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Relating riparian habitat variables to incidence of whirling disease

Jack McWilliams
The University of Montana

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RELATING RIPARIAN HABITAT VARIABLES

TO INCIDENCE OF WHIRLING DISEASE

by

Jack McWilliams

B.S., Humboldt State University - *Arcata*, CA 1981

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Dean, Graduate School

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Relating Riparian Habitat Variables To Incidence Of Whirling Disease

Director: Paul L. Hansen

This study attempts to determine which of nine quantified riparian habitat variables are important to incidence of whirling disease. It also attempts to determine whether the total health score (sum of the riparian habitat variables) can be used to predict the presence of whirling disease. Spearman's rank order correlation coefficient analysis, a non-parametric procedure, was used to establish correlations between the nine riparian habitat variables, total health score and benthic macro invertebrate data indicative of disturbance in streams.

Six of the nine riparian habitat variables were found to be significantly tied to disturbance in streams and hence to the incidence of whirling disease. These six variables were ranked with the number of statistically significant correlations to disturbance as the criterium for determining relative importance. A logistic regression model with presence or absence of whirling disease as the binary response, and scores from the six significantly correlated riparian habitat variables as independent variables was modeled. Results of this model were used to devise a form that will enable managers to determine the probability of a stream having, or being capable of supporting, whirling disease. Suggestions for future research are given to increase the predictive power of the logistic model and possibly establish *Helicopsyche borealis* , a caddis fly, as an indicator species.

TABLE OF CONTENTS

INTRODUCTION....1
Specific Study Goals....3
METHODS AND MATERIALS....5
Data....5
Collection times....5
Site selection....5
Riparian habitat variables....7
Sampling procedure....7
Analysis Methods....9
Method # 1....9
Method # 2....10
RESULTS....13
Analysis Method # 1....13
Analysis Method # 2....17
DISCUSSION....18
Percent of Riparian Zone Covered by Noxious Weeds (Variable # 3)....19
Percent of Site Covered by Disturbance-Induced Undesirable Species (Variable # 4)....20
Degree of Browse Utilization of Trees And Shrubs (Variable # 5)....21
Woody Species Establishment and Regeneration (Variable # 6)....22
Percent of Site With Human Caused Bare Ground (Variable # 7)....24
Incisement (Vertical Stability of the Channel) (Variable # 9)....24
Riparian Habitat Variables and Disturbance....25
Logistic Regression....27
SUMMARY....30
Analysis Method # 1....30
Analysis Method # 2....30
Additional Conclusions....30
SUGGESTIONS FOR FUTURE RESEARCH....31
ACKNOWLEDGEMENTS....32
LITERATURE CITED....33

LIST OF TABLES

1. Riparian habitat variables used in statistical analysis....................................... 7

2. Positive and negative sites used in logistic regression and record ID number.. 12

3. Riparian habitat variables and P-value for last level of statistical significance 13

4. Riparian habitat variables and significantly related taxa groupings with tied P and Rho values.. 14

5 Riparian habitat variables, and P-value for last level of statistical significance.. 14

6. Riparian habitat variables and significantly related taxa (taxonomic level) with tied P and Rho values.. 15

7. Riparian habitat variables and number of statistically significant correlations.. 16

8. Riparian habitat variables and number of statistically significant correlations.. 16

9. Riparian habitat variables and number of significantly significant correlations.. 17

10. Habitat variables, parameter estimate, standard error, Chi square value and probability of obtaining a chi square value greater than shown for logistic model.. 17

11. Suggested format of page to added to currently used lotic health assessment form to determine probability of WD in a reach of stream......... 28

LIST OF FIGURES

1. Map of sites used for Spearman's rank-order correlation coefficent analysis.. 6

APPENDICES

A Sampling sites eliminated from analysis and reason for elimination.. A1

GLOSSARY

Glossary G1

INTRODUCTION

Whirling disease (WD) is the common name for a condition in salmonids caused by infection by the microscopic parasite *Mycobolus cerebralis.* This parasite has a complex life cycle involving two hosts and assumes two different forms. The spore form of the parasite is released when an infected fish dies. Spores are ingested by worms of the genus *Tubifex.* After a few months inside the worm, the parasite changes into a free swimming infective stage called a TAM, and is released into the water column. It infects a fish host to complete its life cycle (Rocky Mountain Fly Fishing Center 1996).

Once inside a young fish, *M. cerebralis* attacks the cartilage. Young fish are most vulnerable because they possess large amounts of soft cartilage., Adult fish are less affected because once cartilage has hardened they show few ill effects of infection. In severe infections, inflammation around the damaged cartilage places pressure on the nervous system, causing the fish to "whirl" when startled. Spinal deformities can also result. Seriously infected fish have a reduced ability to feed or escape from predators (Markiw 1992). Although all species of trout and salmon are susceptible to WD, rainbow trout populations seem to be the most devastated.

The impact of WD on trout fisheries, especially in the western United States, has been devastating. Colorado reports wild rainbow population declines in the Colorado, Gunnison, Arkansas, Rio Grande, South Platte and Poudre rivers (Marlowe and Gardner 1995). Rainbow populations in Montana's Madison River have decreased from 3,300 per mile to 300 per mile (Marlowe and Gardner 1995). WD has been confirmed in Montana's Ruby River, Clark Fork River, Rock Creek and others for a total of 73 rivers, streams and lakes (Montana Fish, Wildlife and Parks 1999). Rivers in other states have been similarly impacted.

Waddington and Laughland (1996) indicate that in Montana, average angler expenditure per day is $66. It is estimated that fishermen spend about 2.3 million angler days per year pursuing trout in Montana (McFarland 1995.) This revenue, much of it spent in local economies, could be seriously impacted by WD.

The 1996 Whirling Disease Conference in Denver, CO, developed a list of five ecological factors which appear to influence presence of the disease and potential for populational impacts. All sites that test positive for WD:

1. Are highly productive, i.e., over 300 pounds/acre (this is standing crop and Wetzel, 1983, defines this as the weight of organic material that can be sampled or harvested at any one time from a given area) and commonly have very high electrical conductivity readings.
2. Have flushing flows less than one out of ten years.
3. Have brown trout present to act as a reservoir for the disease. (The parasite is from Europe where it co-evolved with brown trout. These trout show little symptoms of the disease, but function as carriers.)
4. Are relatively low gradient streams.
5. Have human-altered or enriched habitats which amplify the pathogen.

In a keynote address at the 1999 Whirling Disease Symposium held in Missoula, MT, Allendorf (1999), stated habitat degradation may cause stress on fish. This stress may make fish more susceptible to the disease by weakening them.

Spring creeks, tail water streams and disturbed streams and rivers are considered to be high risk for WD (Gustafson 1996). Conversely, undisturbed Rocky Mountain streams and rivers, warm trout waters (above the critical higher temperature limit for TAMs) and lake outlets (too cold for TAMs) are listed as low risk areas.

Within these general ecological factors influencing the presence or absence of WD, there is much to be discovered. This study attempts to identify the specific ecological factors that support conditions conducive to WD by using benthic macroinvertebrate data indicative of disturbance in streams. Use of benthic macroinvertebrates to monitor and assess biological condition in the Pacific Northwest is an accepted approach (Fore and others 1996.) The goal of such biological monitoring and assessment often is to measure and evaluate the consequences of human actions on biological systems (Karr and Chu 1999).

Determining these factors and being able to scale their importance will provide managers a tool for dealing with WD. Additionally, if criteria for these ecological factors could be used to predict probability of a stream reach supporting WD, evaluation procedures could be developed that would enable managers to evaluate a reach of stream, determine problem areas, and plan remedial efforts accordingly.

Specific Study Goals

Specific goals of this study are:

1. Construct a list of habitat variables ranked in their importance to presence or absence of WD in a stream.
2. Relate the score generated by the ranked habitat variables to incidence of WD.

Whirling disease is a fact of life in America and is here to stay'(Vincent 1999). While WD can not be eliminated, it may be controlled, especially in the small feeder streams where salmonids hatch. If control methods can be developed, fisheries in the western states can be maintained.

Developing effective control methods will involve several steps:

1. Identifying riparian habitat variables that influence presence or absence of WD.
2. Scaling importance of those variables.
3. Designing an assessment procedure that would enable managers to evaluate streams within their purview and estimate probability of those streams supporting WD.

METHODS AND MATERIALS

Data

Data for analyses were provided by the Bureau of Land Management (BLM) and collected by an independent contractor. Data were for 185 sites in the montane ecoregion of western Montana (Omernik and Gallant 1986). A total of 40 sites were eliminated from analysis to maintain data consistency (Appendix A). Figure 1 shows the 145 sites used.

Nilson (1998) describes collection procedures for benthic macroinvertebrates used in this study. Relevant details follow.

Collection times—Late fall and early spring are the appropriate times to collect mature *Tubifex.* Identification of *Tubifex* worms requires mature specimens. The worms mature in late fall and in spring are washed away from their habitat during heavy runoff. *Tubifex* worms were sampled in the spring and fall of 1997, and spring of 1998.

Site selection—BLM 1:100,000 surface management maps were used for site selection. Road access was considered in determining sample sites. The number of sites sampled on a given stream was determined by how much of the stream was on BLM land. Majority of sites sampled were on BLM land but a few additional Forest Service and private sites were sampled. Sites were selected to provide an overall view of a stream. Riparian areas in degraded condition received special attention.

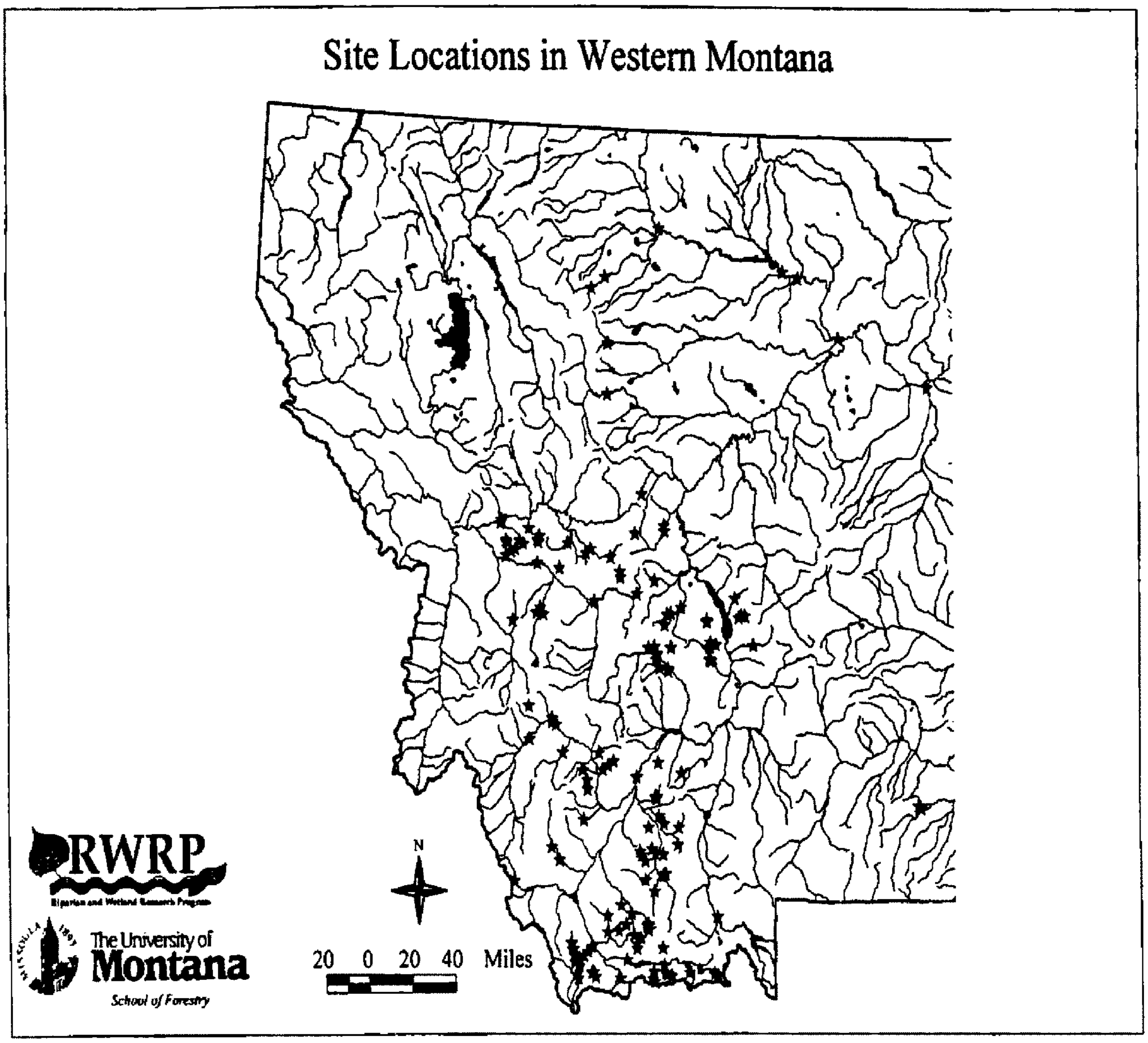

Figure 1. Map of sites used for analysis

Riparian habitat variables—Nine riparian habitat variables were measured and used in statistical analyses. Table 1 provides a list of these riparian habitat variables and numbers.

Table 1. Riparian habitat variables and numbers used in statistical analysis

Habitat Variable	Variable Number
Amount of flood plain and stream banks covered by plant growth	1
Percent of stream bank bound by a deep root mass	2
Percent of riparian zone covered by noxious weeds	3
Percent of the site covered by disturbance-induced undesirable herbaceous species	4
Degree of browse utilization of trees and shrubs	5
Woody species establishment and regeneration	6
Percent of site with human caused bare ground	7
Percent of stream bank structurally altered by humans	8
Incisement (vertical stability of the channel)	9

Sampling procedure—Once a site representative of upstream and downstream reaches was selected, the following data was obtained:

1. A GPS reading, utilizing a Magellan GPS 2000 XL or 3000XL, was taken and additional readings determined 25 ft upstream and downstream to delineate upper and lower boundaries of the sample site.

2. Riparian health assessment form was filled out in accordance with Thompson and others (1998). The following information was added to each assessment form:

- Rosgen stream type (Rosgen 1996)
- Benthic macroinvertebrate sample number (macro ID number)
- Section, township and range
- Macro stream habitat types (shoreline, undercut banks, sediment, gravel, cobble, boulder, bedrock, submerged vegetation, woody debris)
- Comments
- Valley type (subjective description by sampler)

3. At each site an ocular estimation of macro stream habitat types was made. Each stream habitat type was assigned a percentage of total stream sample area and sampled in its representative percentage by kick net.

4. Sample material was placed in sifting buckets and non-benthic macroinvertebrate material removed. Remainder of sample was placed in 500 ml containers and preserved with ethanol.

5. When large macroinvertebrates stopped movement, 50 ml formalin was added to each 500 ml container.

6. Each container received a waterproof identification tag with benthic macroinvertebrate sample number and stream name.

7. Each container was then sealed and labeled again using a strip of freezer paper.

Scores for riparian habitat variables were calculated using the Riparian and Wetlands Research Program's (RWRP) lotic health assessment (stand alone) form in accordance with Thompson and others (1998). These scores provide a quantified health assessment for each site. Riparian habitat variables for each site were scored at the same time and place as benthic macroinvertebrates for each site were collected.

Benthic macroinvertebrates were identified and placed into taxonomic groupings by the National Aquatic Monitoring Center at Utah State University, Logan, Utah. A full description of protocols used to identify and classify benthic macroinvertebrates used for analysis in this study can be found at their web site: www.usu.edu/~buglab under aquatic invertebrate sampling protocols, option two.

Analysis Methods

Method # 1—Spearman's rank-order correlation coefficient analysis, a non-parametric procedure, was utilized to calculate correlations between scores for nine individual habitat variables, total riparian rating and taxonomic category. Statview statistical software (Abacus Concepts 1996) was used to perform this procedure. This statistical procedure investigates whether there is a monotomic relationship between two variables (Sheskin 1997) and requires no assumption of normality or independence of variables.

The correlation coefficient, Rho, provides a measure of strength of the monotomic relationship between two variables even if that relationship is not linear (Ott 1993). Perfect positive relationships show a Rho score of positive one while perfect negative relationships display a Rho score of negative one. Complete lack of correlation is indicated by zero.

Statistical strength of the Rho statistic is defined by a p-value with scores from zero to one. Smaller p-values indicate greater statistical significance. Tied Rho and p-values vs. basic Rho and p-values were utilized to strengthen conclusions of analyses. Use of basic Rho and p-values can cause inflation of statistical significance (Sheskin 1997). Tied values in either scores for riparian habitat variables, overall riparian score or taxonomic data were corrected by assigning to each tied observation the mean rank of the rank positions for which it is tied (Daniel 1990).

A total of 650 Spearman's rank order correlation coefficient analyses were conducted between nine habitat variables, total riparian rating, and 65 taxa groupings indicative of disturbance in streams. A spreadsheet of results for each Spearman analysis was constructed. With 650 values and a p-value of 0.05 set as necessary for significance, as many as 32 spurious correlations could result. To avoid this, a Bonferroni-Dunn test was performed with a familywise Type I error rate set at 0.05 (Sheskin 1997). This procedure adjusts p-values to eliminate spurious correlations as number of correlations increases. As a result of this test, a p-value of less than or equal to 0.0001 was established as necessary to maintain a true p-value of 0.05.

Spearman analyses were also performed between habitat variables, total riparian rating, and individual benthic macroinvertebrate data used to determine taxonomic groupings. This was done to ensure relationships between individual taxa and habitat variables were not masked by inclusion of individual taxa in a taxa grouping. Data for 205 individual taxa were provided and each of these were run against nine habitat variables and an overall score for a total of 2,050 Spearman rank order correlation analyses.

A Bonnferroni-Dunn test with a familywise Type I error set at 0.05 (Sheskin 1997) was utilized to determine the p-value necessary to maintain a true p-value of 0.05 for the 2,050 analyses. This procedure established 0.000024 as the correct p-value to maintain a p-value of 0.05. Statview (Abacus Concepts 1996) sets < 0.0001 as minimum p-value it will compute. However, 0.00001, lowest increment to five decimal places is lower than 0.000024. Using < 0.0001, as provided by Statview, is more rigorous than required.

Method # 2—Because presence or absence of whirling disease is a binary response to habitat variables determined to be significantly tied to disturbance in streams, a logistic regression model is appropriate (Hosmer and Lemeshow 1989). JMP (SAS Institute 1995), a statistical software package, was used to fit a logistic regression model that estimates the probability of stream reaches having or being capable of supporting whirling disease based on habitat variable scores.

Sites used to provide data for logistic regression were selected by matching sites with benthic macroinvertebrate samples provided by the BLM with sites listed as tested for whirling disease by Montana Fish, Wildlife & Parks (1999). A total of 35 data points, 15 with WD and 25 without, were used to construct the logistic model and are included as Table 2.

Table 2. Positive and negative sites used in logistic regression and record ID number

Record ID Number	Positive sites	Negative sites
006	X	
009	X	
010	X	
013	X	
029	X	
031	X	
038	X	
072	X	
108	X	
113	X	
115	X	
142	X	
143	X	
154	X	
174	X	
003		X
011		X
016		X
018		X
023		X
026		X
027		X
070		X
071		X
090		X
091		X
092		X
093		X
094		X
095		X
096		X
097		X
098		X
104		X
106		X
146		X
168		X
173		X
187		X
190		X

RESULTS

Analysis Method # 1

Table 1 provides a list of riparian habitat variables and numbers used in statistical analyses. With a p-value of less than or equal to 0.0001 required to be statistically significant for correlation of taxa groupings with habitat variables, four habitat variables were eliminated (Table 3). None of these variables are correlated with groupings of benthic macroinvertebrates indicative of disturbance in streams with a level of significance satisfying the minimum of less than or equal to 0.0001. Table 3 lists these variables, and p-values for the last level of significance met by the variable before being eliminated.

Table 3. Riparian habitat variables, numbers, and P-value for last level of statistical significance

Habitat Variable	Variable Number	P-value
Percent of stream bank bound by a deep root mass	2	0.1
Percent of stream bank structurally altered by humans	8	0.025
Percent of riparian zone covered by noxious weeds	3	0.005
Amount of flood plain and stream banks covered by plant growth	1	0.001

With a p-value less than or equal to 0.0001, the following five habitat variables remain:

- Percent of the site covered by disturbance-induced undesirable herbaceous species (variable # 4)
- Degree of browse utilization of trees and shrubs (variable # 5)
- Woody species establishment and regeneration (variable # 6)
- Percent of site with human caused bare ground (variable # 7)
- Incisement (vertical stability of the channel) (variable # 9)

Table 4 displays variable number, taxonomic groupings with statistically significant correlations, and tied p and Rho values for the correlation.

Table 4. Habitat variable numbers, significantly related taxa groupings with tied p and Rho values

Variable	Taxa Groupings	P-values	Rho values
4	# intolerant taxa	<0.0001	+0.375
5	# intolerant taxa	<0.0001	+0.408
6	Intolerant taxa abundance	<0.0001	+0.414
	# intolerant taxa	<0.0001	+0.382
	Plecoptera abundance	<0.0001	+0.336
	EPT taxa	0.0001	+0.320
	Mollusca taxa	<0.0001	-0.366
	CTQ:d	<0.0001	-0.387
	# tolerant taxa	<0.0001	-0.422
7	Scraper abundance	<0.0001	+0.389
9	Scraper abundance	<0.0001	+0.370

Values at top and bottom of each variable represents strongest positive and negative correlations where applicable.

With a p-value of < 0.0001 established as minimum for significant correlation between individual benthic macroinvertebrate taxa and habitat variables to be significant, five habitat variables are eliminated. Table 5 shows these habitat variables, and P-values for the last level of significance met by the variable before being eliminated.

Table 5. Riparian habitat variables, and p-value for last level of statistical significance

Habitat Variable	Variable Number	P-value
Percent of streambank structurally altered by humans	8	0.0036
Incisement (vertical stability of the channel)	9	0.0016
Percent of streambank bound by a deep root mass	2	0.0010
Amount of the floodplain and stream banks covered by plant growth.	1	0.0007
Percent of site with human caused bare ground	7	0.0002

With a p-value < 0.0001, four habitat variables remain:

- Percent of the riparian zone covered by noxious weeds (variable # 3)
- Percent of the site covered by disturbance-induced undesirable herbaceous species (variable # 4)
- Degree of browse utilization of trees and shrubs (variable # 5)
- Woody species establishment and regeneration (variable # 6)

Table 6 displays habitat variable number, taxa with statistically significant correlations, and tied p and Rho values for the correlation.

Table 6. Habitat variable numbers, significantly related taxa (taxonomic level) with tied p and Rho values

Habitat Variable Number	Taxa	P-values	Rho values
3	Capniidae	<0.0001	+0.356
	Ephemerella infrequens	<0.0001	-0.520
4	*Ephemerella infrequens*	<0.0001	+0.459
	Nemouridae	<0.0001	+0.366
	Prosimulium	<0.0001	+0.361
5	Nemouridae	<0.0001	+0.360
	Ephemerella infrequens	<0.0001	+0.347
	Epeorus	<0.0001	+0.325
	Ephemerella	<0.0001	-0.418
6	*Epeorus*	<0.0001	+0.421
	Rhyacophila	<0.0001	+0.419
	Sweltsa	<0.0001	+0.396
	Helicopsyche	<0.0001	-0.329
	Physella	<0.0001	-0.348

Values at top and bottom of each variable represent strongest positive and negative correlations where applicable.

Table 7 displays riparian habitat variables, and number of statistically significant correlations to taxa groupings indicative of disturbance in streams for each variable.

Table 7. Riparian habitat variables their numbers, and number of statistically significant correlations

Habitat Variable	Variable Number	Number of statistically significant correlations to taxa groupings indicative of disturbance
Percent of the site covered by disturbance-induced undesirable herbaceous species	4	1
Degree of browse utilization of trees and shrubs	5	1
Woody species establishment and regeneration	6	7
Percent of site with human caused bare ground	7	1
Incisement (vertical stability of the channel)	9	1

Table 8 shows riparian habitat variables, and number of statistically significant correlations to individual taxa.

Table 8. Riparian habitat variables their numbers, and number of statistically significant correlations

Variable	Variable Number	Number of statistically significant correlations to taxa groupings indicative of disturbance
Percent of the riparian zone covered by noxious weeds	3	2
Percent of the site covered by disturbance-induced undesirable herbaceous species	4	3
Degree of browse utilization of trees and shrubs	5	4
Woody species establishment and regeneration	6	5

Table 9 combines results shown in Tables 7 and 8. It displays riparian habitat variables, number of statistically significant correlations to taxa groupings indicative of disturbance in streams for each variable, number of statistically significant correlations to individual taxa for each variable, and total number of statistically significant correlations for each variable.

Table 9. Riparian habitat variable numbers and number of statistically significant correlations

Variable Number	Number of statistically significant correlation to taxa groupings indicative of disturbance in streams	Number of statistically significant correlations to individual taxa	Total number of statistically significant correlations for each variable
3	0	2	2
4	1	3	4
5	1	4	5
6	7	5	12
7	1	0	1
9	1	0	1

Analysis Method # 2

A logistic regression model provided by JMP (SAS 1995) utilizing presence or absence of whirling disease as the dependent variable and scores for habitat variables listed in Tables 4 and 6 as independent variables yielded the results shown in Table 10.

Table 10. Habitat variable numbers, parameter estimate, standard error, Chi square value and probability of obtaining a chi square value greater than shown for logistic model

Variable Number	Parameter Estimate	Standard Error	Chi Square	Prob > Chi Square
3	+ 0.01	0.21	0.00	0.9501
4	+ 0.59	0.85	0.49	0.4862
5	- 0.32	0.39	0.67	0.4137
6	+ 0.36	0.34	1.10	0.2934
7	+ 0.31	0.24	1.59	0.2079
9	- 0.68	0.35	3.60	0.0577

DISCUSSION

There is an association between incidence of WD and human impacted streams (Gustafson 1996). One of five ecological factors identified as associated with WD by the 1996 Whirling Disease Conference is human altered or enriched habitats. Habitat degradation may stress fish making them more susceptible to disease and changes in stream habitat may increase abundance of *Tubifex tubifex,* alternate host of *M. cerebralis.* (Allendorf 1999).

Identifying variables in degraded habitats important to incidence of WD gives managers a tool to help control the disease. Utilizing benthic macroinvertebrate data, in groupings that reflect riparian health is an effective way to validate those variables. Both variables and taxonomic data are measures of disturbance in streams where whirling disease is found. I was able to identify five measurable habitat variables linked with strong statistical significance to benthic macroinvertebrate groupings indicating disturbance in streams (Table 3).

Utilizing individual taxa vs. taxonomic groupings in analysis with habitat variables and overall health scores may yield relationships masked by an individual taxa's inclusion in a group. Such relationships can provide another link between disturbance in a stream and habitat variables, and therefore with whirling disease. I found four riparian habitat variables with strong correlations to individual taxa (Table 5).

Thompson and others (1998) provide instructions for measuring the habitat variables listed in Tables 4 and 6. As riparian health increases, scores for applicable variables increase. Table 4 provides tied Rho values that are measures of correlation between variables and listed taxa groupings indicative of disturbance. Thompson and others (1998) provide instructions for measuring the habitat variables listed in Tables 4 and 6. As riparian health increases, scores for applicable variables increase. Table 4 provides tied Rho values that are measures of correlation between variables and listed taxa groupings indicative of disturbance in streams. Table 6 provides tied Rho values that are measures of correlation between variables and listed individual taxa. Negative correlation values are inversely related to increasing health while positive correlations are directly related to increasing health values.

Table 8 lists each riparian habitat variable that has significant statistical correlations to either taxa groupings indicative of riparian health or individual taxa and total number of significant statistical correlations. Each of these riparian habitat variables and its significant correlations will be discussed in order.

Percent of Riparian Zone Covered by Noxious Weeds (Variable # 3)

This habitat variable has significant statistical links with Capniidae, a family of Plecoptera (stoneflies), and with *Ephemerella infrequens* , a species of Ephemeroptera (mayfly). As the health rating for this particular variable increases, the percent of noxious weeds decreases. Numbers of Capniidae increase as health ratings increase. This positive correlation was expected since Plecoptera are generally associated with cool, clean water (Stewart and Harper 1996) and increasing health scores indicate such conditions.

Congenerics of *Ephemerella infrequens* include 32 species of insects which feed as collector-gatherers and scrapers. *E. infrequens*, ,however, is a shredder (Edmunds and Waltz 1996). Wisseman (1990) states shredders are indicative of a healthy riparian vegetation community; however numbers of *E. infrequens* decrease as riparian health scores for percent of noxious weeds increase. Although we view noxious weeds as undesirable and assign lower health scores as their presence increases, *E. infrequens* uses noxious weeds only as riparian plants. Cummins and others (1989) state shredders do not feed specifically on litter of a particular species, but rather on appropriately conditioned litter, regardless of species. Strong negative correlation with health scores in this instance reflects a strong positive correlation with the presence of riparian vegetation, and is an indicator of good health.

Percent of the Site Covered by Disturbance-Induced Undesirable Herbaceous Species (Variable # 4)

This variable has a positive correlation with number of intolerant taxa. As health score increases, so do the number of intolerant taxa. This relationship was as expected.

Habitat variable # 4 is statistically linked to three individual taxa. Strongest of these statistical links is a positive correlation with *E. infrequens.* Undesirable herbaceous species indicate displacement from potential natural community and are less productive and generally have shallow roots. They poorly perform most riparian functions (Thompson and others 1998). Positive correlation with health scores indicates an increase in *E. infrequens* numbers as health score increases.

Nemouridae, a family of Plecoptera, is positively tied to health scores for variable # 4. Their numbers should and do increase as health score increases.

Riparian health variable # 4 is also positively correlated with *Prosimulium,* a genus of Diptera (true flies). Hilsenhoff (1988) rates Simulidae, the family containing *Prosimulium,* as moderately intolerant, but the same author (1987) listed three species of genus *Prosimulium* as very intolerant and two as moderately intolerant. A positive correlation with health scores is consistent with his ratings for intolerance.

Degree of Browse Utilization of Trees and Shrubs (Variable # 5)

Like riparian habitat # 4, this variable also has a strong positive correlation with number of intolerant taxa. As health score increases, so do number of intolerant taxa.

This riparian habitat variable also has four significant statistical ties to individual benthic macroinvertebrate taxa. For this variable, health scores increase as browse utilization decreases. Three of four taxa linked to this variable have positive correlations while one is negative.

Of three positively correlated taxa, two, Nemouridae (a family of Plecoptera) and *E. infrequens,* have been discussed relative to riparian habitat variable #4. That discussion applies to degree of browse utilization also. The third, *Epeorus,* is in Ephemeroptera in the family Heptageniidae. Winget and Magnum (1979) and Wisseman (1990) both list *Epeorus* spp. as intolerant so their numbers would be expected to increase as health scores increase. The positive correlation found confirms this.

Ephemerella, a genus in the family Ephemerelladae in Ephemeroptera, is negatively correlated to degree of browse utilization. Edmunds and Waltz (1996) describe *Ephemerella*, a genus with 32 species, as mostly collector-gatherers with some scrapers. Wisseman (1990) states collector-gatherers feed on fine sediment enriched with particles of organic matter and that when collector-gatherers increase, it is an indicator of declining water quality. A negative correlation between *Ephemerella* and riparian health is expected.

Woody Species Establishment and Regeneration (Variable # 6)

This riparian habitat variable has seven statistically significant correlations with taxa groupings indicative of disturbance in streams. Four of these correlations are positive and three negative.

Intolerant taxa abundance number and associated number of intolerant taxa have a strong positive correlation to health scores. As health score increases, so does number of intolerant taxa and intolerant taxa abundance. Plecoptera abundance and Ephemerella Plecoptera Tricoptera (EPT) taxa are also related and also show a positive correlation. Bode (1988) and Wisseman (1990) state EPT richness is indicative of disturbance in a stream and is positively tied to health, i.e., EPT taxa richness increases as health increases. Correlation values in Table 3 reflect that relationship.

Mollusca taxa, CTQ:d (a community tolerance quotient), and number of tolerant taxa show negative correlations to riparian health. As health scores increase, these taxa decrease. Mollusca do well in organically enriched habitats with high sediment (Harmon 1974) which indicates disturbance. Winget and Mangum

(1979) list the entire phylum Mollusca as highly tolerant of disturbance. A negative correlation as shown in Table 4, therefore, was expected. CTQ:d and number of tolerant taxa should both decrease as riparian health increases. Negative correlations indicate they do. Of five significant correlations with individual benthic macroinvertebrate taxa, three are positive and two negative.

Epeoreus, a genus of mayflies considered to be intolerant (Winget and Mangum 1979; Wisseman 1990) has the strongest positive correlation. This was expected and is discussed above with respect to habitat variable # 5.,

Rhyacophila, in Tricoptera (caddis flies) and in the family Rhyacophilidae, is tied positively to woody species establishment and regeneration. Hilsenhoff (1988) assigns the family, Rhyacophilidae, a zero tolerance level and Winget and Magnum (1979) rate the genus *Rhyacophila* equal with the family. A strong positive correlation between *Rhyacophila* and habitat variable # 6 indicates this highly intolerant genus increases with increasing health..

A Plecopteran, *Sweltsa*, also shows a strong positive relationship with woody species establishment and regeneration. Hilsenhoff (1988) assigns this genus an intolerant rating and Winget and Magnum (1979) give it a low tolerance rating. Both these ratings are in keeping with the positive correlation that was found.

Helicopsyche, a second Trichopteran genus and *Physella*, a gastropoda in the Phylum Mollusca, both exhibit negative correlations with woody species establishment and regeneration. *Helicopsyche* is a member of the family Heliocopsychidae which Hilsenhoff (1988) lists as relatively intolerant to pollution. However, *Helicopsyche* is considered an algal grazer (Williams and others 1983; Wiggins 1996). Vaughn, (pers. com. 1999) suggests that as woody species grow in riparian zones, they provide shade for streams and shading eliminates or reduces periphyton on rocks. This is completely consistent with the negative correlation with woody shrub establishment and regeneration. As shrubs increase in riparian zones along stream banks, shade increases, periphyton decreases and numbers of *Helicopsyche* will decline.

Winget and Mangum (1979) list the entire phylum Mollusca as highly tolerant. Harmon (1974) describes the family Physidae, which contains Physella , as one of two most resistant families in the entire Phylum. It is no surprise that their numbers decline as health scores for woody establishment and regeneration increase.

Percent of Site With Human Caused Bare Ground (Variable # 7)

This riparian habitat variable has a positive significant correlation to the benthic macroinvertebrate functional group "scrapers." Wisseman (1990) discusses the functional group scrapers and concludes a diverse and numerically well represented scraper community reflects good habitat/water quality. My analyses indicate a positive relationship as is shown in Table 4.

Incisement (Vertical Stability of the Channel) (Variable # 9)

This variable has only one statistically significant correlation and this correlation is a positive one to the functional group "scrapers." As discussed above with respect to variable # 7, a numerically well represented scraper community indicates good habitat/water quality.

Riparian Habitat Variables and Disturbance

Riparian habitat variable #6, woody species establishment and regeneration, was the most important indicator of disturbance in streams. There are 12 significant ties to disturbance indicative benthic macroinvertebrate data. Degree of browse utilization of trees and shrubs (riparian habitat variable # 5) has five significant links with benthic macroinvertebrate data and is second most important. It is clearly tied to woody species establishment and regeneration.

High browse utilization however accomplished, will prevent woody species from establishment and regeneration. Woody plant species increase rapidly when riparian areas are protected from livestock grazing (Schulz and Leninger 1990). Kauffman and others (1983) conclude excess browsing does not allow woody species to regenerate and Kauffman and Krueger (1984) state excessive grazing pressures prevent establishment of seedlings. Sixty-eight percent of statistically significant ties between riparian habitat variables and benthic macroinvertebrate data indicative of stream health are accounted for by these two riparian habitat variables (variables # 5 and # 6).

Habitat variable # 4 (percentage of site covered by disturbance-induced undesirable herbaceous species) accounts for an additional four significant links between riparian habitat variables and disturbance indicative benthic macroinvertebrates. Platts (1978) states livestock trample and compact the soil resulting in loss of high-quality, fibular-rooted plants. These are replaced by shallow rooted annual species or taprooted forbs and shrubs. These generally continue to increase with continued grazing because they are unpalatable. Ohmart (1996) concludes that after a few years of grazing, the herbaceous ground cover mix changes from highly palatable, better soil holding species to less, or even non-palatable, shallow rooted annuals and perennials. They contribute to stream degradation because they do not perform well the role of riparian vegetation (Thompson and others 1998). These four links added to the 12 links associated with habitat variable six and five links with habitat variable five indicate 84 percent of statistically significant ties between benthic macroinvertebrate data and disturbance are associated with grazing.

Habitat variable # 3 (percent of riparian zone covered by noxious weeds) has an additional two links to disturbance indicative benthic macroinvertebrate data. Young and Evans (1989) have tied deteriorated range conditions due to grazing to spread of noxious weeds in Nevada. These two links, added to the above, give 92 percent of riparian habitat variables associated with disturbance as also associated with disturbance caused by grazing.

Percent of human-caused bare ground, habitat variable # 7, is strongly associated with grazing by Schulz and Leininger (1990.) They state grazed areas in their study had four times more bare ground than ungrazed areas. Adding this link to ones discussed above increases percentage of statistically significant links between benthic macroinvertebrate data and disturbance to 96 percent strongly linked to grazing.

Livestock are attracted to riparian areas because of lush foliage, shade and water, especially in hotter arid months (Ohmart 1996; Skovlin 1984). Fleischner (1994) states livestock grazing is the most widespread land management practice in western North America and that 70 percent of this area is grazed. He further states that ecological implications of grazing can be dramatic. Szaro (1989) feels that degradation of many riparian systems and overgrazing by domestic livestock are integrally related. Armour and others (1994) feel overgrazing by domestic livestock was one of the principal factors contributing to damage and loss of riparian and stream ecosystems in the West. Elmore (1992) attributes degradation and elimination of many riparian systems to improper grazing practices.

In addition to general riparian degradation caused by livestock grazing, the specific cause and effect relationship of riparian vegetation removal and increases in stream temperature is critical. As vegetation is removed and streams lose shading, stream temperatures increase (Ohmart 1996; Beschta 1991; Brazier and Brown 1973). WD shows a significant correlation between intensity of infection and daily mean water temperature (Vincent 1999.)

Habitat variable # 6 (woody species establishment and regeneration) and habitat variable #5 (degree of browse utilization of trees and shrubs) have a total of 17 significant links with benthic macroinvertebrate data indicative of disturbance. This represents 68 percent of total significant links and are clearly tied to grazing. If woody species in these areas were allowed to regenerate, stream temperatures would be lowered and infection rate for WD lessened.

Logistic Regression

Parameter estimates listed in Table 9, results of logistic regression modeling, are actually weighting factors for the habitat variables associated with them. Multiplying scores for each habitat variable listed by its parameter provides a weighted and signed (positive or negative) score for that variable. Summing these scores for each site provides the probability that the stream or reach of stream for which the habitat assessment was done, has or will support WD.

Table 11 is a suggested format of a page to be added to the currently used lotic health assessment form that can be used to determine probability of WD in a reach of stream:

Table 11. Suggested format to be added to currently used lotic health assessment form that can be used to determine probability of WD in a reach of stream

Score for habitat variable # 3	_____	X 0.01 =	_____
Score for habitat variable # 4	_____	X 0.59 =	_____
Score for habitat variable # 6	_____	X 0.36 =	_____
Score for habitat variable #7	_____	X 0.31 =	_____
	Subtotal A		_____
Score for habitat variable #5	_____	X 0.32 =	_____
Score for habitat variable #9	_____	X 0.68 =	_____
	Subtotal B		_____
	Subtotal A		_____
	Minus Subtotal B		_____
	Probability of WD		_____

Although the additional page for the lotic health assessment form as illustrated above provides some predictive power, it is limited for the following reasons:

1. Dates Montana Department of Fish, Wildlife and Parks (MTDFWP) determined presence or absence of WD do not coincide with the dates benthic macroinvertebrate samples used in this study were collected. These dates differ by two years in some cases. For meaningful statistical analysis of the presence or absence of WD relative to benthic macroinvertebrate samples, sampling and the determination of WD presence must be accomplished at the same time.
2. Benthic macroinvertebrates were collected on the same streams that MTFWP

determined presence or absence of WD. However, the location of sampling sites for benthic macroinvertebrates and sites used to determine presence of WD were different. For valid ststistical analysis locations must be the same.

Recommendations to improve predictive power of the logistic regression model are included under suggestions for future research.

Unhealthy streams support the presence of WD. The type of disturbance caused by improper grazing appears to be especially critical to the presence or absence of the disease. Changes in composition of the vegetative community and presence of a healthy "woody" component of that community can be directly linked to grazing. Lack of trees and shrubs in a riparian zone also contribute to temperature regulation of a stream and this is crucial to the TAM phase in the life cycle of the parasite.

The assessment protocol developed by logistic regression, when refined by future research, will allow managers to determine if a stream, or reach of stream, will support WD. Additionally, this procedure will provide managers with a list of strong and weak points for remedial action

SUMMARY

Analysis Method # 1

Of nine habitat variables considered in this study, six have significant statistical links with disturbance in streams and hence with WD:

- Woody species establishment and regeneration (variable # 6)
- Degree of browse utilization of trees and shrubs (variable # 5)
- Percent of the site covered by disturbance induced undesirable herbaceous species (variable # 4)
- Percent of the riparian zone covered by noxious weeds (variable # 3)
- Percent of site with human caused bare ground (variable # 7)
- Incisement (vertical stability of the channel) (variable # 9)

Variables are ranked by total number of significant links with benthic macroinvertebrate data indicative of disturbance.

Analysis Method # 2

Logistical regression enables an assessment procedure to be generated that will give the probability that a reach of stream has or will support WD. A suggested format is provided in the discussion section as Figure 2.

Additional Conclusion

There is a strong association between WD and disturbance caused by grazing. Ninety-six percent of statistical links between riparian habitat variables and benthic macroinvertebrate data indicative of disturbance can be attributed to grazing, whether by livestock or wildlife.

SUGGESTIONS FOR FUTURE RESEARCH

1. The logistic regression model developed in this study lacks sufficient predictive power for reasons discussed previously. To overcome this problem, it is recommended that an additional research project be conducted. This project should provide for a site to be assessed according to previously developed methods (Thompson and others 1998.) Benthic macroinvertebrate samples should be collected at the same time and place as the assessment. Additionally, presence or absence of WD at that site at that time should be determined through analysis of gut contents of *T. tubifex* collected at the site for presence of *M. cerebralis*.

2. It was noted during research and analysis of data for this study that the caddisfly, *Helicopsyche borealis*, may well possess attributes allowing it to be used as an indicator species. It is easily recognizable because of its unique snail shaped shell constructed of sand grains and has transcontinental distribution (Resh and others 1984).

As noted in the discussion section, this caddisfly is an algal grazer and increases with increased periphyton. It also has high tolerance for increased thermal conditions (Vaughn 1984). Further, *T. tubifex* is difficult to identify while *H. borealis* is simple. More importantly, *H. borealis* should increase as shrubby riparian vegetation decreases and water temperature and periphyton increases. The combination of these characteristics make larvae of this benthic macroinvertebrate a possible candidate as an indicator species.

When benthic macroinvertebrate samples are collected, correlations between *H. borealis*, *T. tubifex*, and presence or absence of WD should be examined.

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Appendix A

Sampling Sites Eliminated From Analysis Listed By Record ID Number And Reason Eliminated

Record ID Number	Reason Eliminated
000	No health scores, not included in data base
015	no health scores
028	no elevation
029	"
047	no lat/long, elev.
048	"
049	"
070	"
073	"
076	"
084	no Hill's evenness
092	no elevation
096	no lat/long, elev.
105	no elevation
106	"
107	"
108	"
109	"
111	"
112	"
113	"
114	"
115	"
116	"
117	"
118	"
154	no lat/long, elev.
155	"
156	"
157	"
158	"
159	"
160	"
161	"
162	"
163	"
164	"
165	"
166	"
167	"

Glossary

Collector-gatherers. A functional feeding group of benthic macroinvertebrates that feed on fine sediment that is enriched with particles of organic matter.

CTQ:d. A community tolerance quotient used by the Forest Service.

EPT. Ephemeroptera+Plecoptera+Tricoptera (Mayflies+Stoneflies+Caddisflies) are orders of aquatic insects.

Functional feeding group. A group of insects that feed in the same manner.

Gatherers. A functional feeding group of benthic macroinvertebrates that feed on sediments.

Scrapers. A functional feeding group that acquires its food by grazing periphyton (microfloral growth) off hard surfaces found on the bottom of streams.

Shredders. A functional feeding group of benthic macroinvertebrates that consumes large particles of detritus.