LICHEN DATA FROM THE ATHABASCA OIL SANDS REGION

STATISTICAL REPORT

Prepared for

Janet Marsh
Woods Buffalo Environmental Association
Okotoks, Alberta, Canada

June 2005

by

Gregory Brenner

Pacific Analytics
PO Box 219
Albany, OR 97321
(541) 926-0117

LICHEN DATA FROM THE ATHABASCA OIL SANDS REGION

STATISTICAL REPORT

I. TABLE OF CONTENTS

II. EXECUTIVE SUMMARY

This is a report of statistical analysis of the apportionment of variance within the Lichen Data from the Athabasca Oil Sands Region. The report answers three questions of interest. They are:

1. What is the relative error due to laboratory analysis, as measured with laboratory replicates?
2. What is the error due to field collections, as measured by field replicates?
3. What is the error due to pollution deposition among sites?
4. Do the concentrations of elements in lichens change with distance from mine sites?

The report includes an introduction to the analysis, a description of the statistical procedures, and the results of the analyses.

The results are presented in three chapters, one dealing with each question of interest.

III. INTRODUCTION

Lichens are a unique life form, consisting of a relationship between a fungus and a photosynthetic partner, a cyanobacterium or a green alga. The association is said to be symbiotic, such that the fungus provides moisture and shelter for the algal cells allowing them live even in places that otherwise would be unsuitable for them. Due to this symbiotic relationship, lichens are able to live in some of the harshest habitats on earth. Lichens are extremely widespread in nature; they occur from arid desert regions to the Arctic and grow on bare soil, tree trunks, and rocks. Lichens grow very slowly, often less than a millimeter per year.

Lichens lack any outside covering, or cuticle, and consequently are directly exposed to the atmosphere, which they depend upon for their nutrients and water. Moistened lichen tissues act as blotters, soaking up chemicals or materials deposited on their surfaces. This feature has also made them highly susceptible to air pollutants; and lichens are perhaps the plant species most susceptible to sulfur dioxide, heavy metals, and acid rain.

Since lichens are very sensitive to pollutants, they are sometimes used as indicators of air and water pollution. Lichens are investigated at a number of locations surrounding a point or area pollution source, or at a number of locations within an area of interest. Appropriate lichen metrics are recorded at each location and are related to known or inferred pollution levels. The metrics include distribution of individual indicator species, frequency or abundance of individual species, species richness (total number of different species) at each study location, total lichen cover, and element content of lichen samples.

When assessing pollution with lichens it is appropriate to apportion the variance of the data into its various sources. Variability in element content is dependent upon the ability to collect a representative sample, the precision of laboratory measurement, and the natural variability among sites due to environmental variables such as air pollution, rainfall, temperature, and substrate.

Questions of Interest

1. What is the relative error due to laboratory analysis, as measured with laboratory replicates?
2. What is the error due to field collections, as measured by field replicates?
3. What is the error due to elemental deposition among sites?
4. Do the concentrations of elements in lichens change with distance from mine sites?

Species of Interest

Element content was measured from two lichen species:

1 – Evemes = *Evernia mesomorpha*

2 – Hypphy = *Hypogymnia physodes*

Elements

Twenty-eight elements were detected in laboratory analysis:

Percent Nitrogen, Percent Sulfur, parts per million (ppb for Hg) of Al, B, Ba, Be, Ca, Cd, Co, Cr, Cu, Fe, Hg, K, Li, Mg, Mn, Mo, Na, Ni, P, Pb, Rb, Si, Sr, Ti, V, Zn.

IV. STATISTICAL PROCEDURES

The data were compiled and delivered to Pacific Analytics on April 14, 2005 for analysis. The element content of two lichen species were delivered in spreadsheet format with columns for Lab ID # and Sample Name that was coded for Site, Direction to Mine, Distance to Mine, Species acronym, and replicate code.

Estimation of Components of Variance

Variance components models are used when there is interest in variability of one or more variables other than the residual error. One of the goals of this study was to estimate components of variation due to field measurement and to laboratory analysis. For this purpose Sampling Sites are regarded as fixed. When there are several samples within a Site, the model for the response variable for Site (S), and sample (replicate), is:

$$y_{ij} = \mu + \xi_i + \varepsilon_{ij}$$

where ξ is the fixed effect of Site, and ε is an independent random variable with zero mean and variance σ_S^2 (variance of replicate samples within Sites) (Montgomery 1991).

A maximum likelihood method was used to estimate the variance components. The maximum likelihood process constructs a likelihood function, which is a function of the model parameters. The maximum likelihood method makes use of a Newton-Raphson computing algorithm that iterates until the log-likelihood objective function converges (Searle et al., 1992). The maximum likelihood estimators that result are those values of the parameters from the parameter space that maximize the value of the likelihood function (Milliken and Johnson

1992). The restricted maximum likelihood method is similar to the maximum likelihood method, but it first separates the likelihood into two parts; one that contains the fixed effects and one that does not (Patterson and Thompson 1971). A brief review of the restricted maximum likelihood estimators appears in Anderson et al. (1986).

For the tasks presented in this report, restricted maximum likelihood estimators were obtained for each of the data sets. Software used for all statistical analyses was S-Plus 2000 (MathSoft 1988-1999).

Data Reduction

The sampling units in this study are the sites at which lichen samples were collected. In most cases more than one sample of each species was taken at a site. These field replicate samples are sub-samples of the sampling unit and are not appropriate for analysis without accounting for the structure of the sampling.

For this study, the site mean response of the sub-samples for each element for each species was calculated and used as the response variable for that site.

Twenty-eight elements were detected in laboratory analysis. In each of the species, some of the samples had element concentrations below the detectable limit. Statistical analysis was not conducted for those elements that had greater than about 50% of the samples below detectable limits. These include Beryllium (Be), Cadmium (Cd), Cobalt (Co), Lead (Pb), and Rubidium (Rb) for *E. mesomorpha*. For the same reason, statistical analysis was not performed for Cobalt (Co) and Rubidium (Rb) for *H. physodes*.

Linear Regression

Regression analysis describes statistical relationships between a response variable and several explanatory variables. In the case of lichen data, the response variables are the measurements of element concentration in lichen samples taken at several sites. The explanatory variables are cardinal directions and distance from mine sites.

Knowledge about the statistical relationship between variables enables one to determine certain characteristics of the response variable from the measured explanatory variables. In mathematical terms the relationship looks like

$$\mu = \beta_0 + \beta_1 X_1 + \beta_2 X_2$$

where μ = the mean of the response distribution, β_0 is the intercept coefficient, β_1 is the coefficient for the first explanatory variable, X_1, and β_2 is the coefficient for the next explanatory variable, X_2.

The regression coefficients are calculated using Least Squares Estimation. These parameters are estimated and each has its own associated standard error. An estimate for the value of the response variable can be calculated by inserting the values of the explanatory variables into the equation. The difference between the estimated value and the observed measured value is called the residual. The Least Squares Estimation procedure minimizes the squared residuals while estimating the regression coefficients.

There are several conditions under which Least Squares Estimation is most efficient. The first is that the response distributions for each set of explanatory values have approximately the same standard deviations. The next is that the distributions of the response variables are approximately Normal (Gaussian).

The third is that each response is drawn independently of all the other responses.

Least Squares Regression is useful for modeling the fit of a response variable to several continuous explanatory variables. In this study, the distance from the mine is one such continuous variable. Least Squares Regression can also be used to distinguish between two or more groups or categories. Indicator (or Dummy) variables are set up for each of the groups (or levels of a factor) but one. The indicator variables take on values of "1" or "0", indicating the attribute is present or absent. In the previous AOS Statistical Report, this method was used for the analysis of the effect of direction on the concentration of elements in lichens. Because most sampling was done in only one direction the method was not necessary for the analyses in the current report

The fit of the model to the assumptions can be assessed by examination of the distribution of points in residual plots. These plots are the graph of the residuals on the Y axis versus the fitted values on the X axis. Inequalities of standard error are easily detected from these graphs. Usually transformations (e.g., log or square root transformations) will correct for departures from equality. In other cases, it is necessary to explore transformation of the explanatory variables. All determinations for transformation were made after examination of the distribution of the points in residual plots.

Ordination in Element Space

Non-metric multidimensional scaling (NMS) ordination was used to ordinate the lichen tissue samples in elemental space (20 elements) for both lichen species combined. Ordination summarizes complex relationships among samples based on their elemental content. NMS is an iterative technique that is capable of extracting non-linear gradients in elemental space (Kruskal 1964; Mather 1976). Sørensen distance measure was used with the Slow-and-Thorough autopilot settings in PC-ORD 4.0 (40 runs of 400 iterations; McCune & Mefford 1999). The NMS ordination was rotated to maximize the correlation of distance from the mine sites with axis 1.

Only 20 elements were ordinated; elements were not excluded when a high percentage of samples were below the MDL (see Table C2). Elemental concentrations in lichens were log-transformed for the ordination analysis to reduce noise in the data. Multivariate outlier analysis found no extreme outliers (lichen tissue samples > 3 standard deviations from the grand mean) at either locality. The Pearson's correlations of the individual elements and distance from the oil sands facilities with the ordination axes are reported.

Ordination in Lichen Species Space

We used non-metric multidimensional scaling (NMS) ordination to ordinate stands in lichen species space. The ordination was used to assess gradients in lichen community composition and their relationship to air pollution, topographic features and forest stand characteristics. NMS is an iterative technique that is capable of extracting non-linear gradients in species space (Kruskal 1964; Mather 1976). Sørensen distance measure was used with the Slow-and-Thorough autopilot settings in PCORD 4.0 (40 runs of 400 iterations;

McCune & Mefford 1999). Lichen abundance codes (described above) were converted to a scale of 1 to 10 for data analysis. Lichen species present in < 5% of the stands were excluded from the ordination analysis, eliminating 24 rare species to reduce noise in the dataset. After deletion of rare species, multivariate outlier analysis of all Muskeg stands found no stands that were potential outliers.

The correlations of environmental variables (topographic and stand characteristics, species diversity measures, and concentration of elements in Hypogymnia physodes) were assessed with the ordination axes (using Pearson's correlation). Distance from the mine sites and element concentrations in H. physodes were used to represent the potential pollution gradient.

V. QUALITY ASSURANCE ANALYSIS

Detection Limits

The method detection limit (MDL) is 2 times the standard deviation of eleven replicates of a reagent water sample (blank). The percent of samples below the MDL was determined for each element by lichen species. Many elements were below the MDL in lichen samples (Table C2).

All *Evernia mesomorpha* samples were below the MDL for Rubidium (Rb) and Cobalt (Co). Beryllium (Be), Cadmium (Cd), and Lead (Pb) concentrations were below the MDL for > 50% of *E. mesomorpha* samples. Consequently, none of these elements were evaluated in the statistical analyses for the elemental data. Although a large percentage of *E. mesomorpha* samples were below the MDL for Lithium (Li – 48%) and Molybdenum (Mo – 18%) these elements were included in statistical analyses.

Almost all *Hypogymnia physodes* samples were below the MDL for Rubidium (Rb – 98%) and Cobalt (Co – 98%). Consequently, none of these elements were evaluated in the statistical analyses for the elemental data. No other elements were below the MDL for > 50% of *H. physodes* samples. Although a large percentage of *H. physodes* samples were below the MDL for Lithium (Li – 44%) and Beryllium (Be – 48%) these elements were included in statistical analyses.

Table C2. The percentage of samples with element concentrations below the MDL, reported by element for each lichen species. Only elements with samples below the MDL are shown.

Element	***Evernia Mesomorpha***	***Hypogymnia physodes***
Beryllium	92%	48%
Cadmium	74%	6%
Cobalt	100%	98%
Lead	88%	0%
Lithium	48%	44%
Molybdenum	18%	6%
Rubidium	100%	98%

Laboratory contamination

The laboratory contamination was assessed using acid blanks. Element concentrations in blanks were above the MDL for Calcium and Silicon. Concentration of most elements in blanks was < 1% of the grand mean for field samples with the exception of Beryllium, Boron, Cadmium, Cobalt, Copper, Chromium, Molybdenum, Nickel, Rubidium, Sodium, Titanium, and Vanadium (Table C3).

Table C3. Contamination in acid blanks measured by ICP-AES at the UMRAL. Values are in µg/g dry weight. Average values are reported for 2 acid blanks and the grand mean for that element is reported for the lichen samples across all species (n = 125). Percent of the grand mean is: (blank/grand mean) *100.

Element	Symbol	MDL Units (dry wt.)	Average Blank	Grand Mean	% of Grand Mean
Aluminum	Al	3.58 µ/g	3.58	743.15	0.48
Barium	Ba	0.12 µ/g	0.12	15.92	0.75
Beryllium	Be	0.04 µ/g	0.04	0.05	87.61
Boron	B	0.46 µ/g	0.46	3.73	12.32
Cadmium	Cd	0.12 µ/g	0.12	0.19	64.37
Calcium	Ca	0.82 µ/g	2.43	7583.87	0.03
Chromium	Cr	0.28 µ/g	0.28	1.10	25.55
Cobalt	Co	0.24 µ/g	0.24	0.24	100.00
Copper	Cu	0.52 µ/g	0.52	2.18	23.84
Iron	Fe	0.34 µ/g	0.34	1069.90	0.03
Lead	Pb	1.68 µ/g	1.68	2.38	70.70
Lithium	Li	0.40 µ/g	0.40	0.59	68.28
Magnesium	Mg	3.80 µ/g	3.80	512.71	0.74
Manganese	Mn	0.06 µ/g	0.06	114.91	0.06
Mercury	Hg	0.005 µ/g	0.005	154.08	0.03
Molybdenum	Mo	0.22 µ/g	0.22	0.42	52.54
Nickel	Ni	0.44 µ/g	0.44	1.90	23.21
Phosphorus	P	0.70 µ/g	0.70	600.72	0.12
Potassium	K	14.14 µ/g	14.14	2181.65	0.65
Rubidium	Rb	53.0 µ/g	53.00	53.00	100.00
Silicon	Si	1.00 µ/g	2.27	729.38	0.31
Sodium	Na	3.60 µ/g	3.60	31.67	11.37
Strontium	Sr	0.06 µ/g	0.06	9.68	0.62
Titanium	Ti	0.30 µ/g	0.30	12.69	2.36
Vanadium	V	0.36 µ/g	0.36	3.66	9.85
Zinc	Zn	0.14 µ/g	0.14	43.14	0.32

Laboratory replicates.

Replicate analyses of twelve selected sample digests were run. This determined the laboratory precision for repeated measures of the same sample. Relative percent difference (RPD) between the laboratory replicates was determined to assess laboratory precision using the equation:

$$RPD = ABS|(x1 - x2)| \ / \ (mean\ x1, x2) *100$$

where ABS is the absolute value; x1 is the first lab measurement and x2 is the repeated lab measurement. Re-runs of lichen samples were also considered lab replicates and were used to estimate laboratory precision.

Table C5. Laboratory precision is measured by the average relative percent difference, comparing two measurements of the same sample. The mean, median, minimum and maximum relative percent differences are reported (RPD). Sulfur and Nitrogen comparisons were made with 12 lab replicates; and results for all other elements were made with comparisons of 11 lab replicates. RPD was not calculated for samples near or at the MDL or for mercury (Hg).

Element	Units	Mean RPD	Median RPD	Min. RPD	Max. RPD
Nitrogen	%	4.81	3.04	0.27	14.80
Sulphur	%	6.46	3.16	0.09	39.08
Aluminum	ppm	3.66	2.26	0.11	13.07
Barium	ppm	3.78	3.19	0.35	12.47
Beryllium	ppm	3.76	3.67	1.04	6.67
Boron	ppm	4.69	4.81	0.06	9.54
Cadmium	ppm	13.44	3.60	0.97	63.27
Calcium	ppm	3.59	1.01	0.39	20.33
Chromium	ppm	9.94	4.43	0.45	69.48
Copper	ppm	3.84	1.78	0.16	21.08
Iron	ppm	3.12	2.20	0.57	15.01
Lead	ppm	5.69	5.65	1.00	10.27
Lithium	ppm	5.89	4.01	0.35	18.38
Magnesium	ppm	2.16	0.83	0.20	13.02
Manganese	ppm	1.81	0.58	0.02	12.11
Molybdenum	ppm	8.36	4.53	1.10	40.76
Nickel	ppm	9.74	6.23	0.00	42.97
Phosphorous	ppm	0.85	0.83	0.01	2.15
Potassium	ppm	1.14	1.22	0.36	2.00
Silicon	ppm	5.82	4.75	0.42	23.14
Sodium	ppm	3.33	1.98	0.27	9.10
Strontium	ppm	1.79	1.44	0.16	4.39
Titanium	ppm	5.38	2.92	1.02	15.67
Vanadium	ppm	3.71	2.16	0.29	13.28
Zinc	ppm	1.62	1.06	0.35	7.13

VI. VARIANCE COMPONENTS

Presented in Tables F6 and F7 are the estimated (REML) variance components for field replication for *Evernia mesomorpha* and *Hypogymnia physodes* respectively. Reported for each element in Part a. of each table are the Variances of Sites and Field Duplicates, a Total (Sum) of the two variances, and the proportion of the total variance for each component. Reported for each element in Part b. of each table are the Variances of Sites and Laboratory Duplicates, a Total (Sum) of the two variances, and the proportion of the total variance for each component.

Table F6. Part a. Field Replicate Variation of Evernia mesomorpha.

	ELEMENT							
SOURCE OF VARIATION	N (%)	S (%)	Al (ppm)	B (ppm)	Ba (ppm)	Ca (ppm)	Cr (ppm)	Cu (ppm)
Sample Site	0.01	0.00	11184.95	2.13	5.03	2238028.00	0.14	0.54
Field Duplicate	0.00	0.00	270.82	0.40	0.05	607418.50	0.01	0.03
TOTAL VARIATION	0.02	0.00	11455.77	2.54	5.08	2845446.50	0.15	0.57
PROPORTION Site	0.79	0.77	0.98	0.84	0.99	0.79	0.92	0.95
PROPORTION Field Duplicate	0.21	0.23	0.02	0.16	0.01	0.21	0.08	0.05

SOURCE OF VARIATION	Fe (ppm)	K (ppm)	Li (ppm)	Mg (ppm)	Mn (ppm)	Mo (ppm)	Na (ppm)	Ni (ppm)
Sample Site	294036.90	3.66	0.05	8705.56	139.24	0.03	133.49	0.18
Field Duplicate	743.05	39.29	0.01	19.47	68.99	0.00	0.06	0.01
TOTAL VARIATION	294779.95	42.95	0.06	8725.03	208.24	0.03	133.54	0.18
PROPORTION Site	1.00	0.09	0.82	1.00	0.67	0.96	1.00	0.97
PROPORTION Field Duplicate	0.00	0.91	0.18	0.00	0.33	0.04	0.00	0.03

SOURCE OF VARIATION	P (ppm)	Si (ppm)	Sr (ppm)	Ti (ppm)	V (ppm)	Zn (ppm)	Hg (ppb)
Sample Site	4952.18	37583.97	5.09	8.34	0.84	26.34	2982.09
Field Duplicate	12881.98	2363.94	0.02	0.22	0.02	2.83	26.45
TOTAL VARIATION	17834.16	39947.91	5.11	8.56	0.86	29.17	3008.54
PROPORTION Site	0.28	0.94	1.00	0.97	0.98	0.90	0.99
PROPORTION Field Duplicate	0.72	0.06	0.00	0.03	0.02	0.10	0.01

Table F6. Part b. Laboratory Replicate Variation of Evernia mesomorpha.

	ELEMENT							
SOURCE OF VARIATION	**N (%)**	**S (%)**	**Al (ppm)**	**B (ppm)**	**Ba (ppm)**	**Ca (ppm)**	**Cr (ppm)**	**Cu (ppm)**
Sample Site	0.01	0.00	97548.65	2.23	5.40	2585566.00	0.15	0.60
Laboratory Duplicate	0.00	0.00	0.00	0.00	0.00	701.55	0.00	0.00
TOTAL VARIATION	0.01	0.00	97548.65	2.23	5.40	2586267.55	0.15	0.60
PROPORTION Site	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
PROPORTION Laboratory Duplicate	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00

SOURCE OF VARIATION	**Fe (ppm)**	**K (ppm)**	**Li (ppm)**	**Mg (ppm)**	**Mn (ppm)**	**Mo (ppm)**	**Na (ppm)**	**Ni (ppm)**
Sample Site	316719.00	84506.06	0.06	9331.42	155.17	0.03	140.27	0.19
Laboratory Duplicate	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
TOTAL VARIATION	316719.00	84506.06	0.06	9331.42	155.17	0.03	140.27	0.19
PROPORTION Site	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
PROPORTION Laboratory Duplicate	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00

SOURCE OF VARIATION	**P (ppm)**	**Si (ppm)**	**Sr (ppm)**	**Ti (ppm)**	**V (ppm)**	**Zn (ppm)**
Sample Site	9729.37	39441.00	5.27	9.62	0.90	28.24
Laboratory Duplicate	0.00	0.00	0.00	0.00	0.00	0.00
TOTAL VARIATION	9729.37	39441.00	5.27	9.62	0.90	28.24
PROPORTION Site	1.00	1.00	1.00	1.00	1.00	1.00
PROPORTION Laboratory Duplicate	0.00	0.00	0.00	0.00	0.00	0.00

Table F7. Part a. Field Replicate Variation of *Hypogymnia physodes*.

	ELEMENT								
SOURCE OF VARIATION	**N (%)**	**S (%)**	**Al (ppm)**	**B (ppm)**	**Ba (ppm)**	**Be (ppm)**	**Ca (ppm)**	**Cd (ppm)**	**Cr (ppm)**
Sample Site	0.01	0.00	52093.20	0.85	25340740.00	0.00	25340740.00	0.01	0.16
Field Duplicate	0.00	0.00	10.36	0.01	1.00	0.00	1.00	0.00	0.01
TOTAL VARIATION	0.01	0.00	52103.56	0.86	25340741.00	0.00	25340741.00	0.01	0.17
PROPORTION Site	0.98	0.00	1.00	0.99	1.00	0.90	1.00	1.00	0.95
PROPORTION Field Duplicate	0.02	1.00	0.00	0.01	0.00	0.10	0.00	0.00	0.05

SOURCE OF VARIATION	**Cu (ppm)**	**Fe (ppm)**	**K (ppm)**	**Li (ppm)**	**Mg (ppm)**	**Mn (ppm)**	**Mo (ppm)**	**Na (ppm)**	**Ni (ppm)**
Sample Site	0.22	302958.80	41040.03	0.04	23437.04	14617.96	0.02	114.32	0.43
Field Duplicate	0.01	72351.99	74365.44	0.00	16661.18	0.03	0.00	0.14	0.00
TOTAL VARIATION	0.22	375310.79	115405.47	0.04	40098.22	14617.99	0.02	114.45	0.43
PROPORTION Site	0.96	0.81	0.36	1.00	0.58	1.00	1.00	1.00	1.00
PROPORTION Field Duplicate	0.04	0.19	0.64	0.00	0.42	0.00	0.00	0.00	0.00

SOURCE OF VARIATION	**P (ppm)**	**Pb (ppm)**	**Si (ppm)**	**Sr (ppm)**	**Ti (ppm)**	**V (ppm)**	**Zn (ppm)**	**Hg (ppb)**
Sample Site	16957.42	0.29	13343.23	74.48	12.18	1.18	321.43	495.27
Field Duplicate	13305.73	0.02	32431.79	0.19	1.54	0.05	0.15	1.10
TOTAL VARIATION	30263.15	0.31	45775.02	74.67	13.72	1.23	321.58	496.37
PROPORTION Site	0.56	0.93	0.29	1.00	0.89	0.96	1.00	1.00
PROPORTION Field Duplicate	0.44	0.07	0.71	0.00	0.11	0.04	0.00	0.00

Table F7. Part b. Laboratory Replicate Variation of *Hypogymnia physodes*.

	ELEMENT								
SOURCE OF VARIATION	**N (%)**	**S (%)**	**Al (ppm)**	**B (ppm)**	**Ba (ppm)**	**Be (ppm)**	**Ca (ppm)**	**Cd (ppm)**	**Cr (ppm)**
Sample Site	0.01	0.00	92458.14	0.88	69.94	0.00	46371156.00	0.01	0.13
Laboratory Duplicate	0.00	0.00	0.00	0.00	0.00	0.00	35.32	0.00	0.08
TOTAL VARIATION	0.01	0.00	92458.14	0.88	69.94	0.00	46371191.32	0.01	0.21
PROPORTION Site	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	0.61
PROPORTION Laboratory Duplicate	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.39

SOURCE OF VARIATION	**Cu (ppm)**	**Fe (ppm)**	**K (ppm)**	**Li (ppm)**	**Mg (ppm)**	**Mn (ppm)**	**Mo (ppm)**	**Na (ppm)**	**Ni (ppm)**
Sample Site	0.24	358646.00	77017.61	0.04	36408.47	14724.06	0.02	117.08	0.41
Laboratory Duplicate	0.00	0.00	5.84	0.00	0.00	0.00	0.00	0.00	0.04
TOTAL VARIATION	0.24	358646.00	77023.45	0.04	36408.47	14724.06	0.02	117.08	0.44
PROPORTION Site	1.00	1.00	1.00	1.00	1.00	1.00	0.89	1.00	0.91
PROPORTION Laboratory Duplicate	0.00	0.00	0.00	0.00	0.00	0.00	0.11	0.00	0.09

SOURCE OF VARIATION	**P (ppm)**	**Pb (ppm)**	**Si (ppm)**	**Sr (ppm)**	**Ti (ppm)**	**V (ppm)**	**Zn (ppm)**
Sample Site	35920.07	0.32	43367.08	82.23	12.51	1.24	331.86
Laboratory Duplicate	0.00	0.00	0.00	0.00	0.00	0.00	0.00
TOTAL VARIATION	35920.07	0.32	43367.08	82.23	12.51	1.24	331.86
PROPORTION Site	1.00	1.00	1.00	1.00	1.00	1.00	1.00
PROPORTION Laboratory Duplicate	0.00	0.00	0.00	0.00	0.00	0.00	0.00

VII. REGRESSIONS

All regressions were run with natural log-transformed elemental concentrations and natural log-transformed Distance from mine measurements. Transformations were necessary to stabilize the variance. The need for transformation was determined through visual inspection of the residual graphs from the regression analyses. Potential outliers (i.e., lichen tissue samples) were assessed; however, no outliers were severe enough to affect any of the regression analyses. Consequently, all samples above the MDL were included in the analyses.

Results from regression analyses are reported for each species by element. Statistically significant terms in the models were those coefficients with p-values = 0.05. All regression coefficients with p-values between 0.05 and 0.10 were indicative of "suggestive evidence." The intercept and slope are reported for each regression analysis with associated standard errors. The p-values are reported for the distance coefficient in the regression analysis. The tables also include the R-Squared value for the regressions. The R-Squared value is a measure of how well the model fit the data and is generally interpreted as the amount of variation in the data explained by the model. Software used for all regression analyses was S-Plus version 2000 (MathSoft 1999).

Table D2. Results from regression analyses of the effects of distance on elemental concentrations in *Evernia mesomorpha* on the Firebag transect. Results are reported by element. The intercept and slope are also reported for the model, with associated standard errors (SE). The p-values for the Distance coefficient and regression R-Squared values are reported.

Element	Intercept	SE	Distance	SE	T-statistic	P-Value	R-Squared
N	0.130	0.118	-0.047	0.015	-3.205	0.005	0.363
S	-2.213	0.165	-0.042	0.020	-2.065	0.054	0.192
Al	7.762	0.361	-0.220	0.045	-4.943	0.000	0.576
B	0.874	0.413	0.017	0.051	0.327	0.748	0.006
Ba	2.168	0.505	-0.089	0.062	-1.421	0.173	0.101
Ca	7.387	0.284	-0.119	0.035	-3.377	0.003	0.388
Cr	1.476	0.545	-0.229	0.067	-3.409	0.003	0.392
Cu	1.360	0.209	-0.126	0.026	-4.891	0.000	0.571
Fe	8.626	0.404	-0.332	0.050	-6.636	0.000	0.710
K	7.712	0.178	-0.034	0.022	-1.565	0.135	0.120
Mg	6.786	0.204	-0.176	0.025	-6.967	0.000	0.730
Mo	0.324	0.363	-0.194	0.045	-4.330	0.000	0.510
Mn	2.896	0.880	0.050	0.109	0.464	0.648	0.012
Na	4.502	0.523	-0.143	0.065	-2.216	0.040	0.214
Ni	0.983	0.404	-0.126	0.050	-2.528	0.021	0.262
P	6.124	0.367	-0.004	0.045	-0.079	0.938	0.000
Si	7.524	0.321	-0.154	0.040	-3.888	0.001	0.456
Sr	2.438	0.516	-0.236	0.064	-3.700	0.002	0.432
Ti	3.514	0.307	-0.154	0.038	-4.059	0.001	0.478
V	0.960	0.418	-0.043	0.052	-0.824	0.421	0.036
Zn	3.386	0.241	0.002	0.030	0.074	0.942	0.000
Hg	4.636	0.250	0.090	0.031	2.912	0.009	0.320

Table D3. Results from regression analyses of the effects of distance on elemental concentrations in *Evernia mesomorpha* on the Muskeg a and b transects. Results are reported by element. The intercept and slope are also reported for the model, with associated standard errors (SE). The p-values for the Distance coefficient and regression R-Squared values are reported.

Element	Intercept	SE	Distance	SE	T-statistic	P-Value	R-Squared
N	0.300	0.199	-0.066	0.028	-2.322	0.034	0.252
S	-1.989	0.325	-0.053	0.046	-1.135	0.273	0.075
Al	8.054	0.334	-0.182	0.048	-3.814	0.002	0.476
B	2.796	0.230	-0.169	0.033	-5.140	0.000	0.623
Ba	3.361	0.343	-0.199	0.049	-4.066	0.001	0.508
Ca	10.520	0.692	-0.405	0.099	-4.099	0.001	0.512
Cr	1.230	0.275	-0.146	0.039	-3.732	0.002	0.465
Cu	0.605	0.556	0.013	0.079	0.159	0.876	0.002
Fe	8.367	0.431	-0.167	0.062	-2.723	0.015	0.317
K	7.990	0.217	-0.046	0.031	-1.483	0.157	0.121
Li	1.100	0.352	-0.196	0.050	-3.896	0.001	0.487
Mg	6.943	0.279	-0.147	0.040	-3.693	0.002	0.460
Mo	0.192	0.431	-0.130	0.062	-2.116	0.050	0.219
Mn	3.751	0.428	-0.027	0.061	-0.439	0.667	0.012
Na	4.565	0.290	-0.142	0.041	-3.415	0.004	0.422
Ni	1.155	0.339	-0.106	0.048	-2.202	0.043	0.233
P	6.848	0.301	-0.088	0.043	-2.056	0.056	0.209
Si	7.411	0.161	-0.093	0.023	-4.032	0.001	0.504
Sr	3.312	0.467	-0.247	0.067	-3.703	0.002	0.461
Ti	3.168	0.283	-0.093	0.040	-2.306	0.035	0.249
V	1.469	0.392	-0.045	0.056	-0.795	0.439	0.038
Zn	3.643	0.319	-0.037	0.046	-0.809	0.431	0.039
Hg	4.417	0.524	0.083	0.075	1.111	0.283	0.072

Table D4. Results from regression analyses of the effects of distance on elemental concentrations in *Hypogymnia physodes* on the Firebag transect. Results are reported by element. The intercept and slope are also reported for the model, with associated standard errors (SE). The p-values for the Distance coefficient and regression R-Squared values are reported.

Element	Intercept	SE	Distance	SE	T-statistic	P-Value	R-Squared
N	-0.129	0.102	-0.040	0.013	-3.121	0.006	0.351
S	-3.913	0.261	0.107	0.032	3.315	0.004	0.379
Al	7.792	0.302	-0.174	0.037	-4.650	0.000	0.546
B	1.036	0.331	-0.003	0.041	-0.060	0.953	0.000
Ba	3.354	0.457	-0.021	0.056	-0.377	0.711	0.008
Ca	8.776	0.535	0.027	0.066	0.407	0.689	0.009
Cd	-0.879	0.718	-0.072	0.089	-0.813	0.427	0.035
Cr	1.639	0.383	-0.224	0.047	-4.728	0.000	0.554
Cu	1.707	0.270	-0.101	0.033	-3.022	0.007	0.337
Fe	8.511	0.312	-0.229	0.039	-5.947	0.000	0.663
K	8.121	0.186	-0.048	0.023	-2.084	0.052	0.194
Mg	7.217	0.265	-0.106	0.033	-3.232	0.005	0.367
Mo	0.808	0.344	-0.237	0.043	-5.583	0.000	0.634
Mn	4.993	0.742	0.054	0.092	0.587	0.564	0.019
Na	4.609	0.613	-0.173	0.076	-2.282	0.035	0.224
Ni	1.477	0.385	-0.084	0.048	-1.756	0.096	0.146
P	6.959	0.398	-0.054	0.049	-1.108	0.282	0.064
Pb	1.550	0.294	-0.049	0.036	-1.352	0.193	0.092
Si	7.467	0.262	-0.137	0.032	-4.231	0.001	0.499
Sr	2.986	0.682	-0.108	0.084	-1.285	0.215	0.084
Ti	4.002	0.282	-0.189	0.035	-5.417	0.000	0.620
V	1.844	0.218	-0.047	0.027	-1.739	0.099	0.144
Zn	4.024	0.440	0.015	0.054	0.276	0.785	0.004
Hg	5.396	0.232	-0.057	0.029	-1.971	0.064	0.178

Table D5. Results from regression analyses of the effects of distance on elemental concentrations in *Hypogymnia physodes* on the Muskeg a and b transects. Results are reported by element. The intercept and slope are also reported for the model, with associated standard errors (SE). The p-values for the Distance coefficient and regression R-Squared values are reported.

Element	Intercept	SE	Distance	SE	T-statistic	P-Value	R-Squared
N	0.128	0.194	-0.081	0.028	-2.935	0.010	0.350
S	-3.146	0.264	0.027	0.038	0.717	0.484	0.031
Al	8.143	0.239	-0.177	0.034	-5.181	0.000	0.627
B	2.272	0.174	-0.118	0.025	-4.738	0.000	0.584
Ba	3.848	0.593	-0.091	0.085	-1.080	0.296	0.068
Be	-1.568	0.223	-0.188	0.032	-5.912	0.000	0.686
Ca	11.401	0.601	-0.239	0.086	-2.790	0.013	0.327
Cd	-0.805	0.451	-0.135	0.064	-2.099	0.052	0.216
Cr	1.532	0.201	-0.169	0.029	-5.873	0.000	0.683
Cu	1.890	0.165	-0.126	0.024	-5.335	0.000	0.640
Fe	8.659	0.291	-0.178	0.041	-4.286	0.001	0.535
K	8.427	0.154	-0.083	0.022	-3.766	0.002	0.470
Li	1.207	0.362	-0.243	0.052	-4.701	0.000	0.580
Mg	7.482	0.247	-0.108	0.035	-3.054	0.008	0.368
Mo	0.378	0.275	-0.152	0.039	-3.871	0.001	0.484
Mn	4.059	0.449	0.104	0.064	1.621	0.125	0.141
Na	4.270	0.280	-0.122	0.040	-3.045	0.008	0.367
Ni	1.572	0.249	-0.072	0.036	-2.029	0.060	0.205
P	7.396	0.318	-0.128	0.045	-2.825	0.012	0.333
Pb	1.387	0.244	-0.050	0.035	-1.426	0.173	0.113
Si	7.688	0.159	-0.126	0.023	-5.563	0.000	0.659
Sr	4.693	0.677	-0.245	0.097	-2.537	0.022	0.287
Ti	3.400	0.209	-0.093	0.030	-3.116	0.007	0.378
V	1.514	0.334	0.026	0.048	0.549	0.591	0.018
Zn	3.701	0.269	0.023	0.038	0.604	0.555	0.022
Hg	4.376	0.289	0.033	0.041	0.796	0.438	0.038

Scatter plots

Graphs show the variation of element concentrations in tissue samples of the three lichen species with distance from the mines. Sites with field duplicate lichen samples are shown as the average value for the two samples. Linear lines are drawn in the graphs to show correlations of element concentrations with distance from the mines by species. Lichen samples are symbol and color coded by species: *Hypogymnia physodes* and *Evernia mesomorpha*. Values for the element concentrations and distance are shown on the natural log-scale.

The interpretation of the Distance coefficients are provided below the graphs in which the slope was significantly different from zero and where the R-Squared value was greater than 0.500.

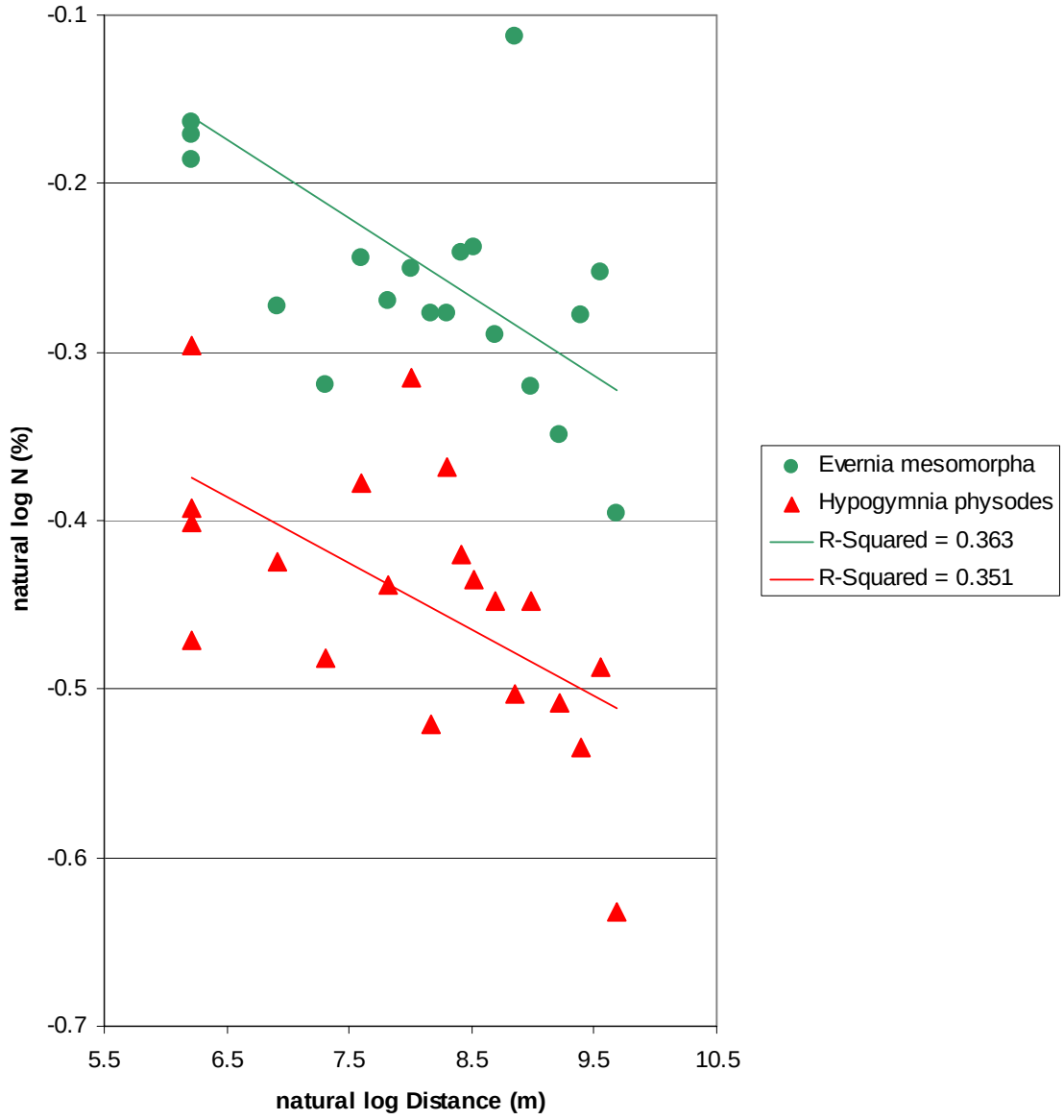

Figure G1. Total percent nitrogen (dry weight) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

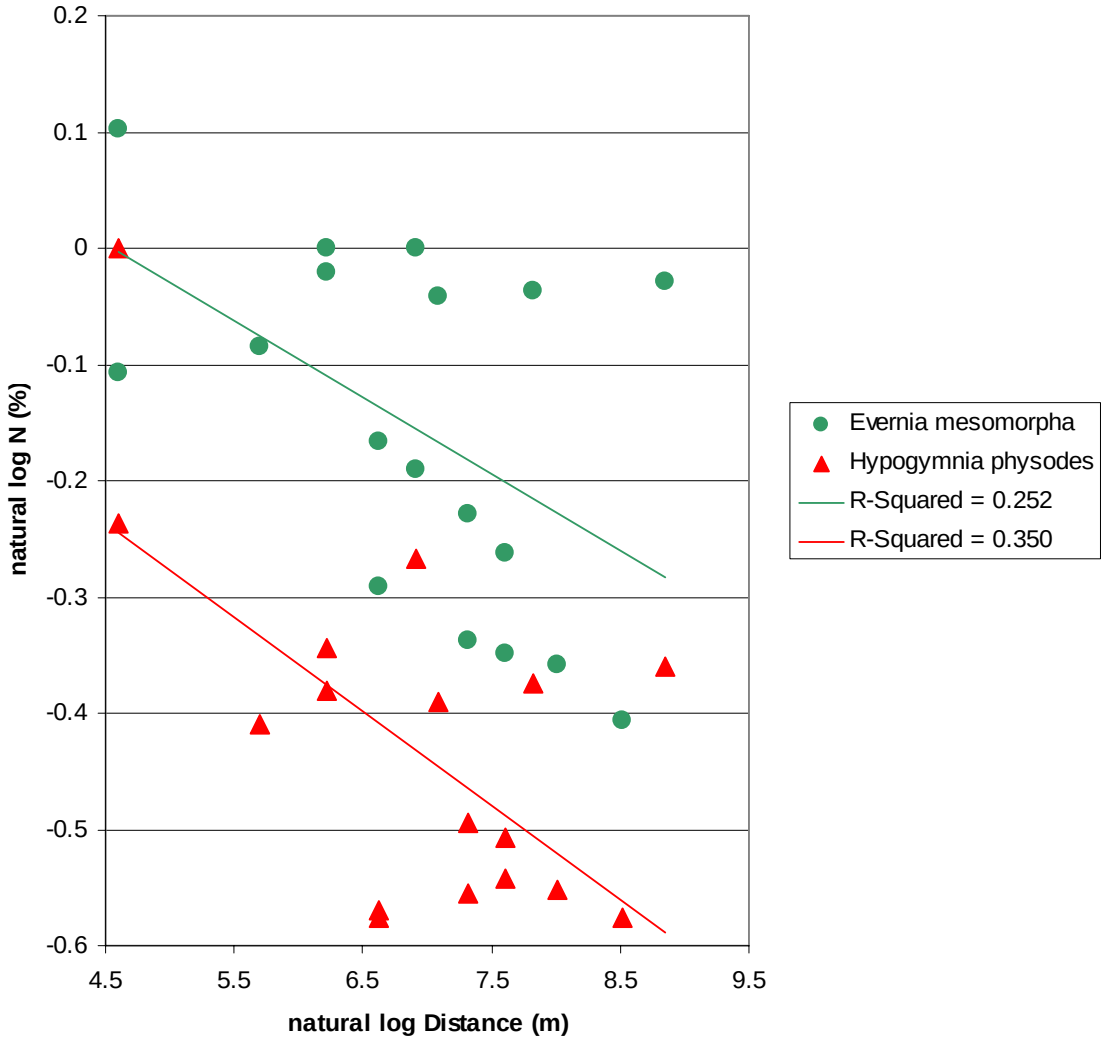

Figure G2. Total percent nitrogen (dry weight) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

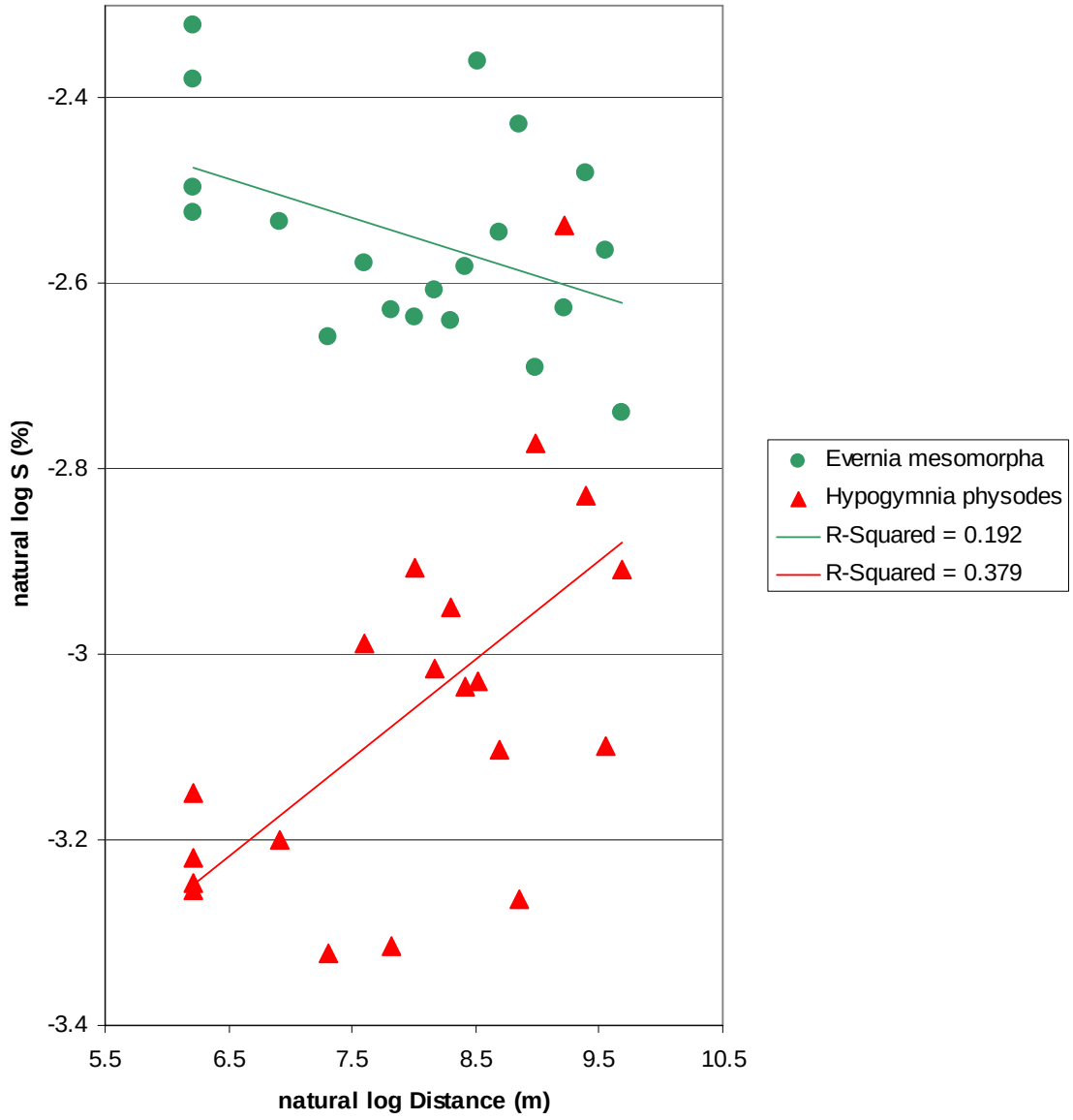

Figure G3. Total percent sulfur (dry weight) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

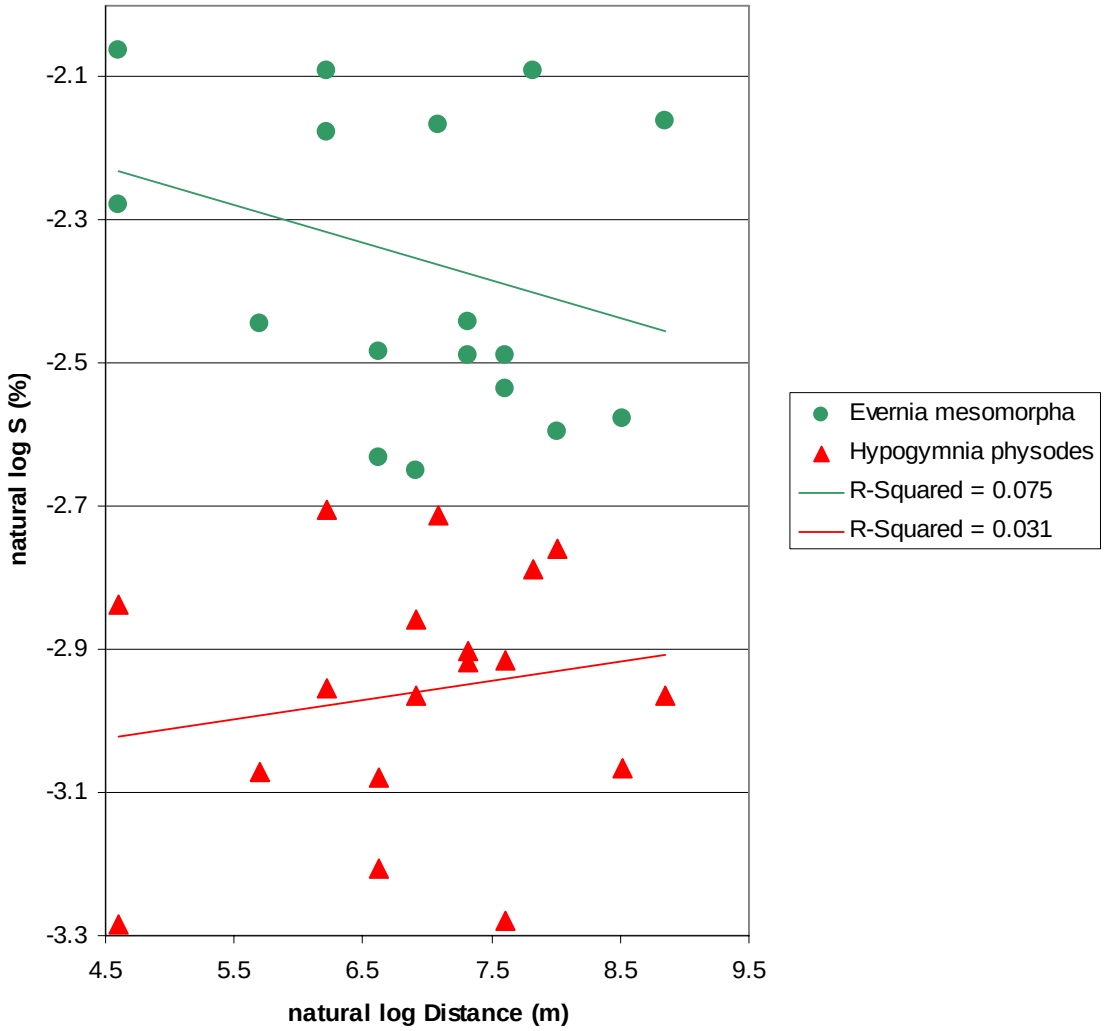

Figure G4. Total percent sulfur (dry weight) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

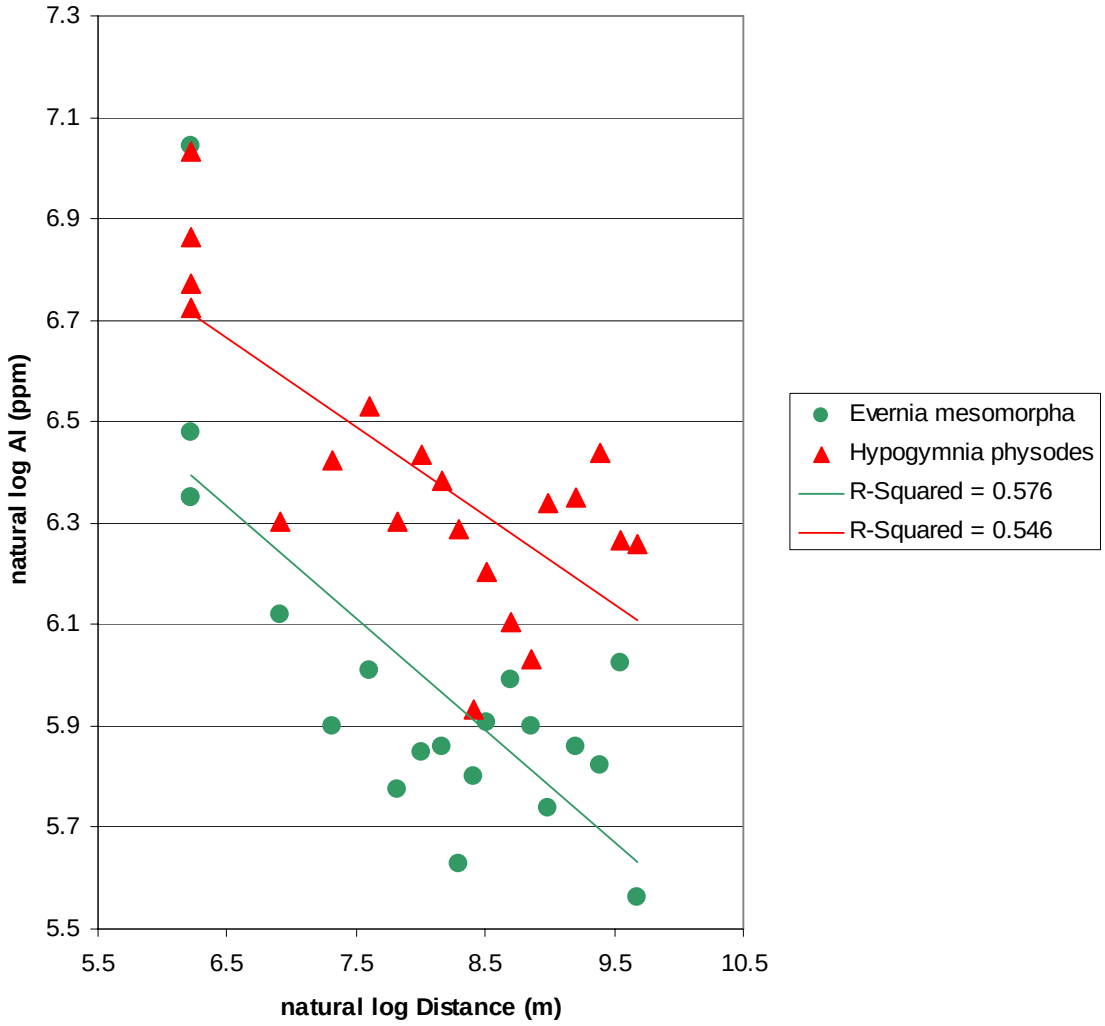

Figure G5. Total Aluminum (ppm) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples. For *E. mesomorpha*, as the distance from the mine doubles the concentration of Al decreases by about 14%. For *H. physodes*, as the distance from the mine doubles the concentration of Al decreases by 13%. For both lichen species, the concentration of Al is much higher within 500 m of the mines.

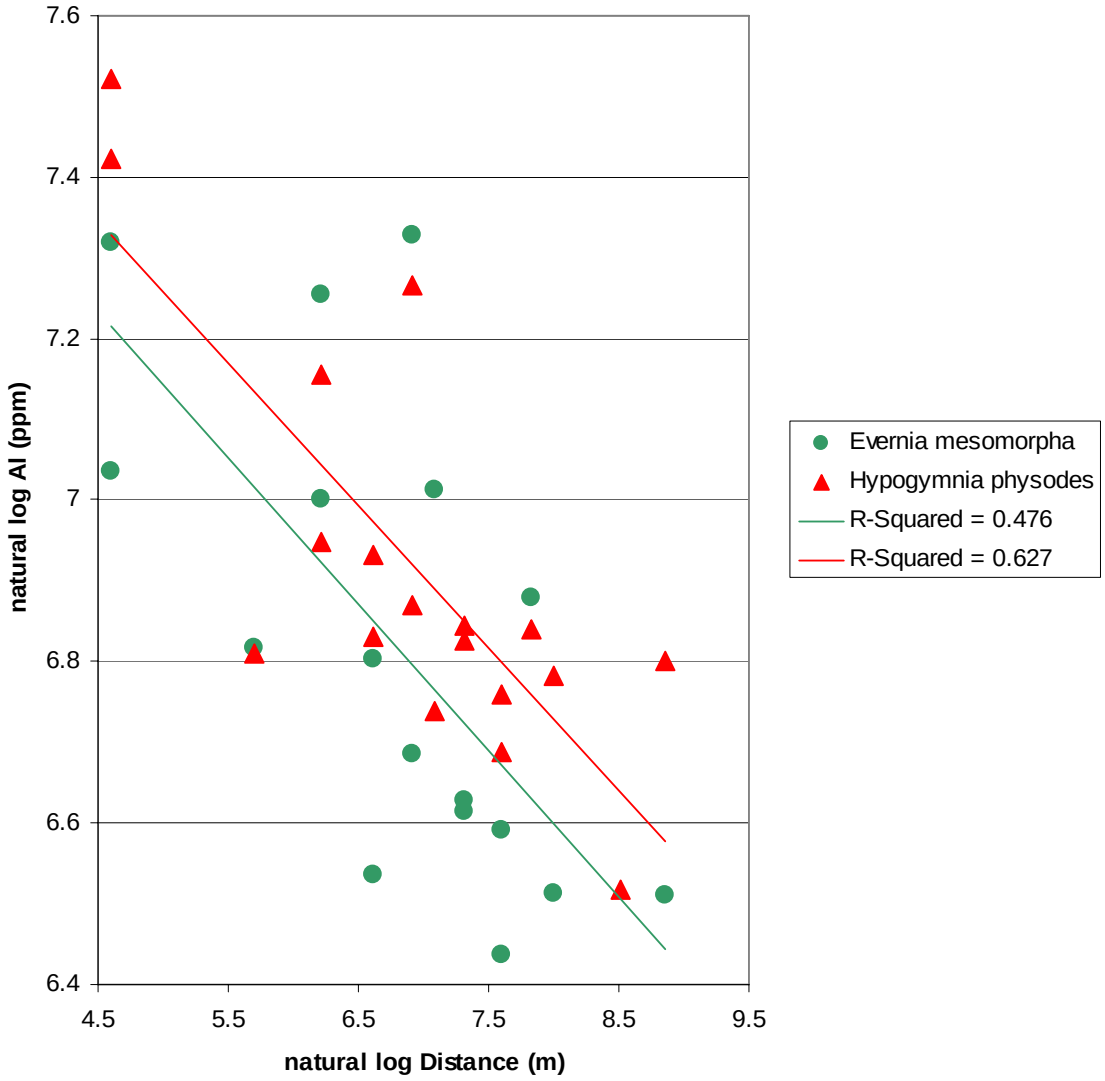

Figure G6. Total Aluminum (ppm) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes* and *Evernia mesomorpha* samples. For *H. physodes*, as the distance from the mine doubles the concentration of Al decreases by 12%. For both lichen species, the concentration of Al is much higher within 100 m of the mines.

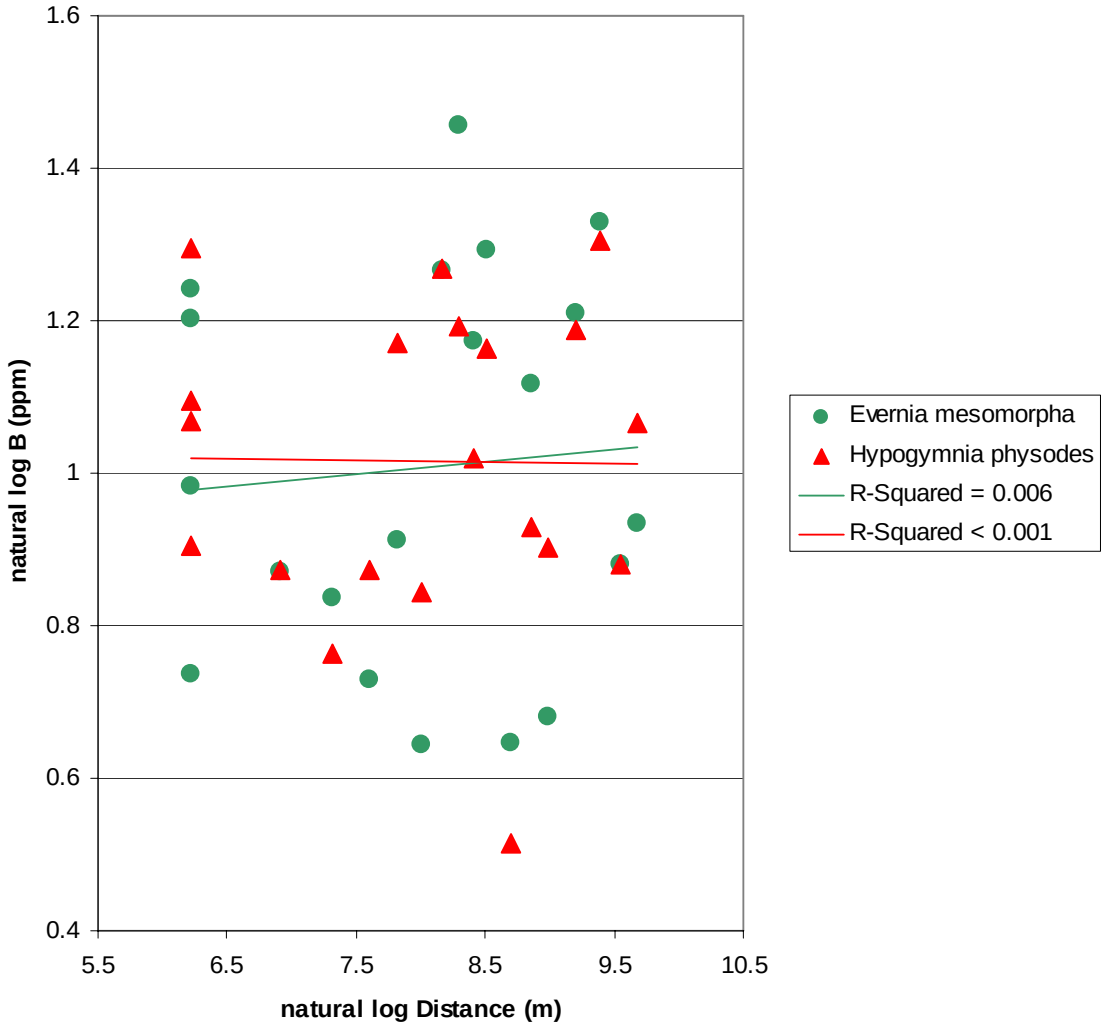

Figure G7. Total Boron (ppm) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

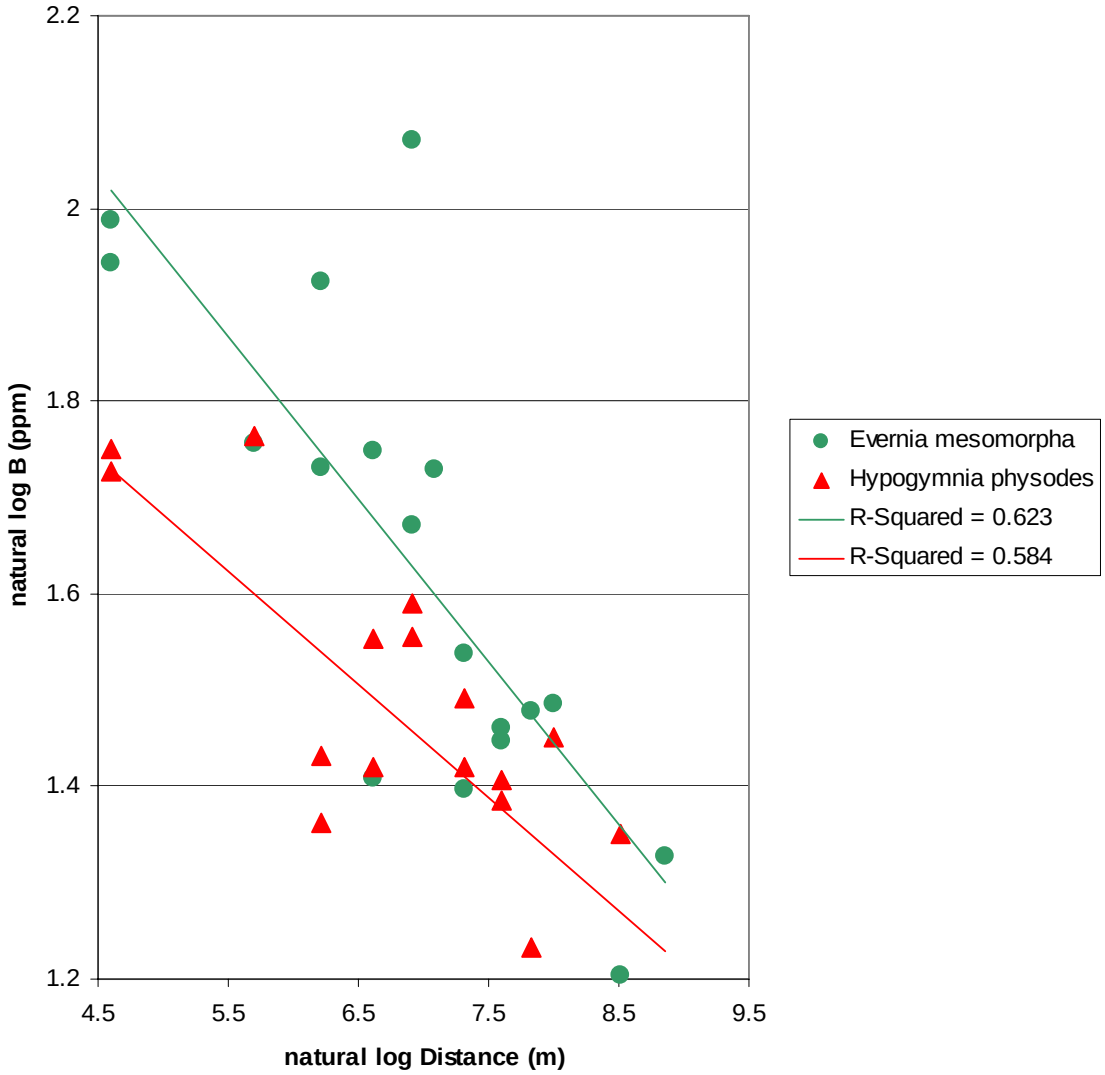

Figure G8. Total Boron (ppm) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes* and *Evernia mesomorpha* samples. For *E. mesomorpha*, as the distance from the mine doubles the concentration of B decreases by 11%. For *H. physodes*, as the distance from the mine doubles the concentration of B decreases by 8%.

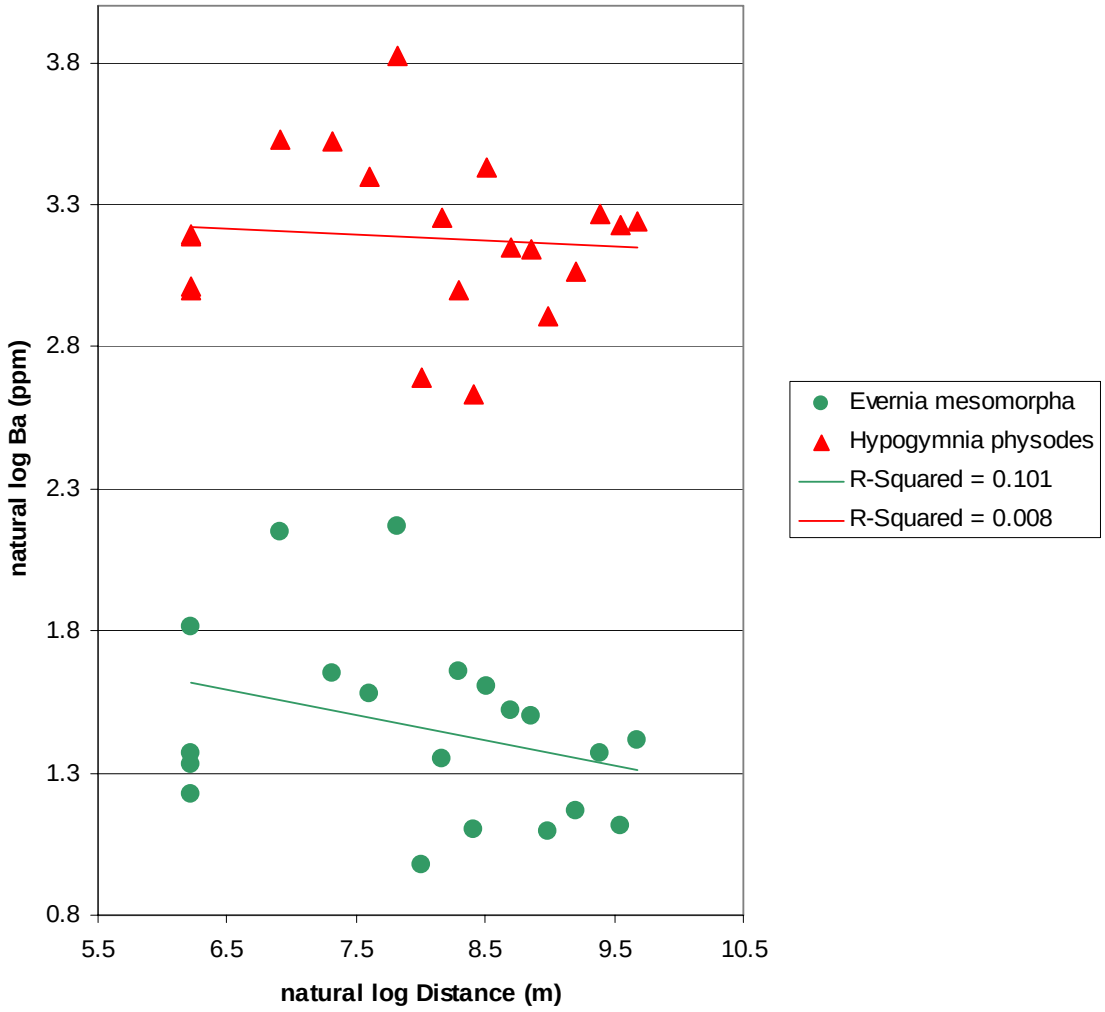

Figure G9. Total Barium (ppm) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

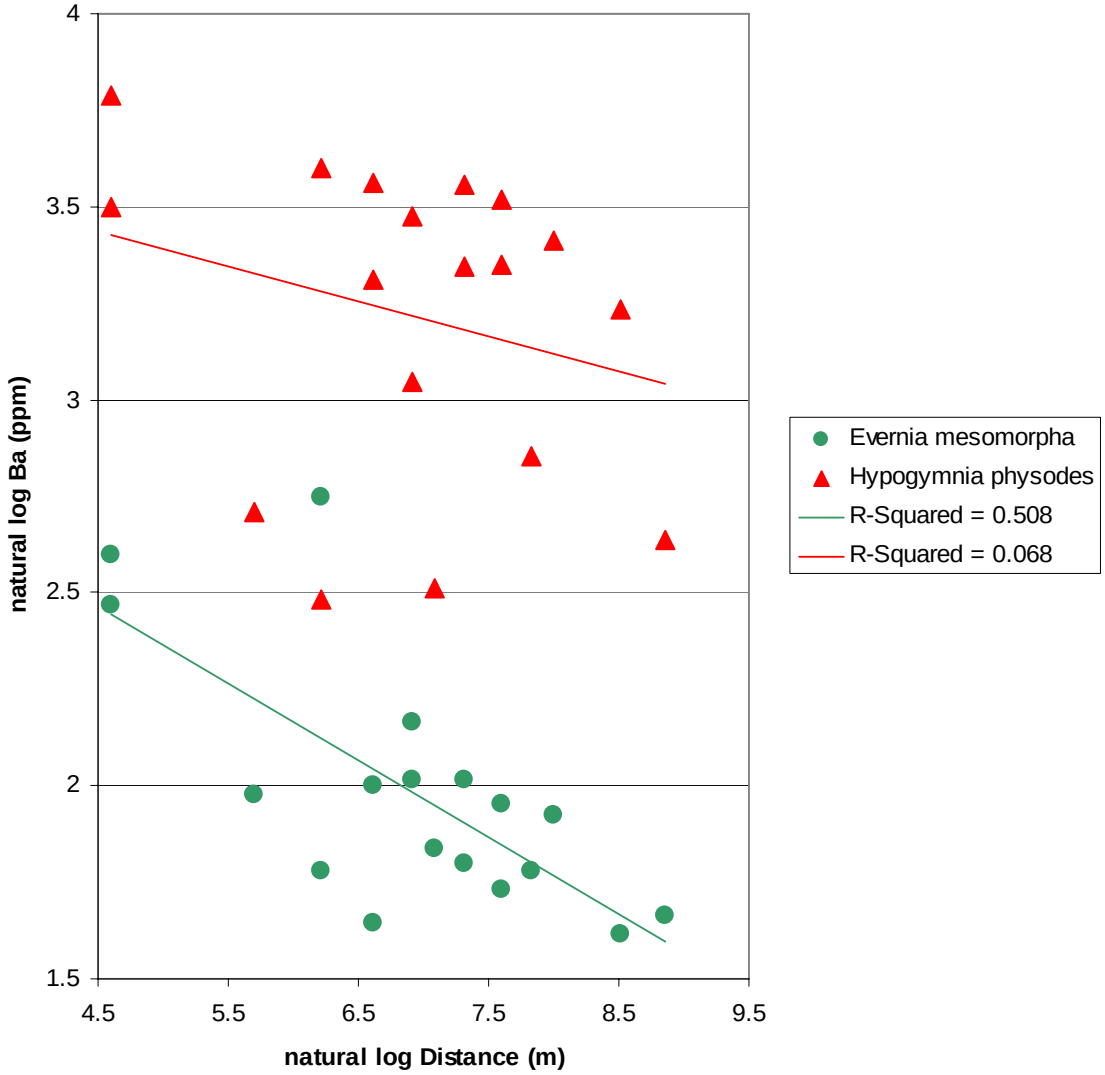

Figure G10. Total Barium (ppm) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes* and *Evernia mesomorpha* samples. For *E. mesomorpha*, as the distance from the mine doubles the concentration of Ba decreases by 13%. For both lichen species, the concentration of Ba is much higher within 100 m of the mines.

Figure G11. Total Beryllium (ppm) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples. All samples were below MDL therefore no regression was analyzed.

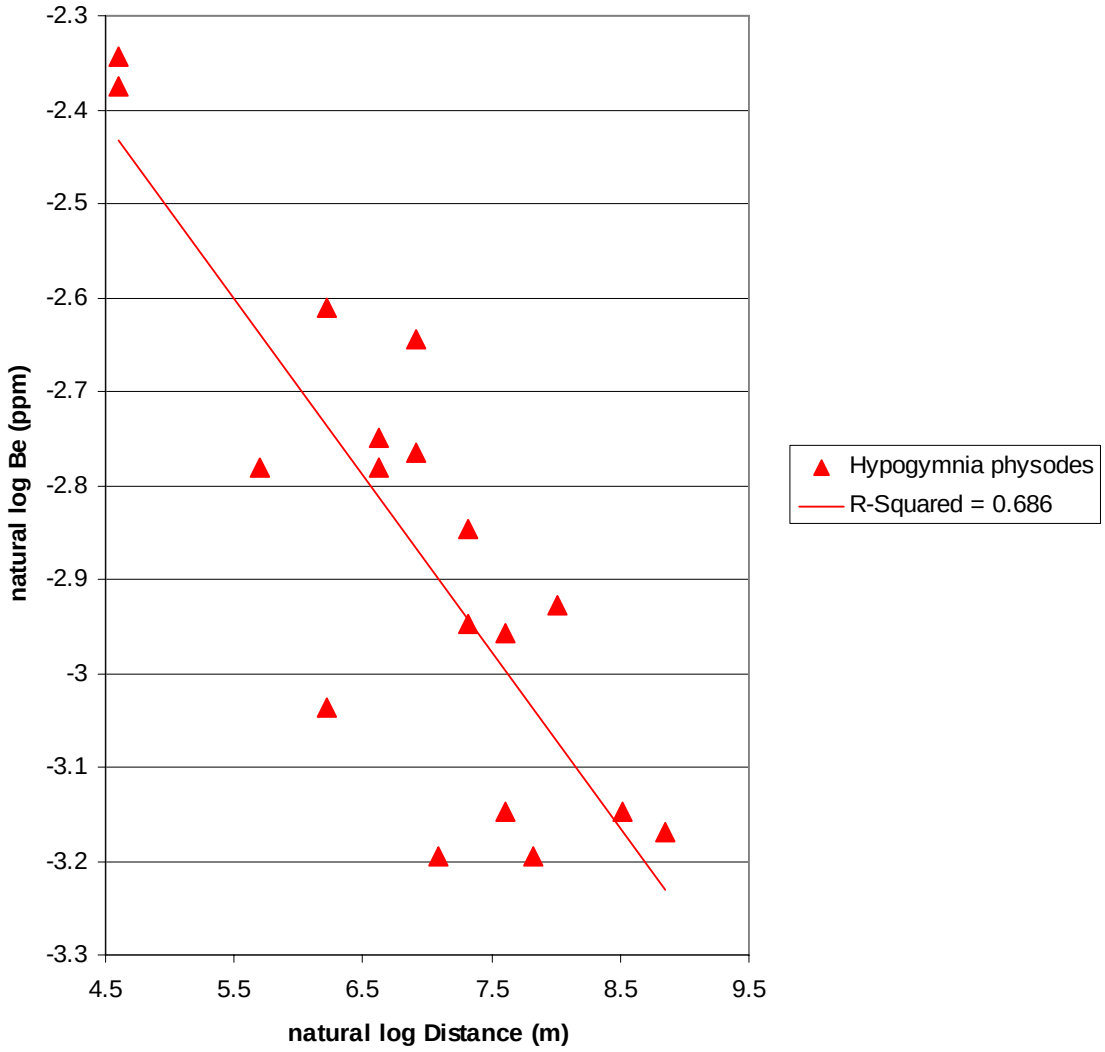

Figure G12. Total Beryllium (ppm) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes.* All samples of *Evernia mesomorpha* were below the MDL and no regression was analyzed. For *H. physodes*, as the distance from the mine doubles the concentration of Be decreases by 12%, and the concentration of Be is much higher within 100 m of the mines.

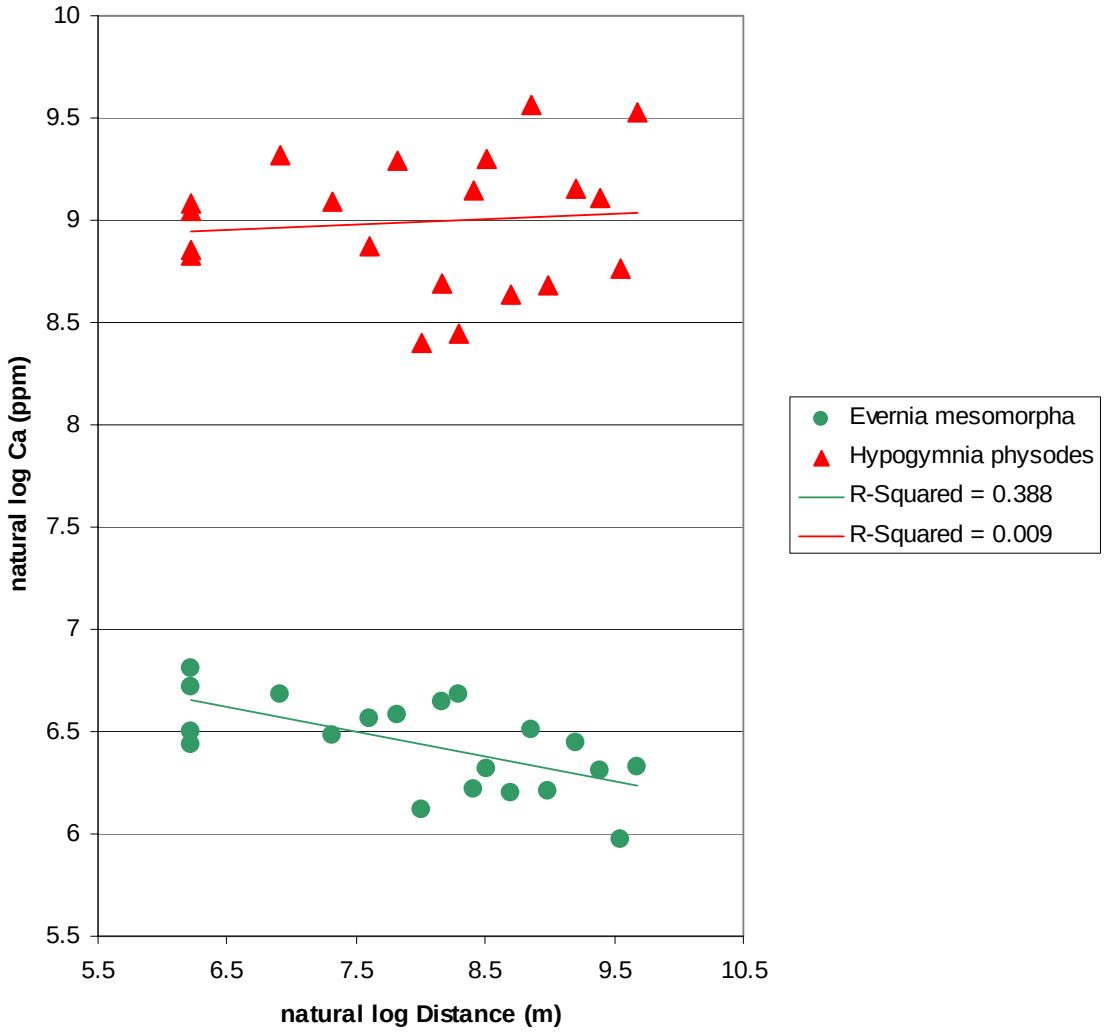

Figure G13. Total Calcium (ppm) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

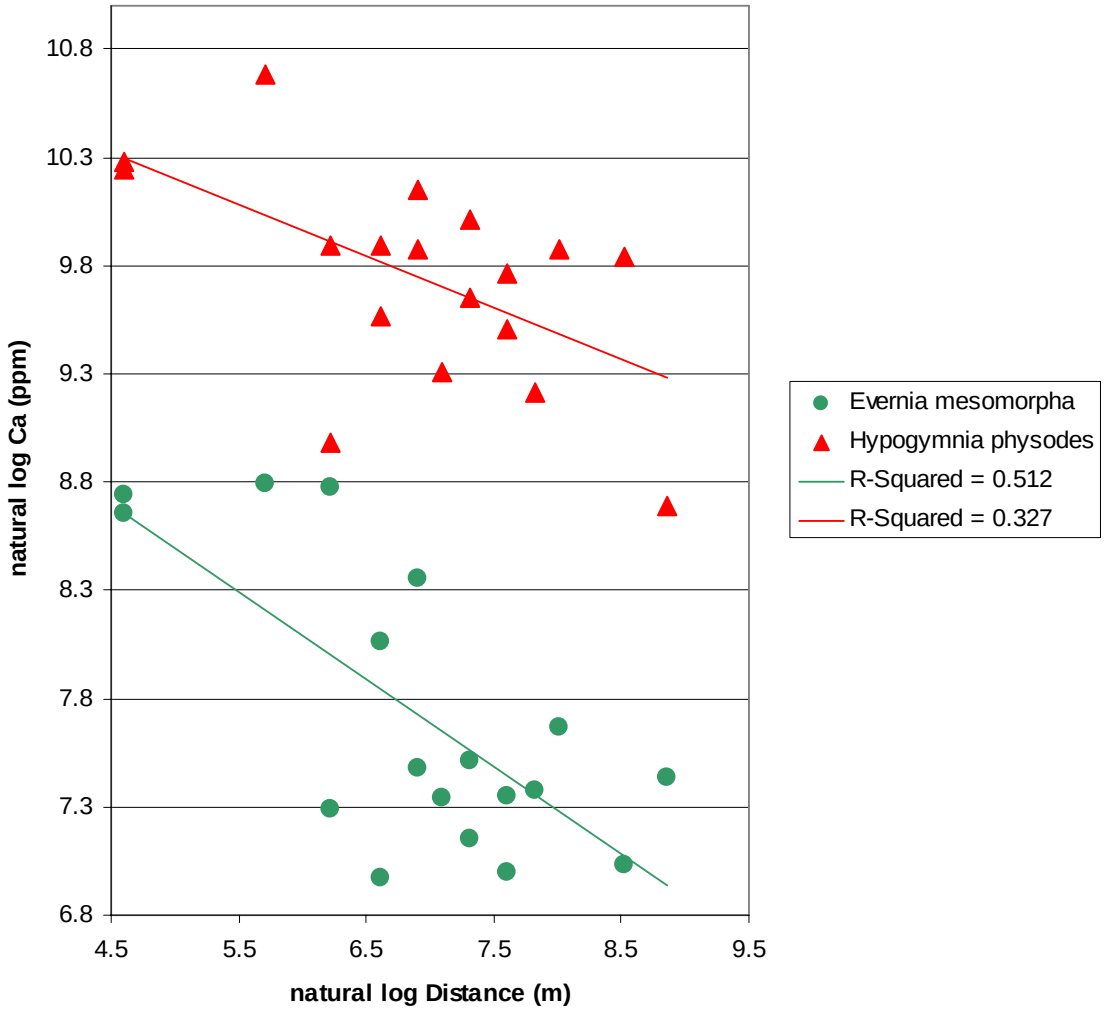

Figure G14. Total Calcium (ppm) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes* and *Evernia mesomorpha* samples. For *E. mesomorpha*, as the distance from the mine doubles the concentration of Ca decreases by 24%.

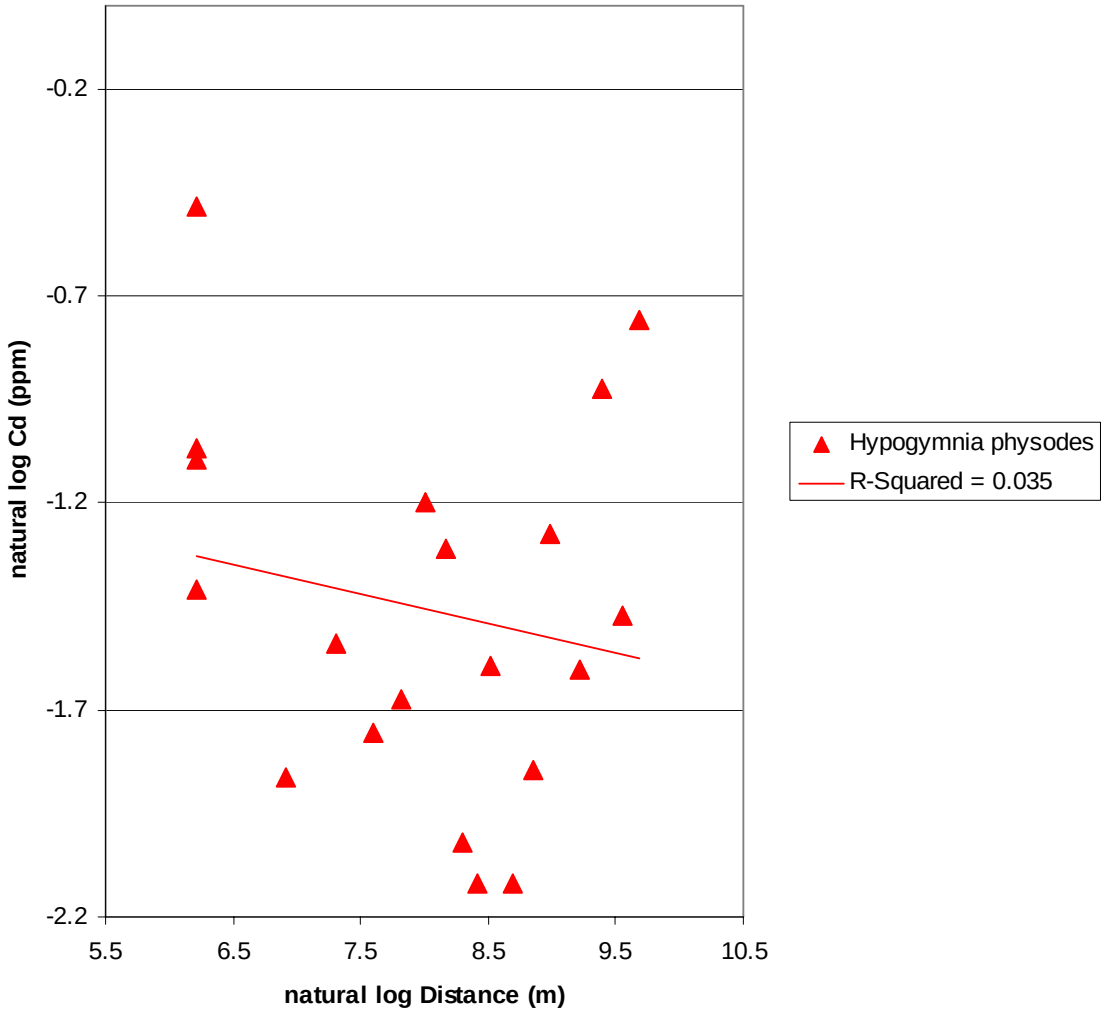

Figure G15. Total Cadmium (ppm) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes.* More than 50% of the samples of *Evernia mesomorpha* had Cd concentrations below the MDL therefore regression was not analyzed.

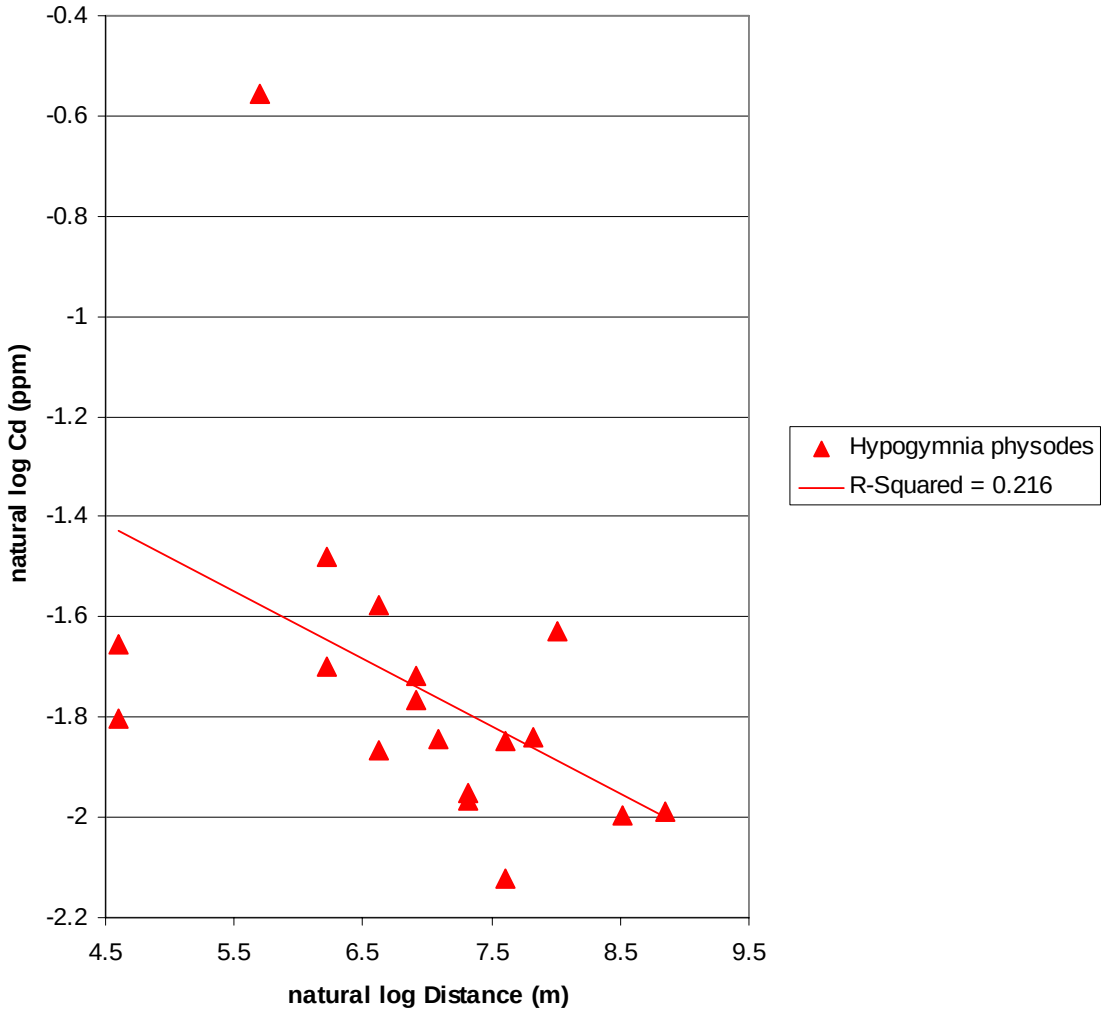

Figure G16. Total Cadmium (ppm) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes*. More than 50% of the samples of *Evernia mesomorpha* had Cd concentrations below the MDL therefore regression was not analyzed.

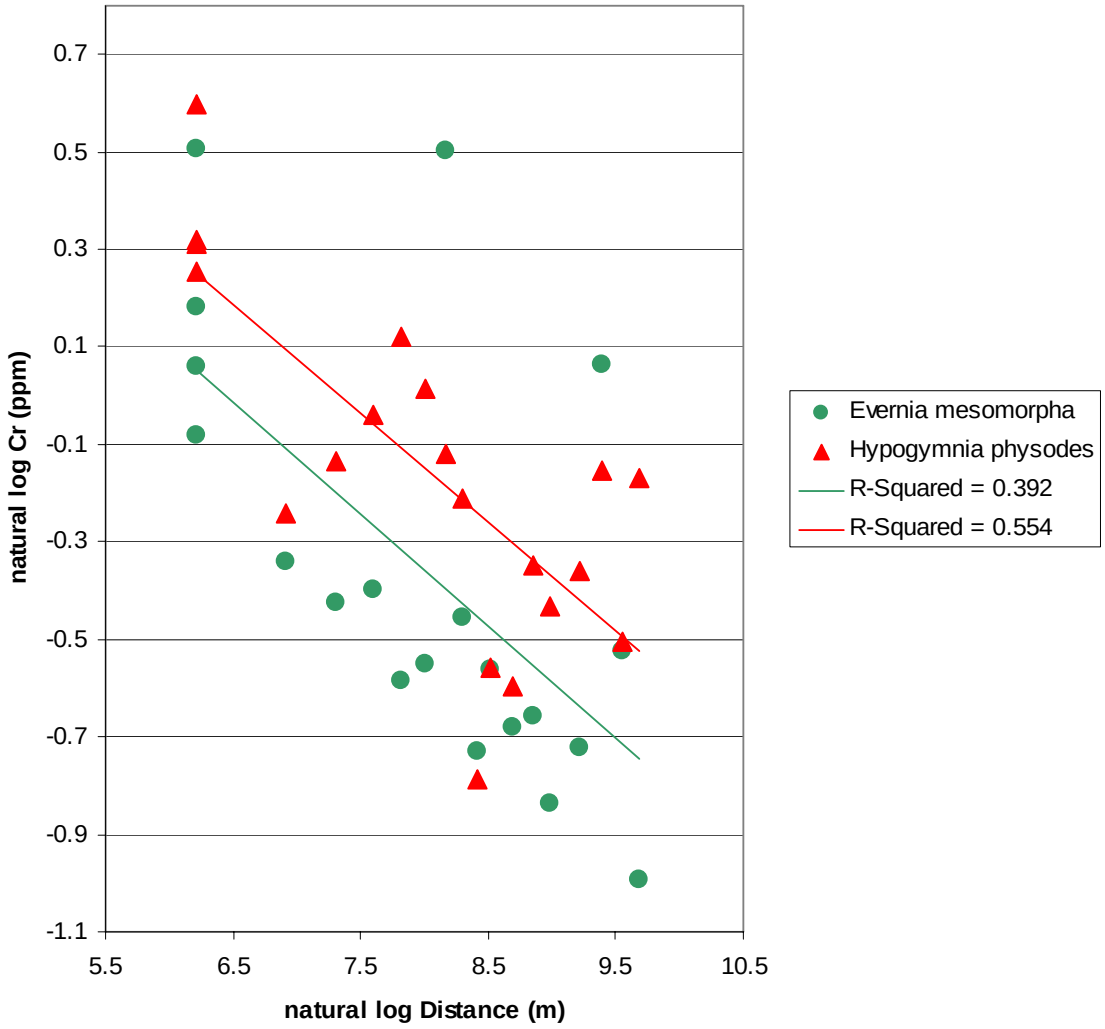

Figure G17. Total Chromium (ppm) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples. For *H. physodes*, as the distance from the mine doubles the concentration of Cr decreases by 14%. For both lichen species, the concentration of Al is much higher within 500 m of the mines.

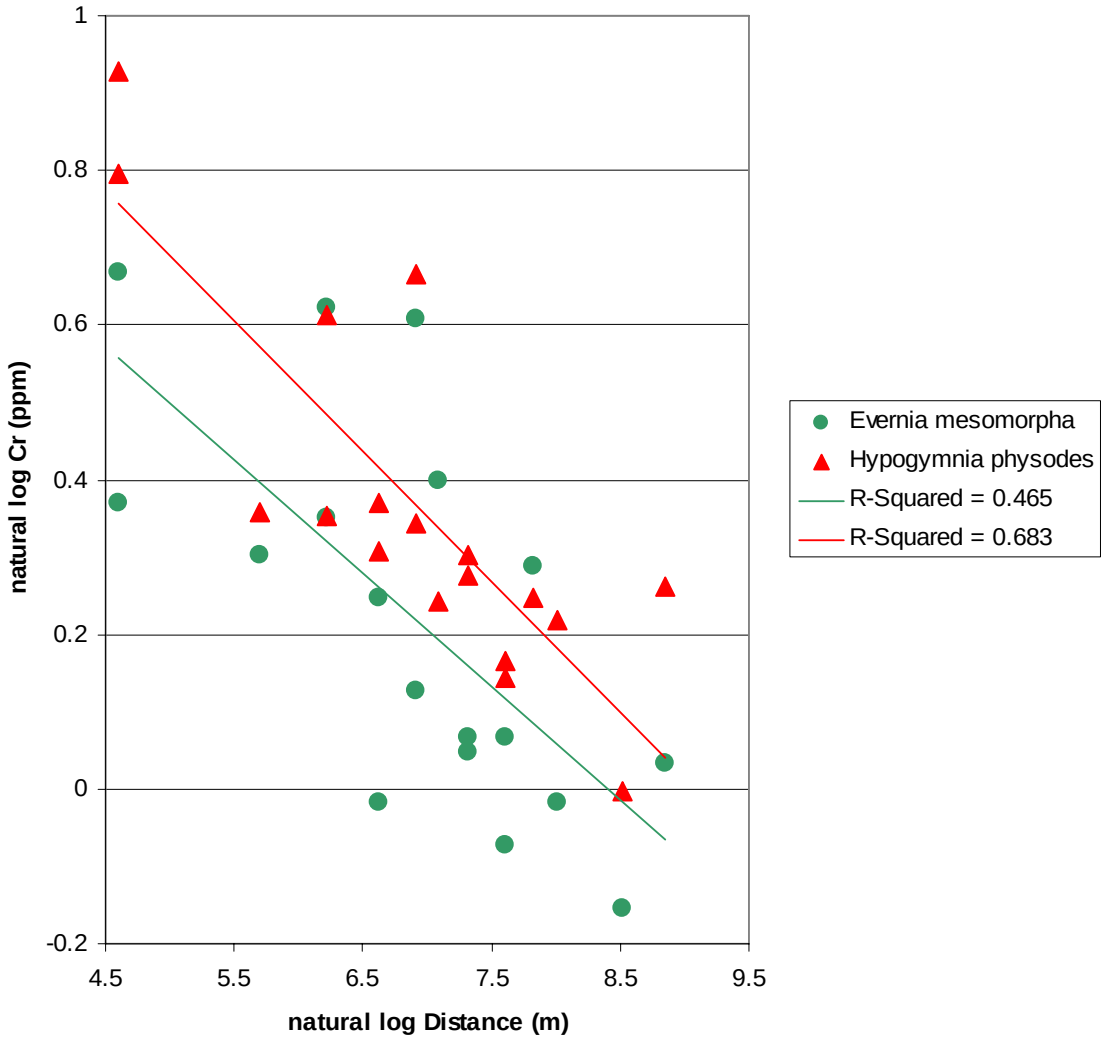

Figure G18. Total Chromium (ppm) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes* and *Evernia mesomorpha* samples. For *H. physodes*, as the distance from the mine doubles the concentration of Cr decreases by 11%. For both lichen species, the concentration of Al is much higher within 500 m of the mines.

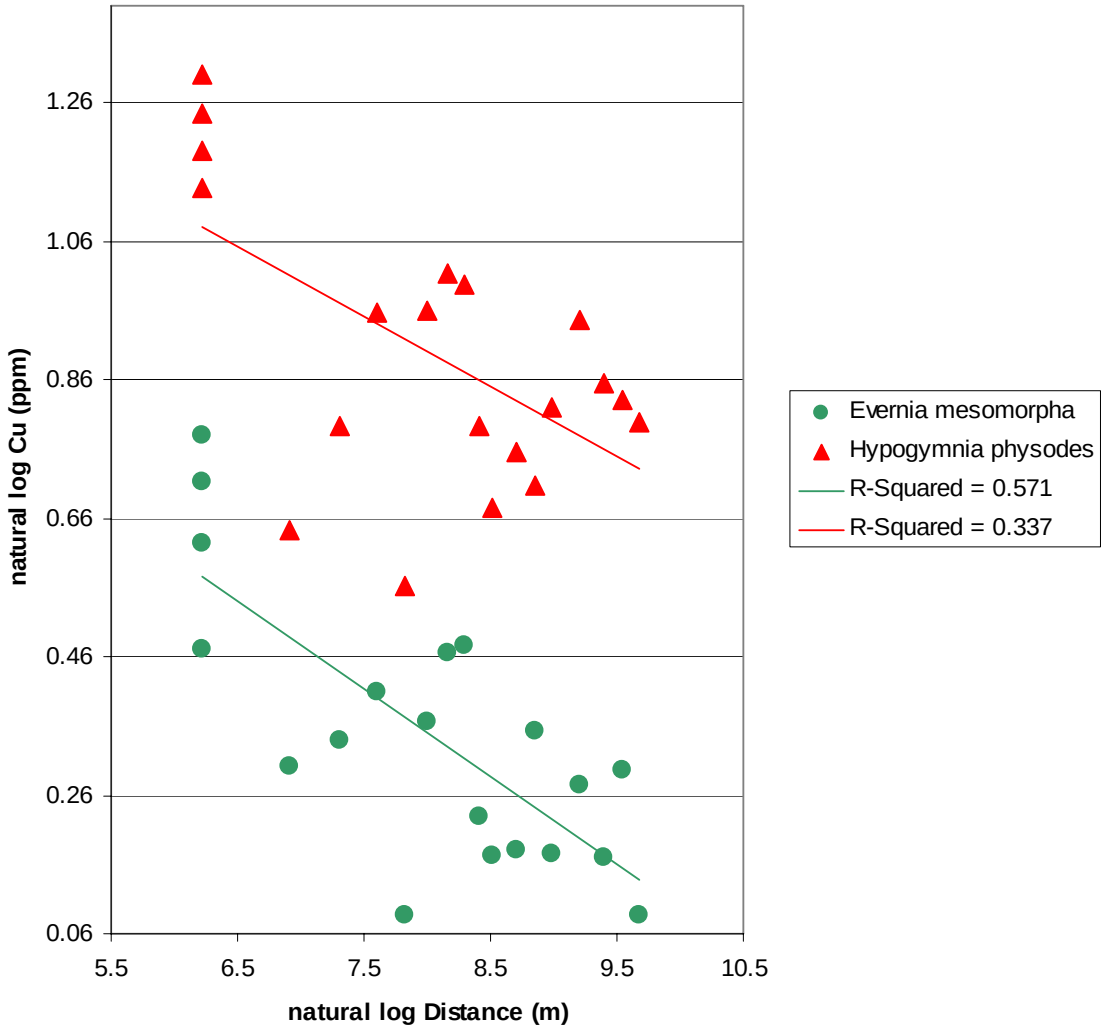

Figure G19. Total Copper (ppm) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples. For *E. mesomorpha*, as the distance from the mine doubles the concentration of Cu decreases by 8%. For both lichen species, the concentration of Cu is much higher within 500 m of the mines.

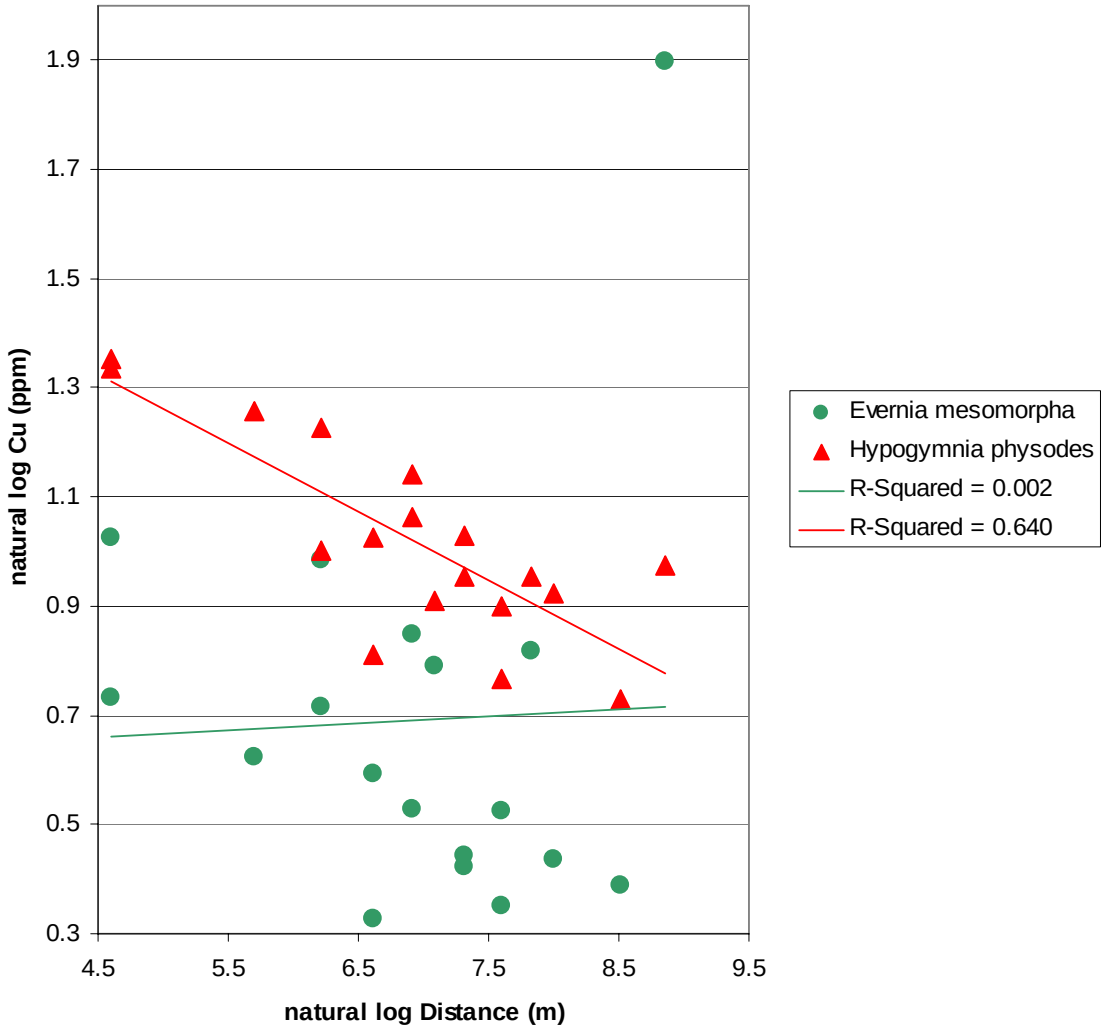

Figure G20. Total Copper (ppm) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes* and *Evernia mesomorpha* samples. For *H. physodes*, as the distance from the mine doubles the concentration of Cu decreases by 8%.

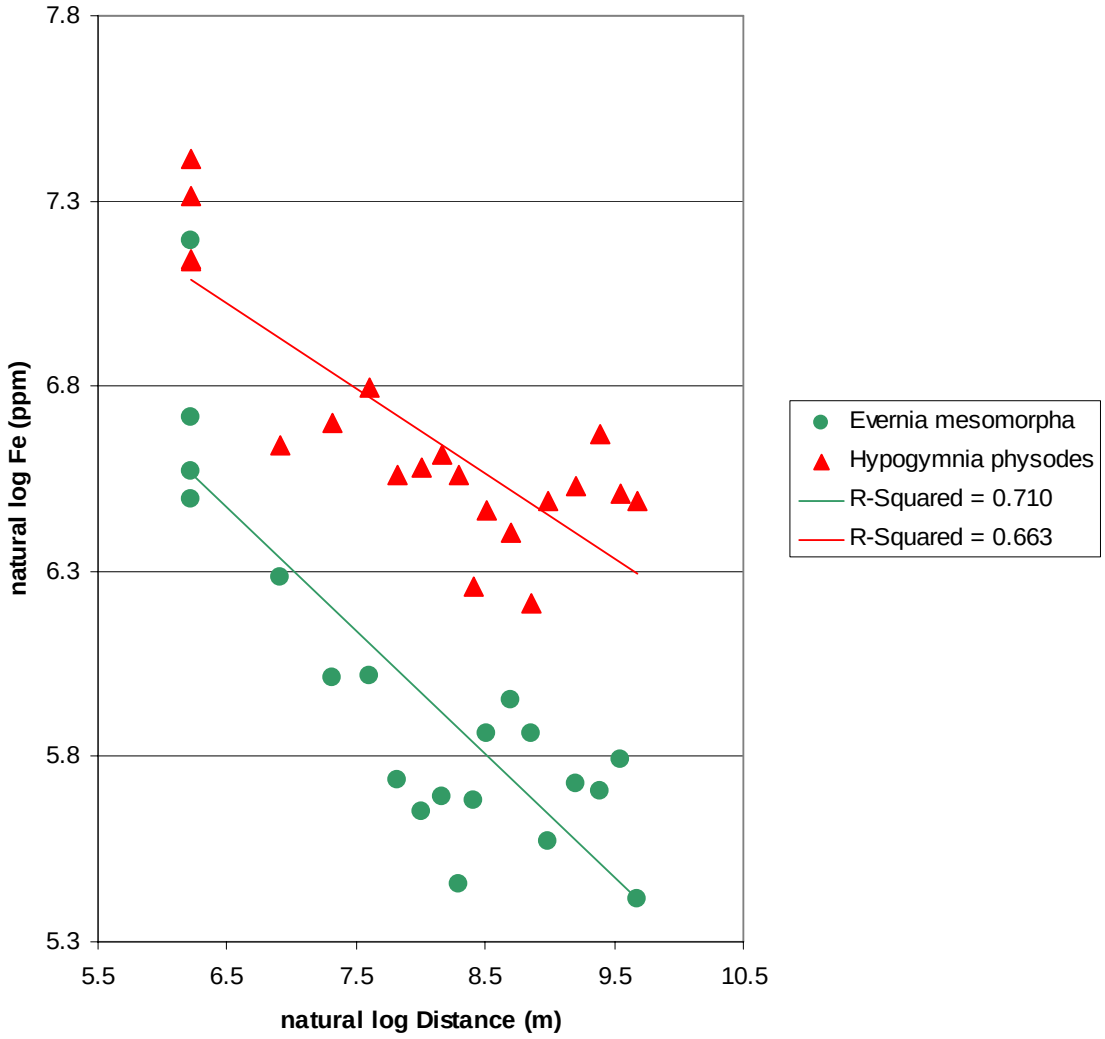

Figure G21. Total Iron (ppm) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples. For *E. mesomorpha*, as the distance from the mine doubles the concentration of Fe decreases by 20%. For *H. physodes*, as the distance from the mine doubles the concentration of Fe decreases by 15%. For both lichen species, the concentration of Fe is much higher within 500 m of the mines.

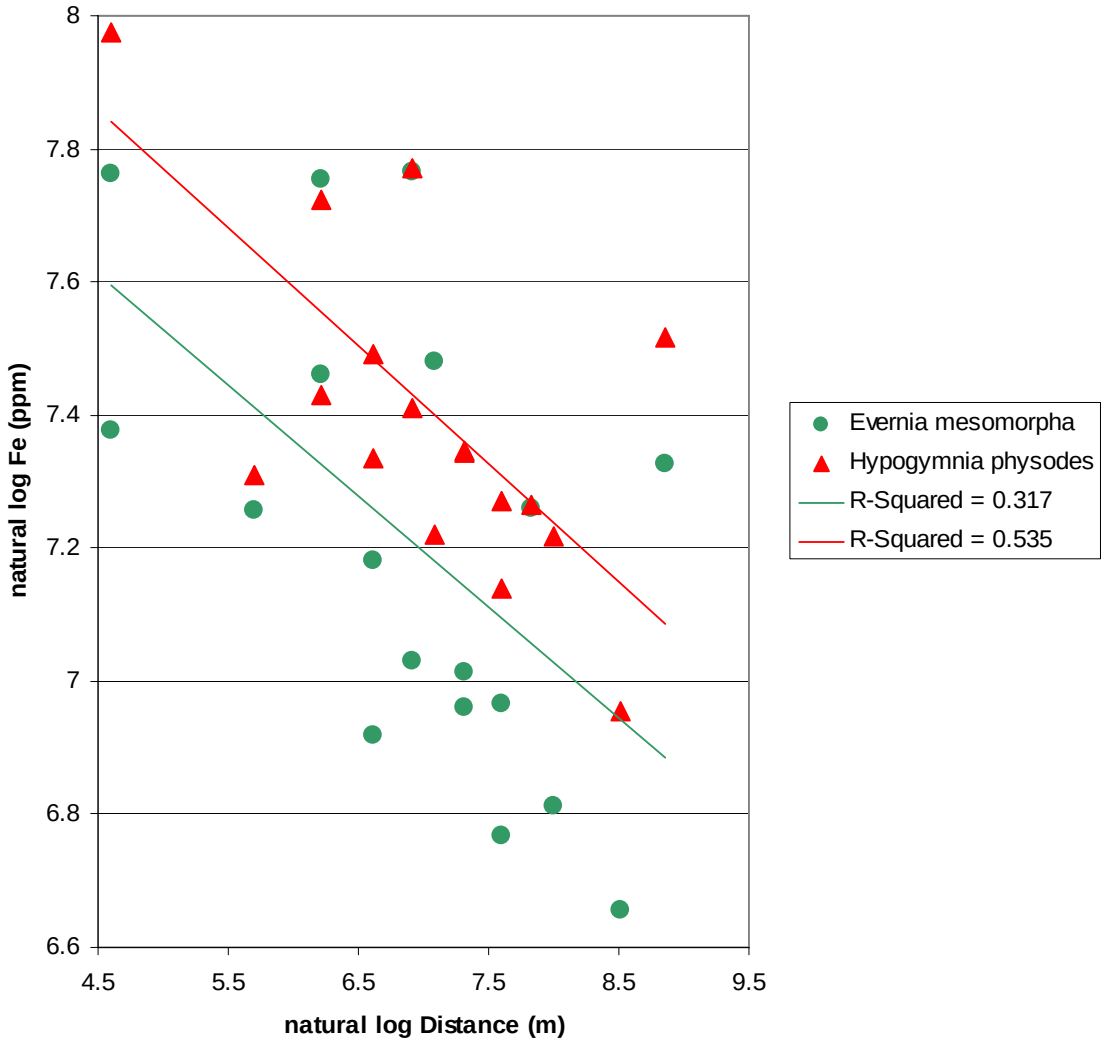

Figure G22. Total Iron (ppm) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes* and *Evernia mesomorpha* samples. For *H. physodes*, as the distance from the mine doubles the concentration of Fe decreases by 12%.

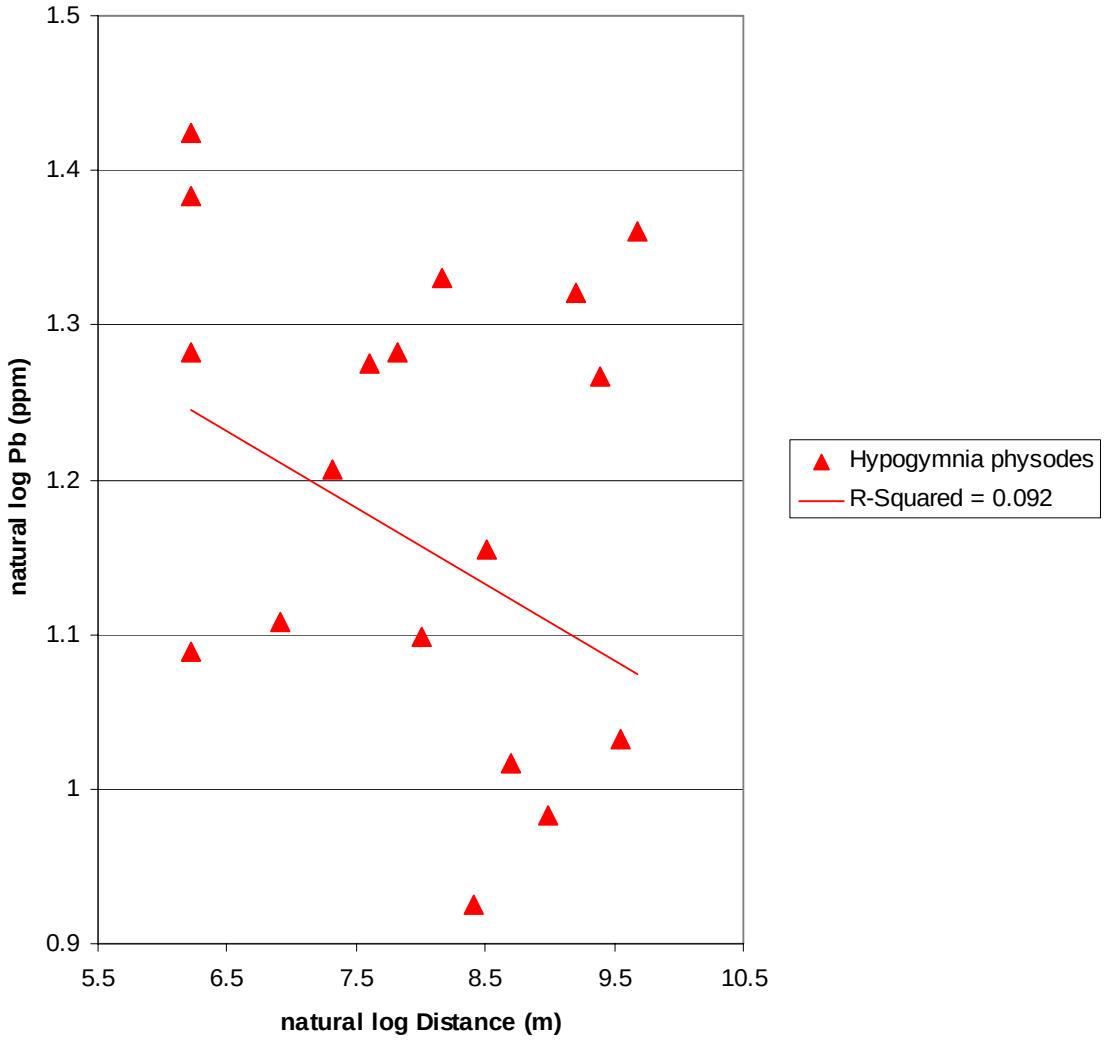

Figure G23. Total Lead (ppm) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes.* More than 50% of the samples of *Evernia mesomorpha* had Pb concentrations below the MDL therefore regression was not analyzed.

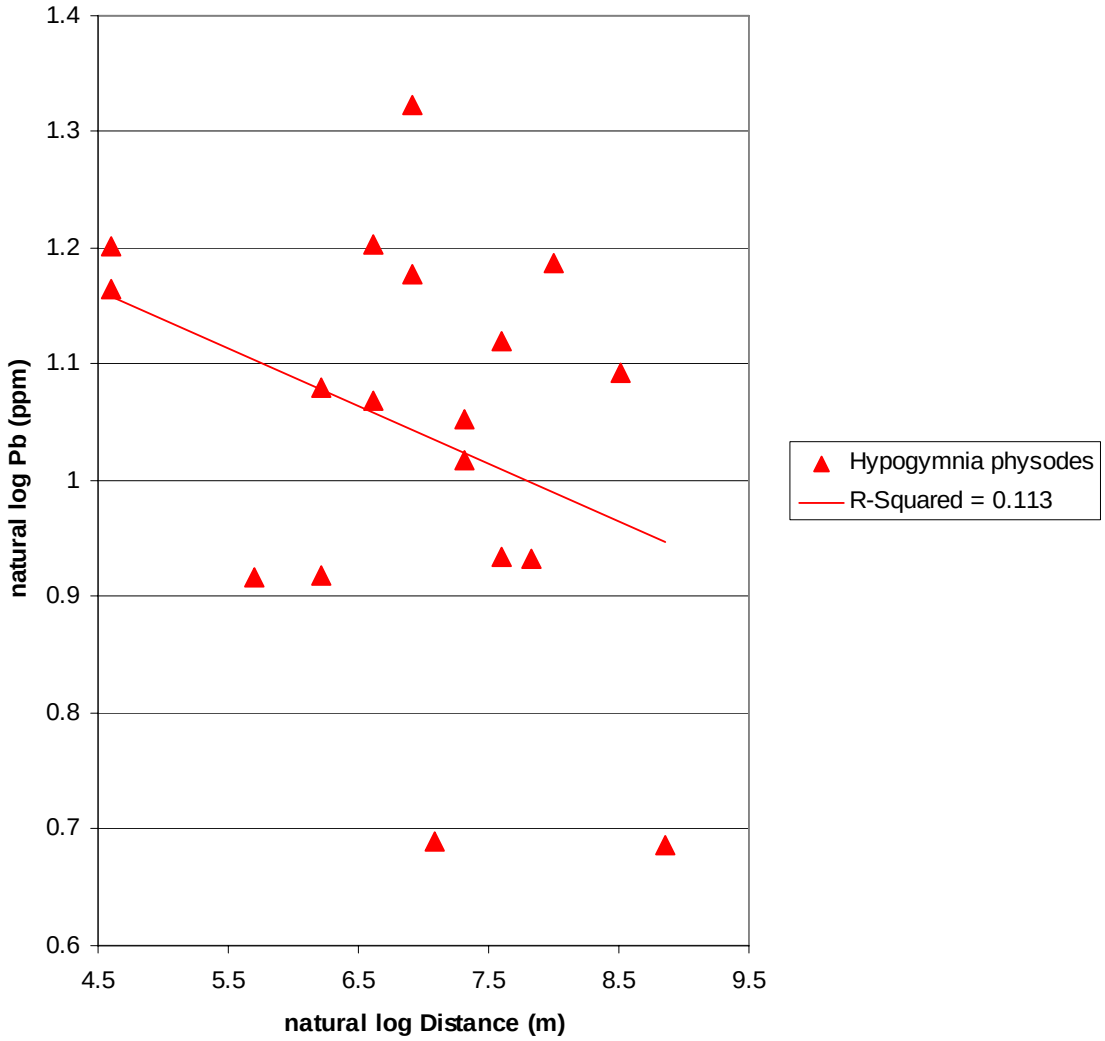

Figure G24. Total Lead (ppm) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes*. More than 50% of the samples of *Evernia mesomorpha* had Pb concentrations below the MDL therefore regression was not analyzed.

Figure G25. Total Lithium (ppm) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples. More than 50% of the samples of both lichens had Li concentrations below the MDL therefore regressions were not analyzed.

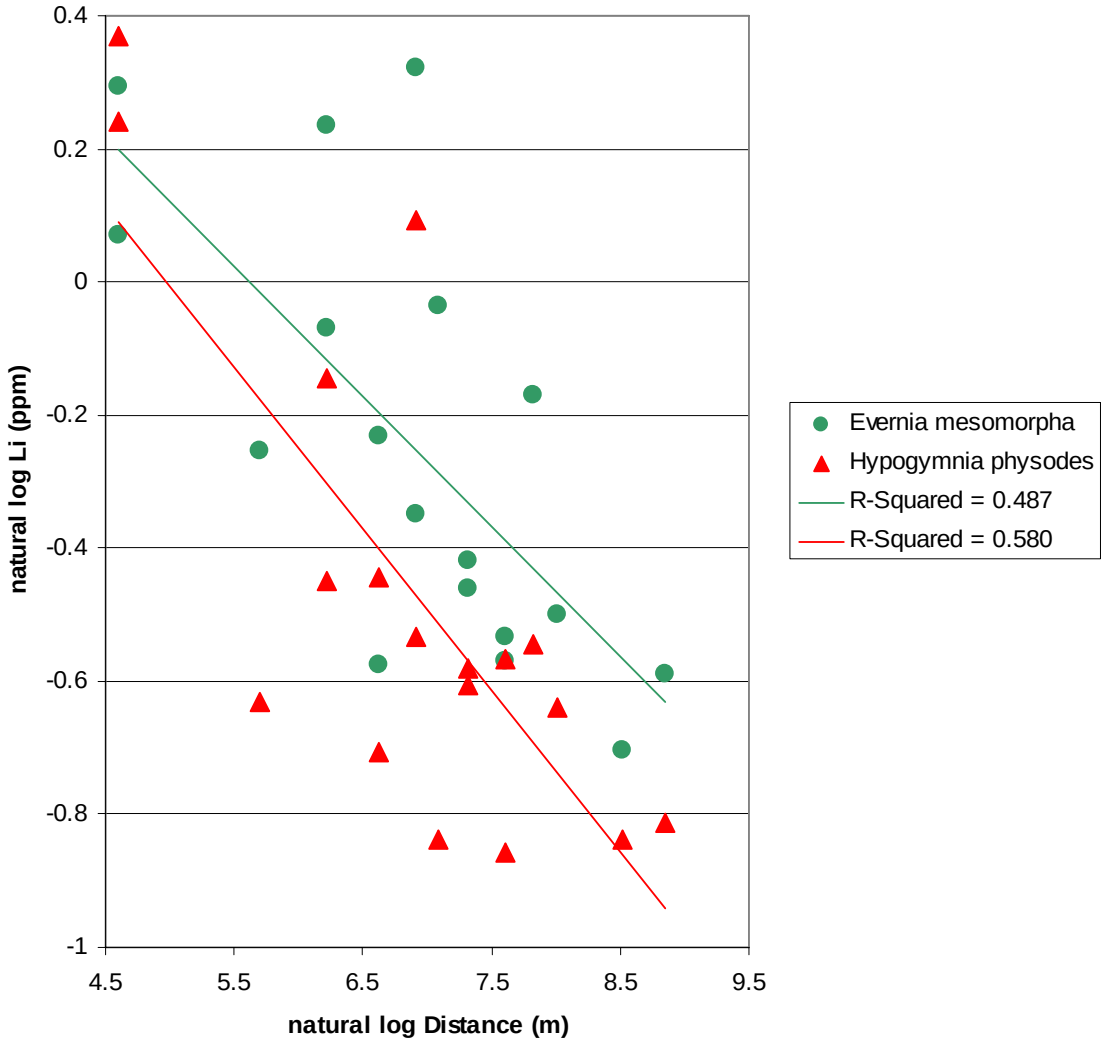

Figure G26. Total Lithium (ppm) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes* and *Evernia mesomorpha* samples. For *H. physodes*, as the distance from the mine doubles the concentration of Li decreases by 15%. For *E. mesomorpha*, the concentration of Li is much higher within 100 m of the mines.

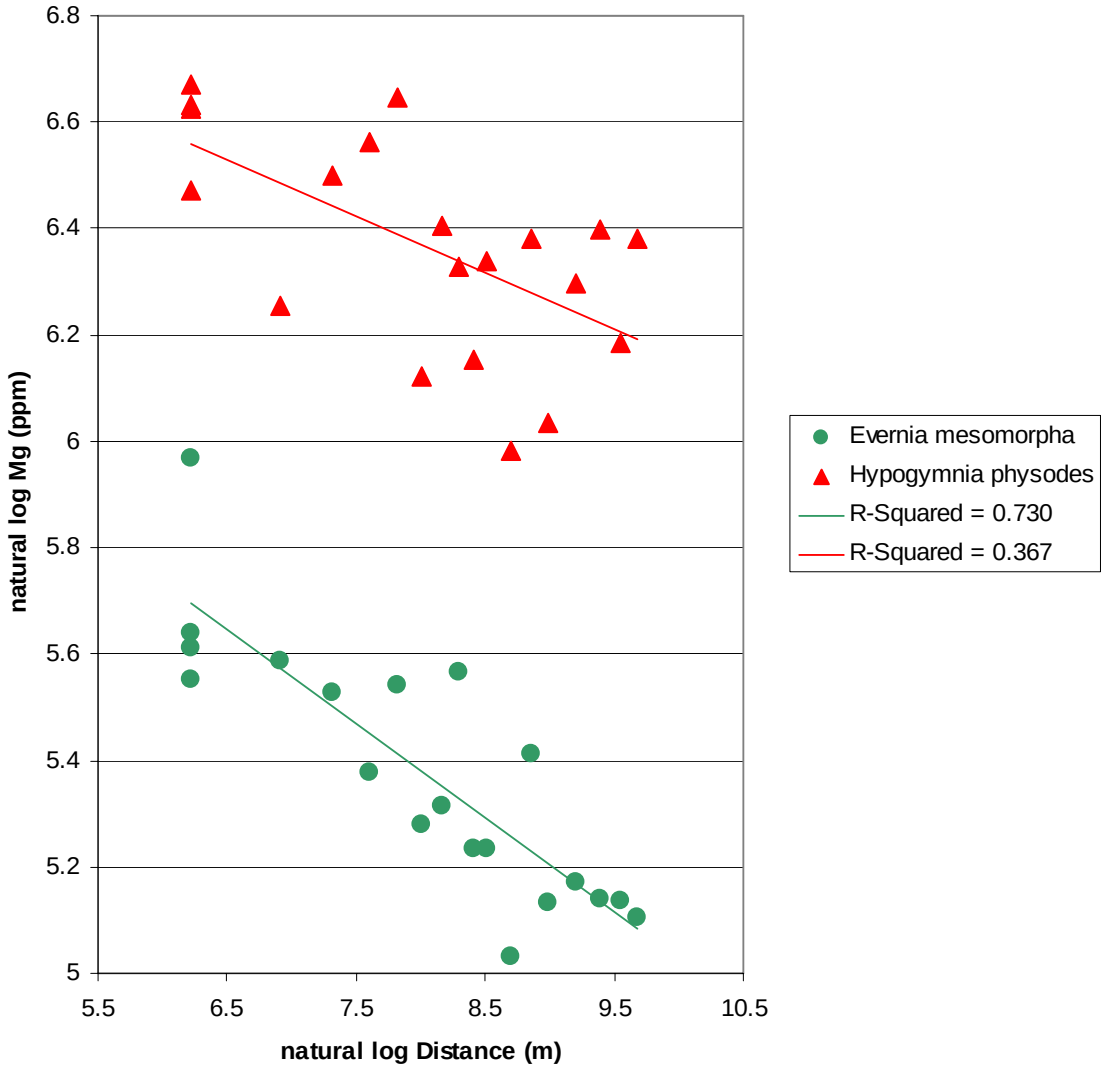

Figure G27. Total Magnesium (ppm) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples. For *E. mesomorpha*, as the distance from the mine doubles the concentration of Mg decreases by 11%.

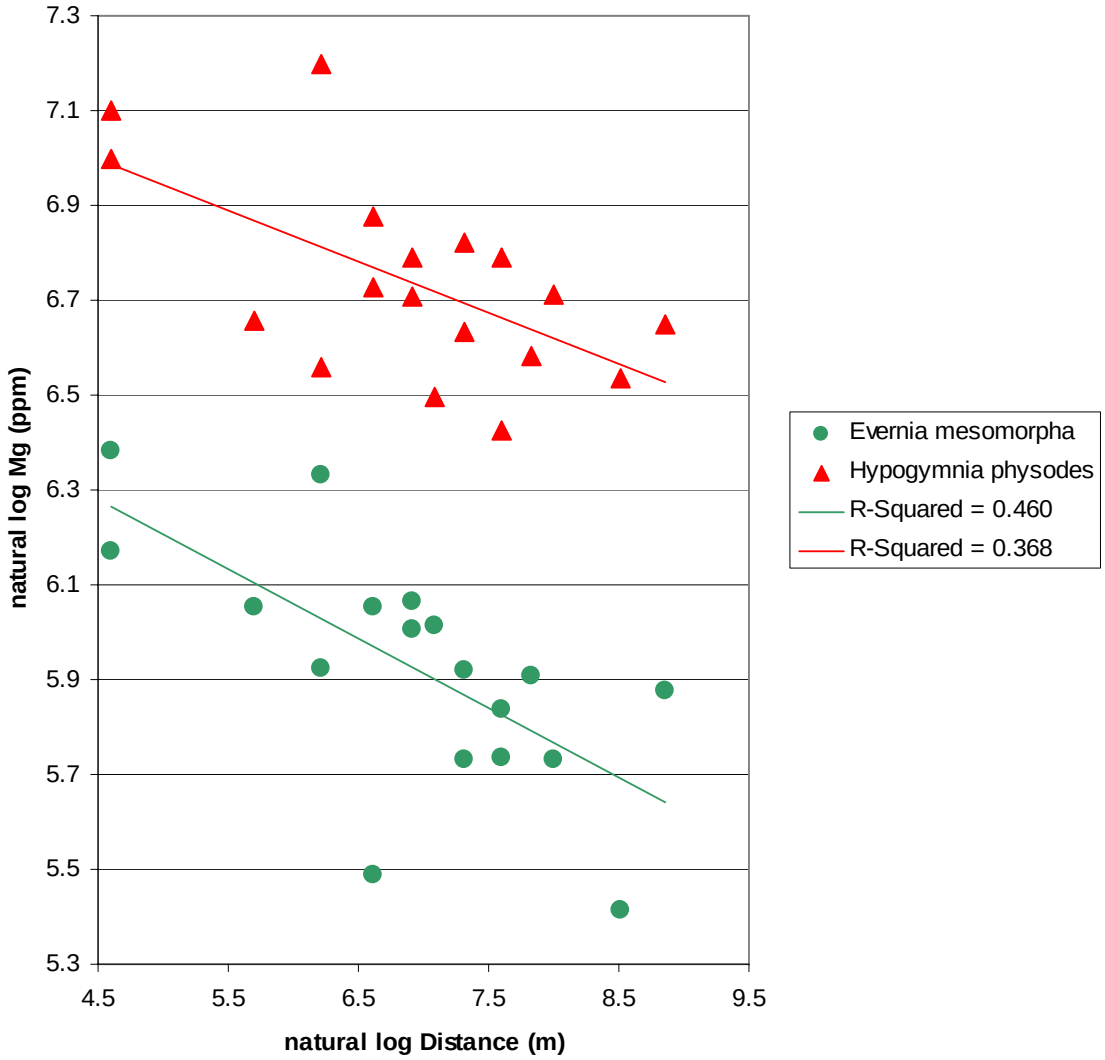

Figure G28. Total Magnesium (ppm) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

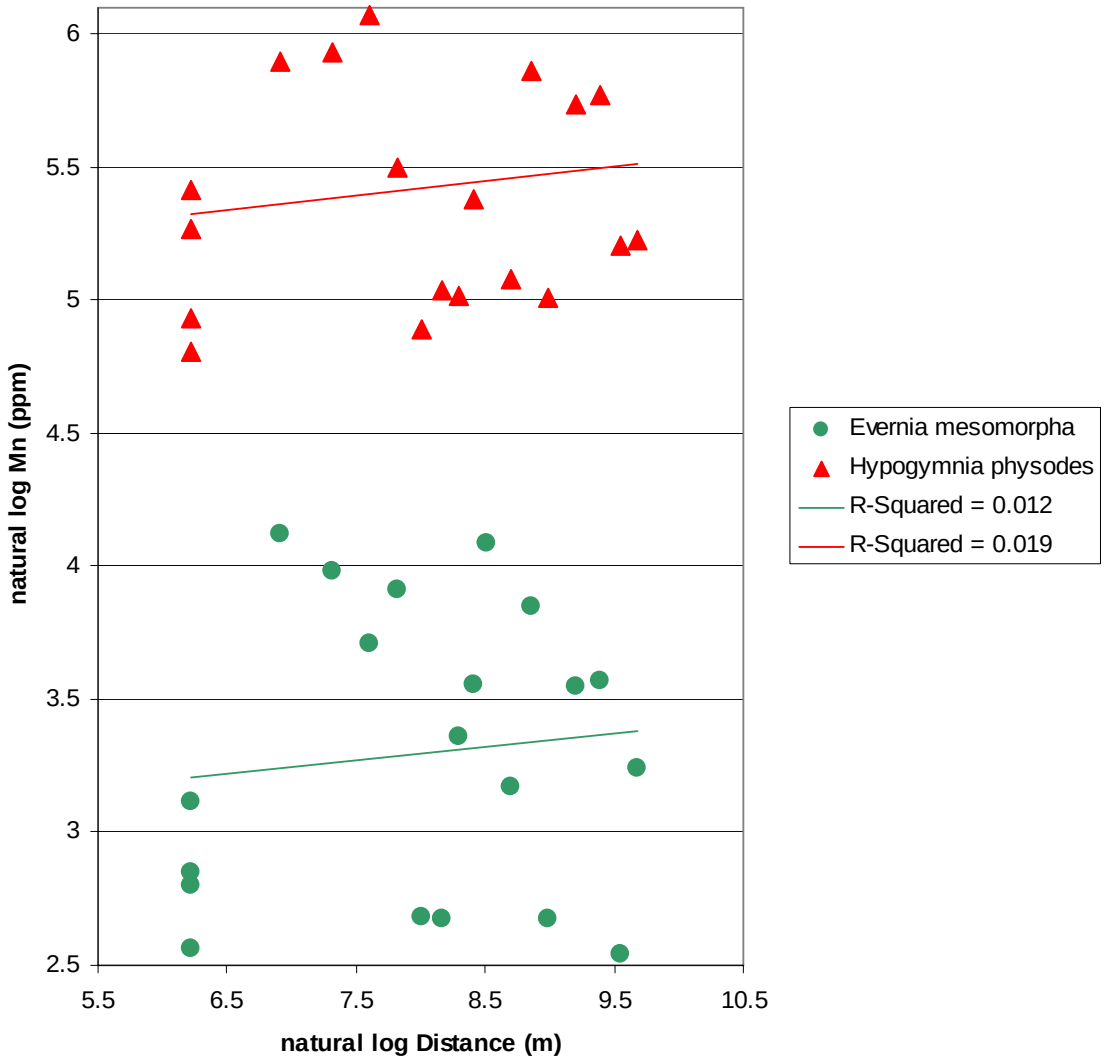

Figure G29. Total Manganese (ppm) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

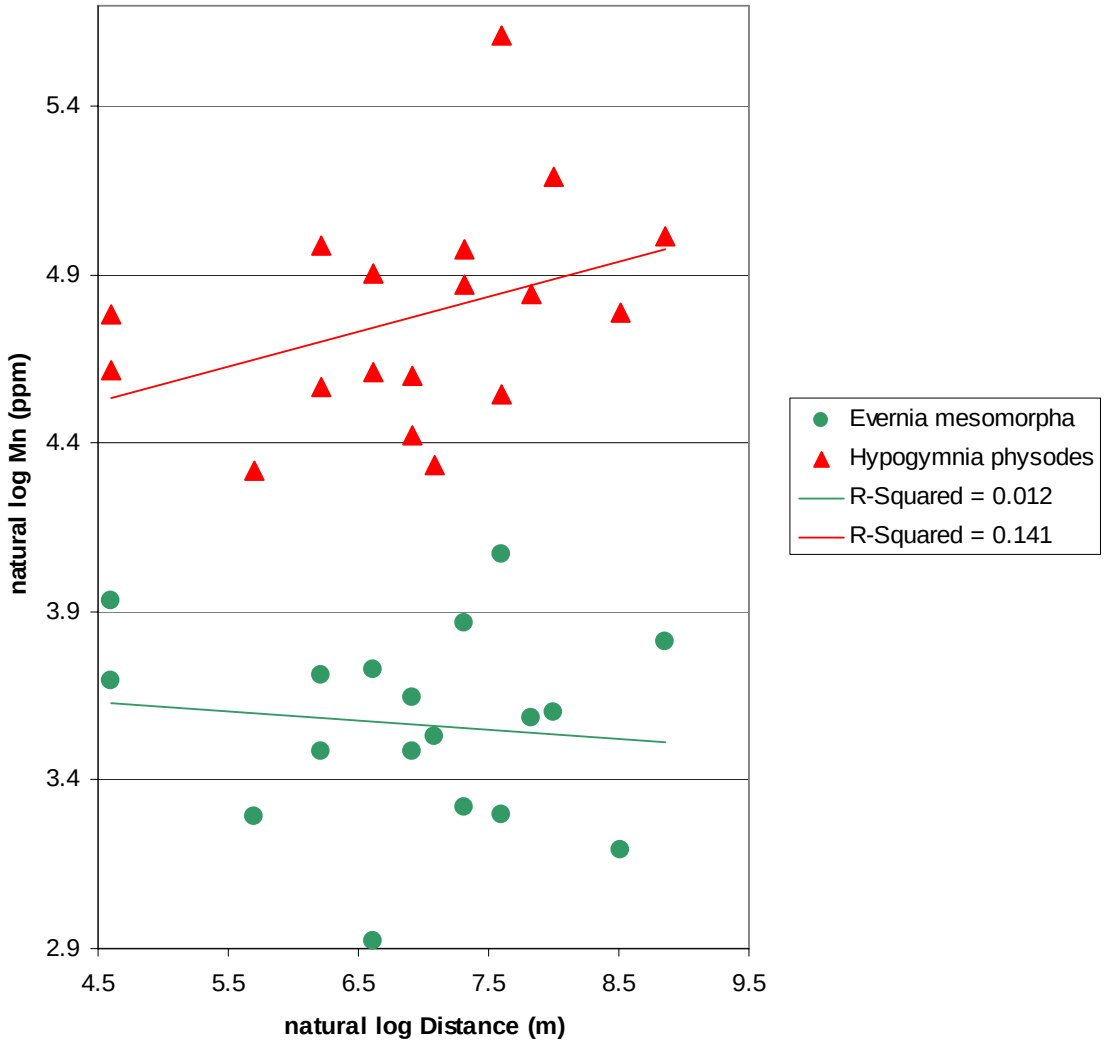

Figure G30. Total Manganese (ppm) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

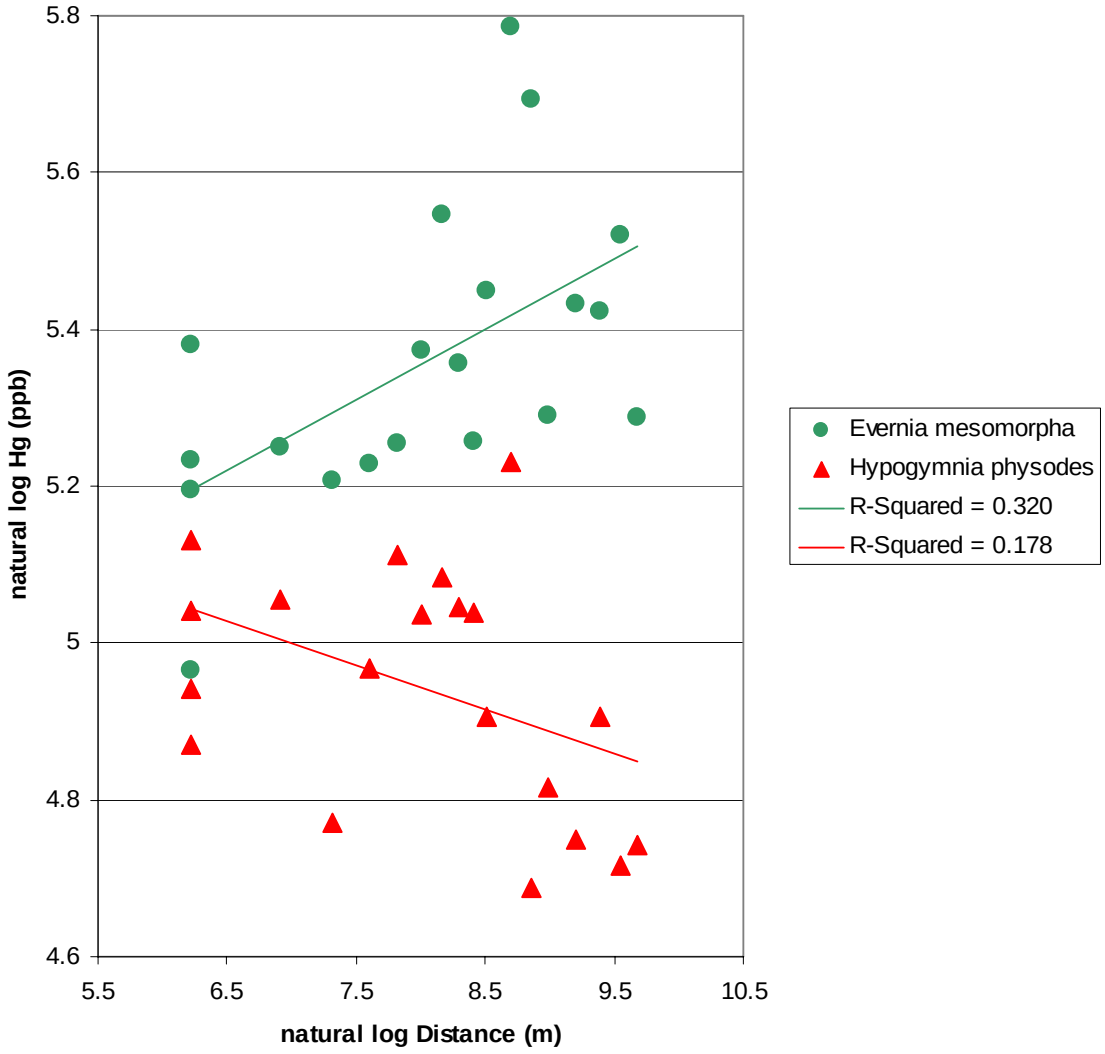

Figure G31. Total Mercury (ppb) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

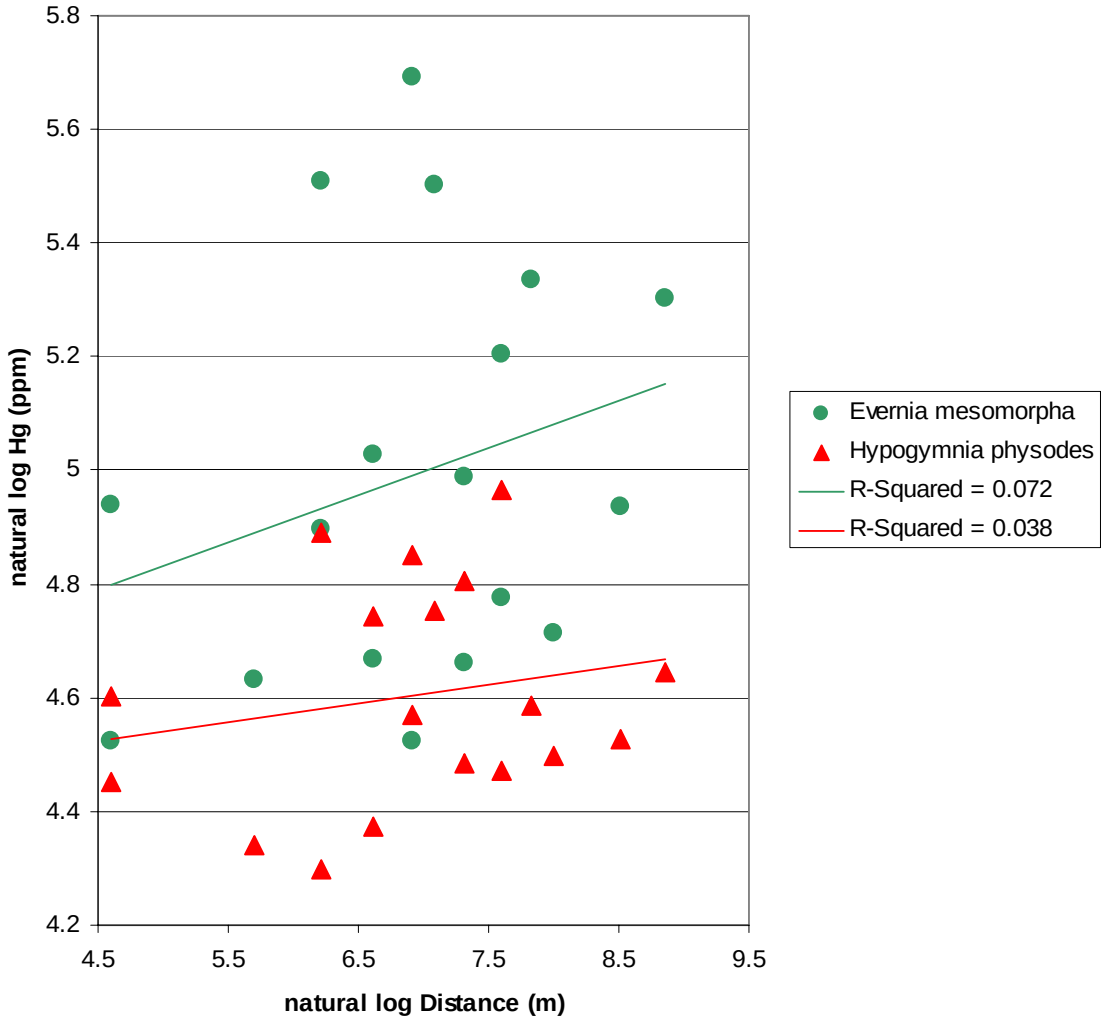

Figure G32. Total Mercury (ppb) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

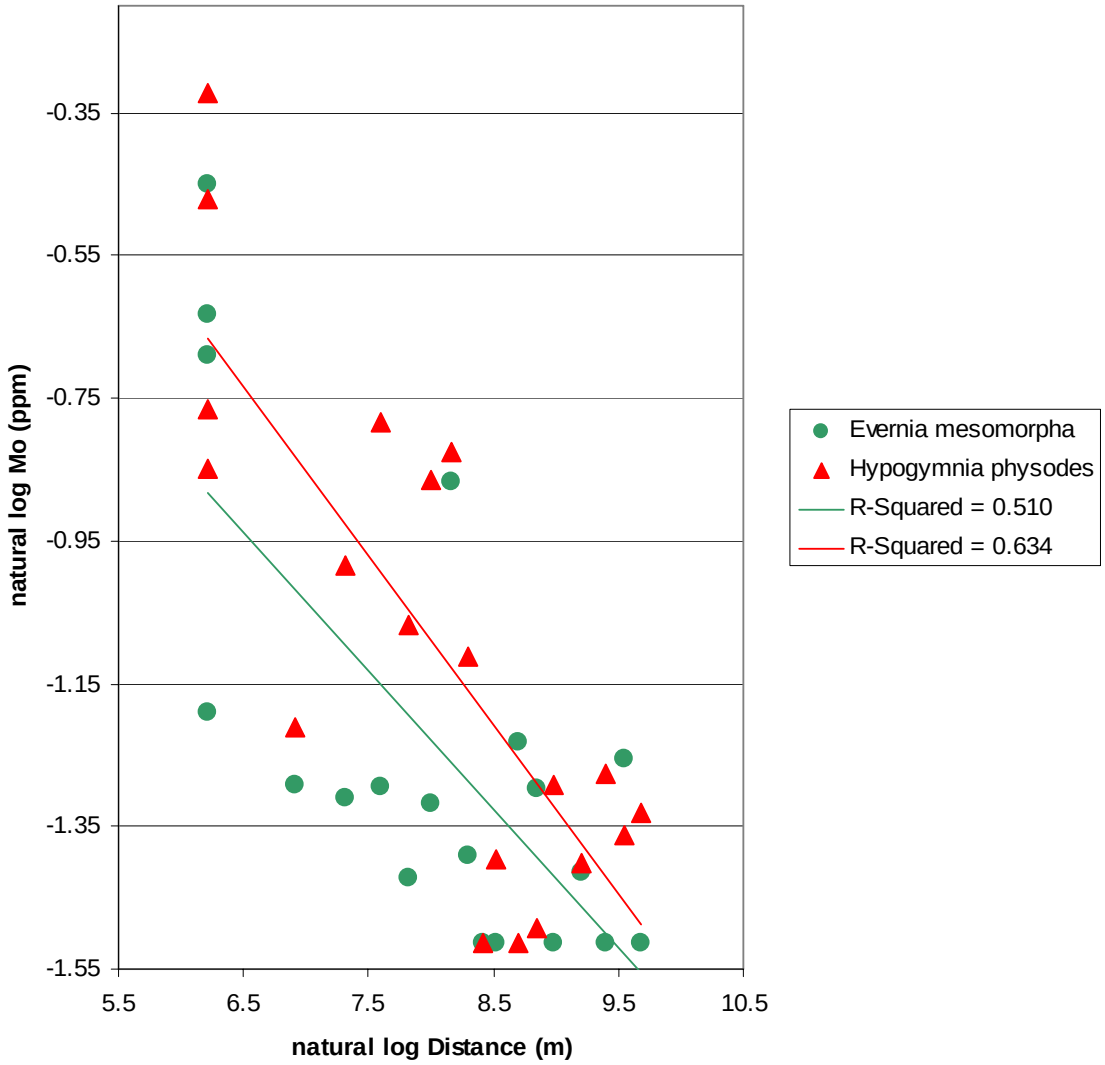

Figure G33. Total Molybdenum (ppm) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples. For *E. mesomorpha*, as the distance from the mine doubles the concentration of Mo decreases by 12%. For *H. physodes*, as the distance from the mine doubles the concentration of Mo decreases by 15%. For both lichen species, the concentration of Mo is much higher within 100 m of the mines.

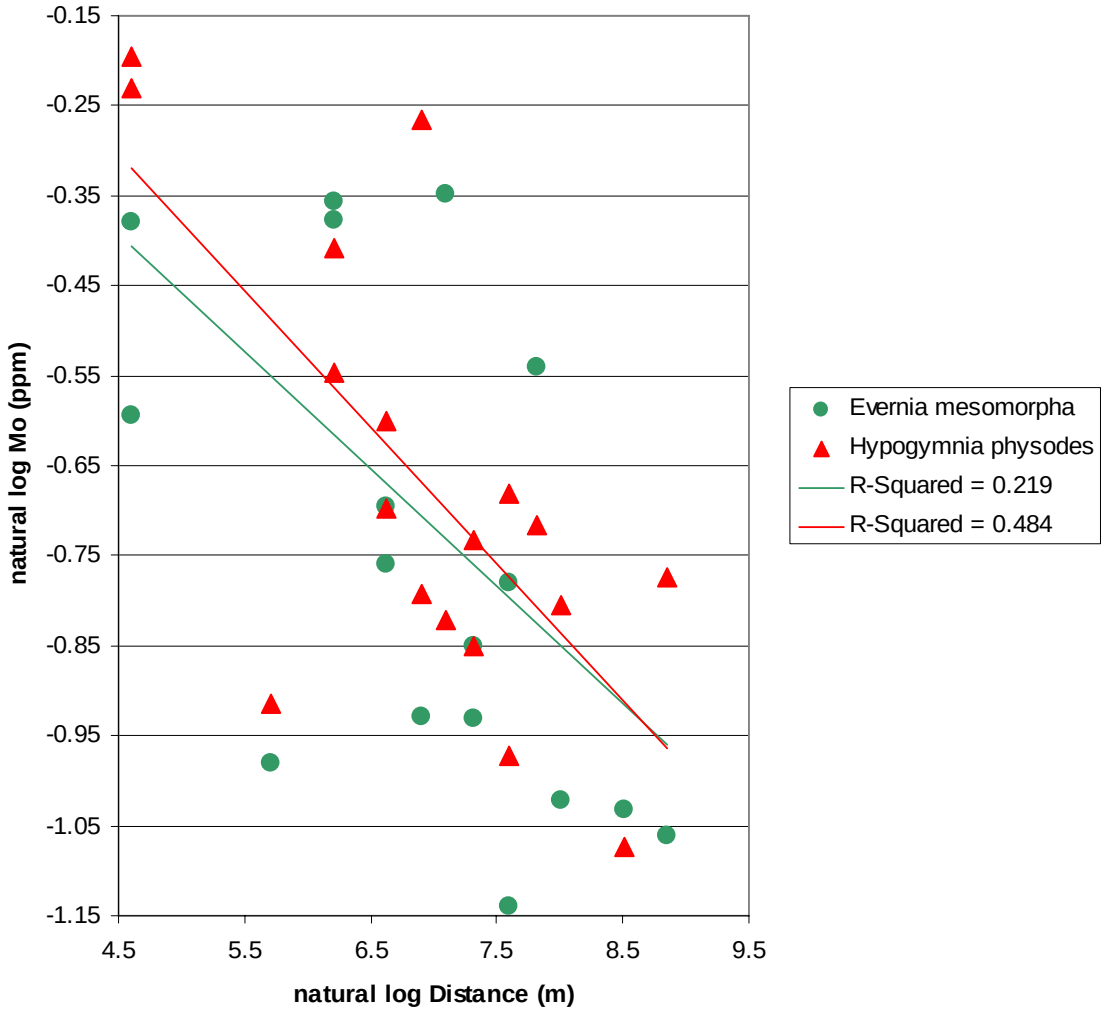

Figure G34. Total Molybdenum (ppm) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

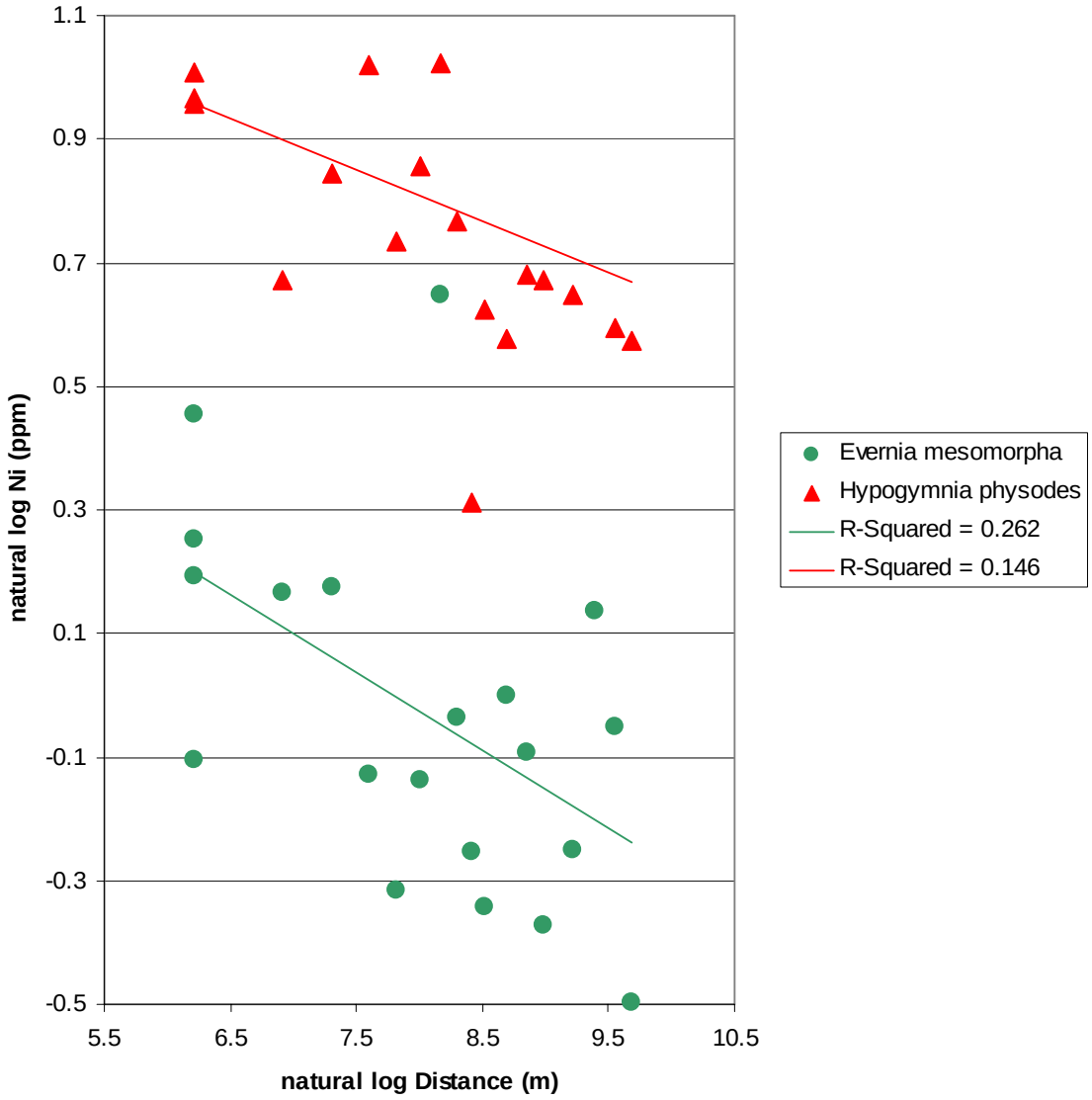

Figure G35. Total Nickel (ppm) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

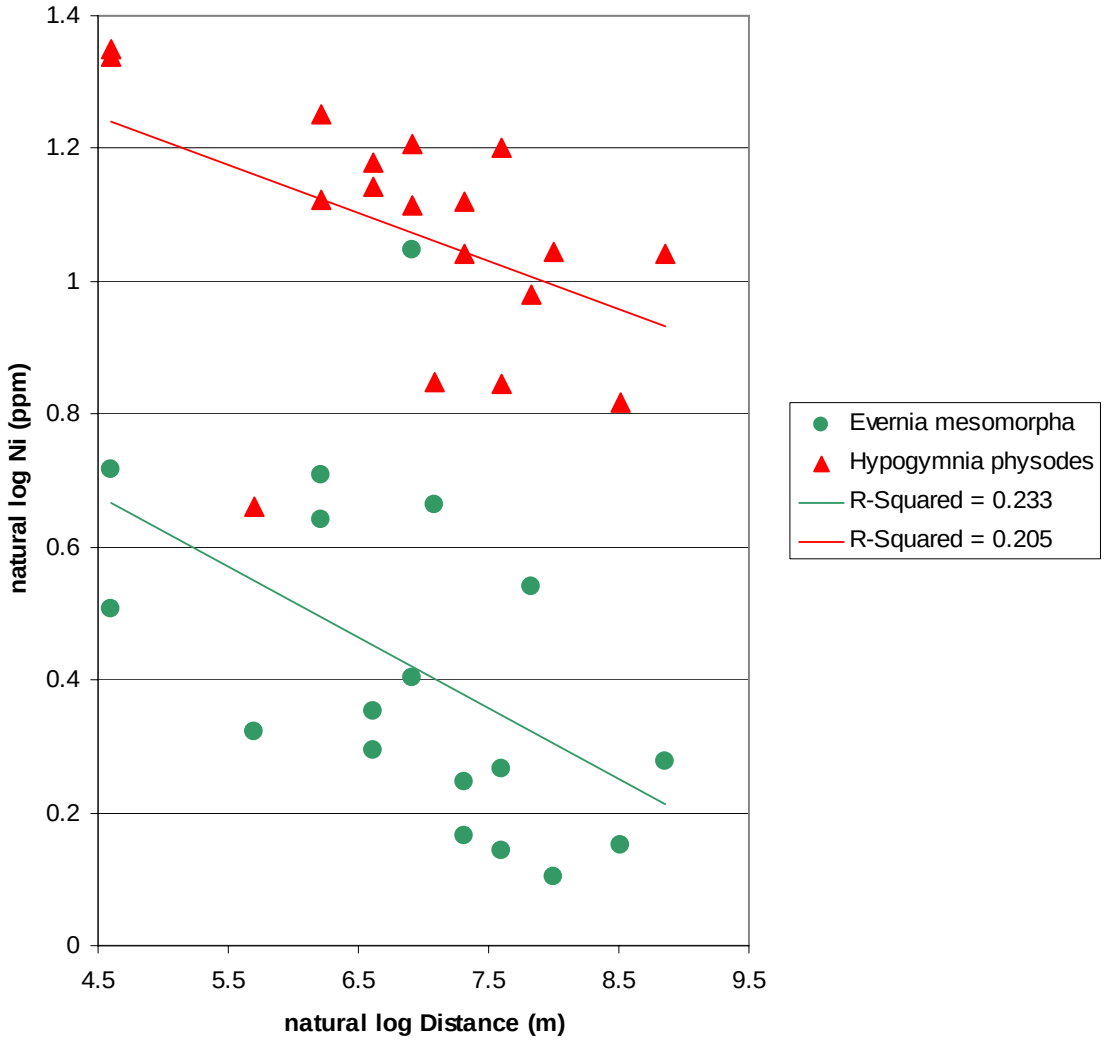

Figure G36. Total Nickel (ppm) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

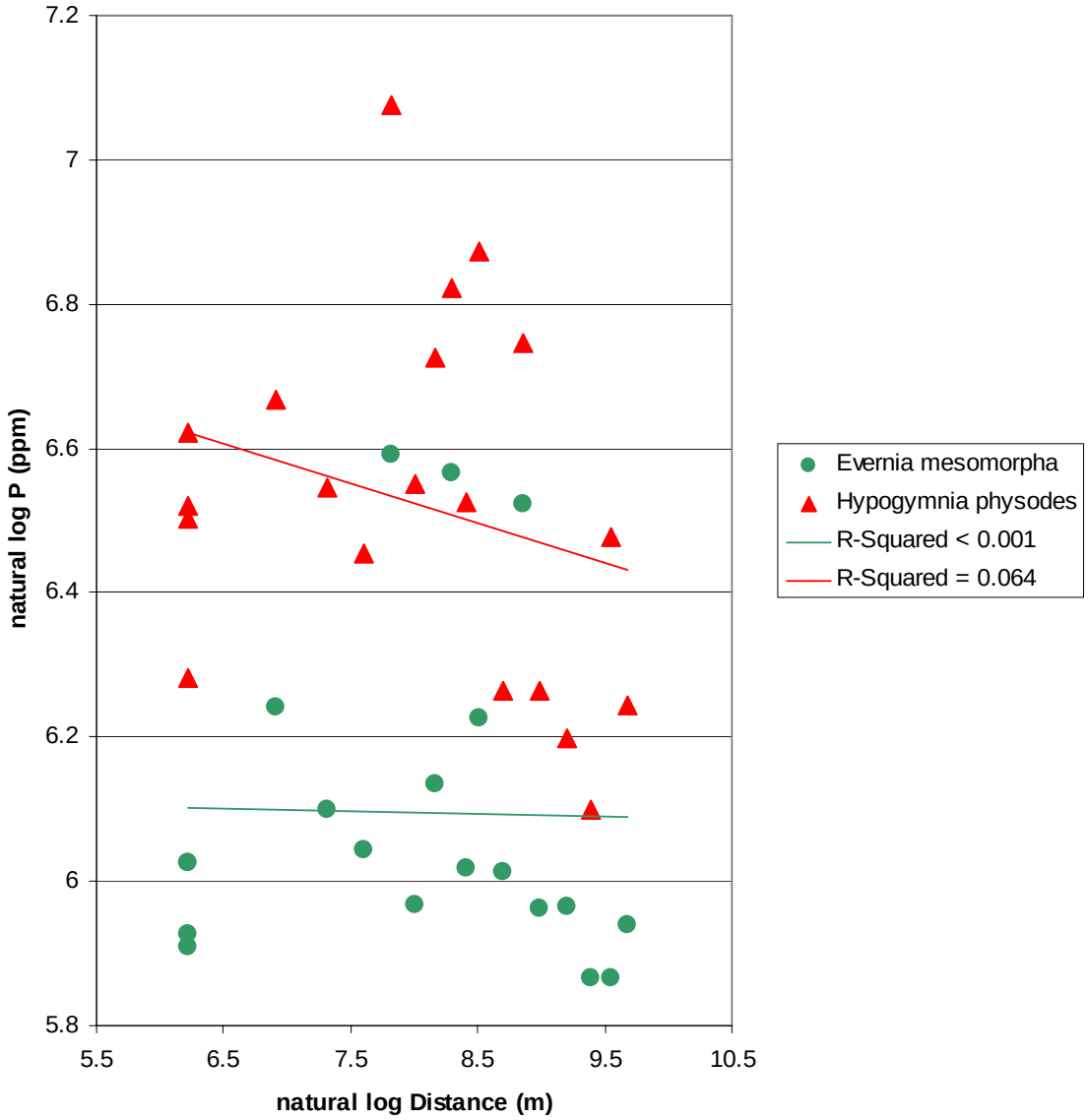

Figure G37. Total Phosphorous (ppm) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

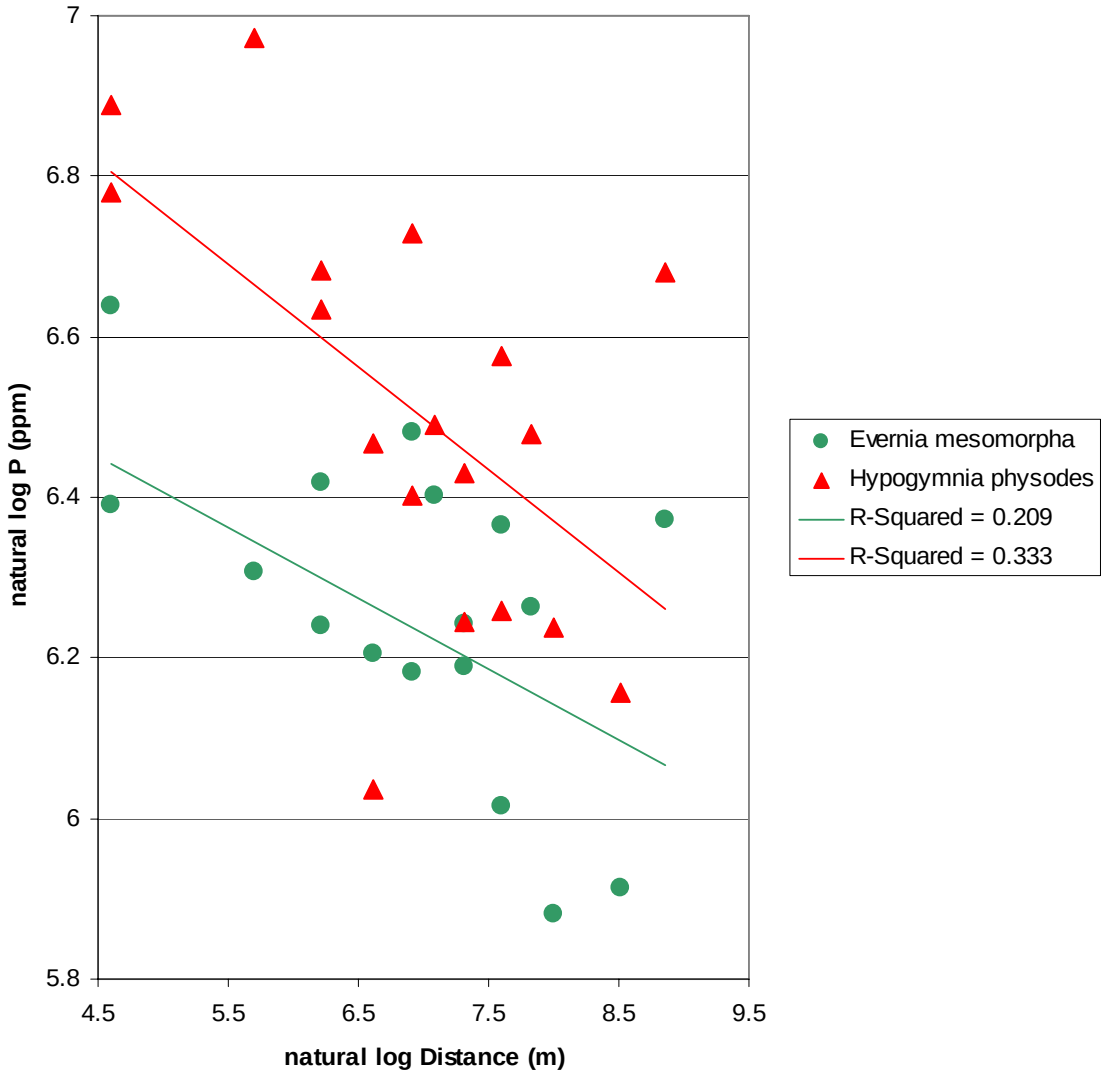

Figure G38. Total Phosphorous (ppm) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

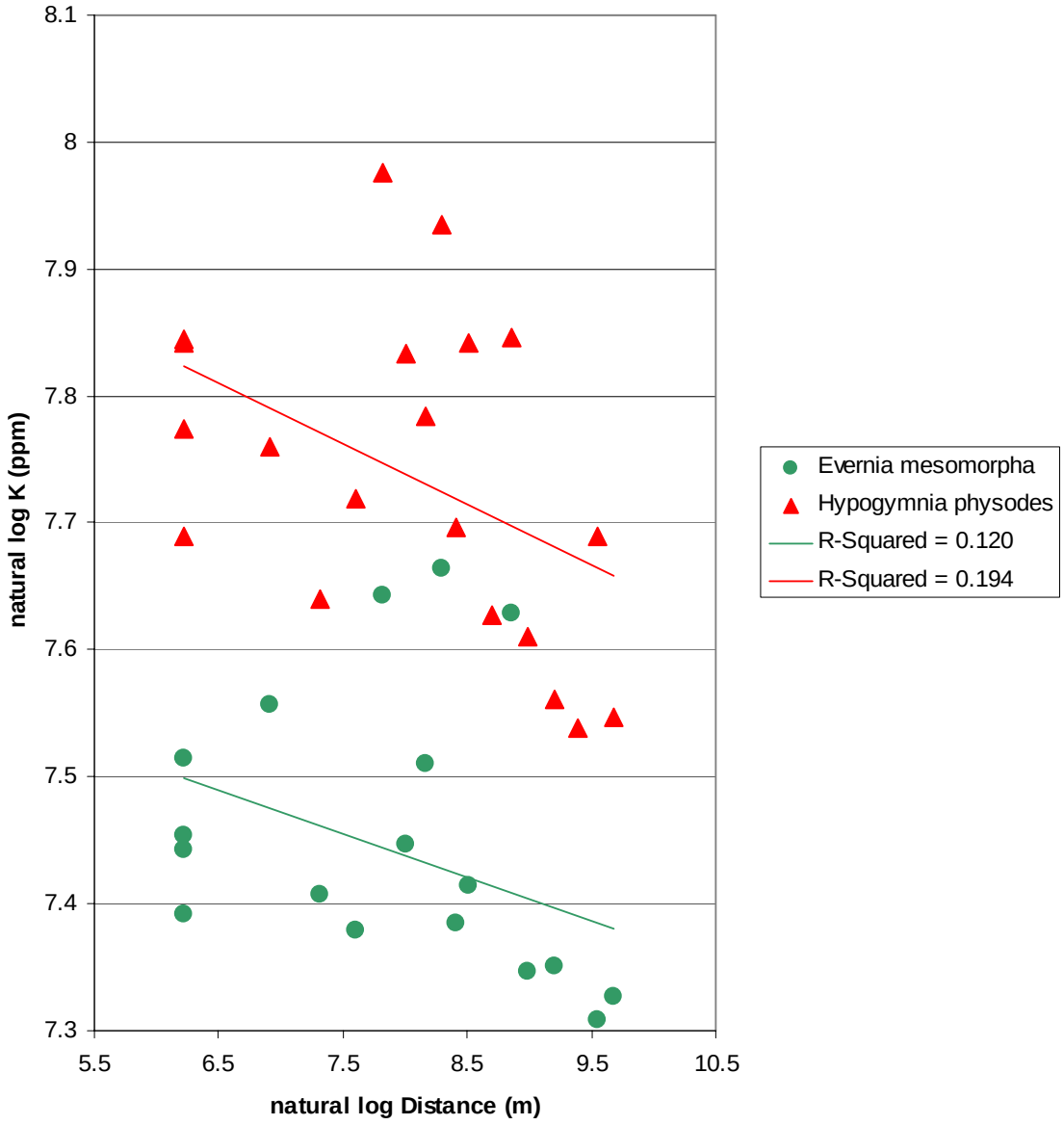

Figure G39. Total Potassium (ppm) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

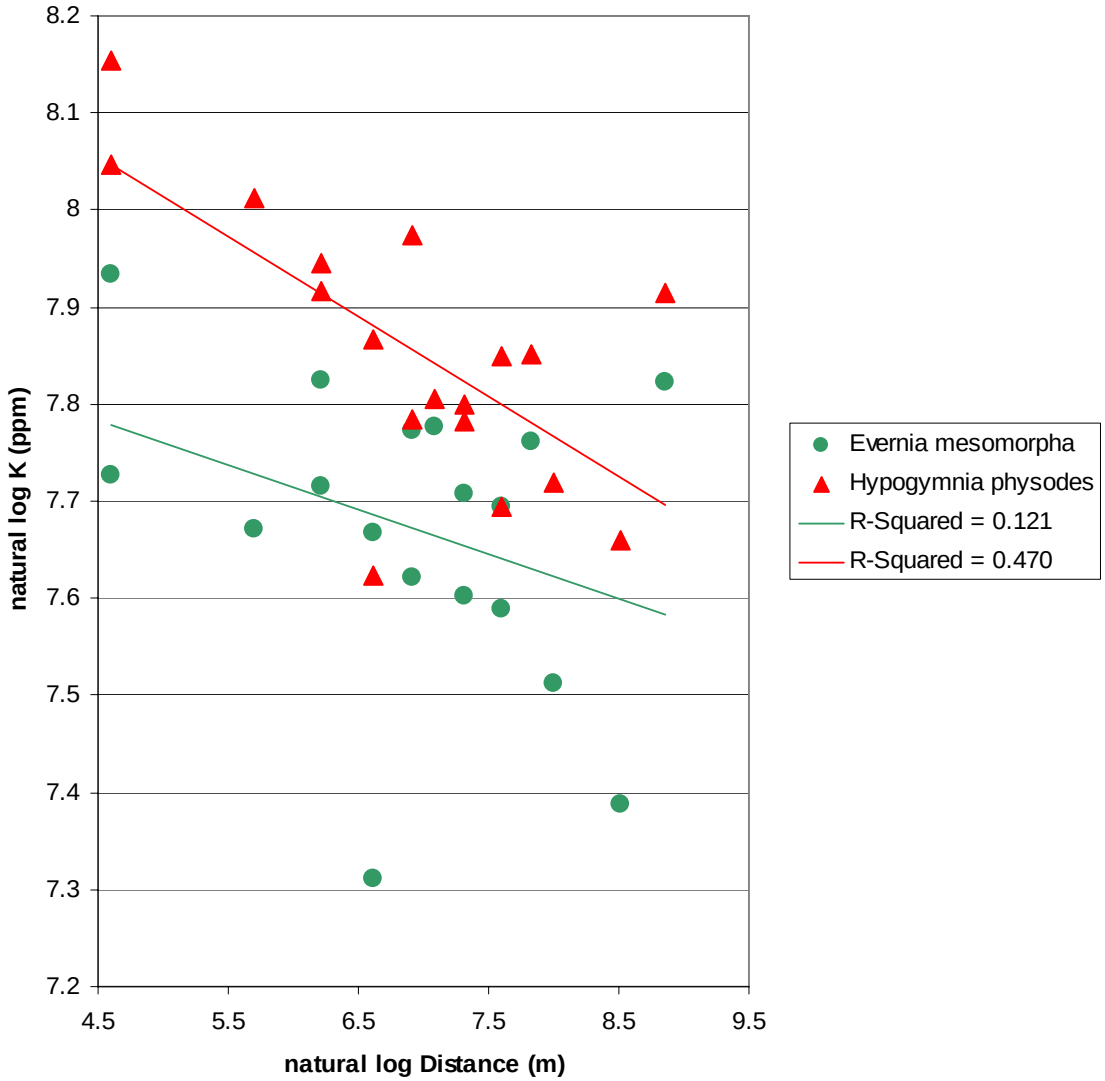

Figure G40. Total Potassium (ppm) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

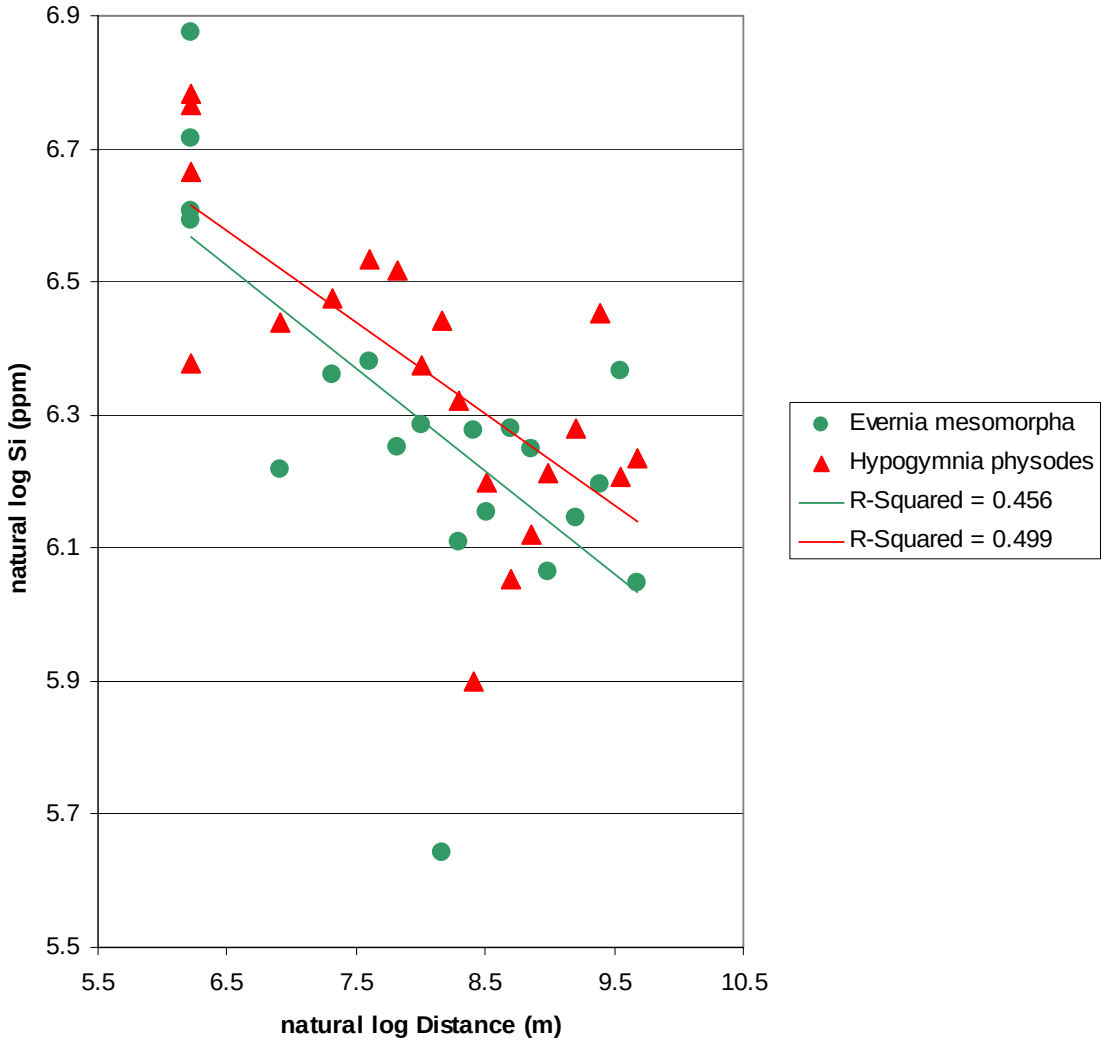

Figure G41. Total Silicon (ppm) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples. For both lichen species, the concentration of Si is much higher within 500 m of the mines.

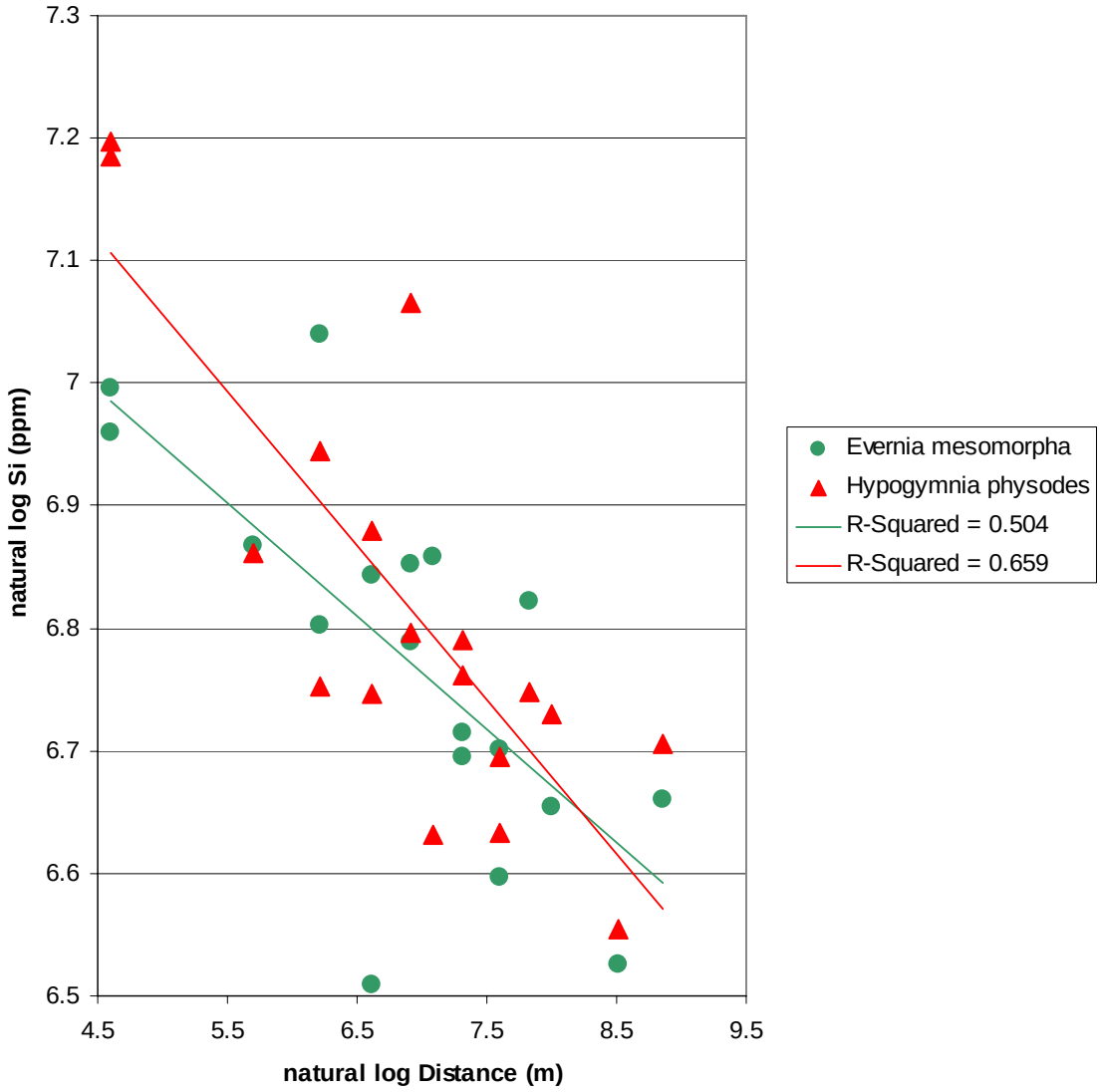

Figure G42. Total Silicon (ppm) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes* and *Evernia mesomorpha* samples. For *E. mesomorpha*, as the distance from the mine doubles the concentration of Si decreases by 6%. For *H. physodes*, as the distance from the mine doubles the concentration of Si decreases by 8%. For both lichen species, the concentration of Si is much higher within 100 m of the mines.

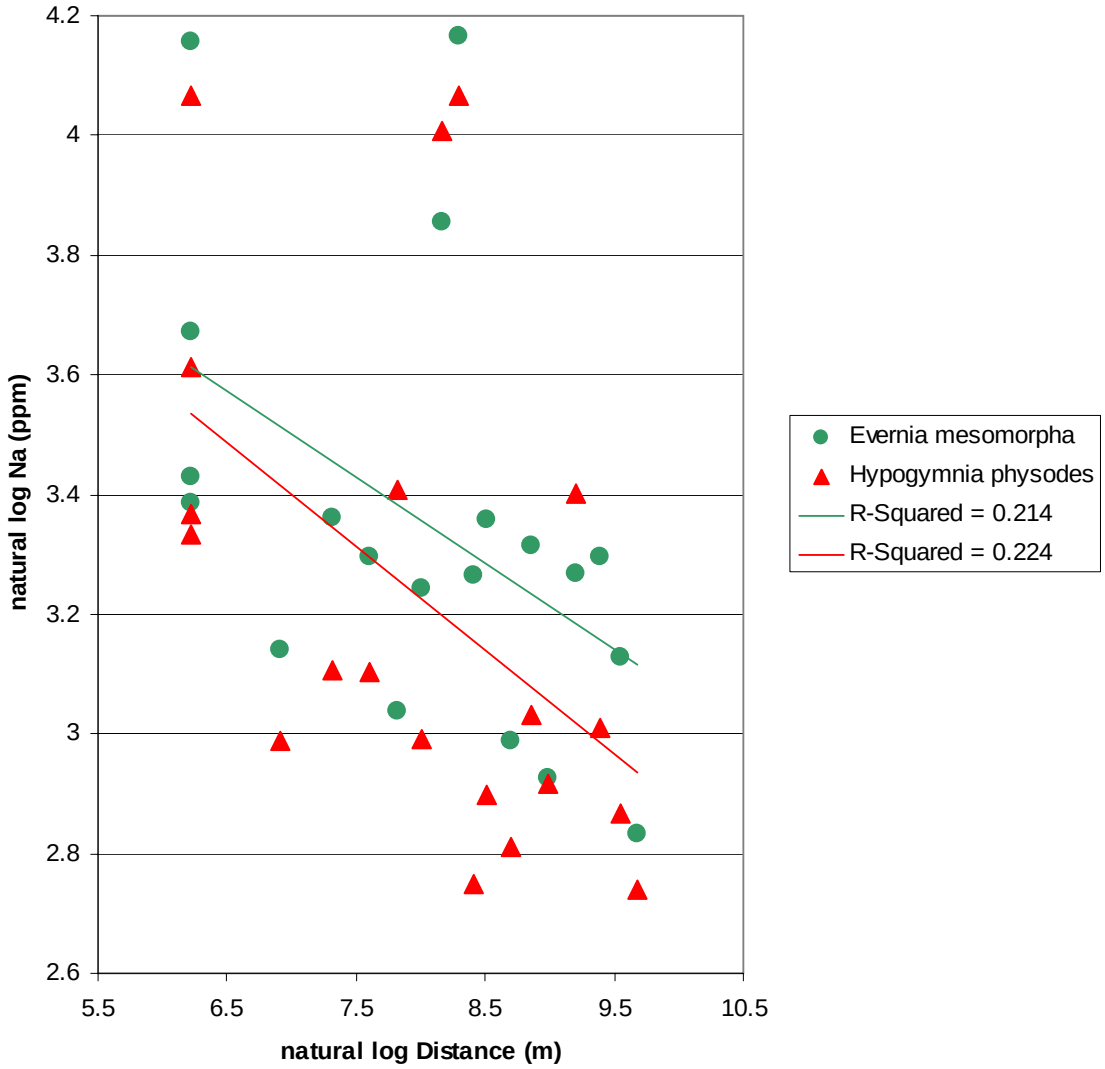

Figure G43. Total Sodium (ppm) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

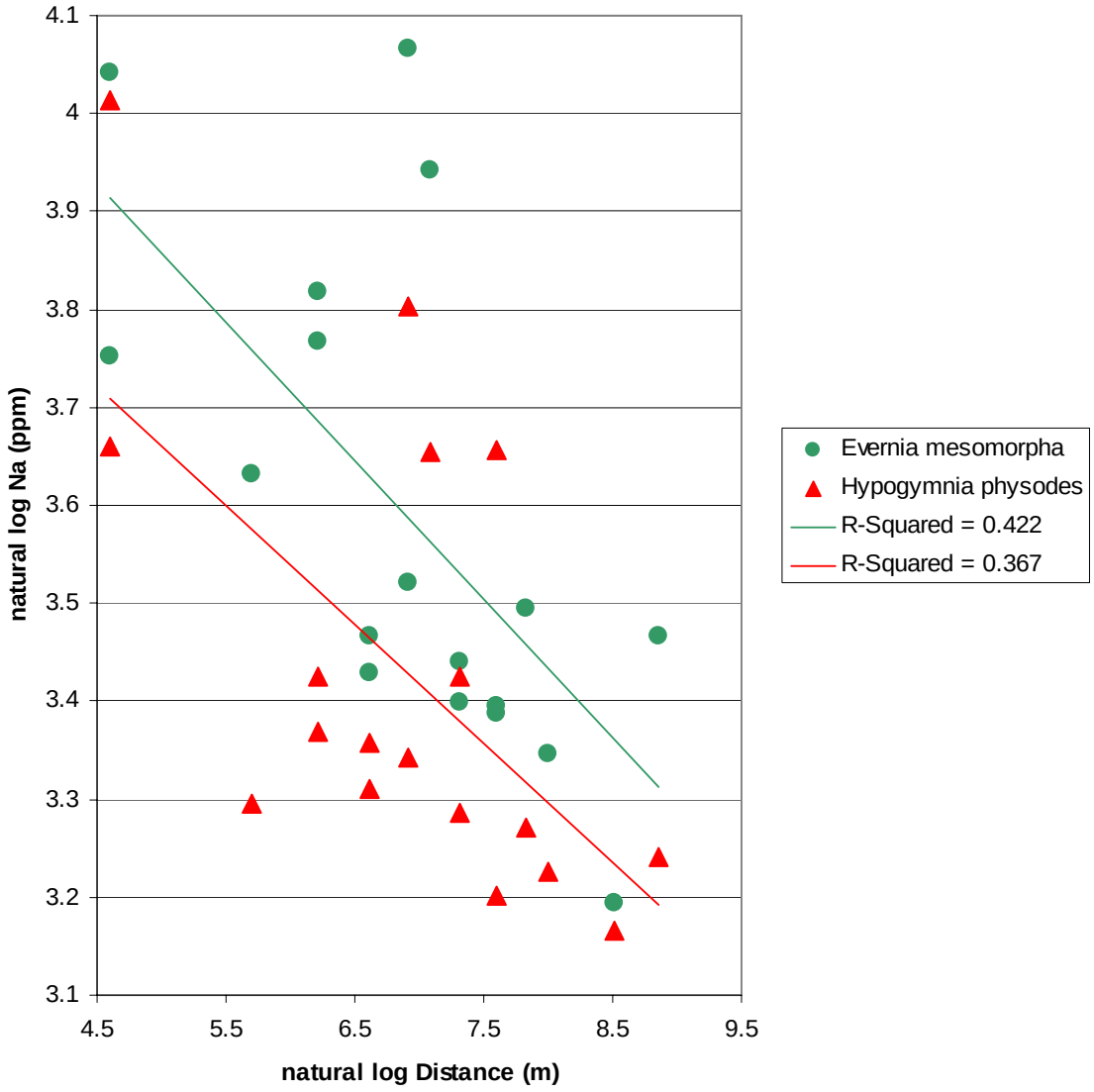

Figure G44. Total Sodium (ppm) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

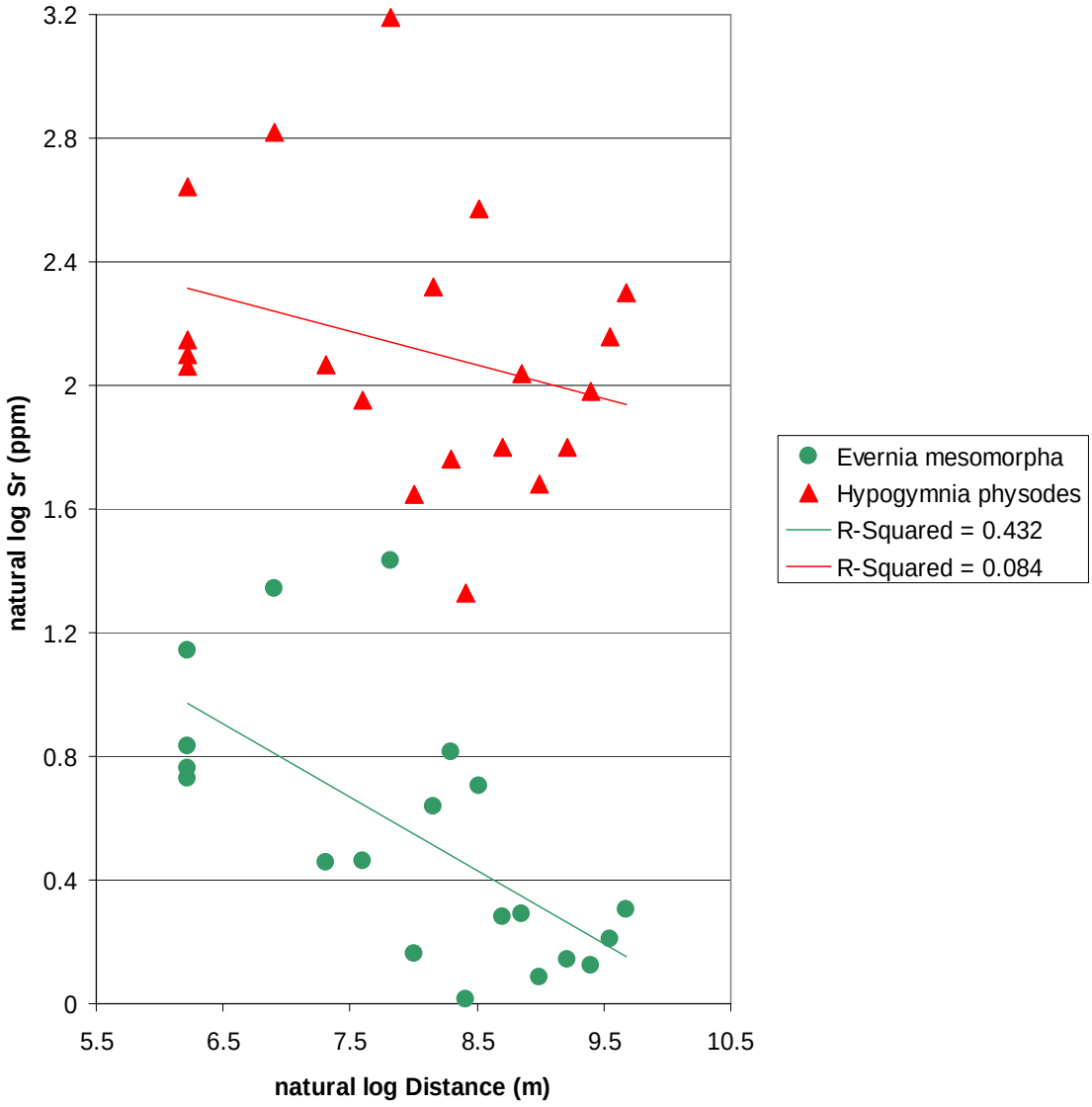

Figure G45. Total Strontium (ppm) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

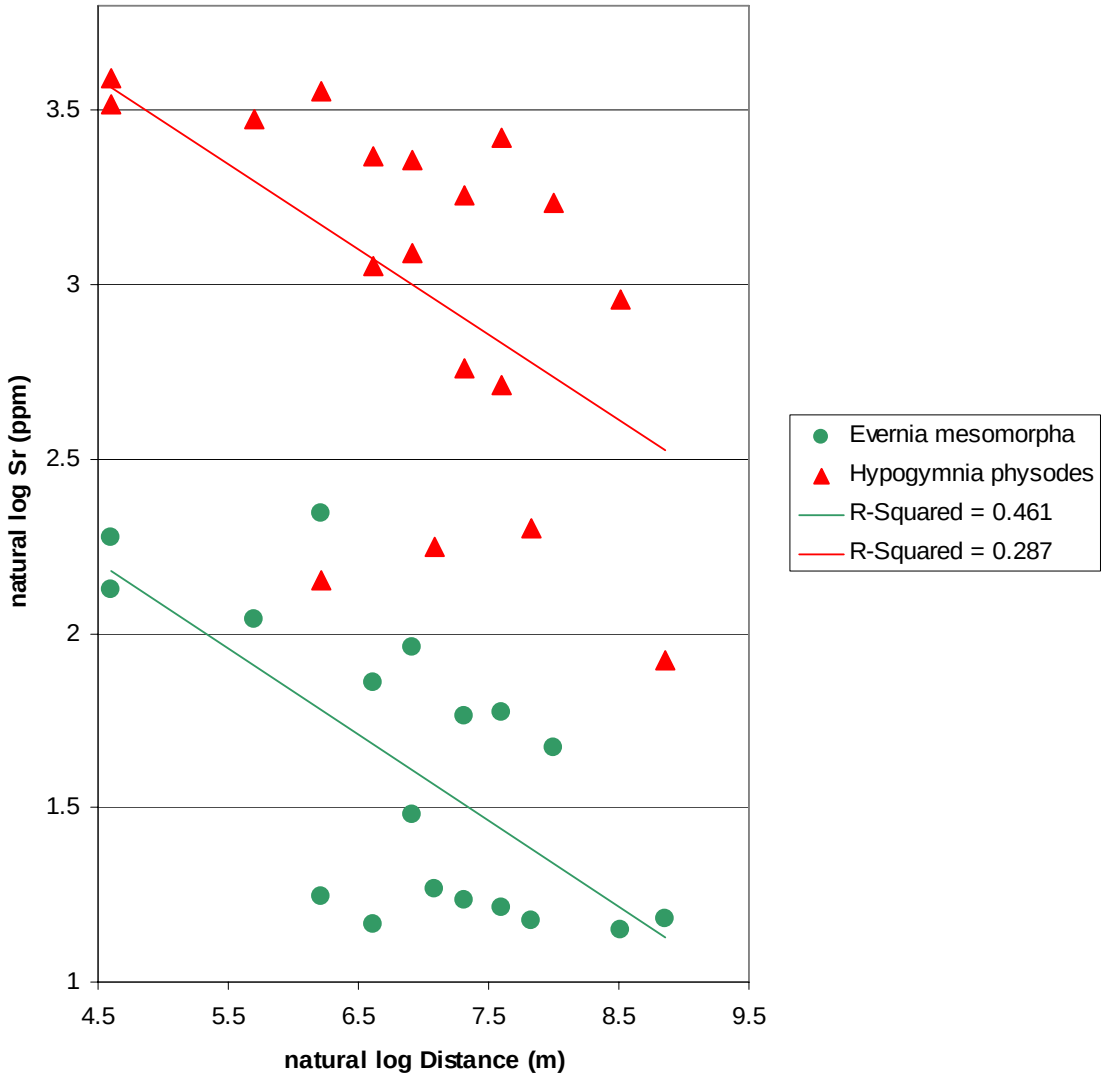

Figure G46. Total Strontium (ppm) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

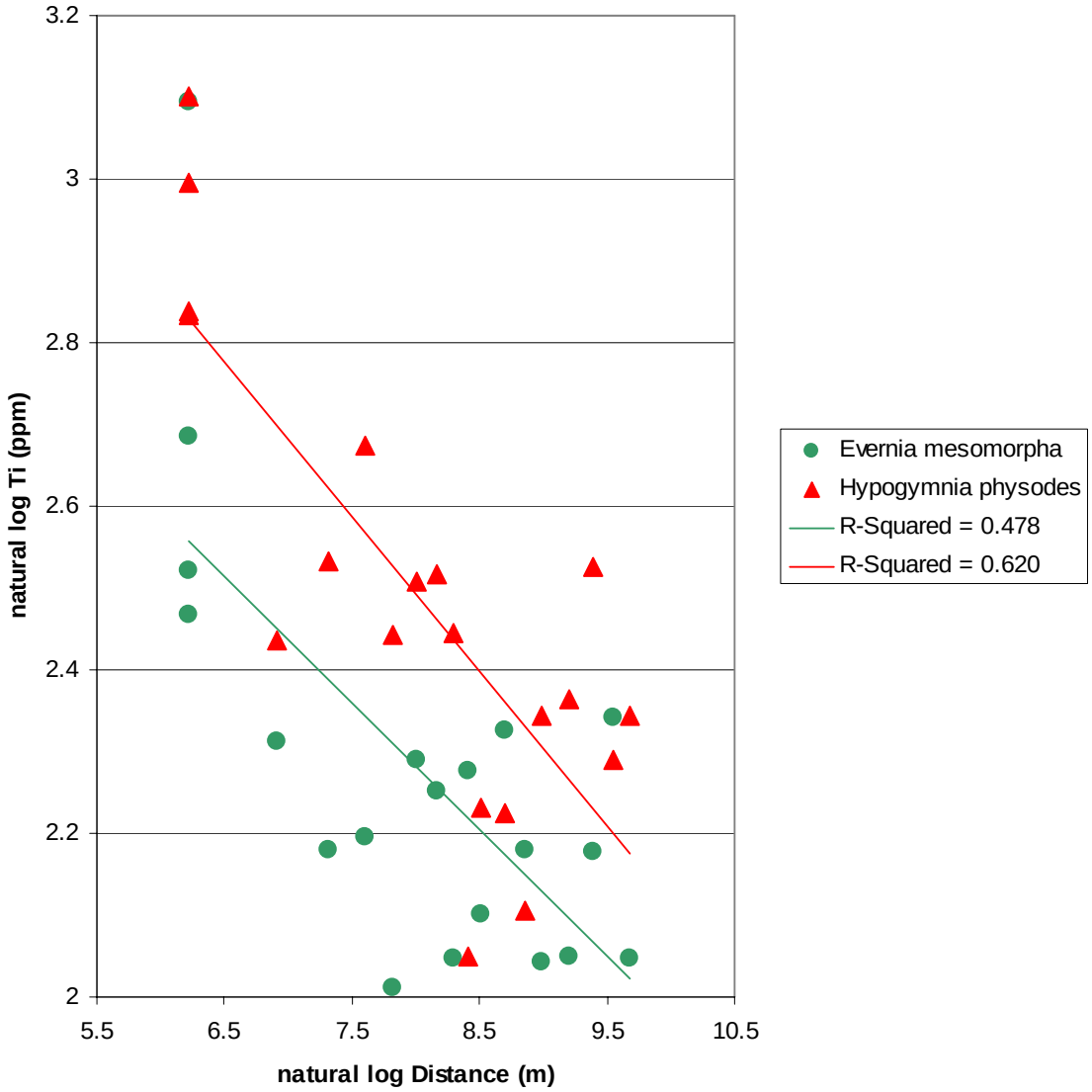

Figure G47. Total Titanium (ppm) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples. For *H. physodes*, as the distance from the mine doubles the concentration of Ti decreases by 12%.

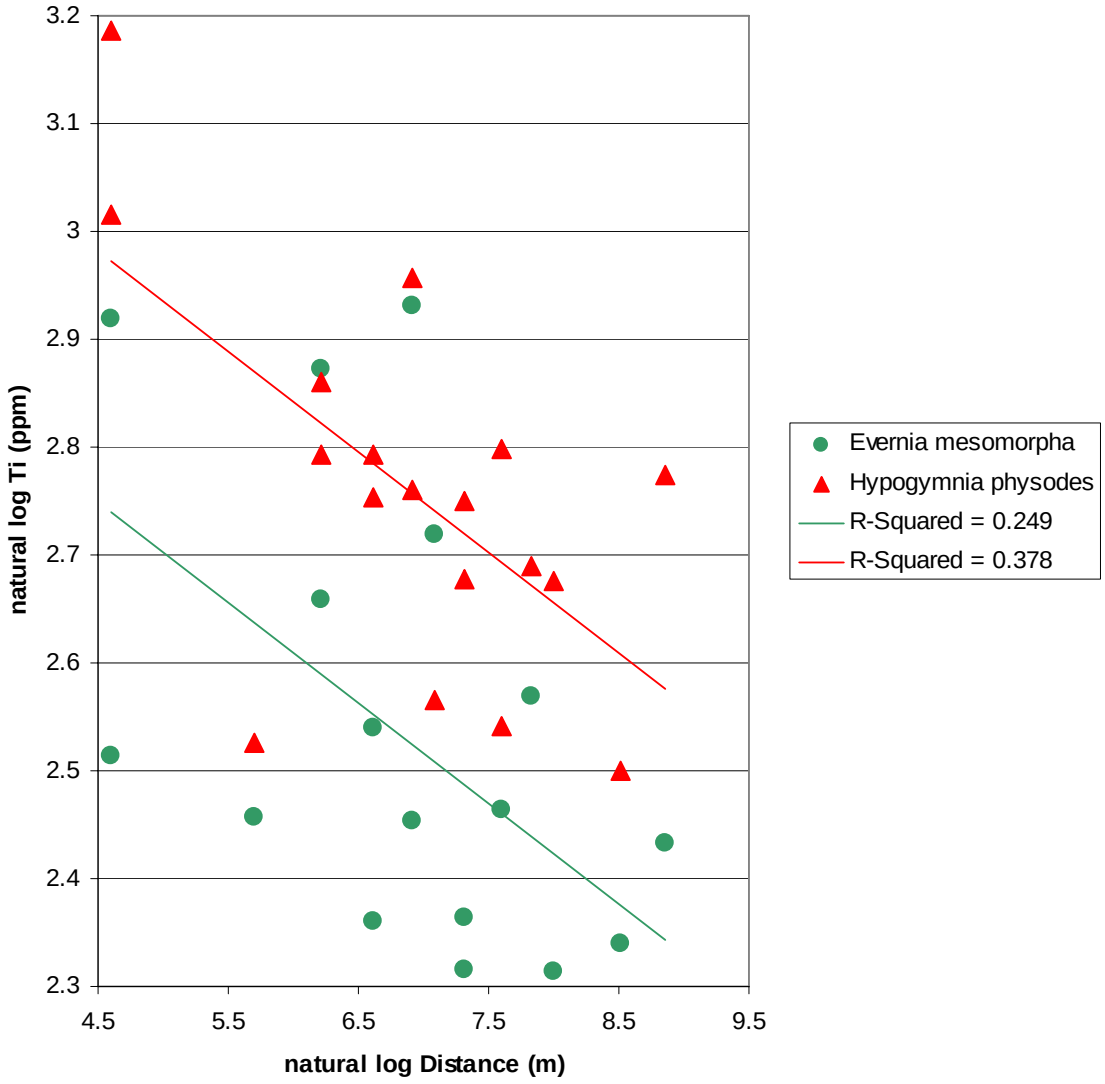

Figure G48. Total Titanium (ppm) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

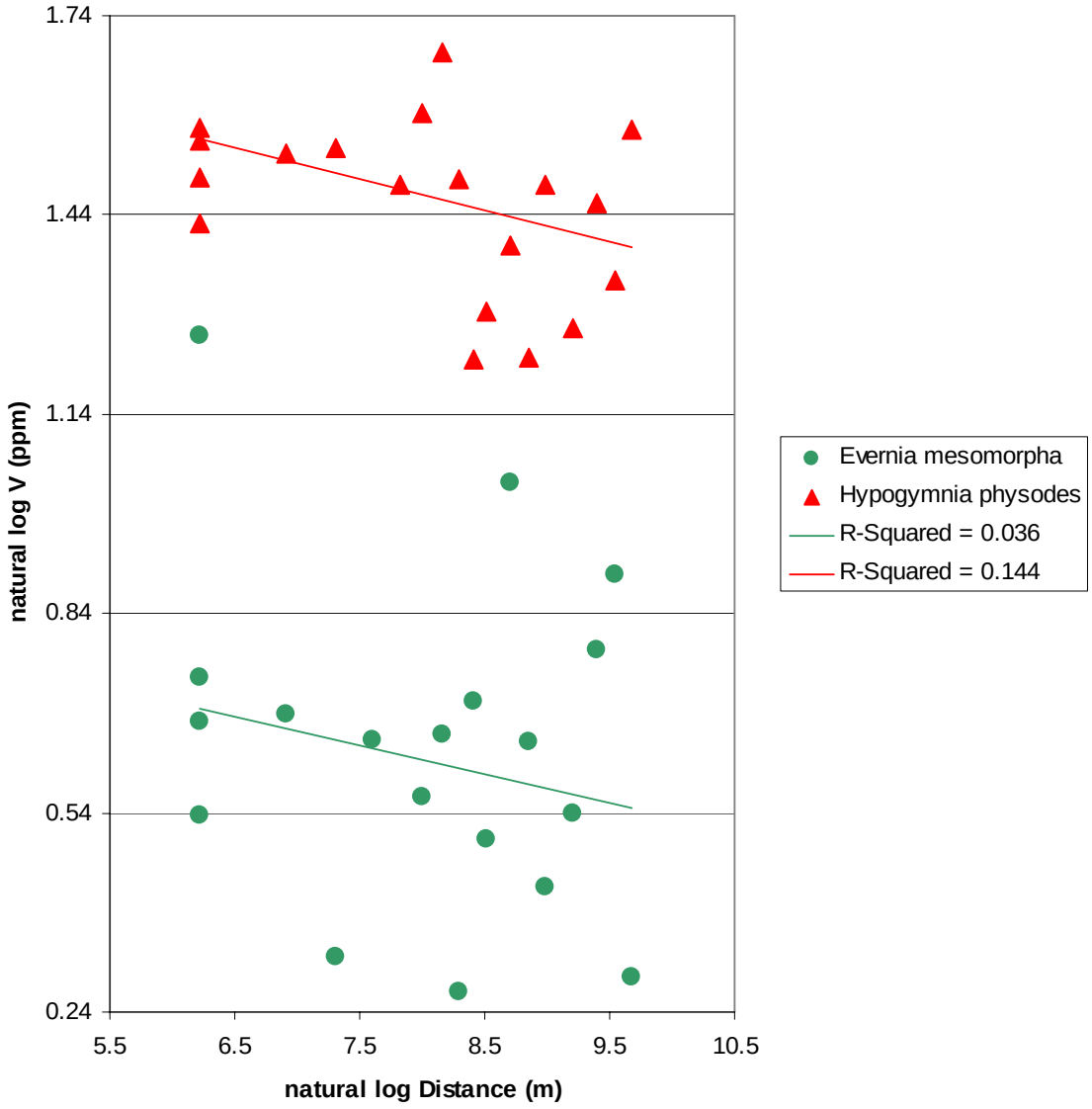

Figure G49. Total Vanadium (ppm) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

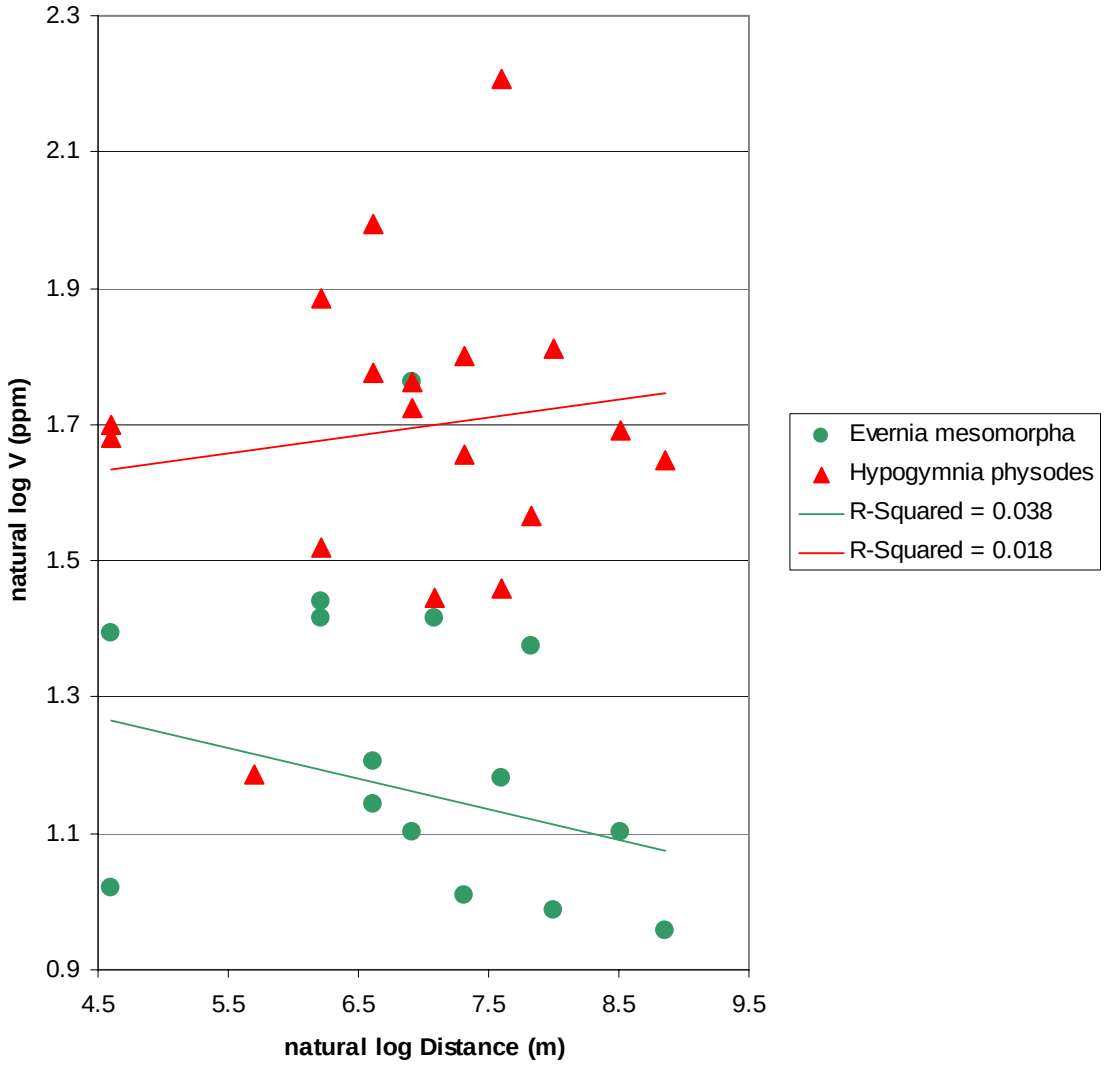

Figure G50. Total Vanadium (ppm) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

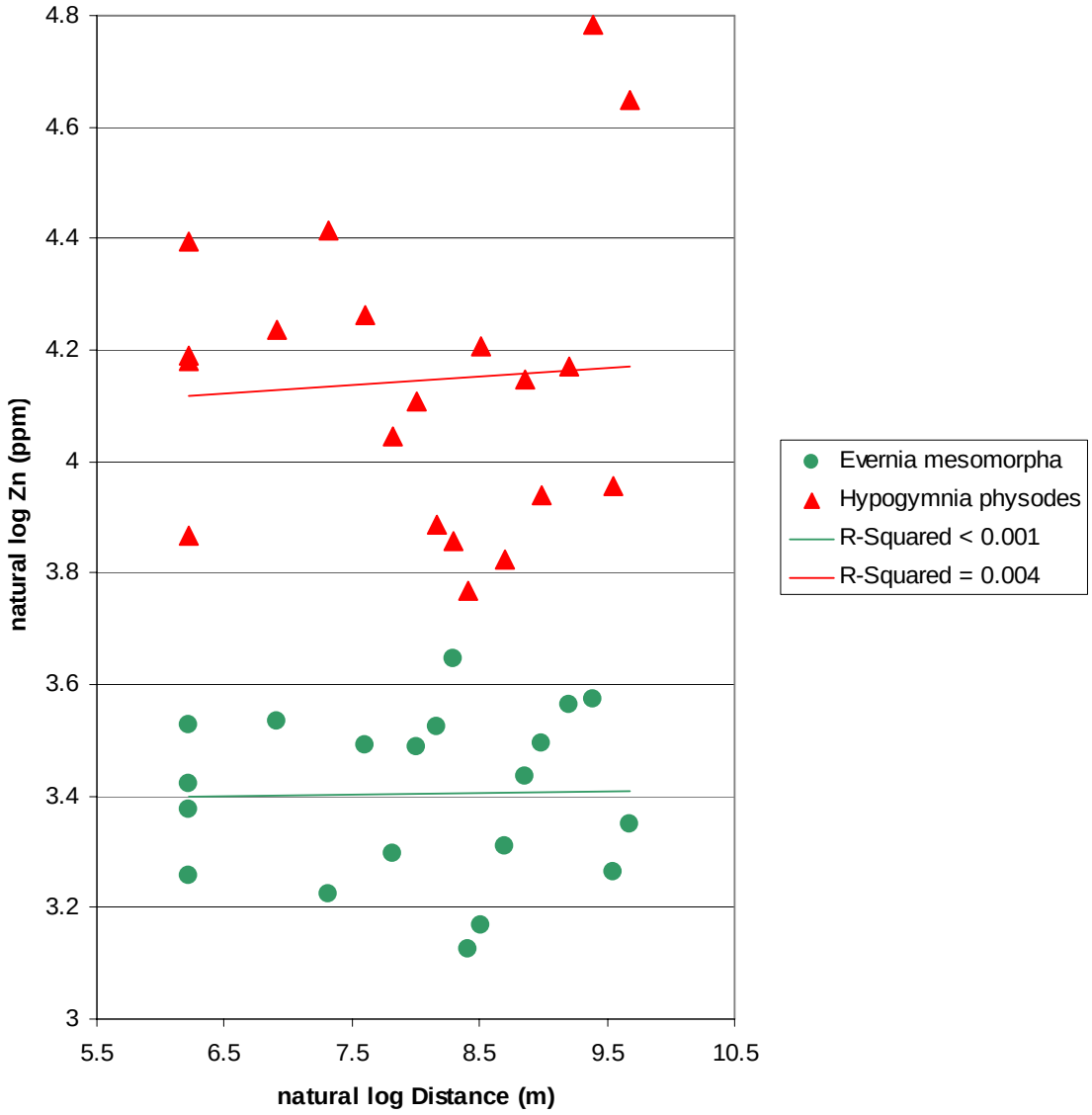

Figure G51. Total Zinc (ppm) in relation to distance (m) from the mines along the Firebag Transect for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

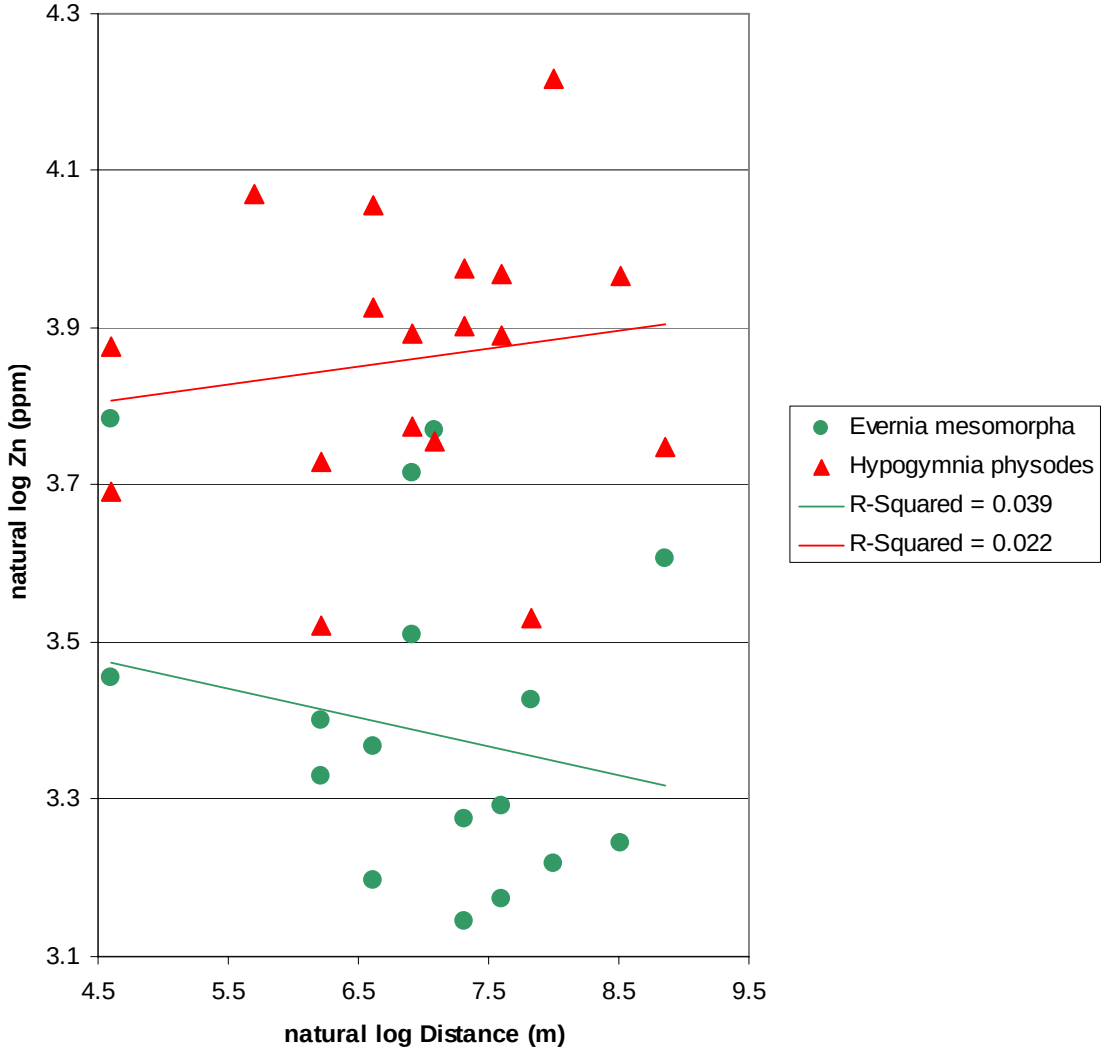

Figure G52. Total Zinc (ppm) in relation to distance (m) from the mines along the Muskeg a and b Transects for *Hypogymnia physodes* and *Evernia mesomorpha* samples.

VIII. SUMMARY STATISTICS

Table F3. Summary statistics for the element concentrations in *Evernia mesomorpha* samples. Total N = number of tissue samples averaged across all field samples (including field replicates); Min. = minimum concentration; Max. = maximum concentration; Median concentration; Std. Dev. = standard deviation; Mean concentration; SE = Standard Deviation of the Mean; 95% LCL = the lower 95% confidence interval of the mean; 95% UCL = the upper 95% confidence interval of the mean. Concentrations of nitrogen and sulfur are in percent dry weight; concentrations of mercury are in ppb; and concentrations of all other elements are in ppm dry weight.

Element	Total N	Min.	Max.	Median	Std Dev	Mean	SE	95% LCL	95% UCL
Al	65	260.730	2269.900	548.450	402.904	663.909	49.974	564.075	763.744
B	65	1.703	9.372	3.376	1.697	3.822	0.210	3.401	4.242
Ba	65	2.639	20.382	5.232	3.137	5.965	0.389	5.188	6.742
Ca	65	394.350	8319.500	797.940	1708.524	1506.147	211.916	1082.795	1929.498
Cr	65	0.370	2.814	0.888	0.494	0.986	0.061	0.864	1.109
Cu	65	1.000	6.662	1.524	0.803	1.744	0.100	1.545	1.943
Fe	65	224.870	3623.400	660.350	699.490	876.905	86.761	703.580	1050.231
K	65	1400.300	3582.800	1831.500	377.123	1910.751	46.776	1817.304	2004.197
Li	34	0.418	2.059	0.741	0.349	0.823	0.060	0.701	0.944
Mg	65	142.980	814.770	263.990	119.530	297.190	14.826	267.572	326.808
Mn	65	12.674	74.456	33.129	14.416	33.996	1.788	30.424	37.568
Mo	53	0.231	1.054	0.375	0.211	0.446	0.029	0.388	0.505
Na	65	16.978	64.647	29.608	13.952	34.353	1.731	30.896	37.810
Ni	65	0.608	2.936	1.155	0.533	1.242	0.066	1.110	1.374
P	65	319.120	1084.400	446.020	138.208	493.381	17.143	459.135	527.628
Si	65	282.380	1342.300	671.530	224.617	699.786	27.860	644.128	755.443
Sr	65	1.014	11.321	3.051	2.424	3.318	0.301	2.717	3.918
Ti	65	6.283	26.620	10.267	4.339	11.478	0.538	10.403	12.553
V	65	1.018	5.861	2.190	1.162	2.525	0.144	2.237	2.813
Zn	65	21.575	45.372	30.011	5.658	30.648	0.702	29.246	32.050
N	65	0.589	1.443	0.786	0.158	0.831	0.020	0.792	0.871
S	65	0.064	0.175	0.080	0.022	0.087	0.003	0.082	0.093
Hg	59	89.780	348.750	188.760	62.091	187.589	8.084	171.408	203.770

Table F3. Summary statistics for the element concentrations in *Hypogymnia physodes* samples. Total N = number of tissue samples averaged across all field samples (including field replicates); Min. = minimum concentration; Max. = maximum concentration; Median concentration; Std. Dev. = standard deviation; Mean concentration; SE = Standard Deviation of the Mean; 95% LCL = the lower 95% confidence interval of the mean; 95% UCL = the upper 95% confidence interval of the mean. Concentrations of nitrogen and sulfur are in percent dry weight; concentrations of mercury are in ppb; and concentrations of all other elements are in ppm dry weight.

Element	Total N	Min:	Max:	Median:	Std Dev	Mean:	SE	LCL	UCL
Al	61	376.830	1847.700	816.480	332.663	827.579	42.593	742.380	912.778
B	61	1.673	5.839	3.655	0.974	3.639	0.125	3.389	3.888
Ba	61	11.831	48.219	25.893	8.974	26.522	1.149	24.224	28.820
Be	33	0.041	0.097	0.057	0.016	0.060	0.003	0.054	0.066
Ca	61	4449.900	43681.000	11410.000	8060.976	14060.125	1032.102	11995.613	16124.636
Cd	58	0.124	0.615	0.193	0.105	0.222	0.014	0.194	0.250
Cr	61	0.455	2.680	1.171	0.502	1.212	0.064	1.084	1.341
Cu	61	1.756	3.873	2.608	0.530	2.646	0.068	2.510	2.782
Fe	61	475.190	3055.100	1261.600	650.427	1275.547	83.279	1108.965	1442.129
K	61	1819.100	3475.400	2451.600	351.311	2470.318	44.981	2380.343	2560.293
Li	35	0.410	1.445	0.574	0.280	0.666	0.048	0.568	0.764
Mg	61	394.190	1381.500	727.210	212.358	742.365	27.190	687.978	796.753
Mn	61	68.142	643.800	150.790	126.965	201.122	16.256	168.605	233.640
Mo	58	0.224	0.823	0.428	0.163	0.449	0.022	0.405	0.492
Na	61	15.500	58.579	26.376	11.039	28.807	1.413	25.979	31.634
Ni	61	1.367	4.287	2.610	0.690	2.593	0.088	2.417	2.770
P	61	418.540	1214.500	698.480	191.951	715.102	24.577	665.941	764.263
Pb	61	1.938	4.914	2.981	0.624	3.076	0.080	2.916	3.235
Si	61	365.040	1337.700	762.030	222.155	760.908	28.444	704.011	817.804
Sr	61	3.771	36.975	13.496	10.271	16.463	1.315	13.832	19.093
Ti	61	7.292	24.203	13.455	3.767	13.985	0.482	13.020	14.950
V	61	2.711	9.079	4.722	1.201	4.861	0.154	4.554	5.169
Zn	61	29.709	119.620	52.280	17.444	56.453	2.233	51.985	60.920
N	62	0.481	1.000	0.619	0.084	0.638	0.011	0.616	0.659
S	62	0.029	0.086	0.051	0.013	0.052	0.002	0.049	0.056
Hg	56	71.800	187.010	114.690	30.120	118.777	4.025	110.711	126.843

IX. ORDINATIONS

Samples in Element Space

Non-metric multidimensional scaling (NMS) ordination was used to determine how all the lichen samples, across both species, were related based on their elemental concentrations (20 elements). The ordination summarizes complex relationships among the lichen samples based on elemental concentrations in the lichens. The ordination axes are synthetic, representing patterns of elemental concentrations in the lichen samples. The rotated two-dimensional NMS ordination solution of lichen samples in elemental space for Muskeg described 97.5% of the variation in patterns of elemental concentrations in lichen samples (axis 1 = 59.7% and axis 2 = 37.8%; Figure 3.1). The rotated two-dimensional NMS ordination solution of lichen samples in elemental space for Firebag described 98.2% of the variation in patterns of elemental concentrations in lichen samples (axis 1 = 5.9% and axis 2 = 92.3%; Figure 3.2).

Samples of both lichen species (i.e., Evernia mesomorpha, and Hypogymnia physodes) separated out in the ordination space (Figure 3.1 and Figure 3.2). The separation of lichen species in the ordination indicates that the concentrations of the 20 elements in the lichen samples were more similar within species than among species.

Muskeg Ordination

For the Muskeg locality, Distance (km) from the mines was correlated with axis 1 (R^2 = 0.33; Table 3.4), such that those lichen samples positioned to the left on axis 1 were collected from sites nearer to the mines than lichen samples further to the right on axis 1 (Figure 3.1). Those samples collected from stands near the mines contained highest concentrations of elements for both lichen species. The elements most highly correlated only with axis 1 (R^2 > 0.5) included: aluminum, chromium, iron, nickel, magnesium, calcium, barium, strontium, titanium, potassium, phosphorous, manganese, zinc, copper, vanadium, and silicon (Table 3.4). Nitrogen and sulphur were also correlated with axis 1, though the correlations were relatively weak compared to the metals mentioned above (Table 3.4). Some elements were correlated with both axis 1 and axis 2, these included: magnesium, nickel, calcium, barium, strontium, manganese, and zinc (Table 3.4).

The elements magnesium, nickel, calcium, barium, strontium, manganese, and zinc were most highly negatively correlated with axis 2 and nitrogen and sulphur were most highly positively correlated with axis 2. Axis 2 explained relatively little variation in the elemental concentrations in lichens (37.8% variation explained by axis 2) and was not related to distance from the mines (Figure 3.1; Table 3.4).

Table 3.4. Pearson's correlations (R^2) with the two-dimensional NMS ordination axes of lichen samples from Muskeg in elemental space (Figure 3.1). Correlations are shown for the log_{10}-transformed concentrations (dry weight) of 20 elements in the lichen samples and for distance from the mine sites with each ordination axis.

Element	Axis 1	Axis 2
Distance	0.333	0.041
Al	-0.749	-0.126
B	-0.253	0.327
Ba	-0.860	-0.926
Ca	-0.885	-0.887
Cr	-0.802	-0.200
Cu	-0.716	-0.329
Fe	-0.815	-0.205
K	-0.839	-0.397
Mg	-0.945	-0.850
Mn	-0.745	-0.905
Na	-0.212	0.391
Ni	-0.900	-0.732
P	-0.779	-0.339
Si	-0.687	-0.099
Sr	-0.855	-0.897
Ti	-0.849	-0.404
V	-0.691	-0.656
Zn	-0.732	-0.708
N	0.171	0.755
S	0.480	0.882

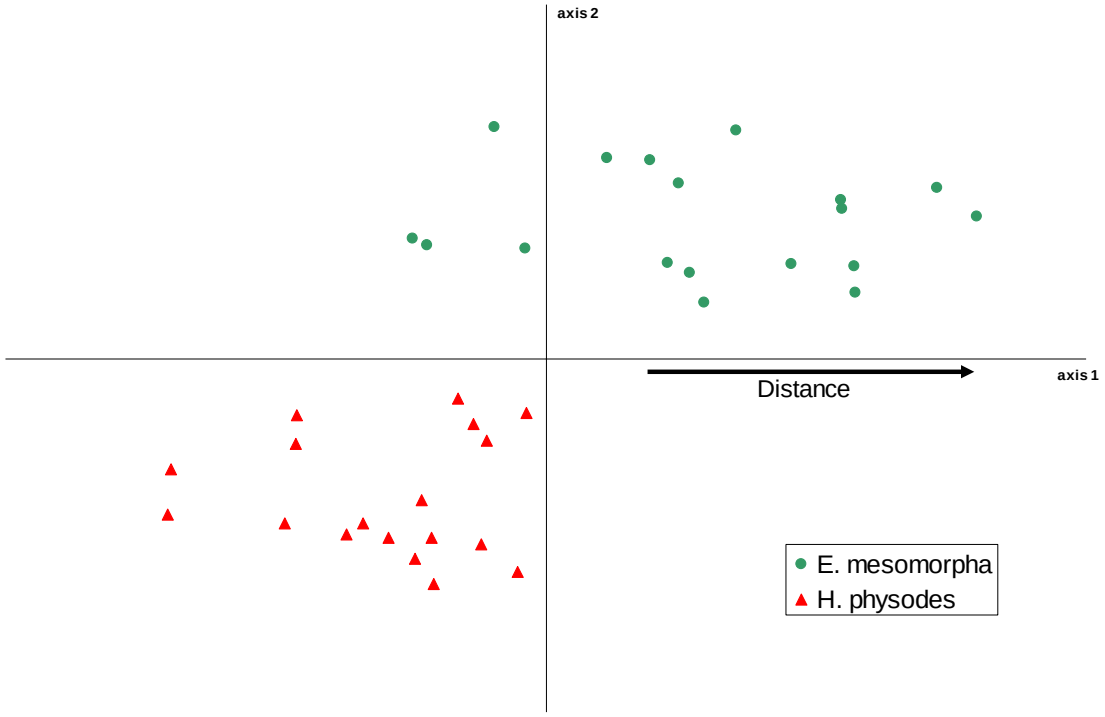

Figure 3.1. Two-dimensional NMS ordination of lichen tissue samples from Muskeg in elemental space (20 elements). Lichen samples are represented by symbols, coded by lichen species. Samples collected nearer to the mines are positioned to the left on axis 1 and samples from farther distances are to the right on axis 1. Lichen samples that are close together in the ordination space have similar concentrations of the elements, while samples that are farther apart in the ordination space have less similar elemental content.

Firebag Ordination

For the Firebag locality, Distance (km) from the mines was correlated with axis 1 (R^2 = 0.38; Table 3.5), such that those lichen samples positioned to the left on axis 1 were collected from sites nearer to the mines than lichen samples further to the right on axis 1 (Figure 3.2). Those samples collected from stands near the mines contained highest concentrations of elements for both lichen species. The elements most highly correlated only with axis 1 (R^2 > 0.5) included: aluminum, chromium, iron, nickel, calcium, barium, strontium, titanium, potassium, magnesium, zinc, copper, vanadium, and silicon (Table 3.5). Sulphur was also correlated with axis 1, although its correlation was positive compared the negative correlations of the other elements mentioned. Nitrogen had a relatively weak positive correlation to axis 1 (Table 3.5). Some elements were correlated with both axis 1 and axis 2, these included: aluminum, iron, nickel, calcium, barium, strontium, potassium, magnesium, zinc, copper, and vanadium (Table 3.5).

The elements magnesium, nickel, calcium, barium, strontium, manganese, vanadium, potassium, copper, phosphorous, iron, aluminum, and zinc were most highly negatively correlated with axis 2 and nitrogen and sulphur were most highly positively correlated with axis 2. Axis 2 explained a majority of the variation in the elemental concentrations in lichens (92.3% variation explained by axis 2) but was not related to distance from the mines (Figure 3.2; Table 3.5).

Table 3.5. Pearson's correlations (R^2) with the two-dimensional NMS ordination axes of lichen samples from Firebag in elemental space (Figure 3.1). Correlations are shown for the $\log_{10}$-transformed concentrations (dry weight) of 20 elements in the lichen samples and for distance from the mine sites with each ordination axis.

Element	Axis 1	Axis 2
Distance	0.381	0.086
Al	-0.953	-0.580
B	-0.181	-0.064
Ba	-0.650	-0.986
Ca	-0.669	-0.985
Cr	-0.817	-0.306
Cu	-0.892	-0.794
Fe	-0.949	-0.704
K	-0.578	-0.829
Mg	-0.815	-0.954
Mn	-0.498	-0.946
Na	-0.345	0.133
Ni	-0.869	-0.846
P	-0.347	-0.745
Si	-0.624	-0.221
Sr	-0.703	-0.944
Ti	-0.890	-0.388
V	-0.817	-0.870
Zn	-0.637	-0.881
N	0.220	0.741
S	0.509	0.842

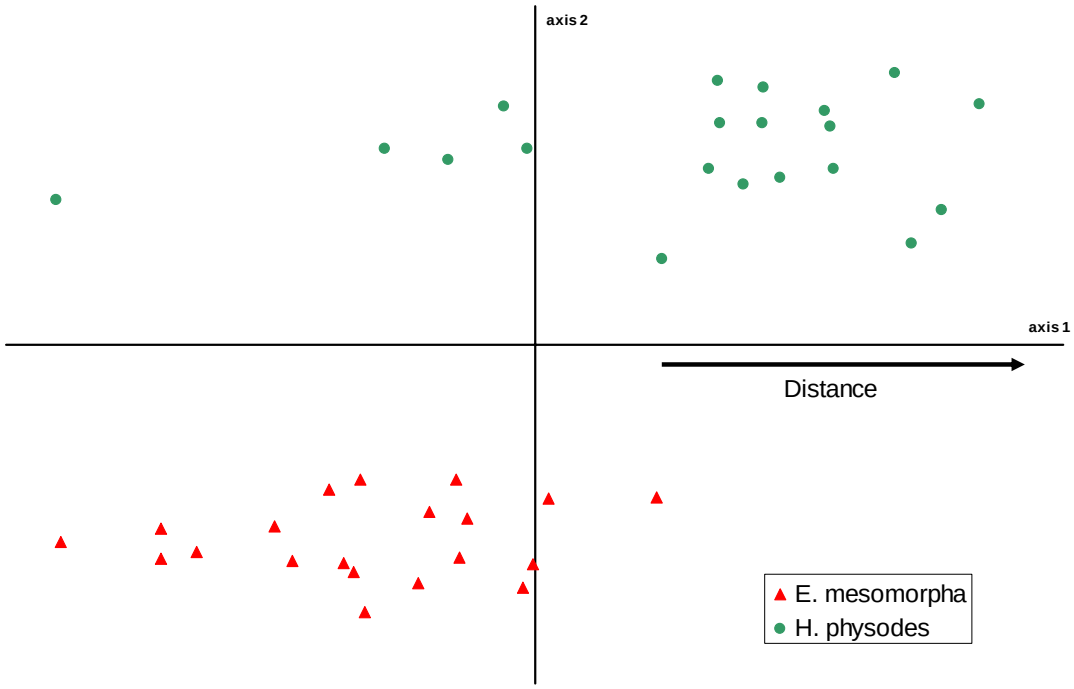

Figure 3.1. Two-dimensional NMS ordination of lichen tissue samples from Firebag in elemental space (20 elements). Lichen samples are represented by symbols, coded by lichen species. Samples collected nearer to the mines are positioned to the left on axis 1 and samples from farther distances are to the right on axis 1. Lichen samples that are close together in the ordination space have similar concentrations of the elements, while samples that are farther apart in the ordination space have less similar elemental content.

Sites in Lichen Space

Muskeg Species Ordination

Differences in lichen communities in the Muskeg locality were most strongly related to species richness and diversity, canopy cover and distance (km) from the mining operations. The two-dimensional NMS ordination of stands in lichen species space described 91.7% of the cumulative variance in the community structure (axis 1 = 70.6% and axis 2 = 21.1 %, Figure 3.18). Distance and diversity were correlated with axis 1 (R^2 0.60 and 0.61 respectively). Nitrophiles were negatively correlated with axis 1 (R^2 = -0.52). Total species richness was correlated with axis 2 (R^2 = 0.58) (Table 3.21). More diverse stands were located furthest from the mines.

Table 3.21. Pearson's correlations (R^2) of environmental variables, lichen diversity measures, and selected elemental concentrations in Hypogymnia physodes with Muskeg NMS ordination axes (Figure 3.18).

Variable	Axis 1	Axis 2
Plot distance to mine site (m)	0.603	0.144
Elevation (m)	0.158	0.078
Average tree age	0.233	0.276
Tree canopy, % Cover of ground area	-0.455	0.372
Total species richness	0.181	0.578
Shannon's Diversity Index	0.610	0.366
Species richness of nitrophiles	-0.520	0.314
Total % sulphur in H. physodes	0.153	0.284
Aluminum (µg/g) in H. physodes	-0.551	-0.385
Nickel (µg/g) in H. physodes	-0.315	-0.360
Vanadium (µg/g) in H. physodes	0.219	-0.117
Ba (µg/g) in H. physodes	-0.226	-0.797
Cu (µg/g) in H. physodes	-0.741	-0.343
Mn (µg/g) in H. physodes	0.131	-0.297
P (µg/g) in H. physodes	-0.669	0.112
Pb (µg/g) in H. physodes	-0.052	-0.606
Zn (µg/g) in H. physodes	0.130	-0.346
Total % Nitrogen in H. physodes	-0.318	-0.020
Hg (ppb) in H. physodes	0.287	0.503

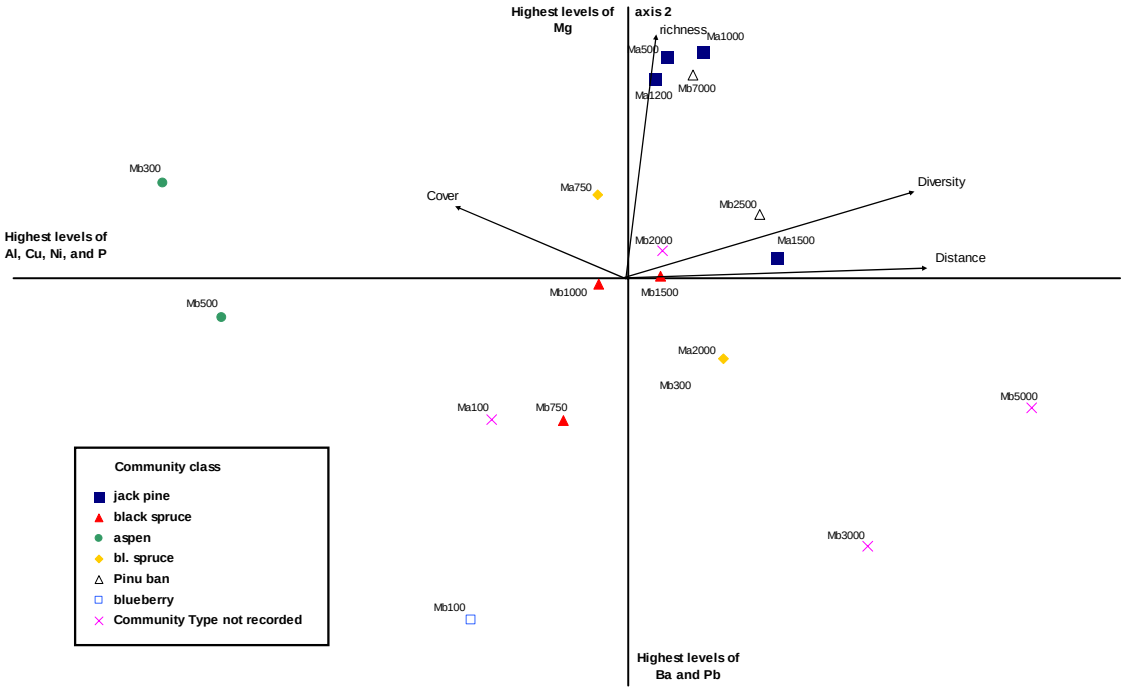

Figure 3.18. NMS ordination of Muskeg stands in lichen species space. Symbols represent the stands and are coded for the different forest community classes. Stands are labeled by plot number, where the letter indicates direction (north, south, east or west) and the number indicates the distance interval. The black vectors represent the relationship of distance to the mines (km), species diversity, species richness, and percent canopy cover with axis 1 and 2. The length of the vector indicates the strength of the correlation with the axis. Stands that are close together in the ordination space have more similar lichen communities than samples that are farther apart in the ordination space.

Firebag Species Ordination

Differences in lichen communities in the Firebag locality were most strongly related to species diversity, canopy cover and average tree age. The two-dimensional NMS ordination of stands in lichen species space described 89.1% of the cumulative variance in the community structure (axis 1 = 51.2% and axis 2 = 37.8 %, Figure 3.19). None of the environmental variables were strongly correlated with axis 1 (R^2 < 0.50, Table 3.22). Species diversity and canopy cover were negatively correlated with axis 2 (R^2 = -0.62 and -0.51 respectively).

Table 3.22. Pearson's correlations (R^2) of environmental variables, lichen diversity measures, and selected elemental concentrations in Hypogymnia physodes with Firebag NMS ordination axes (Figure 3.19).

Variable	Axis 1	Axis 2
Plot distance to mine site (m)	-0.066	0.351
Elevation (m)	-0.043	-0.086
Average tree age	-0.296	0.429
Tree canopy, % Cover of ground area	0.121	-0.508
Total species richness	0.134	-0.058
Shannon's Diversity Index	0.223	-0.622
Species richness of nitrophiles	0.208	0.365
Total % sulphur in H. physodes	0.312	0.601
Aluminum (µg/g) in H. physodes	0.491	-0.530
Nickel (µg/g) in H. physodes	0.253	-0.155
Vanadium (µg/g) in H. physodes	0.115	-0.346
Ba (µg/g) in H. physodes	-0.670	-0.433
Cu (µg/g) in H. physodes	0.618	-0.225
Mn (µg/g) in H. physodes	-0.557	0.014
P (µg/g) in H. physodes	-0.387	-0.227
Pb (µg/g) in H. physodes	0.094	-0.400
Zn (µg/g) in H. physodes	-0.093	0.034
Total % Nitrogen in H. physodes	0.373	-0.333
Hg (ppb) in H. physodes	0.042	-0.425

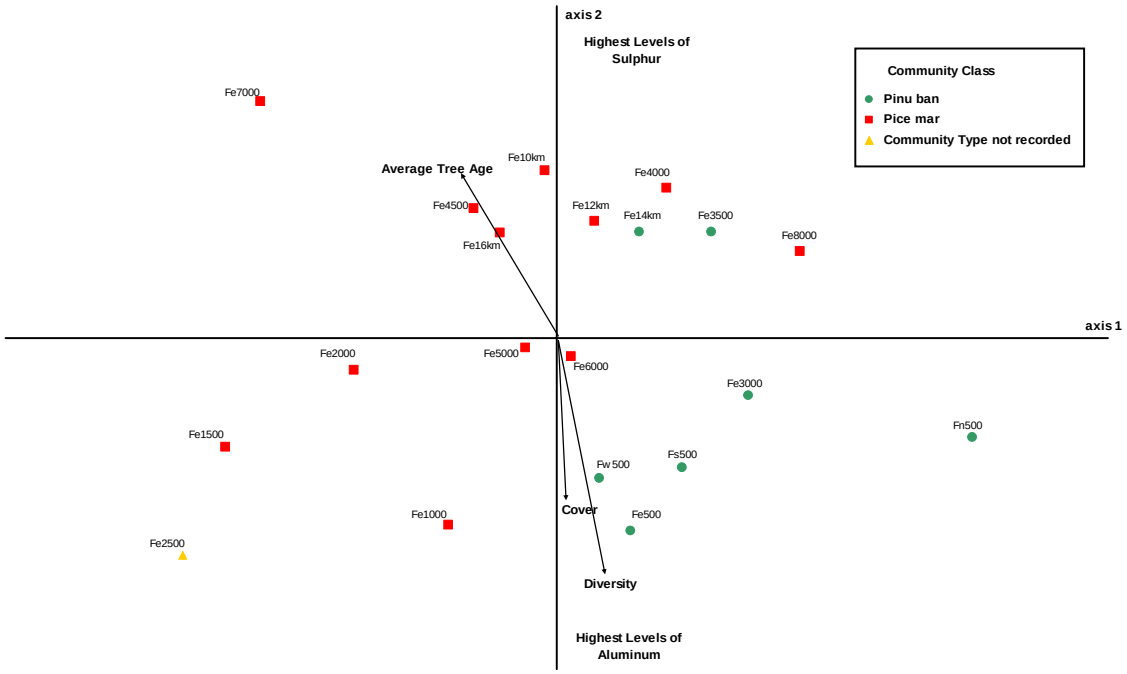

Figure 3.19. NMS ordination of Firebag stands in lichen species space. Symbols represent the stands and are coded for the different forest community classes. Stands are labeled by plot number, where the letter indicates direction (north, south, east or west) and the number indicates the distance interval. The black vectors represent the relationship of species diversity, percent canopy cover and average tree age with axis 1 and 2. The length of the vector indicates the strength of the correlation with the axis. Stands that are close together in the ordination space have more similar lichen communities than samples that are farther apart in the ordination space.

X. SPECIES DIVERSITY

Shannon's species diversity index was calculated for each of the sampling sites

Muskeg

Site	Shannon's Diversity
Ma100	2.237
Ma500	2.386
Ma750	2.390
Ma1000	2.521
Ma1200	2.456
Ma1500	2.289
Ma2000	2.278
Mb100	2.410
Mb300	2.138
Mb500	2.126
Mb750	2.153
Mb1000	2.348
Mb1500	2.374
Mb 2000	2.337
Mb2500	2.565
Mb3000	2.277
Mb5000	2.471
Mb7000	2.485
AVERAGE	**2.347**

Firebag

Site	Shannon's Diversity
Fe1000	2.506
Fe10km	2.422
Fe12km	2.491
Fe14km	2.416
Fe1500	2.441
Fe16km	2.471
Fe2000	2.562
Fe2500	2.487
Fe3000	2.512
Fe3500	2.475
Fe4000	2.458
Fe4500	2.405
Fe500	2.539
Fe5000	2.581
Fe6000	2.438
Fe7000	2.318
Fe8000	2.498
Fn500	2.460
Fs500	2.565
Fw500	2.608
AVERAGE	**2.483**

VII. LITERATURE CITED

Anderson, B.M., T.W. Anderson, and I. Olkin. 1986. Maximum likelihood estimators and likelihood ratio criteria in multivariate components of variance. Annals of Statistics 14:405-417.

Kruskal, J.B. 1964. Nonmetric multidimensional scaling: a numerical method. Psychometrika 29: 1-27.

Mather, P.M. 1976. Computational methods of multivariate analysis in physical geography. J. Wiley and Sons, London. 532 pp.

MathSoft. 1988-1999. S-Plus 2000. Data Analysis Products Division, Seattle, WA.

McCune, B. & M.J. Mefford. 1999. PC-ORD. Multivariate Analysis of Ecological Data, Version 4. MjM Software Design, Gleneden Beach, Oregon, USA.

Milliken, G.A. and D.E. Johnson. 1992. Analysis of Messy Data, Volume 1: Designed Experiments. Chapman & Hall, New York. 473 pp.

Montgomery, D.C. 1991. Design and Analysis of Experiments. Third Edition. John Wiley& Sons, New York. 649 pages.

Patterson, H.D. and R. Thompson. 1971. Recovery of inter-Treatment Area information when Treatment Area sizes are unequal. Biometrika 58:545-554.

Searle, S.R., G. Casella, and C.E. McCulloch. 1992. Variance Components. John Wiley and Sons, Inc., New York.