

AN ABSTRACT OF THE THESIS OF

Elisa B. Di Meglio for the degree of Master of Science in Botany and Plant Pathology presented on August 5, 2019.

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Abstract approved:

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The Red Dog Mine transports concentrated zinc and lead ore in northwestern Alaska through Cape Krusenstern National monument along a haul road. High metal levels in the moss *Hylocomium splendens* along the haul road have been attributed to ore dust released during transport. The mine has implemented several pollution abatement measures to reduce fugitive ore dust since the original study in 2001, and a subsequent remeasure in 2006.

In 2017, we remeasured moss metal concentrations and lichen and vegetation communities with the purpose of documenting potential effects of metal deposition, and climate change to the local ecosystem from 2006 to 2017. Our results indicate significant changes in lichen and vegetation communities as seen through a loss in diversity and a gain in mean total cover. In both years lichen diversity and cover had negative relationships to metal deposition and distance from the haul road. We observed a decrease in lichen diversity and a doubling in cover over time. Vegetation

diversity was nearly constant but similarly doubled in cover over time. Changes in metal content of mosses during the study period were not significant as evident from the subset of data from 60 plots available. The lichen community was correlated to metal content, which suggests a degree of sensitivity and their appropriate use as biomonitors in this system. We found no significant differences in communities between the north and south sides of the haul road. Changes observed through the study period suggest a relationship to climate change. This was observed through increases in total lichen and vegetation cover as likely mediated by regional climate, biome, temperature and drying in wetlands. Overall, pronounced changes were documented within a mere decade, indicating that continued monitoring into the future is necessary and will be an invaluable resource for future generations.

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Decadal Changes in Lichen and Vegetation Communities in Relation to Metal
Deposition and Climate in Northern Alaska

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Elisa B. Di Meglio

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I understand that my thesis will become part of the permanent collection of Oregon State University libraries. My signature below authorizes release of my thesis to any reader upon request.

Elisa B. Di Meglio, Author

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Chapter 1: Introduction

The effects of atmospheric deposition on human health and the loss of ecosystem services (e.g., drinking water and clean air) have increased demand for reliable, accessible and cost-effective methods for environmental evaluation. Many traditional monitoring methods are limited by problems of high cost and availability of adequate sampling. Biomonitoring is defined as the use of organisms and biomaterials to obtain information on characteristics of the biosphere (Szczepaniak and Biziuk, 2003). Mosses and lichens are frequently used as biomonitors for air quality. They indicate poor air quality through declining diversity (Davies et al., 2007), physiological changes (Boquete et al., 2013), and bioaccumulation of pollutants (Wolterbeek, 2002). Attributes which make these organisms ideal biomonitors are the lack of a waxy cuticle, long lifespans, slow growth rates, drought tolerance and the acquisition of water and nutrients primarily from atmospheric deposition (Bargagli, 2016). The use of vascular plants in conjunction with lichens as biomonitors is limited (Nali et al., 2007; Smodiš et al., 2004) and even more limited is the comparison of community level relationships of vascular plants and lichens to metal deposition and climate change. The present study aims to address this knowledge gap and contribute ideas for future investigations that integrate both community analyses and environmental chemistry as effective methods to monitor anthropogenic pollution sources and climate change.

The arctic tundra biome covers about 20% of the earth's surface and is characterized by cold temperatures, high winds and permafrost. This biome receives low precipitation averaging about 20 - 60 cm per year and is dominated by low dwarf shrubs, grasses, forbs, mosses and lichens (Campbell and Reece, 2005). This region is particularly exposed to a changing climate in

part due to a synergistic feedback of both the amount and persistence of sea ice in the Arctic Ocean combined with the alteration of historic snow cover resulting in changes in the albedo of land surfaces (Bigelow et al., 2003). Other potential drivers of change in the Arctic are increased average air temperatures which are changing at a rate two times faster than global averages (Taylor et al., 2017), changes in permafrost depth and water, and nutrient and soil moisture regimes (Serreze et al., 2000).

Consequences of this vulnerability result in the conversion of arctic tundra from a carbon sink to a carbon source (e.g., Sturm et al., 2001; Walker et al., 2006; Deslippe and Simard, 2011), as well as the alteration of plant communities shifting from arctic tundra to shrublands through a ‘greening’ process (Pearson et al., 2013). This ‘greening’ process was documented over a 50-year time-period by Sturm et al. (2001), who commented on the link between increased shrub cover and an increase in net primary productivity. According to Hassol et al. (2004), changes in net primary productivity would initially increase atmospheric carbon uptake; however, ultimately the decrease in land surface reflectivity would outweigh this positive effect by causing more warming overall. In addition, this ‘greening’ process would likely be accompanied by shifts in vegetation zones moving northward with boreal forests replacing tundra and tundra vegetation replacing polar deserts (Hassol et al, 2004). The arctic system is complex and for this reason “...there needs to be continued research, which examines the complexity of the system more closely and that is backed by long-term, integrated and multidisciplinary observational networks” (Hinzman et al., 2005).

The present study was conducted at Cape Krusenstern National Monument (CAKR) and National Historic Landmark in northwestern Alaska. The Monument supports traditional Arctic

peoples through its unique flora and fauna and is globally important due to its sensitive arctic tundra biome. About 40 km northeast of CAKR is the Red Dog Mine which is one of the largest zinc (Zn) and lead (Pb) mines in the world. Since 1989 the Red Dog Mine, has transported concentrated Zn and Pb ore 75 km along the DeLong Mountain Transportation System (DMTS) haul road to a marine port facility on the west side of the Monument. The last 32 km of the DMTS haul road passes through CAKR.

In the year 2000, the National Park Service measured heavy metal concentrations in the moss *Hylocomium splendens* from six transects. Three were north and three were south of the haul road at distances 3 m, 50 m, 100 m, 250 m, 1000 m and two sites at 1600 m. They found moss metal levels as high as: Zn at > 1800 mg/kg, Pb at > 400 mg/kg and Cd at > 12 mg/kg (Ford and Hasselbach, 2001). Median concentrations of heavy metals throughout northern Alaska are much lower for Zn at 37.4 mg/kg, Pb at 0.62 mg/kg and Cd at 0.15 mg/kg (Ford, 1995). Heavy metal concentrations in 2000 near Red Dog were substantially higher than the median regional levels. In 2000, the elevated metal levels were attributed to fugitive dust from the transport of ore concentrates in trucks covered with tarps along the DMTS haul road (Ford and Hasselbach, 2001). Between 2001 and 2003 the mine operator purchased a fleet of new trucks with hydraulically-sealed lids, improved the truck loading and unloading facilities, and upgraded the barges and ship loaders (Exponent, 2007). In 2006 a network of 104 new plots were permanently installed and measured, with the goal of using the distance-to-road as a proxy for the metal deposition gradient. The results of these preceding studies are in Ford (1995), Ford and Hasselbach (2001), Hasselbach et al. (2005) and Neitlich et al. (2017).

Here, we assess decadal change in arctic tundra communities between the years 2006 and 2017 addressing the following questions:

- (1) How have lichen community metrics (species richness and composition) changed between 2006 and 2017 in relation to distance to the haul road?
- (2) How have vegetation community metrics (species richness and composition) changed between 2006 and 2017 in relation to distance to the haul road?
- (3) Do lichens and vegetation have different relationships to the metals gradient and if so, which are more sensitive to metal contamination?
- (4) Do vegetation communities differ between the north and south sides of the haul road?
- (5) Are vegetation changes from 2006 to 2017 far from the haul road (≥ 2000 m) related to climate change?

Chapter 2: Methods

2.1. Field methods

The overall goal of this study was to evaluate change in Arctic tundra communities and metals at CAKR from 2006 to 2017. This was achieved by using the moss *Hylocomium splendens* as a biomonitor of heavy metal concentrations in conjunction with community-level assessment of Arctic tundra lichens and vegetation.

104 plots were permanently placed in 2006 using GPS location and metal rebar stake installation to physically mark the plots, and were 4 x 8 m in size. The plot locations were selected to control for differences in vegetation community and elevation while detecting patterns of metal accumulation in relation to the DMTS haul road (Neitlich et al., 2017). Sampling was therefore restricted to two of the most common vegetation types, Upland Moist Dwarf Birch-Ericaceous Shrub and Upland Moist Dwarf Birch-Tussock Shrub (Jorgenson et al., 2004). The plot locations selected were between 60 and 240 m elevation to avoid confounding the data with alpine communities. With these restrictions in mind, twelve transects crossing the haul road were selected using a stratified-random design from the two vegetation classes and distance classes of 10, 50, 100, 300, 1000, 2000 and 4000 m from the DMTS haul road. Each transect included one “autocorrelation plot”, a plot 10 to 20 m from a 1000 m or greater plot. Ten additional reference plots were chosen at random in CAKR south of the haul road at distances of 40 km or greater and permanently installed (Neitlich et al., 2017).

At each plot, point intercept surveys were used to estimate total cover by species. Points were arrayed at vertices of a grid with 40 x 80 cm cells using meter tapes and pre-labeled PVC pipes. At each point, a laser attached to a PVC staff was suspended over the vegetation and all

lichens and vegetation at that point were tallied each with an abundance of 1.0. Overlapping vegetation was recorded as multiple hits per point. This yielded plots with a total abundance potentially greater than 100%. Bryophytes were recorded as 1.0 if hit by the laser point and added to the total cover. The basal-most point or ground layer was recorded as litter (dead plant material), basal vegetation (woody, or perennial vegetation), standing water, rock or bare ground. Often, a moss species was recorded above the ground layer such as *Sphagnum* moss with an assumed ground layer of litter below. Species not hit by the laser during the point intercept survey, but observed on the plot, were given a trace abundance of 0.1 during an ocular survey. Any specimens with unknown identity were collected to be identified in the laboratory.

2.2. Data assembly

Field collections of species not recognized in the field were identified in the laboratory and added to the raw field data. We combined the 2006 data (Neitlich et al., 2017) with our 2017 data, totaling 208 plot-year combinations (104 plots x 2 years). The raw data were then compiled into two separate data matrices using Excel Microsoft 2016 and PC-ORD 7.02 (McCune and Mefford, 2016); also used for all ordinations and diversity metrics. Two non-linear models were generated using JMP® Pro 14.1.0 data analysis software. One species matrix contained cover by species for lichens (194 plots x 119 species) and the second species matrix contained cover by species for other vegetation (“Vegetation”, 208 plots x 57 species, including mosses and vascular plants). The difference in plot number for the lichen species matrix is due to the deletion of plots with no lichens present, because we applied a proportional distance measure Sørensen or Bray-Curtis that cannot be calculated with empty rows (described in more detail below). Four subset

matrices were also used by selecting only plots 2000 m and greater which made up the “Lichen Far” matrix (112 plots x 114 species), the “Vegetation Far” matrix (112 plots x 56 species), the “Lichen Close” matrix (122 plots x 97 species) and the “Vegetation Close” matrix (136 plots x 40 species).

Environmental matrices were generated with the same number of plots as the species matrices. The environmental matrices contained quantitative and categorical environmental, total cover and diversity variables (Table A2), as well as 2006 *Hylocomium splendens* metal concentration data. Two of the variables in the environmental matrix were calculated using HyperNiche Version 2.3 (McCune and Mefford, 2009), Heat Load and Potential Direct Incident Radiation (PDIR).

To compare the years 2006 and 2017, some taxonomic decisions had to be made to coordinate the data. For example, two species might require lumping into one if they had been given different taxonomic treatments each year but looked the same in the field. A final list of taxonomic decisions made to coordinate the two data sets is given in an appendix (Table A1).

2.3. Data adjustments

Non-metric Multidimensional Scaling (NMS; Kruskal, 1964) ordination is a multivariate method particularly suited to ecological data with non-normal distributions and non-linear relationships among variables (McCune and Grace, 2002). Initial investigation using scatterplots of one species abundance versus another revealed non-normal and non-linear distributions throughout the data set. Therefore, NMS was better suited to our data than Principal Component Analysis (PCA) and other methods based on linear models. NMS ordinations of plots in species

space for each species matrix were performed to measure the strength of relationships of the major community gradients to the 2006 metals gradient and to reveal patterns in species composition. The Sørensen or Bray-Curtis distance measure was used for all multivariate analyses. Although we considered that retaining these plots and using Euclidean distance might better represent the metals gradient, we found the opposite, that Euclidean distance measure yielded ordinations considerably less sensitive to the metals gradient (lower Pearson's correlation coefficient).

Rare species were not deleted from either of the two species matrices, as an evaluation of the species abundance curves with and without rare species yielded only small differences. Outlier analyses were performed on both species matrices. Outlier analysis using the average Sørensen distance among plots was calculated for each plot. This yielded one outlier in the lichen species matrix (plot T6S10_17), which was peripheral with repulsion in the ordination with an average distance of 3.24 standard deviations above the grand mean. Upon further investigation, this plot only had one lichen species with the lowest abundance possible. This plot was removed in the "Lichen" matrix due to the low number of species present and the resulting distortion of the main point cloud in the NMS ordination. In addition, plots with no lichens present in either year, were removed from the Lichen data set. These steps resulted in the total of 194 plots.

In the environmental matrix the distance to the haul road was \log_{10} transformed (logDist) to linearize the function of deposition against distance. To summarize the large number of analytes measured in the 2006 moss tissue samples (Pb, Cd, Cr, Zn, Ni, P, Cl, Al, B, Ca, Cu, Fe, K, Mg, Mn, Na) we generated 3 synthetic variables using PCA to represent the overall trends in the analytes measured. We chose PCA because the relationships among variables could be

linearized. All analytes except potassium (K) and phosphorus (P) which had linear relationships to log distance, were \log_{10} transformed to reduce the skew in distributions and improve linear relationships among analytes. This was followed by a PCA which used a correlation cross products matrix and yielded an ordination with 68% of the variance explained by axis 1, 9% by axis 2 and 6% by axis 3. The environmental matrix was then augmented with plot scores on the first three axes. We also, however, retained $\log(\text{Pb})$, $\log(\text{Zn})$ and $\log(\text{Cd})$ as separate variables in the environmental matrix, because these elements were of particular interest.

2.4. Data analysis

NMS ordinations were based on the “slow and thorough autopilot” settings, and were used for the “Vegetation”, “Lichen Far” and “Vegetation Far” matrices. This included a starting number of 6 axes, 250 runs with real and randomized data, a maximum of 500 iterations, an instability criterion of 0.0000001 and no penalization for ties. The “Lichen” NMS ordination originally suggested a 2-axis solution. Because adding a third axis reduced stress by 4.8 units, which nearly met the arbitrary 5 stress unit cutoff for recommending an additional axis, we forced a 3-axis solution. Joint plots of the environmental, total cover and diversity variables were overlaid on each NMS ordination to show correlations with each of the axes. Axes of each ordination were rotated to facilitate comparisons and interpretation.

A blocked permutation-based multivariate analysis of variance (perMANOVA; Anderson, 2001) was calculated for the “Lichen”, “Vegetation”, “Lichen Far” and “Vegetation Far” matrices to test the null hypothesis of no difference in community composition (measured by Sørensen dissimilarities) between years while controlling for plot as a random effect. The

number of rows for each matrix were maintained at those mentioned previously. Settings used for this were “randomized complete blocks” with the block of “plot” and the grouping variable of “Year”. A one-way perMANOVA for each independent year was used to test the null hypothesis of no difference between the north and south sides of the road. To balance the number of plots on the north vs. south sides of the road for the “Lichen” data set all 20 reference plots were removed in addition to T6S4000A. This yielded a total of 86 plots. For the “Vegetation” data set all 20 reference plots were removed in addition to 4 replicate plots T6S4000A, T4S1000A, T3S1000A and E6S2000A; this yielded 90 plots total.

Univariate blocked or permutation-based analysis of variance (perANOVA) compared the means between years for each diversity metric (species richness, Shannon-Wiener index), and total cover. We also calculated β_D average half changes, a measure of beta diversity for lichens and for vegetation for each data set. The number of rows for each were maintained at 194 for the “Lichen” data set and 208 for the “Vegetation” data set. PerANOVA is identical to perMANOVA, except that the response matrix contains a single variable and is based on Euclidean distances. Using a permutation-based method for calculating the p -value for the null hypothesis of no difference in the response variable between groups avoids the distributional assumptions associated with an analysis of variance (ANOVA).

A blocked indicator species analysis (ISA) was performed on both data sets to compare individual species performance in a pairwise fashion between each plot grouped by year (Root et al., 2010). Blocking by plot focused the comparison on indicator species differences between years. This method calculates a proportional abundance of each species in the two groups (Year) and a proportional frequency of each species per group. These values are then multiplied

yielding an indicator value (IV) for each species, and finally the statistical significance of the IV values are evaluated with a randomization test of 1000 runs. This randomization tests the null hypothesis that the maximum IV is no larger than would be expected by chance (McCune and Grace, 2002).

2.5. Moss samples

Moss samples were processed at Oregon State University (OSU) Keck Collaboratory and the University of British Columbia (UBC) Pacific Centre for Isotopic and Geochemical Research (PCIGR). In the present manuscript, a subset of the total moss concentration data set for 2017 is discussed, as the whole data set was not yet available.

2.5.1. Sample collection:

Hylocomium splendens moss was collected as close to each plot as possible (not from within the plot), with some collections extending up to 20 meters in either direction parallel to the road. Moss was collected using powder-free nitrile gloves and SpecIPAK saran bags coated with polyester and polyethylene manufactured by Ampac. Collections were made from several locations within the desired sample area to be representative of the metal concentrations present at each site. At 10% of the total plots, additional duplicate moss samples were collected.

2.5.2. Laboratory preparation:

Once collected, samples were frozen at -20 °C at the end of each field day and finally transported in coolers to a freezer in Corvallis Oregon at the end of the field expedition. Frozen moss samples were dried in acid-cleaned Pyrex dishes at 60 °C in a drying oven overnight. Once dry, the whole sample was placed on a cleaned plastic tray and ¼ of the bag was cleaned (debris

removed, *Hylocomium splendens* selected) and then homogenized using an IKA Tube Mill 100 and disposable grinding chambers composed of polypropylene with a stainless steel beater.

2.5.3. Sample acid digestion:

Homogenized samples of about 0.1 g were weighed into 15 mL PFE Savillex® vials. Samples were then digested using open-vessel acid digestion. The digestion protocol was as follows: (1) 5 mL ~15 M trace metal grade nitric acid (HNO₃) at 90 °C, (2) 5 mL ~15M HNO₃ at 110 °C, (3) 3.5 mL ~15 M HNO₃ and 0.5 mL ~29 M hydrofluoric acid (HF) at 120 °C, (4) 4 mL ~15 M HNO₃ and 0.5 mL 30% hydrogen peroxide (H₂O₂) at 130 °C, and (5) 5 mL ~15 M HNO₃. Once digested, samples were dried down, brought up in 1% HNO₃, heated at 100 °C for 1 hour and sonicated for half an hour in preparation for metal concentration analysis.

2.5.4. Metal concentration measurement:

Measurements were made on a Thermo X-Series II Inductively Coupled Plasma Mass Spectrometer (ICP-MS) at the OSU Keck Collaboratory and on a high resolution ELEMENT2 ICP-MS at UBC PCIGR. Element concentrations of (Pb, Zn, Ni, Cd, Cr, Cu and As) were quantified using multi-element calibration curves and rhenium (Re) as an internal standard. Standards including Re were prepared from 100 or 1000 µg/mL single or multi-element solutions from High Purity Standards, Inc (USA), Specpure Plasma (Alfa Aesar, USA) and plasmaCAL (SCP Science, Canada). To ensure accuracy and access precision a 1ppb multi-element solution was measured every 10 samples and certified reference materials BCR-482 (lichen) and IAEA-336 (lichen) were measured during each analytical session. All sample levels were well above detection limits (0.002 for Pb, 0.013 for Zn, 0.002 for Ni, 0.003 for Cd, 0.010 for Cr, 0.037 for Cu and 0.017 for As) (Rutila, 2018).

2.6. Wetland indicator status analysis

Preliminary results suggested a possible relationship between change over time and the hydrologic status of the sites. We therefore extracted wetland indicator values for each plot at each date. Using the National Wetland Plant List (NWPL), indicator status ratings were scored for each vascular plant species in our data set (U.S. Army Corps of Engineers, 2016; Lichvar et al., 2016). The rating system has five categories, which we converted to a numerical rating of 1 through 5 (Table 1). Species that could not be assigned a rating were given a neutral value based on the average of known values or were assigned an average of two estimated ratings from expert opinion.

For this analysis, a wetland indicator value for each point (plot) was calculated for each species by averaging the weights of those present at each plot by individual NWPL status ratings. In addition, we calculated a blocked univariate perANOVA which tested for differences in wetland indicator values between the year 2006 and 2017 while controlling for plot as a random effect.

Table 1. National Wetland Plant List indicator status ratings.

Indicator Status	Abrv.	Definition	Numerical Rank
Obligate	OBL	Almost always occur in wetlands.	5
Facultative Wetland	FACW	Usually occur in wetlands, but may occur in non-wetlands.	4
Facultative	FAC	Occur in wetlands and non-wetlands	3
Facultative Upland	FACU	Usually occur in non-wetlands, but may occur in wetlands.	2
Upland	UPL	Almost never occur in wetlands	1

2.7. Regional climate data

Average daily temperature and precipitation climate data from two weather stations were used, Kivalina and Kotzebue Alaska. These data were acquired from ACIS, NOAA Regional Climate Centers (Regional Climate Centers, 2017).

Chapter 3: Results

3.1. Differences in communities through time — All diversity measures differed significantly between years (blocked univariate perANOVA) (Table 2). Mean total cover of vegetation had the greatest F -statistic followed by vegetation mean Shannon diversity, indicating strong differences from 2006 to 2017. Mean species richness decreased from 26 in 2006 to 16 in 2017 for the “Lichen” community while “Vegetation” increased only slightly. “Lichen” community β diversity (average half changes) increased through time while the “Vegetation” community decreased slightly to remaining almost static (Table 2).

Blocked ISA evaluated which species contributed to the significant differences between years in each data set (Table A3, Table A4). Species whose IV correspond to group 2017 were deemed “increasers” and those that correspond to group 2006 were deemed “decreasers”.

Table 2. Summary statistics for the community data sets (Lichens and Vegetation) for the years 2006 and 2017 and blocked univariate perANOVA, testing for differences in community metrics between 2006 and 2017.

	2006	2006	2017	2017	blocked perANOVA <i>F</i> (<i>p</i>)	
	Lichens	Vegetation	Lichens	Vegetation	Lichens	Vegetation
Species richness, mean	26.1	12.1	15.8	12.5	73.4 (0.0002)	11.2 (0.0014)
Shannon diversity, H', mean	2.6	1.9	1.8	2.2	66.7 (0.0002)	150.3 (0.0002)
Total Cover, mean	6.31	92.9	15.64	155.9	40.4 (0.0002)	532.5 (0.0002)
Nonzero Range	0.1-7.0	0.1-57.0	0.1-20.0	0.1-59.0	n.a.	n.a.
Whittaker's β_w Diversity	3.5	3.5	6.4	3.3	n.a.	n.a.
β_D Diversity, Ave. $\frac{1}{2}$ Changes	1.8	0.9	2.2	0.8	n.a.	n.a.

3.2. *Gradients in species composition* — The 3-axis “Lichen” NMS ordination yielded a final stress of 16.0 and an instability of <0.00001 after 95 iterations. The final stress was less than expected by chance ($p = 0.005$). The three axes represent 74% of the variance in the distance matrix. The ordination was rotated -45 degrees to align axis 1 with the metals and distance gradient (Figure 1). Environmental, total cover and diversity variables with the strongest R^2 correlation to axis 1 were: analyte concentration PCA axis 1 (PC_Axis1), 2006 log₁₀ Zn concentration (log(Zn_06)), 2006 log₁₀ Cd concentration (log(Cd_m_06)), 2006 log₁₀ Pb concentration (log(Pb_m_06)), log₁₀ distance to the haul road (logDist). Axis 2 was correlated to macrolichen total cover (Macr_Poi), lichen total cover (Sum_Li), Shannon-Wiener index of lichens and vegetation (H'), total lichen and vegetation cover (Sum) and sample year 2006 or 2017 (Year). Axis 3 had no strong correlations to any measured environmental variable.

For the “Vegetation” ordination, NMS autopilot suggested a 3-axis solution yielding a final stress of 16.9 and an instability of <0.00001 after 100 iterations. The final stress was less than expected by chance ($p = 0.004$). The three axes represented 79% of the variance in the distance matrix. The ordination was rotated -65 degrees in the plane of axes 1 and 2 to align with previous ordinations (Figure 1). Environmental, total cover and diversity variables with the strongest correlations to axis 1 were: Shannon-Wiener index of lichens and vegetation (H'), vegetation species richness (S_{Veg}), Shannon-Wiener index of vegetation (H'_{Veg}), \log_{10} distance to haul road ($\log\text{Dist}$), ocular lichen cover (Lich) and 2017 ocular crustose lichen cover (Crust_{17}). Axis 2 was correlated to moss total cover (Sum_{Mos}), total lichen and vegetation cover (Sum), vegetation total cover (Sum_{Veg}) and ocular sedge cover (Sedge). Axis 3 was correlated to ocular 2017 ocular sphagnum moss cover (sphag_{17}).

NMS ordinations of each community data set provided visual representations of the change in species composition between the two years. Pearson’s correlation coefficients (r) greater than ± 0.5 are bolded in (Table 3). In the “Lichen” NMS ordination, the two years overlap but show a greater spread of plots in species space for the year 2017. Axis 1 has a strong correlation to the metal concentrations gradient, ranging from high levels closest to the haul road (left-hand side) to low levels (right-hand side) (Figure 1). Low metal deposition is associated with high values of lichen species richness (S_{Li}), total lichen cover (Sum_{Li}) and \log_{10} distance to haul road ($\log\text{Dist}$) (Table 3).

In the lichen community, we compared the combined years and each individual year to lichen species richness (S_{Li}). This involved a comparison of three separate ordinations from the lichen data set. In each ordination, we rotated to align axis 1 with the variable log distance

(logdist). Pearson's correlation coefficients for lichen species richness to axis 1 were: 2006 and 2017 ($r = 0.353$); 2006 ($r = 0.672$); and 2017 ($r = 0.567$) (Figure 1, bottom row).

The "Vegetation" NMS ordination shows a shift towards higher total lichen cover in the year 2017 and decreasing ocular sedge cover. The vegetation community changed over time to a greater degree when compared to the lichen community (blocked perMANOVA; lichen: $F = 31$ vs. vegetation: $F = 73$). Environmental variables, total cover and diversity variables associated with axis 1 are restricted to vegetation species richness (S_Veg), Shannon-Wiener index of vegetation (H' _Veg) and Shannon-Wiener index of lichens and vegetation (H'), while metal levels are not strongly correlated to axis 1 as seen in the lichen community (Table 3).

Species exhibited stronger relationships when correlated to their own group, such as lichen species were more associated with the "Lichen" ordination. Specific lichens with the strongest relationship to axis 1 were *Cladonia gracilis*, *Cetraria laevigata* and *Cladonia amaurocraea* (Table 4). Because these are positive correlations, in the ordination these species would fall in the region of low metal levels and may represent species most sensitive to high metal deposition. Species most associated with axis 2 were *Flavocetraria cucullata*, *Cladonia stygia*, and *C. laevigata*. As these have negative correlations these species would be located in the bottom half of axis 2 which is associated with an increase in total lichen cover (Sum_Li) in 2017. Vegetation most associated with axis 1 were *Eriophorum angustifolium*, *Vaccinium uliginosum* and *Empetrum hermaphroditum*. Axis 2 was most correlated to *Ledum decumbens*, *E. angustifolium* and *E. hermaphroditum*.

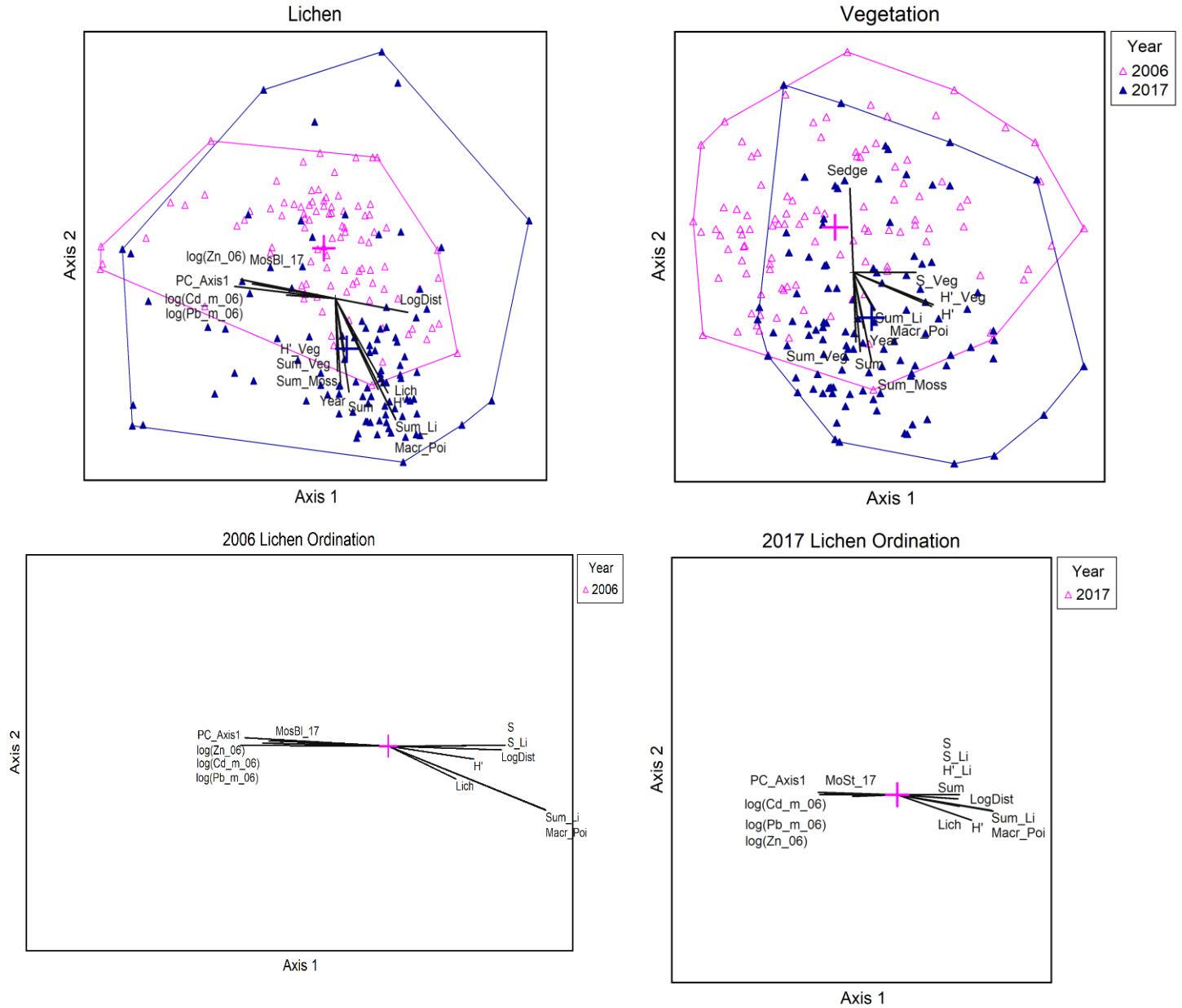


Figure 1. Top row: NMS ordinations of plots in species space for the “Lichen” and “Vegetation” data sets. Included are joint plot overlays of environmental, total cover and diversity variables grouped by year with convex hulls and the centroids of the average position of points for each group. Bottom row: 2006 and 2017 joint plots only, rotated to align axis 1 with log distance (LogDist).

Table 3. Comparison of Pearson's correlation coefficients (r) from NMS ordinations with Axis 1 and 2 for selected environmental, total cover and diversity variables. Pearson's correlation coefficients greater than +/- (0.5) are bolded.

	Environmental Variable	Lichens	Vegetation
Axis 1	S_Li (lichen species richness)	0.418	0.137
	S_Veg (vegetation species richness)	0.170	0.551
	S (lichen + vegetation species richness)	0.420	0.225
	Sum_Li (total lichen cover)	0.530	0.323
	Sum_Veg (vegetation total cover)	0.126	0.115
	Sum (lichen + vegetation total cover)	0.250	0.186
	H'_Li (Shannon-Wiener index of lichens)	0.294	0.024
	H'_Veg (Shannon-Wiener index of vegetation)	0.204	0.624
	H' (Shannon-Wiener index of lichens + vegetation)	0.495	0.615
	logDist (log ₁₀ distance to haul road)	0.582	0.389
	Sedge (ocular sedge cover)	-0.107	-0.129
	PCA axis 1 synthetic variable (PC_Axis1)	-0.686	-0.221
	Log ₁₀ (Pb_m_06) (2006 Pb concentrations)	-0.621	-0.311
	Log ₁₀ (Cd_m_06) (2006 Cd concentrations)	-0.623	-0.281
	Log ₁₀ (Zn_06) (2006 Zn concentrations)	-0.664	-0.239
Axis 2	Sedge (ocular sedge cover)	0.206	0.640
	S_Li (lichen species richness)	0.092	0.027
	Gram_17 (ocular 2017 graminoid cover)	0.090	0.424
	Macr_Poi (total macrolichen cover)	-0.706	-0.432
	Year (year as a quantitative variable)	-0.593	-0.519
	Sphag_17 (2017 ocular Sphagnum moss cover)	-0.134	-0.323
	PCA axis 1 synthetic variable (PC_Axis1)	0.220	0.188
	Log ₁₀ (Pb_m_06) (2006 Pb concentrations)	0.246	0.124
	Log ₁₀ (Cd_m_06) (2006 Cd concentrations)	0.249	0.110
	Log ₁₀ (Zn_06) (2006 Zn concentrations)	0.280	0.213

Table 4. Pearson's correlation coefficients (r) of individual species with axes 1 and 2 for the "Lichen" and "Vegetation" NMS ordinations. Vegetation species have an orange background and Pearson's correlation coefficients greater than +/- (0.5) are bolded.

Binomial	Lichen Ordination		Vegetation Ordination	
	Axis 1	Axis 2	Axis 1	Axis 2
<i>Cladonia gracilis</i>	0.420	-0.444	0.290	-0.300
<i>Cetraria laevigata</i>	0.404	-0.491	0.320	-0.329
<i>Cladonia amaurocraea</i>	0.412	-0.239	0.036	-0.197
<i>Cladina arbuscula/mitis</i>	0.372	-0.400	0.157	-0.280
<i>Flavocetraria cucullata</i>	0.350	-0.613	0.133	-0.361
<i>Cladonia stygia</i>	0.298	-0.496	0.119	-0.312
<i>Peltigera polydactylon</i>	0.257	-0.205	0.178	-0.142
<i>Cetraria islandica</i>	0.224	-0.250	0.233	-0.079
<i>Eriophorum angustifolium</i>	-0.264	0.281	-0.657	0.531
<i>Vaccinium uliginosum</i>	0.246	-0.303	0.550	-0.133
<i>Empetrum hermaphroditum</i>	0.157	-0.318	0.484	-0.305
<i>Carex sp.</i>	-0.240	0.221	-0.336	0.000
<i>Petasites frigidus</i>	0.119	-0.189	0.346	-0.203
<i>Hylocomium splendens</i>	-0.055	-0.318	0.232	-0.259
<i>Ledum decumbens</i>	0.064	-0.219	-0.407	-0.559
<i>Alnus crispa</i>	0.215	-0.127	0.276	-0.024

We evaluated a possible tradeoff in total cover of lichens and vegetation with bivariate scatterplots for 2006 and 2017 (Figure 2). If vegetation was increasing and outcompeting lichens, we would expect an increase in vegetation and a decrease in total lichen cover. However, we found that cover of both lichens and vegetation increased about two-fold in 2017 compared to 2006 (Figure 2). Therefore, we are not seeing the common trend documented by others of an increase in vegetation and a decrease in lichens (e.g. Walker et al., 2006; Elmendorf et al., 2012b; Lang et al., 2012; Fraser et al., 2014; Cornelissen et al., 2001; Joly et al., 2009).

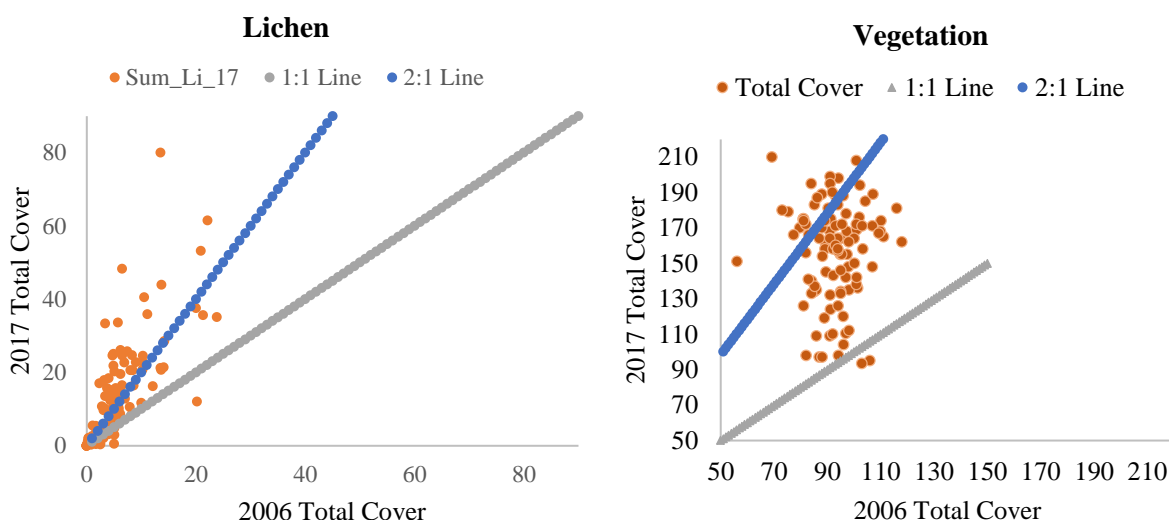


Figure 2. Scatterplots of total Lichen and Vegetation cover comparing 2006 to 2017 data. Included are 1:1 and 2:1 relationship lines. Each point is a plot.

3.3. Changes in moss metal concentrations — Metal concentration data was available for both sampling years for a subset of 60 plots of the total 104 plots (Table A5). A comparison on 3 analytes Pb, Zn and Ni showed a strong correlation between years in analyte concentrations. In this subset, Pb concentrations decreased in 2017, especially for plots that were high in Pb in 2006. Zn stayed about the same and Ni decreased slightly for plots that were high in Zn in 2006 (Figure 3). Because this subset of metal concentration data showed little change from 2006 to 2017 we felt comfortable assuming that the 2006 concentrations represent well the metal concentrations throughout the study period.

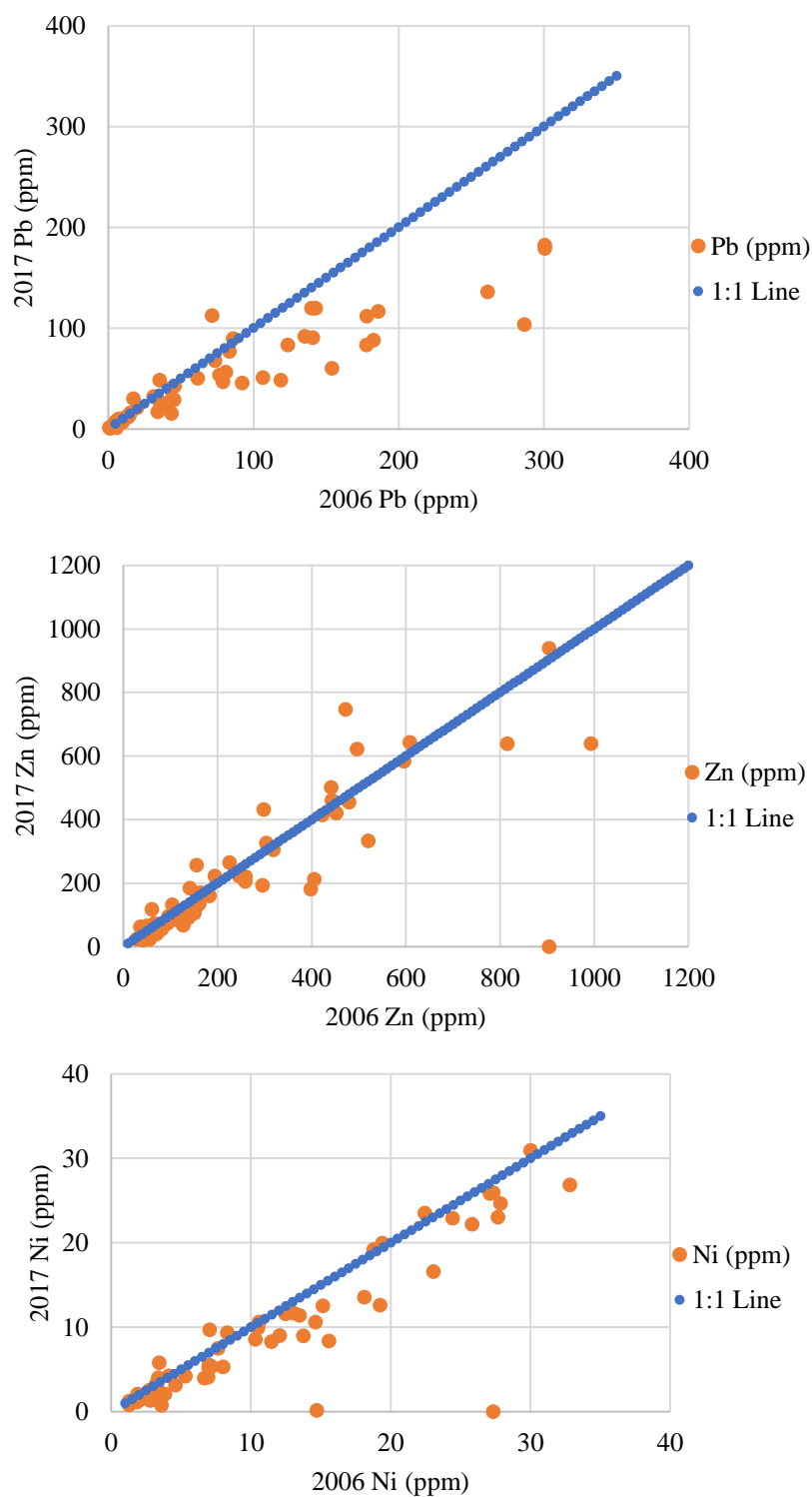


Figure 3. Comparison of 2006 and 2017 metal concentrations for Pb, Zn and Ni. Each shows the observed values and a 1:1 line of no change between years.

3.4. Community differences relative to the haul road — Species richness of lichens and vegetation both increased with respect to \log_{10} distance from the haul road (Figure 4). A stronger relationship appears to exist between lichens and \log_{10} distance when compared to vegetation, however, a sigmoidal logistic 3 parameter model fit lichen species richness best (2006 $R^2 = 0.63$; 2017 $R^2 = 0.54$). Both lichen species richness and \log_{10} distance to the haul road have a negative relationship to heavy metal concentrations in moss (e.g., $\log_{10}(\text{Pb concentration})$) in this case) (Figure 4).

The “Lichen” community had a borderline indication of community difference from the north and south side of the road for each year; and the “Vegetation” community did not differ in species composition for side of the road in either year (perMANOVA “Lichen”: $F = 1.5$ in 2006 ($p = 0.09$) and $F = 1.8$ in 2017 ($p = 0.04$) for side of road; “Vegetation”: $F = 1.5$ in 2006 ($p = 0.16$) and $F = 1.2$ in 2017 ($p = 0.26$) for side of road).

Species composition differed among plots and among years for the “Lichen” and “Vegetation” communities (Blocked perMANOVA “Lichen”: $F = 1.7$ for plot ($p = 0.0002$) and $F = 31.1$ for year ($p = 0.0002$); “Vegetation”: $F = 3.7$ for plot and ($p = 0.0002$) and $F = 75.3$ for year ($p = 0.0002$)).

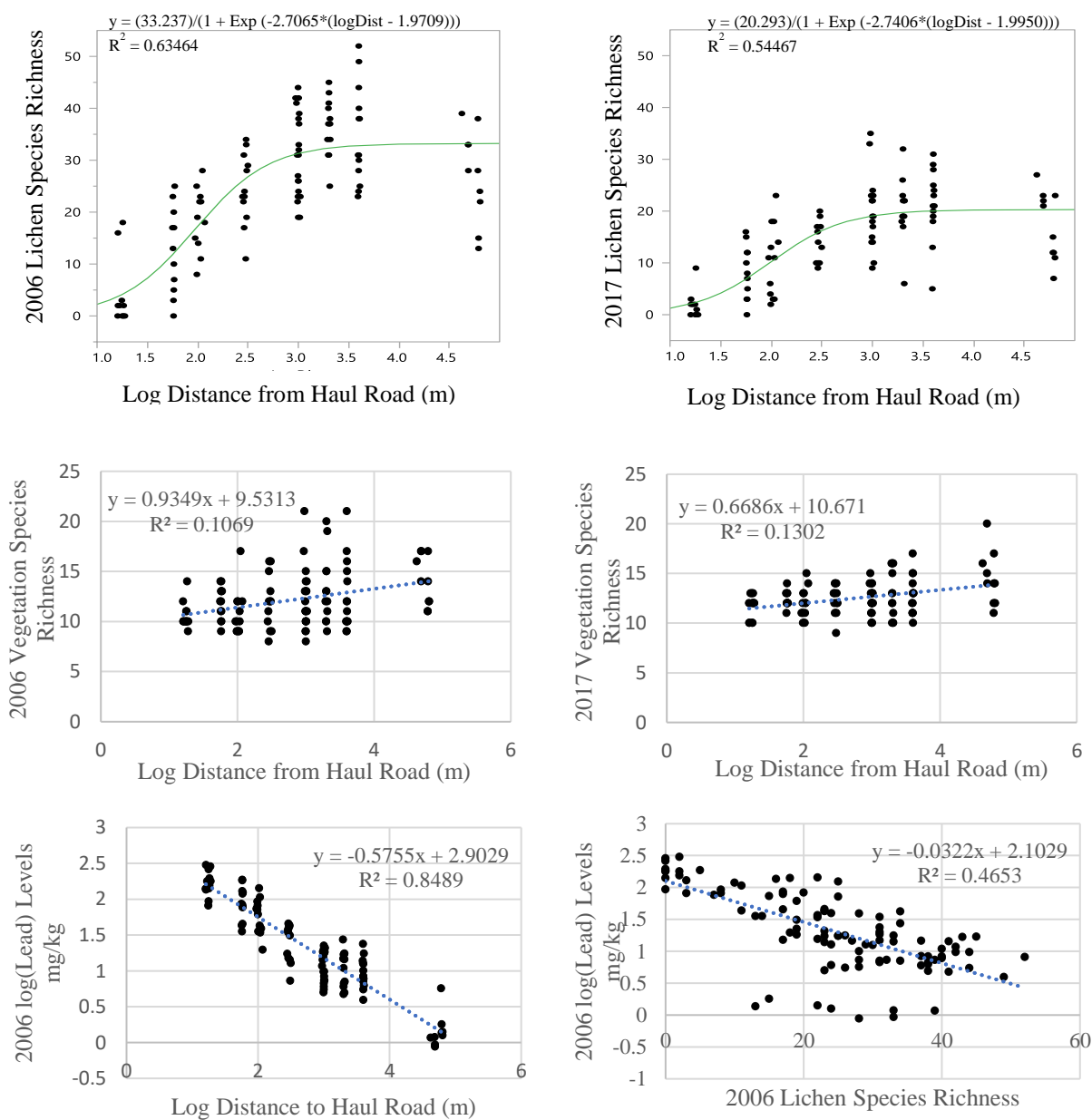


Figure 4. The relationship of \log_{10} distance to the haul road in 2006 and 2017 to species richness in lichens and vegetation, and 2006 lead levels to $\log_{10}(\text{distance})$ and mean lichen species richness. Each include a line of best fit with the equation and R^2 value.

3.5. *Community changes, distant from the haul road* — To further investigate the source of change between years, we selected plots in the “Lichen” and “Vegetation” data sets which were 2000 m and greater from the haul road (72 plots total, 36 per year). This enabled us to analyze community differences through time with little or no influence of metal deposition from the haul road. A temporal change was detected in both communities (blocked perMANOVA “Lichen”: $F = 1.67$ for plot ($p = 0.0002$) and $F = 20.7$ for year ($p = 0.0002$); “Vegetation”: $F = 3.9$ for plot ($p = 0.0002$) and $F = 39.8$ for year ($p = 0.0002$)).

An initial autopilot setting suggested two dimensions for the “Lichen Far” ordination similar to the “Lichen” ordination. However, the difference from 2 to 3 dimensions also yielded a decrease in stress but was slightly under the arbitrary 5-stress unit cutoff. Therefore, the ordination was forced to three axes and the “Lichen Far” ordination yielded a final stress of 14.5 and an instability of <0.00001 after 74 iterations. The final stress was less than expected by chance ($p = 0.005$). The three axes represented 78% of the variance in the distance matrix. The ordination was rotated 75 degrees to align with previous ordinations (Figure 5, left).

Environmental, total cover and diversity variables with the strongest correlations to axis 2 were: year, total lichen and vegetation cover (Sum), Shannon-Wiener index of lichens (H'_{Li}), total lichen cover (Sum_Li), ocular lichen cover (Lich) and total microlichen cover (Macr_Poi). In this ordination of plots far from the haul road the metals gradient is no longer apparent (Figure 5). However, pronounced temporal shifts are apparent based on trajectories of plots in species space (Figure 5, right). With one exception (plot T6S4000A_17), plots shifted in a similar direction in species space, suggesting similar changes in lichen communities across plots.

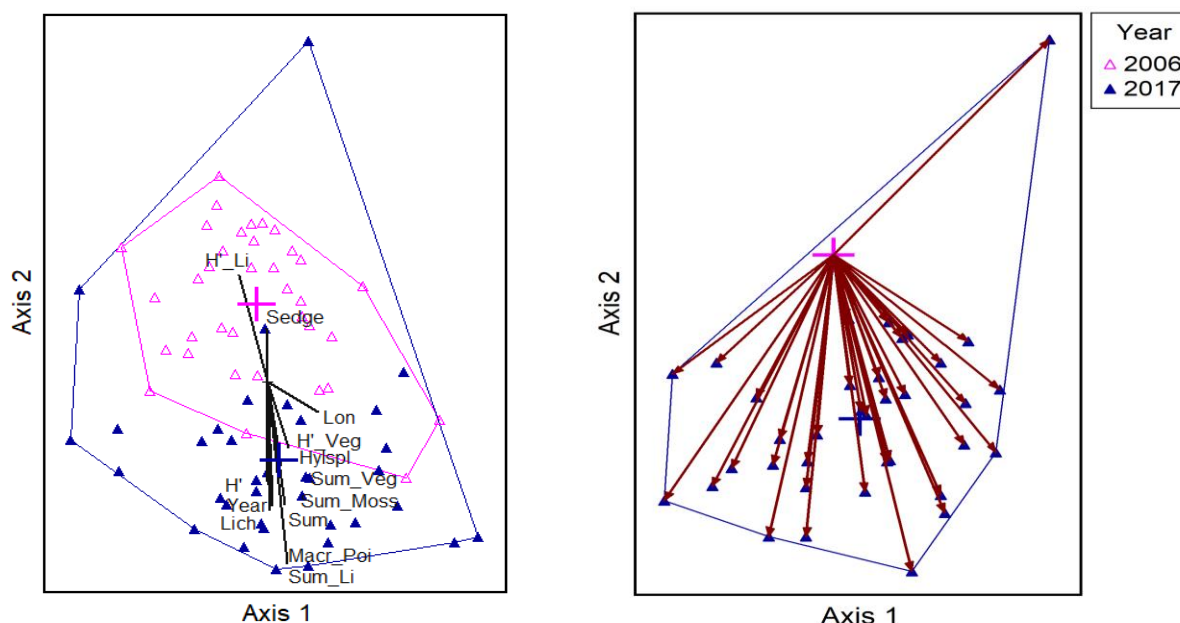


Figure 5. NMS ordinations of far plots only (≥ 2000 m) in the “Lichen” data set. The left-hand ordination includes joint plot overlays of environmental, total cover and diversity variables, grouped by year with convex hulls and the centroids of the average position of points for each group. The right-hand ordination is of successional vectors translated so tails lie on the centroid.

The “Vegetation Far” ordination used a 3-dimensional solution and yielded a final stress of 13.8 and an instability of <0.00001 after 73 iterations. The final stress was less than expected by chance ($p = 0.004$). The three axes represented 85% of the variance in the distance matrix. The ordination was rotated 65 degrees in the plane of axes 1 and 2 to align with previous ordinations (Figure 6). Environmental, total cover, and diversity variables with strongest correlations to axis 1 were: Shannon-Wiener indices for lichens and vegetation (H'), vegetation species richness (S_Veg), Shannon-Wiener index of vegetation (H'_Veg) and ocular 2017 shrub cover ($Shrub_17$). Axis 2 was associated with year, total moss cover (Sum_Mos), total lichen and vegetation cover (Sum), total vegetation cover (Sum_Veg), ocular sedge cover ($Sedge$) and total macrolichen cover ($Macr_Poi$). This subset of the vegetation data displayed patterns similar

to the full vegetation data set, with negligible correlation to metals but still with a correlation to vegetation diversity metrics and increased total macrolichen cover in 2017.

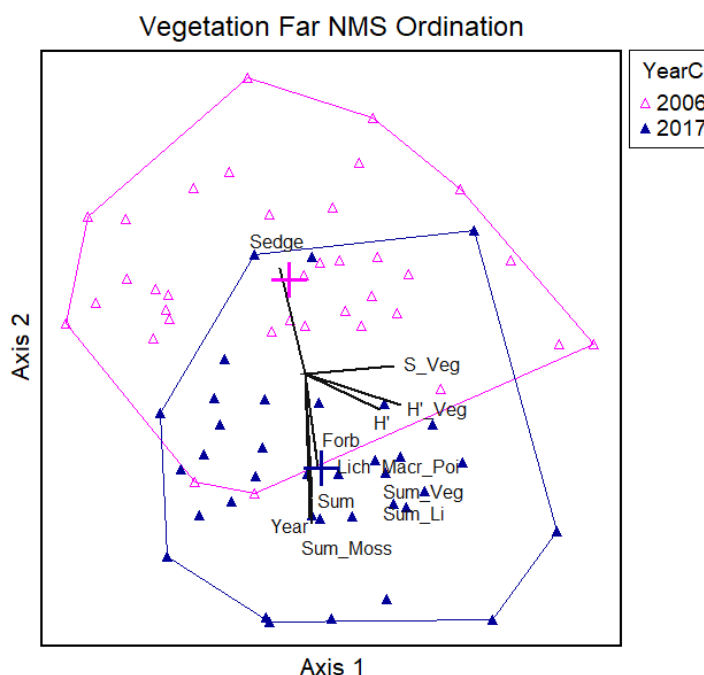


Figure 6. NMS ordination of far plots only (≥ 2000 m) in the “Vegetation” data set. Included are joint plot overlays of environmental, total cover and diversity variables, grouped by year with convex hulls and the centroids of the average position of points for each group.

3.6. Temporal and spatial differences for dominant species and groups — To contrast the behavior of individual species and species groups we plotted the abundance of species along the metals gradient for each year of sampling, selecting dominant species and totals for species groups. Included in this are common lichen species with a positive relationship to distance from the haul road, particularly *Cetraria laevigata*, *Cladonia amaurocraea*, *Cladonia gracilis* and *Flavocetraria cucullata* (Figure 7). These species may be relatively sensitive to metal deposition as their cover is low to none near the haul road. In contrast, *Thamnolia subuliformis* could

represent a more tolerant species in this system as it has a more uniform distribution across distance. Cover increased from 2006-2017 for most lichen species (Figure 7).

Vascular plant species that had a positive relationship to distance from the haul road included *Carex bigelowii*, *Empetrum hermaphroditum*, *Vaccinium uliginosum* and *Vaccinium vitis-idea* (Figure 8). These species might be slightly more sensitive to metal deposition as seen by their lower total cover near the haul road with greater metal exposure. Potentially tolerant species are *Betula nana*, *Eriophorum angustifolium*, *Ledum decumbens* and *Rubus chamaemorus* which demonstrate more of an even distribution across distances. Between 2006-2017 *Ledum decumbens*, *Betula nana*, *Empetrum hermaphroditum* and *Vaccinium vitis-idea* increased (Figure 8).

Finally, if we look at the major groups and their relationship to distance and time, we can see a clear positive relationship of lichens and *Sphagnum* moss to distance from the haul road, potentially denoting their overall sensitivity to metal deposition (Figure 9). All groups when examined individually “Moss”, “Lichen”, “Vascular Plants” and “*Sphagnum*” demonstrate about a doubling in total cover from 2006 to 2017. This pattern is the most apparent in the “All Groups” plot (Figure 9).

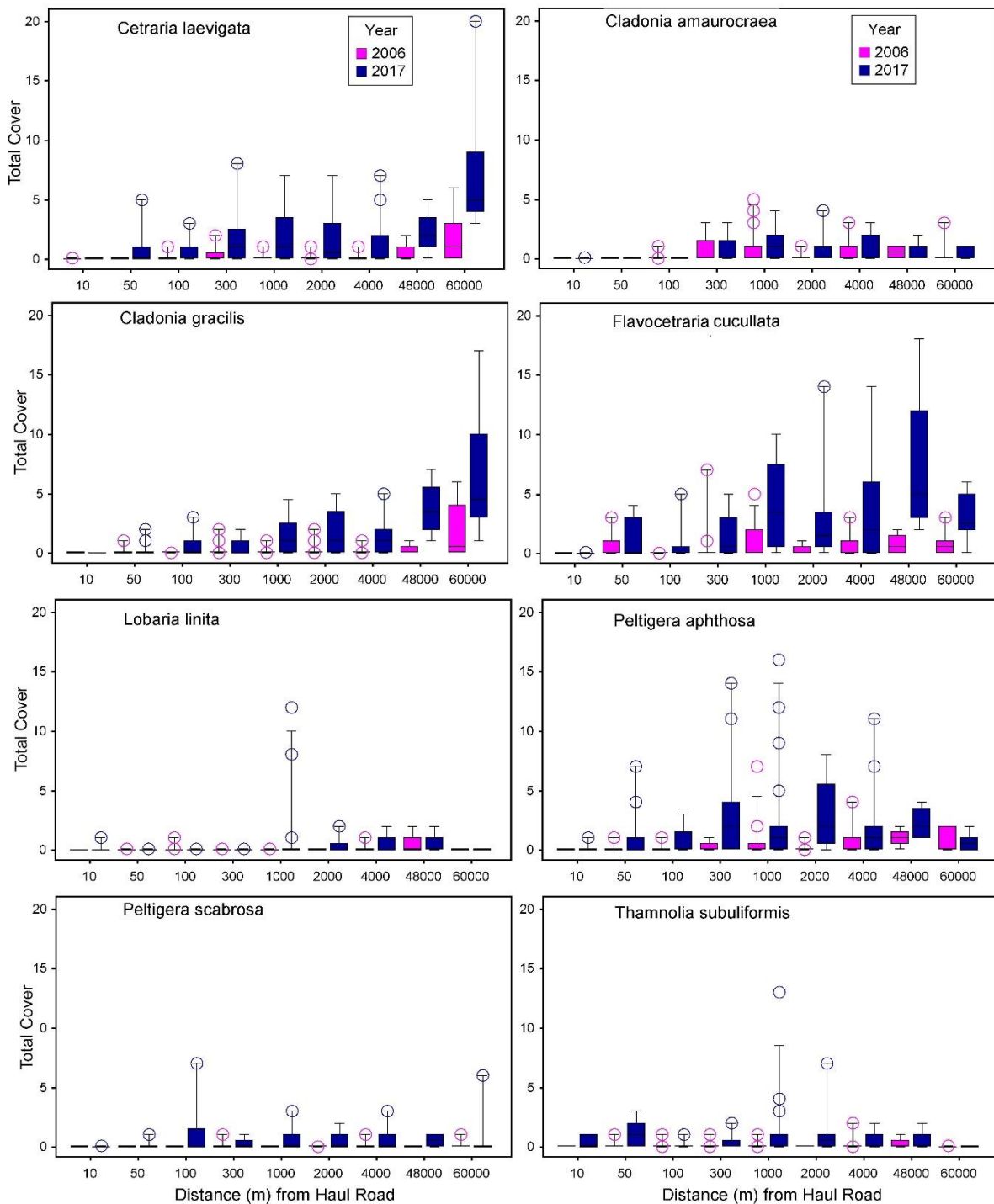


Figure 7. Plots of total lichen cover, comparing year and distance (m) from the Haul Road. The bars represent the interquartile range from the 25th to the 75th percentiles, the whiskers the 5th and

95th percentiles and the circles are outliers which are values that fall from the median 1.5 times outside of the interquartile range.

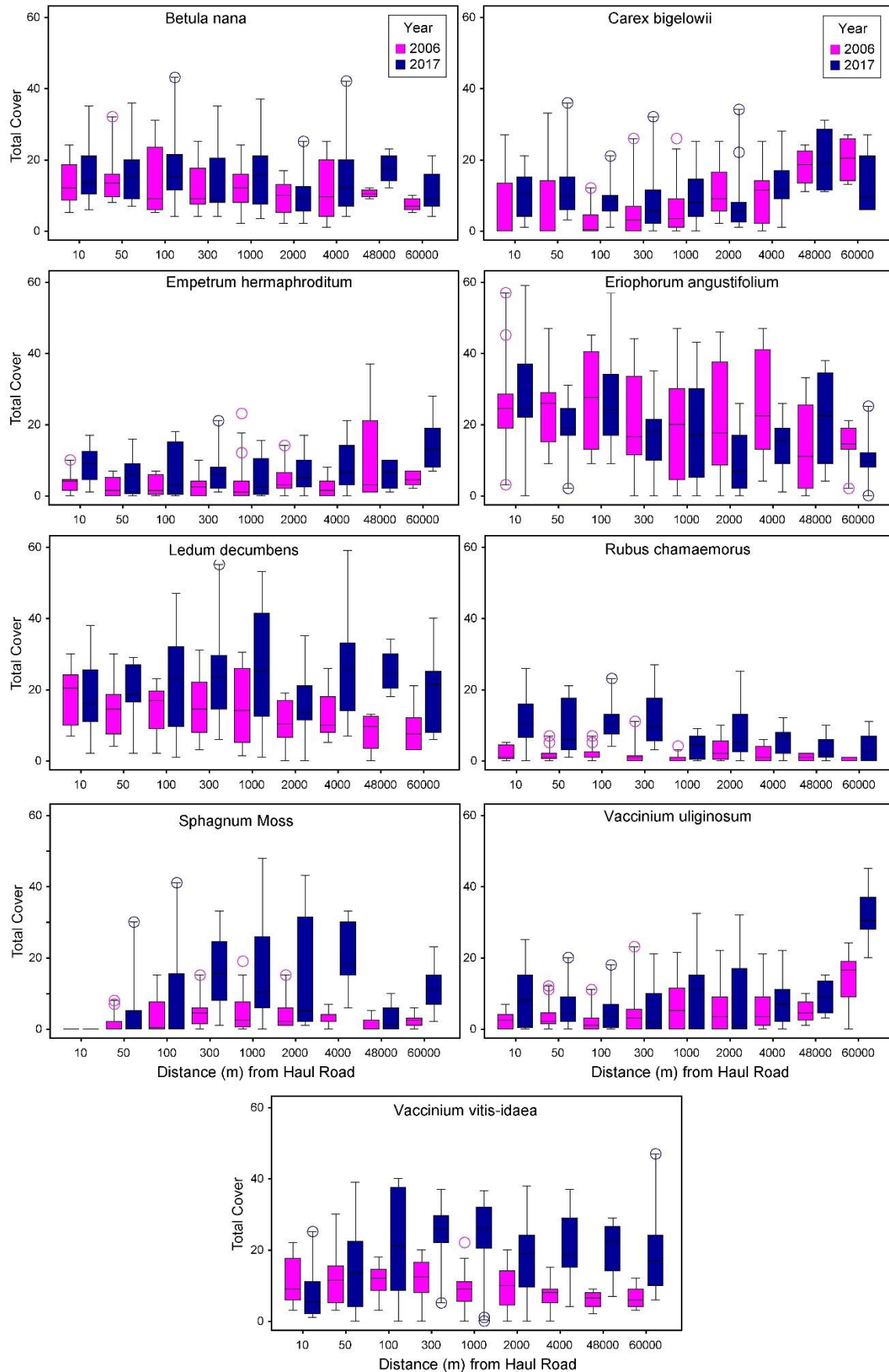


Figure 8. Plots of total vascular plant and total *Sphagnum* moss cover, comparing year and distance (m) from the Haul Road. The bars represent the interquartile range from the 25th to the 75th percentiles, the whiskers the 5th and 95th percentiles and the circles are outliers which are values that fall from the median 1.5 times outside of the interquartile range.

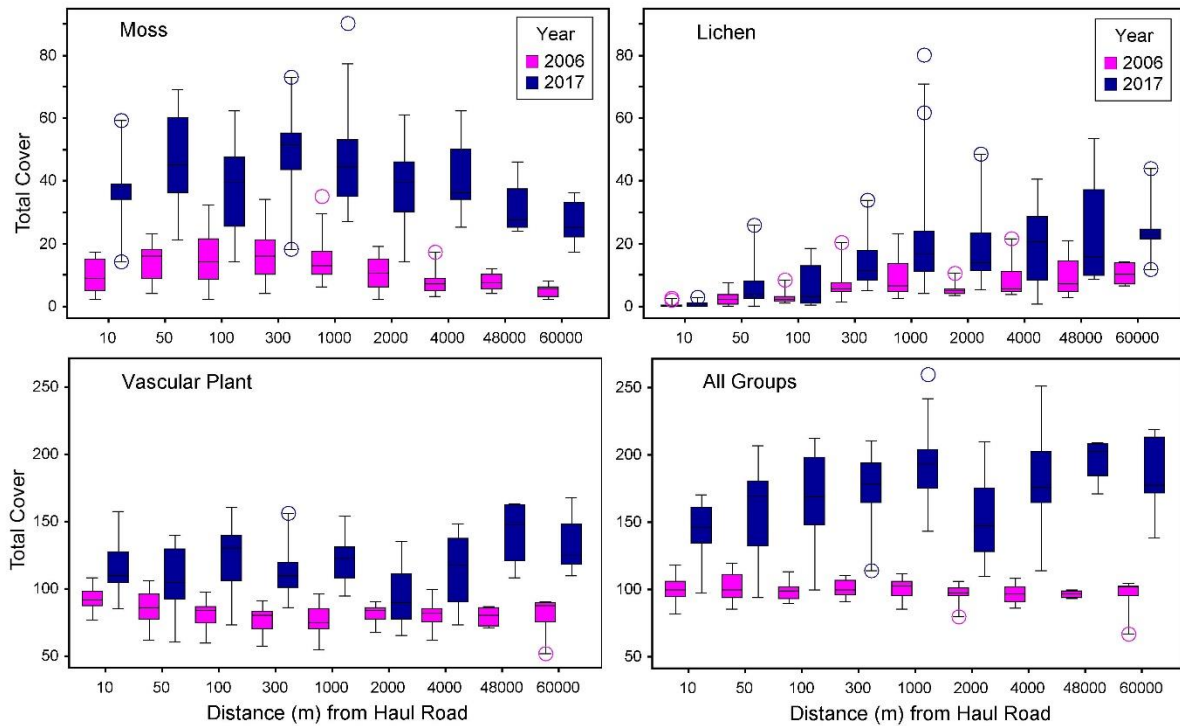


Figure 9. Plots of total cover for three major groups and all groups (total covers of mosses, lichens and vascular plants); comparing year and distance (m) from the Haul Road. (Notice different Y axis scales). The bars represent the interquartile range from the 25th to the 75th percentiles, the whiskers the 5th and 95th percentiles and the circles are outliers which are values that fall from the median 1.5 times outside of the interquartile range.

3.7. *Temporal changes in wetland indicators* — Wetland indicator values for plots were positively related between years but showed a general decrease through time (Figure 10). These results suggest a temporal drying and an increase in upland associated plants from 2006 to 2017 (Figure 10) (blocked perANOVA: $F = 4.8$ for plot ($p = 0.0002$) and $F = 32$ for year ($p = 0.0002$)).

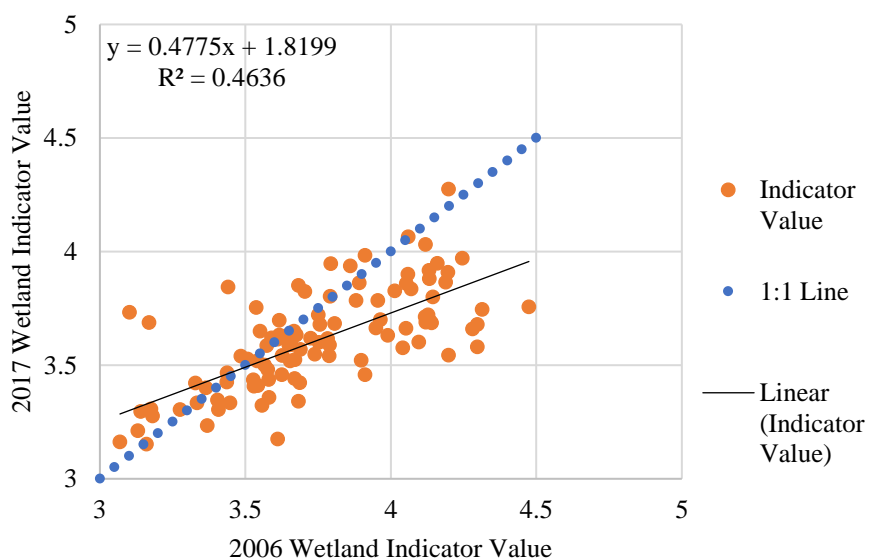


Figure 10. Scatterplot of 2006 vs. 2017 wetland indicator values with a 1:1 line and a line of best fit with the equation and R^2 value.

Chapter 4: Discussion

4.1. Sources of apparent change in lichens and vegetation

We considered multiple explanations for the strong differences between the two years observed in the two communities. For each source of apparent change, we qualitatively evaluate its likely importance, given the available evidence. For some we suggest additional evidence needed for further evaluation. Note that the sources of apparent change are not mutually exclusive.

4.1.1. Changes in metal deposition — An increase, decrease or no change in metal deposition from the haul road to the surrounding environment could be a source of the change which was observed. If moss metal levels increased from 2006 to 2017 we would expect to see an overall decrease in abundance of lichens within 300 m of the road, and a greater change closer to the road than farther from the road. If moss metal levels decreased we would expect no change or a slight increase in abundance of lichens within 300 m of the road as the lichens slowly recover, moving toward the community composition observed at plots farther than 2000 m from the road. We found that there was an increase in lichen and vegetation abundance from 2006 to 2017 both near and far from the haul road, along with a decrease in lichen species richness and an increase in vegetation species richness. Variation in lichen communities far from the haul road did not show an appreciable correlation to metal deposition ($R^2 < 0.25$), in contrast to the strong relationship between lichen communities and metals for the whole data set.

In addition, we compared the degree of community change between the far plots (≥ 2000 m) and the close plots (≤ 1000 m). Lichen communities at the far plots experienced more change than those close to the haul road, even though metal levels were about the same, suggesting that changes in metals were not primarily responsible for the lichen community changes. The lichen community near the haul road could be experiencing less change due to persistently higher metal levels, which are maintaining a somewhat static lichen community.

On the other hand, vegetation, while showing pronounced change everywhere, exhibited the opposite pattern, with the close plots exhibiting somewhat more change between years than far plots (Table 5). Given the relative insensitivity of the vascular vegetation to metals, we do not attribute the differences in rates of vegetation change to a direct effect of metal deposition.

Table 5. Change from 2006 to 2017, blocked perMANOVA of close, far and all plots.

	blocked perMANOVA F of Year (p)	
	Lichens	Vegetation
Close Plots ($\leq 1000\text{m}$)	$F = 16.3$ (0.0002)	$F = 51.8$ (0.0002)
Far Plots ($\geq 2000\text{m}$)	$F = 20.7$ (0.0002)	$F = 39.8$ (0.0002)
All Plots (10m-Reference)	$F = 31.1$ (0.0002)	$F = 75.3$ (0.0002)

4.1.2. Climate change effects on vegetation — The community differences observed between years could be due to the effects of climate change on vegetation. If this were true, based on current literature (Nelson et al., 2015; Myers-Smith, 2011; Myers-Smith, 2015; Martin et al., 2017, Myers-Smith et al., 2019) we would expect an increase in total vascular plant cover, particularly shrubs, and a decrease in total lichen cover. Our results (Figure 8) agree with other studies that have documented increases in Arctic regions through time in vegetation cover (e.g. Walker et al., 2006; Elmendorf et al., 2012a; Lang et al., 2012; Fraser et al., 2014; Cornlissen et al., 2001; Joly et al., 2009).

To evaluate potential climatically-driven changes in lichens and vegetation with low levels of metal pollution, we analyzed the subset of plots ≥ 2000 m from the haul road. Metal levels are low at those distances compared to near the haul road. Analysis of this subset also revealed a strong temporal shift (Figure 2,5,6; Table 8) in species composition of both lichen and vegetation communities. Because there was no indication of widespread disturbance to this ecosystem in the intervening period, and no evidence of a strong successional pattern we cannot exclude the hypothesis that a substantial amount of the changes in lichens and vegetation are induced by climatic changes. Predictions and effects of a changing climate on arctic tundra ecosystems have been extensively documented and match the trends seen here of increased shrub, sedge and graminoid cover through a “greening” process (excluding lichen cover) (e.g.

Elmendorf et al., 2012b; Fraser et al., 2014; Hassol, 2004; Jia et al., 2009; Pearson et al., 2013; Stow et al., 2004). However, the observed increase in cover for most lichen species (Figure 7), is the opposite of what we expected based on previous studies which have documented an overall decrease through time in total lichen cover (e.g. Walker et al., 2006; Elmendorf et al., 2012b; Lang et al., 2012; Fraser et al., 2014).

4.1.3. Hydrologic changes — At Arctic wetland sites lichens tend to be poorly represented as compared to uplands (Shaver and Chapin, 1991; Williams and Rastetter, 1999). Slowly drying as suggested by the decreasing wetland indicator scores that we observed over time, could explain the observed increases in both lichen and vascular plants. This seems to be the most logical explanation for the observed changes through time in lichen and vegetation communities.

The observed change in wetland associated plants at our study site could be tied to changes in climate as permafrost wetlands in the Arctic will likely be altered by changes in temperature and hydrology (Burkett and Kusler, 2000). The Arctic Climate Impact Assessment (ACIA) stated that as temperatures rise and permafrost thaws, the summer active layer increases and drains water from the landscape. Subsequently “these processes are likely to lead to an initial greening, followed by desertification in some areas as warming continues” (Hassol, 2004). Potential consequences of climate change through alterations in temperature and hydrology were documented from 1950 to 1996 across the Kenai Peninsula Lowlands. They observed an increase in woody plant cover, water shrinkage, wetland drying and invasion of woody and upland species in lake beds (Klein et al., 2005). These studies support the idea that long-term climate change could be inducing an overall drying of our study region as reported by an increase in

upland-associated vascular plants and an overall increase in temperature throughout our study site (Figure 10, 11).

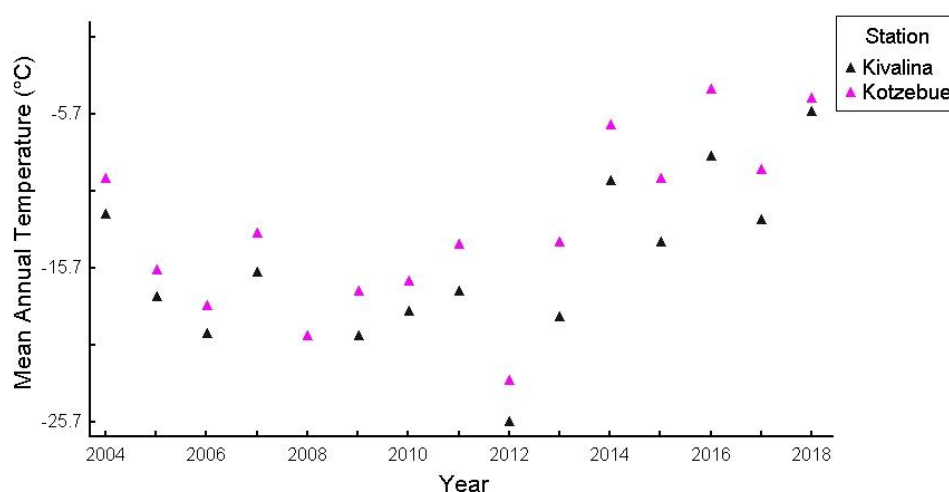


Figure 11. Mean annual temperature at Kivalina and Kotzebue from 2004 to 2018.

4.1.4. Inconsistent taxonomic treatment — Systematic differences in identification of the same species between the two years may form a bias that creates apparent change through time. If this were true we would expect an artificially high F ratio for year from perMANOVA. We evaluated this problem in two ways. (a) Species revealed by blocked Indicator Species Analyses to be significant indicators of sampling year should be taxonomically challenging or unstable in some way. Yet we found no obvious association between problematic species and being an indicator species. (b) We repeated the perMANOVA test for difference in date while retaining only easy-to-identify species. We found this yielded a higher F statistic, suggesting that if anything taxonomic inconsistencies appeared to weaken the difference between dates. Therefore, we conclude that taxonomic inconsistencies did not appear to be an important contributor to the differences in communities between years.

4.1.5. Observer bias — It is possible that individual field personnel were biased either up or down in their point-sampling hits. If this were true we would expect modest 2006 to 2017 changes commensurate with that bias. However, many differences in abundance were much larger than could be attributable to observer bias. For example, from the ISA analysis, about four times as much *Rubus chamaemorus* was recorded in 2017 than in 2006. Furthermore, this particular species is unmistakable in the field, which supports the apparent difference as a real change through time. Therefore, we conclude that although observer error will always be present to some degree, the differences in abundances are much larger than observer error.

We suggest three methods to further evaluate this source of error:

- i. Resample several arctic tundra plots with multiple observers, then calculate the error associated with this sampling as the pooled within-plot variation among observers. These data would help to document the expected observer error in other applications of this methodology in arctic tundra.
- ii. Compare amounts of bare soil, litter, basal vegetation, rock and standing water between years. If vegetative cover is increased dramatically, as indicated for 2017, then bare soil hits should decrease proportionately. Because these layers were not recorded at points with lichen and vegetative cover in 2006, but were recorded for all points in 2017, it would be necessary to filter 2017 data, by selecting only the subset of points with no vegetation or lichen hits. This would put the bare soil percentages on a comparable basis between years.
- iii. Evaluate the magnitude of differences with the Arctic Network's Vegetation Vital Sign "Node" dataset (Swanson and Neitlich, 2016) from Cape Krusenstern in 2010 and 2011.

4.1.6. Caribou grazing — Grazing on lichens by caribou (*Rangifer tarandus*) could affect the

abundance of lichens. In particular, if grazing was lower in the decade preceding 2017, as compared to the decade preceding 2006, we would expect an increase in both lichen and vegetation cover. We found that both lichens and vegetation increased from 2006 to 2017 consistent with a reduction in herd size from 2006 to 2017 (350,000 to 200,000). However, overall use of this area by caribou is relatively small, about 5% of the total records for the region, therefore minimal effects by caribou grazing can be inferred (K. Joly, personal communication, May 23, 2018).

4.1.7. Phenological differences — Local geography and short-term seasonal climate differences influence the phenology of vascular plant leaf development, such that vegetative cover could appear to be lower if sampling was begun before full expansion of summer foliage. Field conditions were cold and wet at the beginning of the 2006 field season. If this influenced our cover estimates we would expect increasing vegetative cover relative to date, especially for deciduous vascular plants. To evaluate this, we regressed total cover against Julian day of year for the vegetation data set and compared that to average temperature at two regional weather stations (Kivalina and Kotzebue) and precipitation of Kotzebue (Figure A1). When the two field seasons are superimposed on the same graph, the differences in total cover between years is apparent, as expected, but the trend was inconsistent between years. The 2006 field season had a decrease in total cover relative to date and the 2017 field season had an increase in total cover relative to date. Therefore, we conclude that short term weather-induced phenological differences did not lead to the observed trends of changing total cover through time (Figure A1).

4.1.8. Effects of CaCl_2 dust palliative on the haul road — The use of calcium chloride (CaCl_2) to diminish fugitive dust from the haul road could effect the lichens and or vegetation through the

addition of Ca^{2+} and Cl^- to the system. If this were the primary cause of increasing lichen and vegetation cover, then we would expect larger changes in vegetation and lichens near the haul road than far from the haul road. As described above, the opposite is true: changes in cover were larger in the more distant plots. Therefore, we conclude that CaCl_2 dust additions were not a primary cause of the increasing lichen and vegetation cover from 2006 to 2017.

4.1.9. Effects of the haul road dust as a nutrient source — Many of the minerals from the road dust could act as macronutrients to lichens and vegetation along the haul road, supplying nitrogen, phosphorus, potassium and calcium. If this were the primary cause of increasing vegetative and lichen cover, then we would expect larger changes in vegetation and lichens near the road than far from the road. As described above, the opposite is true: changes in cover were larger in the more distant plots. Therefore, we conclude that macronutrients in dust additions were not a primary cause of increasing vegetative and lichen cover from 2006 to 2017.

Chapter 5: Conclusions

In the summer of 2017, 104 plots located in Cape Krusenstern National Monument NW Alaska were remeasured. The goals were to document potential effects of metal deposition, and climate change to the local ecosystem as manifest in changes in lichen and vegetation communities. In the decade from 2006 to 2017, the lichen community at our study site changed significantly, with a loss of diversity and a gain in mean total cover. Lichen diversity and covers had negative relationships to metal deposition and distance from the haul road. In comparison, vegetation species richness remained nearly constant, but with a mean doubling of cover through the study period.

The observed correlation of lichen communities to metal deposition suggest sensitivity to metal deposition and reinforces their use as biomonitors in this system. A strong correlation of vegetation to metal deposition was not detected, however, suggesting that vascular vegetation is less sensitive to this particular stress. Community structure did not differ significantly between the north and south sides of the haul road.

Changes documented through time in the present study suggest a relationship to climate change as mediated through increasing temperature, site drying, and loss of wetland indicators. The observed increases in both total lichen and vegetation cover, do not agree with the vast majority of literature, that suggests increasing vascular vegetation at the expense of lichens. Variables such as regional climate, biome, temperature and wetland status appear to be significant in determining climate change effects. Our results indicate a likely drying of the study area and a trend of increasing average temperature through time. These are likely leading to the large increases in total cover observed over a mere decade. With the swift changes in Arctic

ecosystems documented both here and in other studies, it is vital that research in these regions continue so as to inform policy makers and future generations.

Each of the questions posed in the introduction are answered below.

5.1. How have lichen community metrics (species richness and composition) changed between 2006 and 2017 in relation to distance to the haul road?

Lichen communities changed significantly from 2006 to 2017. This is expressed in every lichen community metric examined. From 2006 to 2017 lichen communities decreased in species richness, increased in β diversity (average half changes), and more than doubled in lichen mean total cover. These data suggest that 2006 species-rich plots had low lichen cover and transitioned in a decade to species-poor plots with high lichen cover (lichen mean total cover 6% to 16%; species richness 26 to 16 (Table 2). When each year was examined separately, we saw a strong correlation in both years to lichen species richness (2006 $r = 0.672$; and 2017 $r = 0.567$). However, when the two years were combined as in the overall analyses, that relationship was diminished (both years $r = 0.353$) (Figure 1, bottom row).

Lichen communities were strongly related to metal deposition from the haul road, as lichen species richness increased with distance from the haul road (Figure 4) and metal deposition in 2006 was strongly correlated with the ordination of plots in lichen species space (Figure 1, Table 3).

5.2. How have vegetation community metrics (species richness and composition) changed between 2006 and 2017 in relation to distance to the haul road?

Vegetation communities changed significantly from 2006 to 2017. This is expressed in most of the vegetation community metrics examined. Vegetation mean total cover appears to have almost doubled from 2006 to 2017, however vegetation β_D Diversity (average half changes) remained static in the 10-year time period (Table 2). Unlike the lichen community, a strong relationship to metal deposition was not detected. Metal deposition, as measured through concentrations in the moss *Hylocomium splendens*, was only weakly correlated with the vegetation ordination (Figure 1, Table 3) and vegetation species richness increased slightly with distance from the haul road (Figure 4).

5.3. Do lichens and vegetation have different relationships to the metals gradient and if so, which are more sensitive to metal contamination?

Differences in metal deposition in this system are spatially pronounced. Lichen communities exhibited a much stronger relationship to metal deposition than did vegetation. Lichen communities appear to be effective biomonitors of heavy metal pollution in this particular system. These data support past research documenting lichens as effective biomonitors (e.g. Rühling and Tyler, 1969; Sloof, 1995; Wolterbeek, 2002; Conti and Cecchetti, 2001).

The entirety of the 2017 metal deposition data are not yet available. For the purposes of this manuscript, we considered differences in metal levels between 2006 and 2017 to be small compared to the changes in lichens and vegetation.

5.4. Do vegetation communities differ between the north and south sides of the haul road?

Neither lichen nor vegetation communities differed significantly between the north and south sides of the haul road. This offers an opportunity to simplify further analyses by ignoring that factor.

5.5. Are vegetation changes from 2006 to 2017 far from the haul road (≥ 2000 m) related to climate change?

A shift in vascular plant community from wetland to upland associated plants was documented through time (Figure 10). These results support overall predictions of a warming climate in the arctic leading to vegetation shifts northwards (Hassol, 2004). Further potential evidence of regional influences from climate change are seen in the doubling of total vegetation cover as well as total lichen cover (Figure 2). The trend of increasing vegetation cover through time has been associated with changes in climate, particularly due to a rapid warming observed in the Arctic (e.g. Elmendorf et al., 2012b; Fraser et al., 2014; Hassol, 2004; Jia et al., 2009; Pearson et al., 2013; Stow et al., 2004). Elmendorf et al. (2012a) suggested that the most significant climate-induced shrub expansion will take place in about 66% of arctic tundra regions that have ‘warm’ summer temperatures, tall shrubs and in particular areas that are wet and moist. This idea agrees with our results of the vegetation community shifting from wetland associated to upland associated species, potentially due to a warming climate.

Most of these studies, however, document an associated decrease in lichen cover with the increase in vascular vegetation, attributed to vascular vegetation suppressing lichens. Some studies do agree however with our results, such as Daniëls et al., (2011), who observed an increase in both vegetation and lichen cover in southern Greenland over a 40-year period from 1968 to 2007. They attributed these increases to a decrease in substrate moisture, likely a product of climate warming. Daniëls and de Molenaar (2011) similarly recorded an increase in lichen cover and richness from 1900 to 2007. They concluded that the observed changes were most conspicuous in wet and temporarily moist habitats, possibly due to a warming climate and less snow accumulation in winter.

In conclusion, it is evident from the variety in responses of lichens and vegetation to climate change, that we cannot expect one uniting trend. In fact, Elmendorf et al., (2012a) expects some regional heterogeneity, as there are many climate change moderators other than temperature and precipitation which have unknown relationships to vegetation and climate change.

Chapter 6: Bibliography

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Appendix- Supplementary Content

Table A1. Detailed list of taxonomic decisions made to synonymize the 2006 and 2017 data sets.

Species Lumped Together or Previous Name Used	Composite/Revised Name Used for Analysis
Lichens	
<i>Cladina arbuscula</i> , <i>Cladina mitis</i>	<i>Cladonia arbuscula</i>
<i>Cladina stygia</i>	<i>Cladonia stygia</i>
<i>Cladonia albonigra</i> , <i>Cladonia cryptochlorophaea</i> , <i>Cladonia merochlorophaea</i> , <i>Cladonia grayi</i>	<i>Cladonia albonigra</i>
<i>Cladonia bacilliormis</i> , <i>Cladonia cyanipes</i>	<i>Cladonia cyanipes</i>
<i>Cladonia chlorophaea</i> , includes some melanotic specimens	<i>Cladonia chlorophaea</i>
<i>Cladonia coccifera</i> , <i>Cladonia borealis</i>	<i>Cladonia coccifera</i>
<i>Cladonia crispata</i> , <i>Cladonia crispata</i> var. <i>crispata</i>	<i>Cladonia crispata</i>
<i>Cladonia maxima</i> , <i>Cladonia gracilis</i> , <i>Cladonia gracilis</i> ssp. <i>vulnerata</i> , <i>Cladonia gracilis</i> ssp. <i>elongata</i> , <i>Cladonia ecmocyna</i>	<i>Cladonia gracilis</i>
<i>Cladonia phyllophora</i> , <i>Cladonia stricta</i>	<i>Cladonia phyllophora</i>
<i>Cladonia squamosa</i> , <i>Cladonia squamosa</i> var. <i>squamosa</i>	<i>Cladonia squamosa</i>
<i>Cladonia subfurcata</i> , <i>Cladonia crispata</i> var. <i>cetrarriiformis</i>	<i>Cladonia subfurcata</i>
<i>Dactylina arctica</i> , <i>Dactylina beringica</i>	<i>Dactylina arctica</i>
<i>Cetraria cucullata</i>	<i>Flavocetraria cucullata</i>
<i>Lobaria linita</i> , <i>Lobaria pseudopulmonaria</i>	<i>Lobaria linita</i>
<i>Peltigera polydactylon</i> , <i>Peltigera neopolydactyla</i>	<i>Peltigera polydactylon</i>
<i>Physcia aipolia</i>	<i>Physcia alnophila</i>
<i>Stereocaulon grande</i> , <i>Stereocaulon tomentosum</i> , <i>Stereocaulon alpinum</i> , <i>Stereocaulon groenlandicum</i>	<i>Stereocaulon alpinum</i>
<i>Thamnolia subuliformis</i> , <i>Thamnolia vermicularis</i>	<i>Thamnolia subuliformis</i>
Vascular Plants	
<i>Arctostaphylos rubra</i> , <i>Arctostaphylos alpina</i>	<i>Arctostaphylos alpina</i>
<i>Betula glandulosa</i> , <i>Betula nana</i>	<i>Betula nana</i>
<i>Dryas integrifolia</i> , <i>Dryas integrifolia</i> ssp. <i>integrifolia</i>	<i>Dryas integrifolia</i>
<i>Empetrum nigrum</i> , <i>Empetrum hermaphroditum</i>	<i>Empetrum hermaphroditum</i>
<i>Eriophorum angustifolium</i> , <i>Eriophorum angustifolium</i> ssp. <i>subarcticum</i> , <i>Eriophorum vaginatum</i>	<i>Eriophorum angustifolium</i>
<i>Lupinus arcticus</i> , <i>Lupinus</i> sp.	<i>Lupinus arcticus</i>
<i>Pedicularis langsдорфii</i> , <i>Pedicularis labradorica</i> , <i>Pedicularis capitata</i> , <i>Pedicularis</i> sp.	<i>Pedicularis</i> sp.

<i>Petasites frigidus</i> , <i>Petasites frigidus</i> ssp. <i>frigidus</i> , <i>Petasites</i> sp.	<i>Petasites frigidus</i>
<i>Poa arctica</i> ssp. <i>lanata</i> , <i>Calamagrostis canadensis</i> , <i>Arctagrostis latifolia</i> , <i>Festuca altaica</i>	Unknown Grass
<i>Polygonum bistortum</i> , <i>Bistorta plumosa</i>	<i>Polygonum bistortum</i>
<i>Saxifraga punctata</i> ssp. <i>nelsoniana</i> , <i>Saxifraga</i> sp.	<i>Saxifraga</i> sp.

Table A2. Description of variables used in the environmental matrix.

Variable Name	Description	Units	Range
Plot	Plot name	Categorical	104 plots within CAKR
Year	Sampling year	Quantitative	2006 or 2017
YearC	Sampling year	Categorical	2006 or 2017
Transect	Transect number	Categorical	12 transects, on north and south side of haul road
Plot_Type	Plot type	Categorical	T=Transect, R=Reference, K=Kotzebue Surveyors from 2006 and 2017
L_Name	Lichen surveyor name	Categorical	2017
Sum_Moss	Total moss cover	Quantitative	104 plots for both years
Sum_Vasc	Total vascular plant cover	Quantitative	104 plots for both years
S	Species richness	Quantitative	104 plots for both years
H'	Shannon-Wiener index	Quantitative	104 plots for both years
Sum	Total Lichen and Vegetation cover	Quantitative	104 plots for both years
S_Li	Lichen species richness	Quantitative	104 plots for both years
H'_Li	Lichen Shannon-Wiener index	Quantitative	104 plots for both years
Sum_Li	Total lichen cover	Quantitative	104 plots for both years
S_Veg	Vegetation species richness	Quantitative	104 plots for both years
H'_Veg	Vegetation Shannon-Wiener index	Quantitative	104 plots for both years
Sum_Veg	Total vegetation cover	Quantitative	104 plots for both years
DistRd	Distance to haul road in m	Categorical	104 plots for both years
SideRd	South or north side of road	Categorical	104 plots for both years
Topo	Topographic Position (Upper, Lower, Mid Slope)	Categorical	104 plots for both years
Slope	Slope in degrees	Quantitative	104 plots for both years
PDIR	Potential Direct Incident Radiation	Quantitative	104 plots for both years
HeatI	Heat load	Quantitative	104 plots for both years
E	Cover type Ericaceous, yes or no	Categorical	104 plots for both years
LogDist	log10 distance to road	Quantitative	104 plots for both years
TallS	Ocular tall shrub cover	Quantitative	104 plots for both years
DwS	Ocular dwarf shrub cover	Quantitative	104 plots for both years
Forb	Ocular forb cover	Quantitative	104 plots for both years
Sedge	Ocular sedge cover	Quantitative	104 plots for both years
Grass	Ocular grass cover	Quantitative	104 plots for both years
Bryo	Ocular bryophyte cover	Quantitative	104 plots for both years
Lich	Ocular lichen cover	Quantitative	104 plots for both years
BareSoil	Ocular bare soil cover	Quantitative	104 plots for both years
SumSoilCov	Total soil cover	Quantitative	104 plots for both years
DuffOM	Ocular duff organic matter cover	Quantitative	104 plots for both years
Rock	Ocular rock cover	Quantitative	104 plots for both years

StH2O	Ocular standing water cover	Quantitative	104 plots for both years
Hylspl	Ocular Hylocomium splendens cover	Quantitative	104 plots for both years
Macr_Poi	Total macrolichen cover	Quantitative	104 plots for both years
Cr_poi	Total crustose lichen cover	Quantitative	104 plots for both years
Vasc_17	2017 ocular vascular cover	Quantitative	104 plots for both years
Shrub_17	2017 ocular shrub cover	Quantitative	104 plots for both years
MSH_17	2017 ocular mid shrub cover	Quantitative	104 plots for both years
SSh_17	2017 ocular sub shrub cover	Quantitative	104 plots for both years
DwSS_17	2017 ocular dwarf sub shrub cover	Quantitative	104 plots for both years
Gram_17	2017 ocular graminoid cover	Quantitative	104 plots for both years
Sphag_17	2017 ocular Sphagnum cover	Quantitative	104 plots for both years
Crust_17	2017 ocular crustose lichen cover	Quantitative	104 plots for both years
Distur_17	2017 ocular disturbed cover	Quantitative	104 plots for both years
Crater_17	2017 ocular cratered disturbance cover	Quantitative	104 plots for both years
Pell_17	2017 ocular pelleted disturbance cover	Quantitative	104 plots for both years
MosBl_17	2017 ocular moss blackening cover	Quantitative	104 plots for both years
LichBl_17	2017 ocular lichen blackening cover	Quantitative	104 plots for both years
LichSt_17	2017 ocular lichen stressed cover	Quantitative	104 plots for both years
MoSt_17	2017 ocular moss stressed cover	Quantitative	104 plots for both years
VasSt_17	2017 ocular vascular stressed cover	Quantitative	104 plots for both years
Microf_17	2017 microfeature (tussocks/earth hummocks)	Categorical	104 plots for both years
Veg_1_17	2017 Vegetation class	Categorical	104 plots for both years
Lon	Longitude	Quantitative	104 plots for both years
Lat	Latitude	Quantitative	104 plots for both years
PC_Axis1	2006 metals Principal component analysis axis 1	Quantitative	104 plots from 2006 field season
PC_Axis2	2006 metals Principal component analysis axis 2	Quantitative	104 plots from 2006 field season
PC_Axis3	2006 metals Principal component analysis axis 3	Quantitative	104 plots from 2006 field season
log(Pb_m_06)	2006 log10 Pb concentration	Quantitative	104 plots from 2006 field season
log(Cd_m_06)	2006 log10 Cd concentration	Quantitative	104 plots from 2006 field season
log(Zn_06)	2006 log10 Zn concentration	Quantitative	104 plots from 2006 field season

Table A3. Blocked Indicator Species Analysis for the “Lichen” data set. Increases are in orange and decrease have no color. Terms used: RF (Relative Frequency) and RA (Abundance).

Species	Max Group	IV	p*	RF 2006	RF 2017	RA 2006	RA 2017
<i>Cladonia rangiferina</i>	2006	66.6	0.001	78	28	85	15
<i>Cetraria sepincola</i>	2006	65.1	0.001	79	34	82	18
<i>Cladonia fimbriata</i>	2006	62.9	0.001	65	3	97	3
<i>Flavocetraria cucullata</i>	2017	58.3	0.001	93	87	33	67
<i>Peltigera aphthosa</i>	2017	58.1	0.001	73	79	27	73
<i>Cetraria laevigata</i>	2017	55.2	0.001	75	76	28	72
<i>Melanelia septentrionalis</i>	2006	55.1	0.001	73	31	75	25
<i>Thamnolia subuliformis</i>	2017	53	0.008	87	86	38	62
<i>Cladonia ochrochlora</i>	2006	50.5	0.001	51	0	100	0
<i>Cladonia amaurocraea</i>	2006	48.9	0.034	90	72	55	45
<i>Cladonia gracilis</i> s.l.	2017	48.8	0.034	85	74	34	66
<i>Parmeliopsis hyperopta</i>	2006	47.5	0.001	52	7	92	8
<i>Cladonia phyllophora</i>	2006	46.3	0.001	53	9	88	12
<i>Cladonia uncialis</i>	2006	46.2	0.001	60	23	77	23
<i>Cetraria pinastri</i>	2006	44.8	0.001	66	40	68	32
<i>Cladonia stygia</i>	2017	44.3	0.099	79	72	39	61
<i>Cladonia sulphurina</i>	2006	42.5	0.001	56	25	76	24
<i>Cladonia arbuscular</i>	2017	41.4	0.704	88	72	43	57
<i>Cladonia cornuta</i>	2006	40.7	0.045	73	57	56	44
<i>Cetraria islandica</i>	2006	40	0.001	59	29	68	32
<i>Parmelia sulcata</i>	2006	36.2	0.001	39	5	92	8
<i>Cladonia carneola</i>	2006	35.1	0.001	35	0	100	0
<i>Cladonia deformis</i>	2006	34.5	0.001	42	12	82	18
<i>Cladonia squamosa</i>	2006	34.2	0.002	46	21	74	26
<i>Peltigera scabrosa</i>	2017	31.4	0.307	57	52	39	61
<i>Cladonia</i> sp.	2006	30	0.001	34	6	88	12
<i>Parmeliopsis ambigua</i>	2006	29.2	0.001	34	8	86	14
<i>Cladonia pyxidata</i>	2006	28.9	0.001	30	2	97	3
<i>Cladonia pleurota</i>	2017	27.9	0.001	26	41	32	68
<i>Cladonia cyanipes</i>	2006	27.8	0.076	47	36	59	41
<i>Cladonia coccifera</i>	2006	27.4	0.002	35	12	78	22
<i>Cetraria inermis</i>	2006	26.7	0.001	33	9	81	19
<i>Peltigera polydactylon</i>	2006	24	0.801	48	44	50	50
<i>Cladonia coniocraea</i>	2006	23.2	0.001	24	1	98	2
<i>Cladonia albonigra</i>	2017	21.6	0.091	26	35	38	62
<i>Cladonia gracilis</i> ssp. <i>turbinata</i>	2006	19.8	0.001	22	2	91	9
<i>Hypogymnia subobscura</i>	2006	18.6	0.001	19	0	100	0

<i>Sphaerophorus globosus</i>	2006	18.4	0.04	30	20	61	39
<i>Nephroma arcticum</i>	2017	18.3	0.169	26	30	39	61
<i>Psoroma hypnorum</i>	2006	18.3	0.001	22	5	85	15
<i>Nephroma expallidum</i>	2006	16.8	0.095	28	19	60	40
<i>Hypogymnia physodes</i>	2006	16.2	0.013	22	9	75	25
<i>Cladonia subfurcata</i>	2006	15.6	0.002	18	3	89	11
<i>Cladonia crispata</i>	2006	15.3	0.029	21	8	74	26
<i>Dactylina arctica</i>	2006	14.9	0.058	24	15	63	37
<i>Peltigera leucophlebia</i>	2006	14.5	0.014	19	5	78	22
<i>Lobaria linita</i>	2017	14.2	0.833	29	26	45	55
<i>Parmelia omphalodes</i>	2017	13.8	0.387	19	24	42	58
<i>Cladonia transcendens</i>	2006	13.4	0.001	13	0	100	0
<i>Cladonia scabriuscula</i>	2006	13.2	0.004	15	3	85	15
<i>Bryocaulon divergens</i>	2006	13.1	0.024	18	7	75	25
<i>Peltigera didactyla</i>	2006	12.6	0.002	14	2	88	13
<i>Peltigera extenuata</i>	2017	12.4	0.001	0	12	0	100
<i>Cetraria nivalis</i>	2006	12.3	0.805	24	22	52	48
<i>Asahinea chrysantha</i>	2006	11.5	0.135	18	12	66	34
<i>Ochrolechia frigida</i>	2006	11.3	0.002	11	0	100	0
<i>Cladonia bellidiflora</i>	2006	10.9	0.009	12	2	88	12
<i>Peltigera horizontalis</i>	2017	10.3	0.001	0	10	0	100
<i>Cladonia chlorophaea</i>	2006	10.2	0.686	19	15	55	45
<i>Peltigera malacea</i>	2006	9.7	0.483	16	12	59	41
Unknown crust	2017	9.3	0.003	0	9	0	100
<i>Stereocaulon paschale</i>	2006	9	0.115	13	7	67	33
<i>Stereocaulon alpinum</i>	2017	8.9	0.047	5	11	22	78
<i>Peltigera membranacea</i>	2017	8.8	0.064	4	11	22	78
<i>Peltigera rufescens</i>	2017	8.8	0.111	6	12	29	71
<i>Cladonia cenotea</i>	2017	7.5	1	13	14	48	52
<i>Cladonia decorticata</i>	2006	7.2	0.022	7	0	100	0
<i>Pannaria pezizoides</i>	2006	7.2	0.013	7	0	100	0
<i>Peltigera kristinssonii</i>	2006	7.2	0.02	7	0	100	0
<i>Alectoria ochroleuca</i>	2006	6.5	0.084	8	2	79	21
<i>Sphaerophorus fragilis</i>	2006	6.2	0.024	6	0	100	0
<i>Pertusaria dactylina</i>	2017	5.6	0.604	6	9	40	60
<i>Cladonia digitata</i>	2006	5.2	0.05	5	0	100	0
<i>Peltigera canina</i>	2006	4.6	0.327	6	2	75	25
<i>Cladonia metacorallifera</i>	2017	4.3	0.754	5	7	41	59
<i>Peltigera occidentalis</i>	2017	4.1	0.13	0	4	0	100
<i>Bryoria simplicior</i>	2006	3.9	0.397	5	2	75	25
<i>Alectoria nigricans</i>	2006	3.3	0.37	4	1	80	20
<i>Peltigera sp.</i>	2006	3.3	0.375	4	1	80	20

<i>Protopannaria pezizoides</i>	2017	3.3	0.374	1	4	20	80
<i>Cetrelia alaskana</i>	2006	3.1	0.253	3	0	100	0
<i>Cladonia cervicornis</i>	2017	3.1	0.238	0	3	0	100
<i>Cladonia macrophylla</i>	2017	3.1	0.252	0	3	0	100
<i>Dactylina ramulosa</i>	2006	3.1	0.247	3	0	100	0
<i>Parmelia saxatilis</i>	2006	3.1	0.272	3	0	100	0
<i>Cladonia pocillum</i>	2006	2.6	0.49	3	1	83	17
<i>Masonhalea richardsonii</i>	2006	2.6	0.502	3	1	83	17
<i>Phycia alnophila</i>	2006	2.3	0.621	3	1	75	25
<i>Cladonia singularis</i>	2006	2.1	0.503	2	0	100	0
<i>Cladonia stellaris</i>	2006	2.1	0.514	2	0	100	0
<i>Icmadophila ericetorum</i>	2006	2.1	0.522	2	0	100	0
<i>Melanohalea olivacea</i>	2006	2.1	0.501	2	0	100	0
<i>Pilophorus robustus</i>	2017	2.1	0.504	0	2	0	100
<i>Stereocaulon sp.</i>	2006	2.1	0.489	2	0	100	0
<i>Solorina saccata</i>	2017	1.5	1	1	2	25	75
<i>Bryoria nitidula</i>	2017	1.4	1	1	2	33	67
<i>Cetraria sp.</i>	2006	1	1	1	0	100	0
<i>Cetraria ericetorum</i>	2017	1	1	0	1	0	100
<i>Cetraria fastigiata</i>	2006	1	1	1	0	100	0
<i>Cetraria nigricans</i>	2017	1	1	0	1	0	100
<i>Cladonia acuminata</i>	2017	1	1	0	1	0	100
<i>Cladonia bacillaris</i>	2017	1	1	0	1	0	100
<i>Cladonia macrophyllodes</i>	2006	1	1	1	0	100	0
<i>Cladonia macroceras</i>	2006	1	1	1	0	100	0
<i>Cladonia wainioi</i>	2006	1	1	1	0	100	0
<i>Hypogymnia sp.</i>	2006	1	1	1	0	100	0
<i>Hypogymnia austerodes</i>	2006	1	1	1	0	100	0
<i>Hypogymnia bitteri</i>	2017	1	1	0	1	0	100
<i>Lasallia pensylvanica</i>	2006	1	1	1	0	100	0
<i>Lobaria scrobiculata</i>	2006	1	1	1	0	100	0
<i>Nephroma sp.</i>	2006	1	1	1	0	100	0
<i>Parmelia sp.</i>	2006	1	1	1	0	100	0
<i>Peltigera britannica</i>	2017	1	1	0	1	0	100
<i>Phycia sp.</i>	2006	1	1	1	0	100	0
<i>Ramalina roesleri</i>	2006	1	1	1	0	100	0
<i>Solorina bispora</i>	2006	1	1	1	0	100	0

Table A4. Blocked Indicator Species Analysis for the “Vegetation” data set. Increases are in orange and decrease have no color. Terms used: RF (Relative Frequency) and RA (Abundance).

Species	Max Group	IV	p*	RF 2006	RF 2017	RA 2006	RA 2017
Pleurocarpous moss	2017	78.8	0.001	20	87	9	91
<i>Rubus chamaemorus</i>	2017	69.5	0.001	82	88	21	79
<i>Hylocomium splendens</i>	2017	67.5	0.001	65	84	19	81
Acrocarpous moss	2017	67.1	0.001	92	97	31	69
<i>Carex bigelowii</i>	2017	61	0.001	69	97	37	63
<i>Empetrum hermaphroditum</i>	2017	60.4	0.001	78	88	31	69
<i>Vaccinium vitis-idaea</i>	2017	60.4	0.001	94	96	37	63
<i>Ledum ducumbens</i>	2017	59.3	0.001	99	99	40	60
<i>Sphagnum</i> moss	2017	55.7	0.001	63	71	22	78
<i>Betula nana</i>	2017	55.6	0.001	100	100	44	56
<i>Eriophorum angustifolium</i>	2006	52.1	0.027	93	94	56	44
<i>Vaccinium uliginosum</i>	2017	44.5	0.007	72	72	38	62
<i>Salix planifolia</i> ssp. <i>pulchra</i>	2017	33.1	0.333	63	58	43	57
<i>Pedicularis</i> sp.	2006	30.6	0.001	34	4	91	9
<i>Carex</i> sp.	2006	26.9	0.001	27	0	100	0
<i>Petasites frigidus</i>	2017	25.3	0.047	37	38	33	67
Unknown grass	2006	13.1	0.003	16	6	80	20
<i>Arctostaphylos alpina</i>	2017	8.2	0.346	12	13	39	61
<i>Saxifraga</i> sp.	2006	7.8	0.008	9	1	90	10
<i>Polygonum bistortum</i>	2006	6.7	0.073	10	4	70	30
<i>Pyrola</i> sp.	2006	6.7	0.013	7	0	100	0
<i>Ranunculus eschschultzii</i>	2006	5.9	0.03	7	1	87	13
Unknown	2017	5.8	0.065	2	7	14	86
<i>Salix</i> sp.	2006	5.6	0.024	6	1	97	3
<i>Salix arctica</i>	2017	4.9	0.137	1	6	14	86
<i>Polytrichum</i> sp.	2006	4.1	1	8	7	53	47
<i>Aulacomnium</i> sp.	2017	3.8	0.116	0	4	0	100
Unknown forb	2006	3.8	0.124	4	0	100	0
<i>Equisetum arvense</i>	2017	3.6	0.246	3	5	25	75
<i>Salix reticulata</i>	2006	3.6	0.815	8	6	47	53
<i>Dryas integrifolia</i>	2017	3.4	0.123	2	4	11	89

<i>Saussurea angustifolia</i>	2006	3.4	0.256	5	2	72	28
Liverwort	2006	3.1	1	6	5	55	45
<i>Andromeda polifolia</i>	2006	2.9	0.241	3	0	100	0
<i>Rhododendron lapponicum</i>	2006	2.9	0.263	3	0	100	0
<i>Lupinus sp.</i>	2006	2	0.468	3	1	70	30
<i>Carex aquatilis</i>	2006	1.9	0.508	2	0	100	0
<i>Polemonium acutiflorum</i>	2017	1.9	0.516	0	2	0	100
<i>Pyrola asarifolia</i>	2017	1.9	0.474	0	2	0	100
<i>Alnus crispa</i>	2017	1.3	1	2	2	34	66
<i>Aconitum sp.</i>	2006	1	1	1	0	100	0
<i>Cerastium beeringianum</i>	2017	1	1	0	1	0	100
<i>Dicranum sp.</i>	2017	1	1	0	1	0	100
<i>Equisetum sp.</i>	2006	1	1	1	0	100	0
<i>Petasites sp.</i>	2006	1	1	1	0	100	0
<i>Potentilla fruticose</i>	2006	1	1	1	0	100	0
<i>Potentilla sp.</i>	2006	1	1	1	0	100	0
<i>Ranunculus sp.</i>	2006	1	1	1	0	100	0
<i>Rhytidium robusta</i>	2017	1	1	0	1	0	100
<i>Rubus araticucis ssp. arcticus</i>	2006	1	1	1	0	100	0
Unknown sedge	2006	1	1	1	0	100	0
<i>Anemone narcissiflora</i>	2006	0.5	1	1	1	50	50

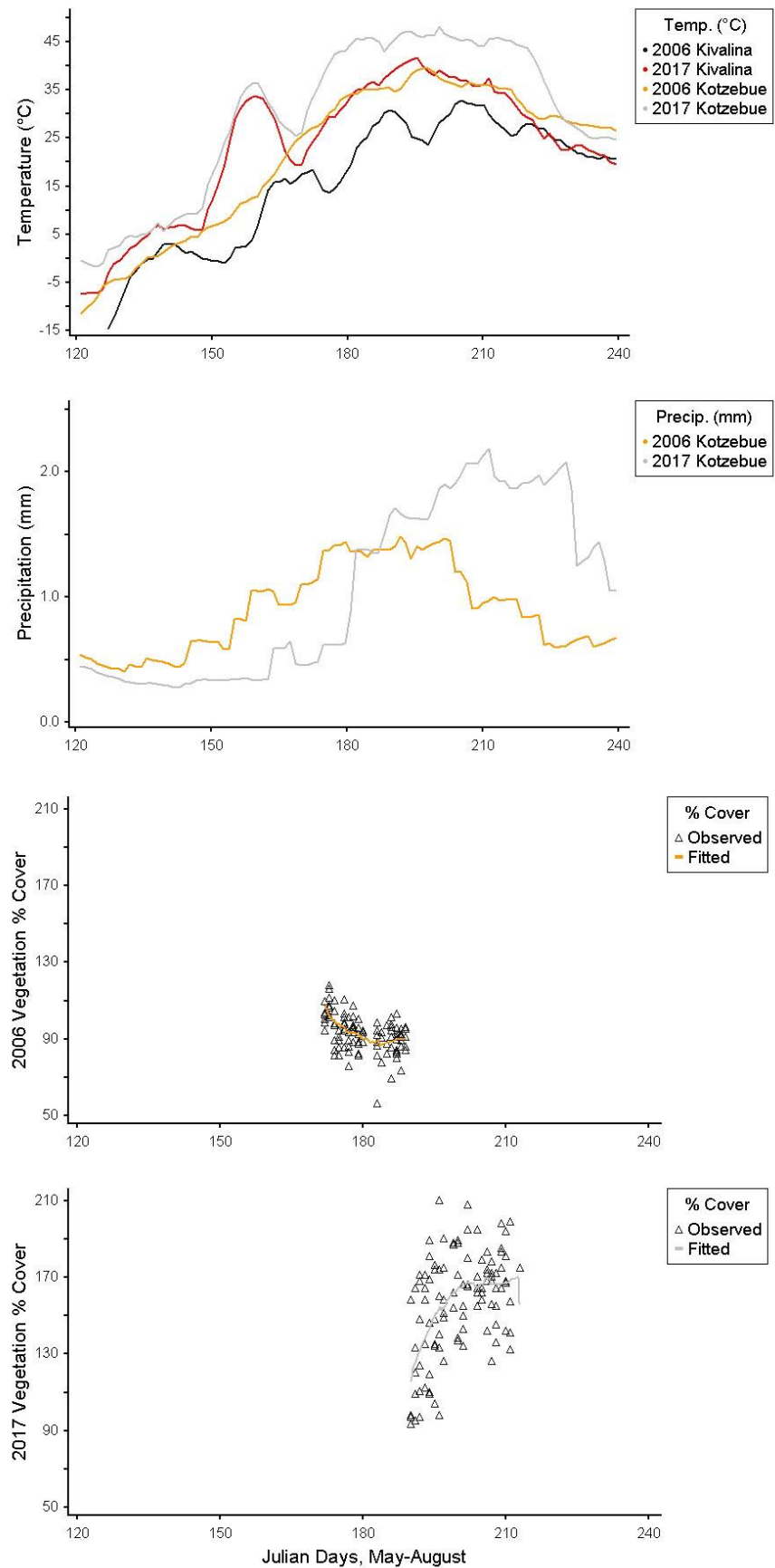


Figure A1. Average air temperature (°C) for 2006 and 2017 at two weather stations, Kivalina and Kotzebue Alaska; Kotzebue precipitation (mm); and total vegetation cover for each year regressed against Julian Days, May through August.

Table A5. Element concentrations of the 60 plots used for analysis.

Plot Name	As ppm	Cd ppm	Pb ppm	Fe ppm	Ni ppm	Cu ppm	Zn ppm
E1S10	4.766861	5.347302	90.67244	15611.82	25.85467	17.25955	746.5169
E1S100	0.991358	1.05515	40.27319	3024.744	5.302572	4.187045	160.0922
E1S1000	0.157474	0.205046	6.924934	337.3984	1.357167	2.011792	58.92903
E1S1000A	0.152441	0.241778	7.107465	349.7438	1.165642	2.363393	55.14257
E1S2000	0.13156	0.148122	6.765702	297.3461	1.876425	2.266961	44.01592
E1S300	0.392037	0.444093	15.82374	1149.943	5.201135	3.102783	95.96521
E1S4000	0.185067	0.162284	7.894095	450.6279	1.685174	2.944081	55.58666
E1S50	2.110111	1.897648	76.93415	7136.179	12.5417	7.425589	305.1079
E2S10	4.922158	3.228712	91.83711	15721.14	22.94294	11.79713	501.653
E2S100	0.722335	0.65071	23.67342	2836.833	5.368991	3.772093	119.6492
E2S300	0.310069	0.276627	13.15452	1019.839	2.668217	2.734602	86.09708
E2S50	1.26069	1.032433	39.48095	4682.449	9.35546	4.549025	170.7964
E3N10	4.108962	2.804194	60.07725	16500.68	24.66706	13.23462	414.4868
E3N100	1.929072	1.462906	50.29095	6650.683	10.61904	6.446449	223.0325
E3N300	1.370756	0.944153	42.34883	6061.964	9.950306	6.455368	184.3463
E3N50	2.362635	1.492126	46.86865	7798.2	10.62557	6.599829	222.6548
E4N10	8.208938	3.811587	111.9258	22799.33	30.9442	16.19377	583.7068
E4N100	2.647348	1.717201	67.78922	8297.475	11.42514	7.637245	265.3178
E4N300	0.722363	0.649972	32.15325	2544.846	4.210101	3.534405	131.7658
E4N50	2.090077	1.216212	48.30309	7871.713	12.64075	7.10774	181.6159
E5N10	7.330409	4.327841	103.6537	18485.89	26.88764	16.07986	638.9179
E5N100	0.977879	2.578505	112.6515	n/a	0.156695	8.299967	432.4489
E5N300	0.650387	0.718422	27.06523	n/a	9.718508	3.894372	134.5554
E5N50	3.588721	2.947383	83.19903	12398.3	16.59722	9.874363	460.7046
E6S100	0.756079	0.410458	21.2859	2484.651	5.550218	3.515509	74.13558
E6S2000	0.083768	0.065748	7.113876	n/a	5.806272	3.019019	32.28685
E6S300	0.285659	0.272415	9.998672	980.4446	3.26278	3.772532	66.19055
E6S4000	0.146382	0.150268	8.65028	311.9014	4.246419	3.096127	34.57167
E6S50	1.453271	0.516535	26.93142	4938.618	8.58479	5.061657	100.6402
EREF1-2	0.118353	0.051384	1.552634	271.9578	1.350173	2.955273	21.29149
EREF2-1	0.073814	0.152373	0.876966	154.3848	2.102601	3.35021	40.70003
EREF2-2	0.137895	0.104756	1.093865	289.7152	1.952109	3.230746	26.83841
EREF2-3	0.09683	0.088053	0.948985	210.0768	1.401545	2.848486	23.51782
T1N10	4.172282	6.391368	179.1916	16475.74	25.94923	16.37459	939.0615
T1N10	1.932732	5.857794	182.3768	n/a	29.52465	16.12032	767.8268
T1N100	2.318471	2.91294	119.8772	7684.52	11.60768	7.819642	421.1235
T1N50	2.708471	3.699126	116.6479	13927.62	19.96223	12.11236	643.4338
T2N10	3.206068	2.738523	83.24284	18649.12	23.53411	13.46522	455.4778
T2N100	1.7594	1.256539	45.67634	6371.546	8.979486	5.597295	193.8788

T2N300	0.62146	0.667058	29.95627	2317.809	4.032402	3.389163	117.7892
T2N50	1.465633	1.191799	53.52905	8357.747	11.66297	6.822	205.179
T3S10	2.326854	1.166871	56.45235	14890.24	19.20915	9.887882	220.7582
T3S100	0.537689	0.307814	17.01855	2144.948	4.118561	3.410708	67.91595
T3S300	0.247733	0.187652	12.47872	797.7879	1.92188	2.66827	73.30829
T3S50	1.314208	0.604139	28.92512	5594.072	8.29507	5.51423	135.4485
T4S10	4.014367	4.746967	136.0619	13526.36	23.04525	13.70125	639.2916
T4S100	1.031091	1.314663	50.88534	3897.82	8.37358	5.243549	212.1805
T4S300	0.37904	0.646537	24.69616	1246.038	3.996017	2.804364	106.2448
T4S50	2.191125	2.169839	87.9706	7745.267	13.57961	8.908434	333.3383
T6S10	3.763958	3.756232	119.9206	17929.97	22.204	13.1976	622.2619
T6S100	1.221162	1.410369	48.43546	5195.049	7.494649	5.47529	256.7183
T6S300	0.482969	0.460664	15.30253	1441.187	3.143457	2.938675	90.5106
T6S4000A	0.184344	0.423791	11.91109	433.097	2.583386	3.083672	101.5611
T6S50	1.913001	2.107283	89.66181	6032.818	9.009865	7.064198	325.9378
TREF1-1	0.155283	0.066751	1.456102	289.0729	1.24969	2.816191	68.97822
TREF1-2	0.107165	0.063681	1.13519	213.2809	1.019175	2.911795	63.00845
TREF1-3	0.107196	0.063755	1.233602	232.2924	0.813052	2.214765	57.36502
TREF2-1	0.115928	0.092902	0.993326	304.9282	2.099315	3.099199	24.06098
TREF2-2	0.103842	0.069298	0.750217	210.9049	1.168285	2.443895	24.14315
TREF2-3	0.059949	0.072852	0.974954	117.9961	0.760528	3.007506	26.22159