	21.00 (± 0.98)	20.25 (± 1.11)	27.50 (± 1.26)	21.25 (± 1.80)	18.00 (± 1.73)
Ephemeroptera Taxa Richness	3.83 (± 0.18)	4.25 (± 0.25)	4.25 (± 0.25)	4.50 (± 0.65)	4.25 (± 0.48)
Plecoptera Taxa Richness	2.54 (± 0.25)	3.50 (± 0.29)	4.00 (± 0.71)	3.50 (± 0.29)	2.75 (± 0.63)
Trichoptera Taxa Richness	4.08 (± 0.27)	4.00 (± 0.71)	4.50 (± 0.50)	2.50 (± 0.65)	3.50 (± 0.50)
EPT Taxa Richness	10.46 (± 0.53)	11.75 (± 0.48)	12.75 (± 0.85)	10.50 (± 1.19)	10.50 (± 0.96)
Proportion Ephemeroptera	0.27 (± 0.03)	0.32 (± 0.05)	0.05 (± 0.02)	0.19 (± 0.05)	0.12 (± 0.02)
Proportion Plecoptera	0.02 (±0.004)	0.05 (± 0.02)	0.01 (± 0.01)	0.01 (± 0.003)	0.02 (± 0.003)
Proportion EPT	0.39 (± 0.03)	0.51 (± 0.02)	0.09 (± 0.02)	0.28 (± 0.11)	0.24 (± 0.03)
Proportion Dominant	0.45 (± 0.04)	0.30 (± 0.04)	0.57 (± 0.04)	0.49 (± 0.12)	0.28 (± 0.02)
Shannon Diversity	1.78 (±0.09)	2.08 (± 0.10)	1.49 (± 0.09)	1.63 (± 0.30)	2.03 (± 0.07)
Density Oligochaetes	7.80E+04 (± 1.51E+04)	7.90E+03 (±1.97E+03)	3.49E+04 (±9.87E+03)	1.68E+04 (±7.95E+03)	2.04E+04 (±9.10E+03)
Density Tubificids	3.66E+04 (± 9.23E+03)	1.54E+03 (±1.32E+03)	2.31E+03 (±7.90E+02)	1.02E+04 (±4.94E+03)	2.21E+03 (±1.00E+03)
Density *T. tubifex*	192.34 (± 79.62)	0.00 (± 0.0)	0.00 (± 0.0)	0.00 (± 0.0)	0.00 (± 0.0)
Density *L. hoffmeisteri*	176.21 (± 96.99)	0.00 (± 0.0)	96.34 (± 96.34)	96.12 (± 96.12)	0.00 (± 0.0)

APPENDIX H

MISSOURI RIVER DRAINAGE – PHYSICOCHEMICAL MEANS BY SITE

Physicochemical means at each site within the Missouri River drainage where data were collected. Discharge, temperature, pH, and dissolved oxygen were quantified from summer 1997 and summer 1998 data. The proportion organic matter in sediment and conductivity were quantified from data collected in the summer 1997, fall 1997 and 1998, and spring 1998 and 1999. Elevation and slope were determined from geospatial data. Data are means ± 1 standard error (± SE); n = sample number.

Physicochemical Parameter	**PR6**	**PR5**	**PR4**	**PR3**	**PR2**	**PR1**	**LY**	**WO2**	**SH**	**DB2**
Discharge (cms)	0.06	0.10	0.82	2.14	2.35	2.14	0.38	0.17	1.23	14.87
	N/A	N/A	N/A	(± 1.43)	(± 1.69)	(± 1.43)	N/A	N/A	(± 0.57)	(± 2.29)
	n = 1	n = 1	n = 1	n = 2	n = 2	n = 2	n = 1	n = 1	n = 2	n = 2
Proportion Organic Organic Matter	0.04	0.04	0.04	0.07	0.09	0.06	0.02	0.02	0.03	0.03
	(± 0.01)	(± 0.01)	(± 0.02)	(± 0.04)	(± 0.01)	(± 0.01)	(± 0.003)	(± 0.001)	(± 0.003)	(± 0.005)
	n = 4	n = 4	n = 4	n = 8	n = 12	n = 12	n = 4	n = 4	n = 12	n = 12
Temperature (°C)	7.73	8.49	9.64	11.36	14.86	14.04	9.51	11.13	15.03	12.23
	(± 0.01)	(± 0.03)	(± 0.03)	(± 1.18)	(± 2.70)	(± 2.94)	(± 0.02)	(± 0.20)	(± 2.72)	(± 1.20)
	n = 2	n = 2	n = 2	n = 3	n = 4	n = 3	n = 2	n = 2	n = 4	n = 4
Conductivity (mS/cm)	0.22	0.26	0.32	0.31	0.29	0.26	0.15	0.28	0.12	0.26
	(± 0.03)	(± 0.04)	(± 0.05)	(± 0.05)	(± 0.03)	(± 0.02)	(± 0.03)	(± 0.02)	(± 0.01)	(± 0.02)
	n = 3	n = 3	n = 3	n = 3	n = 5	n = 4	n = 3	n = 3	n = 5	n = 5
pH	7.87	7.56	8.15	8.32	8.31	8.40	8.15	7.96	7.89	8.27
	(± 0.03)	(± 0.04)	(± 0.02)	(± 0.06)	(± 0.07)	(± 0.16)	(± 0.06)	(± 0.01)	(± 0.08)	(± 0.13)
	n = 2	n = 2	n = 2	n = 3	n = 4	n = 3	n = 2	n = 2	n = 4	n = 4
Dissolved Oxygen (mg/l)	10.36	9.89	10.81	10.23	9.99	10.18	10.84	10.43	9.02	10.16
	(± 0.11)	(± 0.20)	(± 0.08)	(± 0.41)	(± 0.91)	(± 0.45)	(± 0.03)	(± 0.00)	(± 0.55)	(± 0.38)
	n = 2	n = 2	n = 2	n = 3	n = 4	n = 3	n = 2	n = 2	n = 4	n = 4
Elevation (m)	1318	1284	1268	1234	1165	1097	1161	1147	1040	1159
Slope	0.007	0.007	0.007	0.007	0.005	0.004	0.017	0.010	0.012	0.005

APPENDIX I

MISSOURI RIVER DRAINAGE – PHYSICOCHEMICAL MEANS BY REACH

Physicochemical means for each reach within the Missouri River drainage. Reaches are identified by the lowermost site in the reach. Discharge, temperature, pH, and dissolved oxygen were quantified from summer 1997 and summer 1998 data. The proportion organic matter in sediment and conductivity were quantified from data collected in the summer 1997, fall 1997 and 1998, and spring 1998 and 1999. Elevation and slope were determined from geospatial data. Data are means ± 1 standard error (± SE); n = sample number.

Physicochemical Parameter	**PR1**	**LY**	**WO1**	**SH**	**DB2**
Discharge (cms)	1.56	0.38	0.17	1.23	14.87
	(± 0.53)	N/A	N/A	(± 0.57)	(± 2.29)
	n = 9	n = 1	n = 1	n = 2	n = 2
Proportion Organic	0.07	0.02	0.02	0.03	0.03
Organic Matter	(± 0.01)	(± 0.003)	(± 0.001)	(± 0.003)	(± 0.005)
	n = 44	n = 4	n = 4	n = 12	n = 12
Temperature (°C)	11.71	9.51	11.13	15.03	12.23
	(± 1.05)	(± 0.02)	(± 0.20)	(± 2.72)	(± 1.20)
	n = 16	n = 2	n = 2	n = 4	n = 4
Conductivity (mS/cm)	0.28	0.15	0.28	0.12	0.26
	(± 0.01)	(± 0.03)	(± 0.02)	(± 0.01)	(± 0.02)
	n = 21	n = 3	n = 3	n = 5	n = 5
pH	8.16	8.15	7.96	7.89	8.27
	(± 0.08)	(± 0.06)	(± 0.01)	(± 0.08)	(± 0.13)
	n = 16	n = 2	n = 2	n = 4	n = 4
Dissolved Oxygen (mg/l)	10.21	10.84	10.43	9.02	10.16
	(± 0.24)	(± 0.03)	(± 0.00)	(± 0.55)	(± 0.38)
	n = 16	n = 2	n = 2	n = 4	n = 4
Elevation (m)	1097	1161	1147	1040	1159
Slope	0.004	0.017	0.010	0.012	0.005

APPENDIX J

MADISON RIVER DRAINAGE – LAND USE BY SITE

Land use within the entire upstream subbasin and riparian zone at each site within the Madison River drainage.

Land Use	**MA8**	**MA7**	**MA6**	**MA5**	**MA4**	**MA3**	**MA2**	**MA1**	**WFMA2**	**WFMA1**	**SQ**
Subbasin											
Proportion Conifer Forest	0.48	0.48	0.47	0.47	0.47	0.47	0.46	0.46	0.43	0.46	0.48
Proportion Agriculture	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	5.20E-04	4.70E-04	2.08E-03
Proportion Disturbance	0.33	0.33	0.32	0.31	0.26	0.24	0.22	0.20	0.01	0.01	8.40E-04
Proportion Residential	3.25E-04	3.19E-04	3.09E-04	3.08E-04	2.54E-04	2.33E-04	2.18E-04	1.96E-04	0	0	5.7E-05
Road Density	0.29	0.29	0.30	0.30	0.30	0.30	0.31	0.31	0.28	0.30	0.13
Mine Density	3.7E-10	3.7E-10	3.5E-10	3.5E-10	2.9E-10	2.7E-10	5E-10	8.9E-10	0	0	0
Riparian											
Proportion Conifer Forest	0.37	0.37	0.36	0.36	0.38	0.38	0.37	0.36	0.45	0.48	0.20
Proportion Agriculture	0.01	0.01	0.01	0.01	0.01	0.01	9.28E-03	0.01	0	0	0.05
Proportion Disturbance	0.25	0.24	0.24	0.23	0.19	0.17	0.16	0.15	0	0	0
Proportion Residential	6.1E-05	6E-05	5.9E-05	5.8E-05	4.8E-05	4.3E-05	4.1E-05	3.7E-05	0	0	0
Road Density	1.27	1.28	1.27	1.30	1.26	1.20	1.18	1.14	0.94	0.98	0.87
Mine Density	0	0	0	0	0	0	4E-09	4E-09	0	0	0

APPENDIX K

MADISON RIVER DRAINAGE – LAND USE BY REACH

Contributing land use between the upper and lower sites in each reach at the subbasin and riparian scale within the Madison River drainage. Reaches are identified by the lowermost site in the reach.

Land Use	MA5	MA1	WFMA1	SQ
Subbasin				
Proportion Conifer Forest	0.28	0.40	0.72	0.48
Proportion Agriculture	0.01	0.05	0	2.08E-03
Proportion Disturbance	8.0E-05	1.50E-04	0.03	8.40E-04
Proportion Residential	0	1.10E-05	0	5.7E-05
Road Density	0.61	0.32	0.52	0.13
Mine Density	0	4.07E-09	0	0
Riparian				
Proportion Conifer Forest	0.04	0.24	0.82	0.20
Proportion Agriculture	3.13E-03	0.06	0	0.05
Proportion Disturbance	0	0	0	0
Proportion Residential	0	0	0	0
Road Density	2.01	0.82	1.41	0.87
Mine Density	0	0	0	0

APPENDIX L

MADISON RIVER DRAINAGE – BIOASSESSMENT METRIC MEANS BY SITE

Bioassessment metric means (fall 1998 data) at sites where invertebrates were collected within the Madison River drainage. Data are means ± 1 standard error (± SE). n = 6 for oligochaete, tubificid, *T. tubifex*, and *L. hoffmeisteri* density metrics; n = 2 for all other metrics.

Metric	**MA8**	**MA7**	**MA6**	**MA5**	**MA4**	**MA3**	**MA2**
Total Taxa Richness	24.50 (± 0.50)	26.50 (± 1.50)	23.50 (± 0.50)	25.50 (± 2.50)	22.00 (± 1.00)	23.00 (± 3.00)	25.50 (± 2.50)
Ephemeroptera Taxa Richness	3.00 (± 0.00)	3.00 (± 0.00)	3.00 (± 0.00)	3.00 (± 0.00)	3.50 (± 0.50)	3.00 (± 0.00)	3.00 (± 0.00)
Plecoptera Taxa Richness	2.50 (± 1.50)	2.50 (± 0.50)	4.00 (± 0.00)	3.50 (± 0.50)	2.00 (± 0.00)	3.00 (± 1.00)	4.00 (± 0.00)
Trichoptera Taxa Richness	5.00 (± 0.00)	5.50 (± 0.50)	4.50 (± 0.50)	6.00 (± 1.00)	6.50 (± 0.50)	6.00 (± 1.00)	5.50 (± 0.50)
EPT Taxa Richness	10.50 (± 1.50)	11.00 (± 1.00)	11.50 (± 0.50)	12.50 (± 1.50)	12.00 (± 1.00)	12.00 (± 0.00)	12.50 (± 0.50)
Proportion Ephemeroptera	0.25 (± 0.06)	0.13 (± 0.04)	0.12 (± 0.01)	0.13 (± 0.01)	0.08 (± 0.00)	0.16 (± 0.01)	0.17 (± 0.02)
Proportion Plecoptera	0.01 (± 0.01)	0.04 (± 0.01)	0.03 (± 0.00)	0.03 (± 0.00)	0.02 (± 0.01)	0.05 (± 0.01)	0.04 (± 0.00)
Proportion EPT	0.57 (± 0.16)	0.55 (± 0.06)	0.44 (± 0.01)	0.45 (± 0.01)	0.30 (± 0.07)	0.42 (± 0.01)	0.51 (± 0.05)
Proportion Dominant	0.32 (± 0.06)	0.20 (± 0.03)	0.36 (± 0.01)	0.36 (± 0.07)	0.55 (± 0.05)	0.36 (± 0.07)	0.20 (± 0.04)
Shannon Diversity	2.16 (± 0.07)	2.52 (± 0.07)	2.11 (± 0.00)	2.14 (± 0.28)	1.78 (± 0.11)	2.17 (± 0.05)	2.51 (± 0.14)
Density Oligochaetes	1.37E+05 (± 1.43E+04)	1.92E+04 (± 7435.74)	2.10E+04 (± 4993.64)	1.15E+04 (± 3134.06)	1.98E+04 (± 9259.20)	3171.86 (±1121.73)	5.83E+04 (± 2.84E+04)
Density Tubificids	3.29E+04 (±4875.31)	9803.92 (±3807.98)	1.99E+04 (±5472.59)	5094.19 (±1231.65)	6920.42 (±4193.16)	1730.10 (±631.74)	4.16E+04 (±2.25E+04)
Density *T. tubifex*	0.00 (± 0.00)	0.00 (± 0.00)	0.00 (± 0.00)	0.00 (± 0.00)	96.12 (± 96.12)	0.00 (± 0.00)	0.00 (± 0.00)
Density *L. hoffmeisteri*	0.00 (± 0.00)	0.00 (± 0.00)	0.00 (± 0.00)	192.23 (± 192.23)	0.00 (± 0.00)	0.00 (± 0.00)	0.00 (± 0.00)

Metric	MA1	WFMA2	WFMA1	SQ
Total Taxa Richness	19.50 (± 2.50)	21.50 (± 0.50)	21.00 (± 2.00)	18.50 (± 1.50)
Ephemeroptera Taxa Richness	3.00 (± 0.00)	5.00 (± 0.00)	3.00 (± 1.00)	3.50 (± 0.50)
Plecoptera Taxa Richness	3.00 (± 0.00)	3.00 (± 1.00)	1.00 (± 0.00)	4.50 (± 0.50)
Trichoptera Taxa Richness	5.50 (± 0.50)	5.50 (± 0.50)	6.00 (± 0.00)	3.00 (± 1.00)
EPT Taxa Richness	11.50 (± 0.50)	13.50 (± 1.50)	10.00 (± 1.00)	11.00 (± 0.00)
Proportion Ephemeroptera	0.14 (± 0.06)	0.37 (± 0.11)	0.03 (± 0.00)	0.22 (± 0.01)
Proportion Plecoptera	0.02 (± 0.00)	0.02 (± 0.01)	0.00 (± 0.00)	0.23 (± 0.07)
Proportion EPT	0.49 (± 0.01)	0.67 (± 0.10)	0.58 (± 0.03)	0.50 (± 0.07)
Proportion Dominant	0.39 (± 0.01)	0.37 (± 0.01)	0.31 (± 0.03)	0.29 (± 0.09)
Shannon Diversity	1.93 (± 0.00)	2.00 (± 0.11)	1.98 (± 0.01)	2.28 (± 0.06)
Density Oligochaetes	1.81E+04 (±5903.17)	3.95E+04 (±3.07E+04)	5.38E+04 (± 2.03E+04)	1.57E+04 (±6777.02)
Density Tubificids	8938.87 (±6581.73)	96.12 (± 96.12)	3.29E+04 (± 2.23E+04)	96.12 (± 96.12)
Density *T. tubifex*	1345.64 (±1345.64)	0.00 (± 0.00)	0.00 (± 0.00)	96.12 (± 96.12)
Density *L. hoffmeisteri*	768.94 (± 570.26)	0.00 (± 0.00)	96.12 (± 96.12)	0.00 (± 0.00)

APPENDIX M

MADISON RIVER DRAINAGE – BIOASSESSMENT METRIC MEANS BY REACH

Bioassessment metric means (fall 1998 data) for each reach where invertebrates were collected within the Madison River drainage. Data are means ± 1 standard error (± SE). Reaches are identified by the lowermost site in the reach. n = 6, 12, 12 and 24 for oligochaete, tubificid, *T. tubifex*, and *L. hoffmeisteri* density metrics at sites SQ, MA1, WFMA1, and MA5, respectively. n = 2, 4, 4, and 8 for all other metrics at sites SQ, MA1, WFMA1, and MA4, respectively.

Metric	**MA5**	**MA1**	**WFMA1**	**SQ**
Total Taxa Richness	25.00 (± 0.71)	22.50 (± 2.25)	21.25 (± 0.85)	18.50 (± 1.50)
Ephemeroptera Taxa Richness	3.00 (± 0.00)	3.00 (± 0.00)	4.00 (± 0.71)	3.50 (± 0.50)
Plecoptera Taxa Richness	3.13 (± 0.40)	3.50 (± 0.29)	2.00 (± 0.71)	4.50 (± 0.50)
Trichoptera Taxa Richness	5.25 (± 0.32)	5.50 (± 0.29)	5.75 (± 0.25)	3.00 (± 1.00)
EPT Taxa Richness	11.38 (± 0.53)	12.00 (± 0.41)	11.75 (± 1.25)	11.00 (± 0.00)
Proportion Ephemeroptera	0.16 (± 0.02)	0.16 (± 0.03)	0.20 (± 0.11)	0.22 (± 0.01)
Proportion Plecoptera	0.03 (± 0.005)	0.03 (± 0.005)	0.01 (± 0.01)	0.23 (± 0.07)
Proportion EPT	0.50 (± 0.04)	0.50 (± 0.02)	0.63 (± 0.05)	0.50 (± 0.07)
Proportion Dominant	0.31 (± 0.03)	0.29 (± 0.06)	0.34 (± 0.02)	0.29 (± 0.09)
Shannon Diversity	2.23 (± 0.08)	2.22 (± 0.18)	1.99 (± 0.04)	2.28 (± 0.06)
Density Oligochaetes	4.73E+04 (± 11589.00)	3.82E+04 (± 15102.42)	4.67E+04 (± 17687.52)	1.57E+04 (±6777.02)
Density Tubificids	1.70E+04 (± 2953.42)	2.53E+04 (± 1.22E+04)	1.65E+04 (± 1.17E+04)	96.12 (± 96.12)
Density *T. tubifex*	0.00 (± 0.00)	672.82 (± 672.82)	0.00 (± 0.00)	96.12 (± 96.12)
Density *L. hoffmeisteri*	48.05 (± 48.05)	384.47 (± 295.54)	48.06 (± 48.06)	0.00 (± 0.00)

APPENDIX N

MADISON RIVER DRAINAGE – PHYSICOCHEMICAL MEANS BY SITE

Physicochemical means at each site within the Madison River drainage. Discharge, temperature, pH, and dissolved oxygen were quantified from summer 1997 and summer 1998 data. The proportion organic matter in sediment and conductivity were quantified from data collected in the summer 1997 and 1998, fall 1997 and 1998, and spring 1998 and 1999. Elevation and slope were determined from geospatial data. Data are means ± 1 standard error (± SE); n = sample number.

Physicochemical Parameter	**MA8**	**MA7**	**MA6**	**MA5**	**MA4**	**MA3**	**MA2**
Discharge (cms)	1.22	21.15	24.21	4.26	21.14	4.80	20.97
	(± 0.21)	N/A	N/A	(± 0.37)	(± 0.27)	(± 0.11)	N/A
	n = 2	n = 1	n = 1	n = 2	n = 2	n = 2	n = 1
Proportion Organic	0.02	N/A	0.02	0.01	0.03	0.02	0.02
Organic Matter	(±0.002)	N/A	(± 0.004)	(± 0.001)	(± 0.01)	(± 0.01)	(± 0.001)
	n = 12	N/A	n = 12	n = 12	n = 12	n = 11	n = 4
Temperature (°C)	16.10	N/A	17.89	17.56	15.26	16.29	N/A
	(± 0.01)	N/A	(± 0.15)	(± 0.31)	(± 0.80)	(± 0.28)	N/A
	n = 2	N/A	n = 3	n = 4	n = 4	n = 2	N/A
Conductivity (mS/cm)	0.21	0.21	0.23	0.19	0.18	0.17	0.19
	N/A	N/A	(± 0.03)	(± 0.01)	(± 0.01)	(± 0.01)	(± 0.01)
	n = 1	n = 1	n = 4	n = 6	n = 5	n = 3	n = 2
pH	8.45	N/A	8.59	8.71	8.71	8.59	N/A
	(± 0.03)	N/A	(± 0.13)	(± 0.10)	(± 0.05)	(± 0.05)	N/A
	n = 2	N/A	n = 3	n = 4	n = 4	n = 2	N/A
Dissolved Oxygen (mg/l)	8.17	N/A	8.38	8.17	8.50	8.78	N/A
	(± 0.07)	N/A	(± 0.30)	(± 0.15)	(± 0.29)	(± 0.03)	N/A
	n = 2	N/A	n = 3	n = 4	n = 4	n = 2	N/A
Elevation (m)	1891	1867	1815	1792	1780	1724	1659
Slope	0.022	0.006	0.006	0.005	0.005	0.005	0.005

Physicochemical Parameter	**MA1**	**WFMA2**	**WFMA1**	**SQ**
Discharge (cms)	6.79	2.83	2.80	1.15
	N/A	N/A	(± 0.39)	(± 0.37)
	n = 1	n = 1	n = 2	n = 2
Proportion Organic Organic Matter	0.03	0.07	0.05	0.05
	(± 0.01)	(± 0.01)	(± 0.01)	(± 0.01)
	n = 4	n = 4	n = 12	n = 15
Temperature (°C)	N/A	16.92	15.46	9.26
	N/A	(± 0.26)	(± 0.49)	(± 0.73)
	N/A	n = 2	n = 4	n = 2
Conductivity (mS/cm)	0.19	0.20	0.19	0.20
	(± 0.01)	(± 0.03)	(± 0.01)	(± 0.09)
	n = 2	n = 2	n = 5	n = 3
pH	N/A	8.78	8.76	8.11
	N/A	(±0.03)	(± 0.06)	(± 0.04)
	N/A	n = 2	n = 4	n = 2
Dissolved Oxygen (mg/l)	N/A	7.65	8.39	10.26
	N/A	(± 0.10)	(± 0.07)	(± 0.05)
	N/A	n = 2	n = 4	n = 2
Elevation (m)	1572	1838	1793	1777
Slope	0.005	0.013	0.013	0.042

APPENDIX O

MADISON RIVER DRAINAGE – PHYSICOCHEMICAL MEANS BY REACH

Physicochemical means for each reach within the Madison River drainage. Reaches are identified by the lowermost site in the reach. Discharge, temperature, pH, and dissolved oxygen were quantified from summer 1997 and summer 1998 data. The proportion organic matter in sediment and conductivity were quantified from data collected in the summer 1997 and 1998, fall 1997 and 1998, and spring 1998 and 1999. Elevation and slope were determined from geospatial data. Data are means ± 1 standard error (± SE); n = sample number.

Physicochemical Parameter	**MA5**	**MA1**	**WFMA1**	**SQ**
Discharge (cms)	9.39	13.88	2.81	1.15
	(± 4.26)	(± 7.09)	(± 0.22)	(± 0.37)
	n = 6	n = 2	n = 3	n = 2
Proportion Organic Organic Matter	0.02	0.02	0.06	0.05
	(± 0.002)	(± 0.005)	(± 0.006)	(± 0.01)
	n = 36	n = 8	n = 16	n = 15
Temperature (°C)	17.34	N/A	15.94	9.26
	(± 0.27)	N/A	(± 0.44)	(± 0.73)
	n = 9	N/A	n = 6	n = 2
Conductivity (mS/cm)	0.62	0.19	0.19	0.20
	(± 0.41)	(± 0.004)	(± 0.009)	(± 0.09)
	n = 12	n = 4	n = 7	n = 3
pH	8.61	N/A	8.77	8.11
	(± 0.07)	N/A	(± 0.04)	(± 0.04)
	n = 9	N/A	n = 6	n = 2
Dissolved Oxygen (mg/l)	8.24	N/A	8.14	10.26
	(± 0.11)	N/A	(± 0.16)	(± 0.05)
	n = 9	N/A	n = 6	n = 2
Elevation (m)	1792	1572	1793	1777
Slope	0.006	0.005	0.011	0.042

APPENDIX P

ROCK CREEK DRAINAGE – LAND USE BY SITE

Land use within the entire upstream subbasin and riparian zone at each site within the Rock Creek drainage.

Land Use	RO5	RO4	RO3	RO2	RO1	EFRO	MFRO	WFRO	HO	ROSS	ST	WI
Subbasin												
Proportion Conifer Forest	0.72	0.73	0.73	0.74	0.76	0.64	0.85	0.83	0.85	0.94	0.87	0.66
Proportion Agriculture	0.01	0.01	0.01	4.69E-03	3.90E-03	4.10E-03	0	1.11E-03	0	0	0	0.01
Proportion Disturbance	3.40E-04	3.20E-04	3.20E-04	1.36E-03	1.93E-03	2.74E-03	0	0	0	0	0	0
Proportion Residential	0	0	0	0	0	0	0	0	0	0	0	0
Road Density	0.50	0.48	0.48	0.47	0.44	0.48	0.34	0.47	0.01	0.09	0.28	0.62
Mine Density	3.1E-08	3E-08	3E-08	2.6E-08	2.5E-08	5E-09	6E-08	1.3E-08	0	0	8E-08	4E-08
Riparian												
Proportion Conifer Forest	0.50	0.52	0.52	0.55	0.57	0.40	0.81	0.77	0.68	0.93	0.81	0.24
Proportion Agriculture	0.03	0.03	0.03	0.02	0.02	0	0	2.56E-03	0	0	0	0.08
Proportion Disturbance	0	0	0	0	0	0	0	0	0	0	0	0
Proportion Residential	0	0	0	0	0	0	0	0	0	0	0	0
Road Density	1.96	1.85	1.86	1.93	1.90	1.13	2.54	2.84	0.15	0.28	2.58	1.96
Mine Density	9.3E-08	8.8E-08	8.7E-08	7.7E-08	6.5E-08	0	3E-07	1.7E-07	0	0	3E-07	8E-08

APPENDIX Q

ROCK CREEK DRAINAGE – LAND USE BY REACH

Contributing land use between the upper and lower site in each reach at the subbasin and riparian scale within the Rock Creek drainage. Reaches are identified by the lowermost site in the reach.

Land Use	RO1	EFRO	MFRO	WFRO	HO	ROSS	ST	WI
Subbasin								
Proportion Conifer Forest	0.83	0.64	0.85	0.83	0.85	0.94	0.87	0.66
Proportion Agriculture	1.34E-06	4.10E-03	0	1.11E-03	0	0	0	0.01
Proportion Disturbance	5.69E-03	2.74E-03	0	0	0	0	0	0
Proportion Residential	0	0	0	0	0	0	0	0
Road Density	0.31	0.48	0.34	0.47	0.01	0.09	0.28	0.62
Mine Density	1.19E-08	5E-09	6E-08	1.3E-08	0	0	8E-08	4E-08
Riparian								
Proportion Conifer Forest	0.73	0.40	0.81	0.77	0.68	0.93	0.81	0.24
Proportion Agriculture	0	0	0	2.56E-03	0	0	0	0.08
Proportion Disturbance	0	0	0	0	0	0	0	0
Proportion Residential	0	0	0	0	0	0	0	0
Road Density	1.90	1.13	2.54	2.84	0.15	0.28	2.58	1.96
Mine Density	0	0	3E-07	1.7E-07	0	0	3E-07	8E-08

APPENDIX R

ROCK CREEK DRAINAGE – BIOASSESSMENT METRIC MEANS BY SITE

Bioassessment metric means (summer 1998 data) at sites where invertebrates were collected within the Rock Creek drainage. Data are means ± 1 standard error (± SE). n = 6 for oligochaete, tubificid, *T. tubifex*, and *L. hoffmeisteri* density metrics; n = 4 for all other metrics except site EFRO (n=3).

Metric	**RO4**	**RO2**	**RO1**	**EFRO**	**WFRO**	**ROSS**	**ST**
Total Taxa Richness	17.75 (± 1.65)	18.75 (± 1.31)	15.50 (± 0.50)	19.00 (± 2.08)	16.25 (± 1.65)	15.75 (± 1.49)	13.00 (± 1.08)
Ephemeroptera Taxa Richness	3.25 (± 0.25)	4.25 (± 0.48)	4.00 (± 0.41)	4.00 (± 0.58)	3.75 (± 0.48)	4.00 (± 0.41)	4.00 (± 0.41)
Plecoptera Taxa Richness	2.75 (± 0.48)	3.25 (± 0.48)	2.00 (± 0.58)	1.33 (± 0.33)	2.00 (± 0.41)	1.75 (± 0.48)	3.75 (± 0.25)
Trichoptera Taxa Richness	3.00 (± 0.41)	3.75 (± 0.85)	3.50 (± 0.29)	4.00 (± 0.58)	4.00 (± 0.41)	4.25 (± 0.25)	1.75 (± 0.75)
EPT Taxa Richness	9.00 (± 0.71)	11.25 (± 1.25)	9.50 (± 0.29)	9.33 (± 0.88)	9.75 (± 1.03)	10.00 (± 0.71)	9.50 (± 1.04)
Proportion Ephemeroptera	0.36 (± 0.04)	0.37 (± 0.08)	0.48 (± 0.05)	0.39 (± 0.07)	0.48 (± 0.03)	0.17 (± 0.03)	0.71 (± 0.03)
Proportion Plecoptera	0.06 (± 0.01)	0.09 (± 0.03)	0.04 (± 0.01)	0.01 (± 0.005)	0.05 (± 0.01)	0.01 (± 0.01)	0.12 (± 0.03)
Proportion EPT	0.46 (± 0.06)	0.62 (± 0.12)	0.65 (± 0.07)	0.75 (± 0.06)	0.61 (± 0.03)	0.58 (± 0.08)	0.85 (± 0.03)
Proportion Dominant	0.33 (± 0.06)	0.35 (± 0.10)	0.33 (± 0.03)	0.33 (± 0.05)	0.31 (± 0.03)	0.35 (± 0.02)	0.48 (± 0.04)
Shannon Diversity	2.05 (± 0.11)	2.01 (± 0.16)	1.91 (± 0.07)	2.08 (± 0.12)	2.03 (± 0.08)	1.99 (± 0.10)	1.62 (± 0.07)
Density Oligochaetes	1.11E+04 (± 2683.02)	192.23 (± 121.58)	192.23 (± 121.58)	7.54E+04 (± 1.31E+04)	6728.18 (± 4957.99)	768.94 (± 354.46)	480.58 (± 96.12)
Density Tubificids	384.47 (± 285.13)	96.12 (± 96.12)	96.12 (± 96.12)	6.81E+04 (± 1.10E+04)	192.23 (± 121.58)	192.23 (± 121.58)	96.12 (± 96.12)
Density *T. tubifex*	0.00 (± 0.00)	0.00 (± 0.00)	0.00 (± 0.00)	3844.68 (± 3387.90)	0.00 (± 0.00)	0.00 (± 0.00)	0.00 (± 0.00)
Density *L. hoffmeisteri*	0.00 (± 0.00)	96.12 (± 96.12)	0.00 (± 0.00)	1537.87 (± 676.92)	0.00 (± 0.00)	0.00 (± 0.00)	0.00 (± 0.00)

APPENDIX S

ROCK CREEK DRAINAGE – BIOASSESSMENT METRIC MEANS BY REACH

Bioassessment metric means (summer 1998 data) for each reach where invertebrates were collected within the Rock Creek drainage. Data are means ± 1 standard error (± SE). Reaches are identified by the lowermost site in the reach. For all reaches n = 6 for oligochaete, tubificid, *T. tubifex*, and *L. hoffmeisteri* density metrics except reach RO1 (n = 18). n = 4 for all other metrics for all reaches except reach EFRO (n=3) and reach RO1 (n = 12).

Metric	**RO1**	**EFRO**	**WFRO**	**ROSS**	**ST**
Total Taxa Richness	17.33 (± 0.77)	19.00 (± 2.08)	16.25 (± 1.65)	15.75 (± 1.49)	13.00 (± 1.08)
Ephemeroptera Taxa Richness	3.83 (± 0.24)	4.00 (± 0.58)	3.75 (± 0.48)	4.00 (± 0.41)	4.00 (± 0.41)
Plecoptera Taxa Richness	2.67 (± 0.31)	1.33 (± 0.33)	2.00 (± 0.41)	1.75 (± 0.48)	3.75 (± 0.25)
Trichoptera Taxa Richness	3.42 (± 0.31)	4.00 (± 0.58)	4.00 (± 0.41)	4.25 (± 0.25)	1.75 (± 0.75)
EPT Taxa Richness	9.92 (± 0.53)	9.33 (± 0.88)	9.75 (± 1.03)	10.00 (± 0.71)	9.50 (± 1.04)
Proportion Ephemeroptera	0.41 (± 0.04)	0.39 (± 0.07)	0.48 (± 0.03)	0.17 (± 0.03)	0.71 (± 0.03)
Proportion Plecoptera	0.06 (± 0.01)	0.01 (± 0.005)	0.05 (± 0.01)	0.01 (± 0.01)	0.12 (± 0.03)
Proportion EPT	0.58 (± 0.05)	0.75 (± 0.06)	0.61 (± 0.03)	0.58 (± 0.08)	0.85 (± 0.03)
Proportion Dominant	0.34 (± 0.04)	0.33 (± 0.05)	0.31 (± 0.03)	0.35 (± 0.02)	0.48 (± 0.04)
Shannon Diversity	1.99 (± 0.06)	2.08 (± 0.12)	2.03 (± 0.08)	1.99 (± 0.10)	1.62 (± 0.07)
Density Oligochaetes	3844.68 (± 1509.34)	7.54E+04 (± 1.31E+04)	6728.18 (± 4957.99)	768.94 (± 354.46)	480.58 (± 96.12)
Density Tubificids	192.23 (± 104.25)	6.81E+04 (± 1.10E+04)	192.23 (± 121.58)	192.23 (± 121.58)	96.12 (± 96.12)
Density *T. tubifex*	0.00 (± 0.00)	3844.68 (± 3387.90)	0.00 (± 0.00)	0.00 (± 0.00)	0.00 (± 0.00)
Density *L. hoffmeisteri*	38.04 (± 38.04)	1537.87 (± 676.92)	0.00 (± 0.00)	0.00 (± 0.00)	0.00 (± 0.00)

APPENDIX T

ROCK CREEK DRAINAGE – PHYSICOCHEMICAL MEANS BY SITE

Physicochemical means at each site within the Rock Creek drainage where data were collected. Discharge, temperature, pH, and dissolved oxygen were quantified from summer 1998 data. The proportion organic matter in sediment and conductivity were quantified from data collected in the summer 1998 and fall 1999. Elevation and slope were determined from geospatial data. Data are means ± 1 standard error (± SE); n = sample number.

Physicochemical Parameter	**RO4**	**RO2**	**RO1**	**EFRO**	**WFRO**	**ROSS**	**ST**
Discharge (cms)	12.68	9.31	9.35	3.22	5.44	3.67	1.69
	N/A	N/A	N/A	N/A	N/A	N/A	N/A
	n = 1	n = 1	n = 1	n = 1	n = 1	n =1	n = 1
Proportion Organic Matter	0.02	0.01	0.05	0.10	0.03	0.02	0.16
	(± 0.001)	(± 0.001)	(± 0.02)	(± 0.03)	(± 0.02)	(± 0.009)	(± 0.06)
	n = 3	n = 4	n = 4	n = 4	n = 2	n = 3	n = 4
Temperature(°C)	14.67	11.19	N/A	N/A	15.23	15.27	8.52
	(± 0.17)	N/A	N/A	N/A	(± 0.065)	(± 0.39)	(± 0.025)
	n = 2	n = 1	N/A	N/A	n = 2	n = 2	n = 2
Conductivity (mS/cm)	0.13	0.11	0.12	0.23	0.04	0.05	0.07
	(± 0.02)	N/A	N/A	N/A	(± 0.003)	N/A	(± 0.003)
	n = 2	n = 1	n = 1	n = 1	n = 2	n = 1	n = 2
pH	8.39	8.5	N/A	N/A	7.85	8.42	8.6
	(± 0.02)	N/A	N/A	N/A	(± 0.21)	(± 0.32)	(± 0.28)
	n = 2	n = 1	N/A	N/A	n = 2	n = 2	n = 2
Dissolved Oxygen (mg/l)	8.29	9.14	N/A	N/A	8.18	8.36	9.56
	(± 0.03)	N/A	N/A	N/A	(± 0.14)	(± 0.20)	(± 0.025)
	n = 2	n = 1	N/A	N/A	n = 2	n = 2	n = 2
Elevation (m)	1377	1220	1097	1660	1605	1753	1485
Slope	0.006	0.006	0.006	0.012	0.011	0.012	0.022

APPENDIX U

ROCK CREEK DRAINAGE – PHYSICOCHEMICAL MEANS BY REACH

Physicochemical means for each reach within the Rock Creek drainage where data were collected. Reaches are identified by the lowermost site in the reach. Discharge, temperature, pH, and dissolved oxygen were quantified from summer 1998 data. The proportion organic matter in sediment and conductivity were quantified from data collected in the summer 1998 and fall 1999. Elevation and slope were determined from geospatial data. Data are means ± 1 standard error (± SE); n = sample number.

Physicochemical Parameter	**RO1**	**EFRO**	**WFRO**	**ROSS**	**ST**
Discharge (cms)	10.45 (± 1.11) n = 3	3.22 N/A n = 1	5.44 N/A n = 1	3.67 N/A n =1	1.69 N/A n = 1
Proportion Organic Matter	0.02 (± 0.007) n = 11	0.10 (± 0.03) n = 4	0.03 (± 0.02) n = 2	0.02 (± 0.009) n = 3	0.16 (± 0.06) n = 4
Temperature (°C)	13.50 (± 1.16) n = 3	N/A N/A N/A	15.23 (± 0.065) n = 2	15.27 (± 0.39) n = 2	8.52 (± 0.025) n = 2
Conductivity (mS/cm)	0.13 (± 0.008) n = 4	0.23 N/A n = 1	0.04 (± 0.003) n = 2	0.05 N/A n = 1	0.07 (± 0.003) n = 2
pH	8.43 (± 0.38) n = 3	N/A N/A N/A	7.85 (± 0.21) n = 2	8.42 (± 0.32) n = 2	8.6 (± 0.28) n = 2
Dissolved Oxygen (mg/l)	8.57 (± 0.28) n = 3	N/A N/A N/A	8.18 (± 0.14) n = 2	8.36 (± 0.20) n = 2	9.56 (± 0.025) n = 2
Elevation (m)	1097	1660	1605	1753	1485
Slope	0.006	0.012	0.011	0.012	0.022

APPENDIX V

BLACKFOOT RIVER DRAINAGE – LAND USE BY SITE

Land use within the entire upstream subbasin and riparian zone at each site within the Blackfoot River drainage.

Land Use	BL3	BL2	BL1	NF2	NF1	KL	WA	CH	CO
Subbasin									
Proportion Conifer Forest	0.79	0.67	0.70	0.85	0.76	0.63	0.45	0.72	0.71
Proportion Agriculture	0.03	0.05	0.04	0.01	0.05	0.06	0.11	0.00	0.05
Proportion Disturbance	0.01	0.01	0.01	0	1.60E-03	0.03	0	0.01	0.04
Proportion Residential	6.44E-05	2.47E-05	4.58E-05	0	0	0	4.23E-04	0	0
Road Density	0.58	0.55	0.69	0.10	0.24	0.85	1.19	0.55	0.67
Mine Density	3.3E-08	1.50E-08	1.50E-08	0	1E-09	0	0	0	0
Riparian									
Proportion Conifer Forest	0.70	0.56	0.60	0.86	0.71	0.30	0.19	0.79	0.58
Proportion Agriculture	0.08	0.11	0.10	0.02	0.09	0.17	0.12	1.87E-03	0.07
Proportion Disturbance	5.32E-03	2.42E-03	0.01	0	1.60E-04	2.26E-03	0	9.30E-04	2.82E-03
Proportion Residential	1.75E-04	4.99E-05	1.97E-04	0	0	0	0	0	0
Road Density	1.83	1.37	1.66	0.35	0.58	1.40	1.51	0.68	0.99
Mine Density	2.9E-08	1.7E-08	1.50E-08	0	0	0	0	0	0

APPENDIX W

BLACKFOOT RIVER DRAINAGE – LAND USE BY REACH

Contributing land use between the upper and lower site in each reach at the subbasin and riparian scale within the Blackfoot River drainage. Reaches are identified by the lowermost site in the reach.

Land Use	**BL3**	**BL1**	**NF2**	**NF1**	**KL**	**WA**	**CH**	**CO**
Subbasin								
Proportion Conifer Forest	0.79	0.75	0.85	0.76	0.63	0.45	0.72	0.71
Proportion Agriculture	0.03	0.02	0.01	0.05	0.06	0.11	0.00	0.05
Proportion Disturbance	0.01	0.03	0	1.60E-03	0.03	0	0.01	0.04
Proportion Residential	6.44E-05	8.85E-05	0	0	0	4.23E-04	0	0
Road Density	0.58	0.98	0.10	0.24	0.85	1.19	0.55	0.67
Mine Density	3.32E-08	1.42E-08	0	1.23E-09	0	0	0	0
Riparian								
Proportion Conifer Forest	0.70	0.69	0.86	0.71	0.30	0.19	0.79	0.58
Proportion Agriculture	0.08	0.08	0.02	0.09	0.17	0.12	1.87E-03	0.07
Proportion Disturbance	5.32E-03	0.02	0	1.60E-04	2.26E-03	0	9.30E-04	2.82E-03
Proportion Residential	1.75E-04	5.13E-04	0	0	0	0	0	0
Road Density	1.83	2.29	0.35	0.58	1.40	1.51	0.68	0.99
Mine Density	2.9E-08	1.19E-08	0	0	0	0	0	0

APPENDIX X

BLACKFOOT RIVER DRAINAGE – BIOASSESSMENT METRIC MEANS BY SITE

Bioassessment metric means (summer 1998 data) at sites where invertebrates were collected within the Blackfoot River drainage. Data are means ± 1 standard error (± SE). n = 6 for all metrics.

Metric	**BL3**	**BL2**	**BL1**	**NF2**	**NF1**	**KL**	**WA**	**CH**	**CO**
Density Oligochaetes	0.00 (± 0.00)	6.34E+03 (± 2.68E+03)	4.56E+04 (± 1.83E+04)	768.94 (± 243.16)	6.93E+04 (± 3.40E+04)	3.49E+04 (± 7621.41)	2.86E+04 (± 2.70E+04)	8073.82 (± 2068.64)	1826.22 (± 720.56)
Density Tubificids	0.00 (± 0.00)	4.04E+03 (± 2.04E+03)	1.18E+04 (± 5.04E+03)	0.00 (± 0.00)	6.92E+04 (± 3.40E+04)	2.99E+04 (± 6722.82)	2.80E+04 (± 2.69E+04)	6632.06 (± 2171.90)	1730.10 (± 729.48)
Density *T. tubifex*	0.00 (± 0.00)	576.70 (± 364.74)	0.00 (± 0.00)	0.00 (± 0.00)	1.54E+04 (± 9486.30)	0.00 (± 0.00)	0.00 (± 0.00)	576.70 (± 576.70)	0.00 (± 0.00)
Density *L. hoffmeisteri*	0.00 (± 0.00)	288.35 (± 288.35)	0.00 (± 0.00)	0.00 (± 0.00)	2595.16 (± 1472.19)	0.00 (± 0.00)	0.00 (± 0.00)	480.58 (± 275.24)	192.23 (± 121.58)

APPENDIX Y

BLACKFOOT RIVER DRAINAGE – BIOASSESSMENT METRIC MEANS BY REACH

Bioassessment metric means (summer 1998 data) for each reach where invertebrates were collected within the Blackfoot River drainage. Data are means ± 1 standard error (± SE). Reaches are identified by the lowermost site in the reach. n = 6 for all reaches except reach BL1 (n = 12).

Metric	**BL3**	**BL1**	**NF2**	**KL**	**WA**	**CH**	**CO**
Density Oligochaetes	0.00 (± 0.00)	2.60E+04 (± 1.06E+04)	768.94 (± 243.16)	3.49E+04 (± 7621.41)	2.86E+04 (± 2.70E+04)	8073.82 (± 2068.64)	1826.22 (± 720.56)
Density Tubificids	0.00 (± 0.00)	7929.64 (± 2845.80)	0.00 (± 0.00)	2.99E+04 (± 6722.82)	2.80E+04 (± 2.69E+04)	6632.06 (± 2171.90)	1730.10 (± 729.48)
Density *T. tubifex*	0.00 (± 0.00)	288.35 (± 194.41)	0.00 (± 0.00)	0.00 (± 0.00)	0.00 (± 0.00)	576.70 (± 576.70)	0.00 (± 0.00)
Density *L. hoffmeisteri*	0.00 (± 0.00)	144.18 (±144.18)	0.00 (± 0.00)	0.00 (± 0.00)	0.00 (± 0.00)	480.58 (± 275.24)	192.23 (± 121.58)

APPENDIX Z

BLACKFOOT RIVER DRAINAGE – PHYSICOCHEMICAL MEANS BY SITE

Elevation and slope of sites within the Blackfoot River drainage.

Physicochemical Parameter	BL3	BL2	BL1	NF2	NF1	KL	WA	CH	CO
Elevation (m)	1330	1146	995	1270	1242	1272	1212	1188	1197
Slope	0.002	0.003	0.003	0.007	0.007	0.004	0.034	0.017	0.013

APPENDIX AA

BLACKFOOT RIVER DRAINAGE – PHYSICOCHEMICAL MEANS BY REACH

Elevation and slope of each reach within the Blackfoot drainage. Reaches are identified by the lowermost site in the reach.

Physicochemical Parameter	**BL3**	**BL1**	**NF2**	**KL**	**WA**	**CH**	**CO**
Elevation (m)	1330	995	1242	1272	1212	1188	1197
Slope	0.002	0.003	0.007	0.004	0.034	0.017	0.013