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Assessing the Atmospheric Deposition of Heavy Metals Using Biomonitoring of Moss Near a Mine in South-central British Columbia

BY

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ABSTRACT

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Mines are known to cause severe environmental problems. Heavy metals appear in the environment (i.e., during combustion, extraction, and processing), surface water (i.e., through runoff and releases from storage and transport), and in the soil. Air pollution is noted to be the primary concern of the public, in regard to mines. The presence of heavy metals in the air are known to have a drastic effect on human health and cause ecological implications. Moss have become a popular plant in monitoring studies as they have the ability to receive and accumulate heavy metals through absorption from the atmosphere and from their rooting substrate. These plants are very simple morphologically and anatomically and as a result, they are often chosen as an ideal organism for biomonitoring. I collected a total of 90 moss samples from 3 primary radii (North, East, and West) at distances of 1km, 2km, and 5km from the mine boundary for chemical analysis in order to meet two primary objectives: 1) determine whether moss outside the limits of the mine contain heavy metals: Lead (Pb), Zinc (Zn), Copper (Cu), and Nickel (Ni); and 2) analyze the potential use and effectiveness of biomonitoring to assess ecosystem response. Due to COVID restrictions, only samples from East 5km and West 2km were analyzed. Using flame atomic adsorption spectrometry (FAAS), concentrations of Zn and Cu were found to be higher at the East 5km site than at the West 2km site due to elevation, wind trends, and leaching. Concentrations of Pb and Ni were undetectable by the FAAS. Moving forward, biomonitoring is a relatively inexpensive, passive way to assess ecosystem response. Air pollution levels change rapidly over time and moss as biomonitors are capable of monitoring and detecting external conditions averaged over periods of time. The environmental impacts of mines outside their boundaries is poorly studied and future research would be beneficial. A standardized protocol is lacking for sampling, sample preparation, and elemental analysis.

Keywords: biomonitoring, bioindicator, biodiversity, moss, mining, heavy metals, pollution

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INTRODUCTION

Ecosystems are fundamentally comprised of various biotic and abiotic factors that work together to perform ecosystem functions. Without all of these factors, an ecosystem would be unable to function properly. Fauna act as key players ecologically requiring food, thus playing a role in energy transfer. Meanwhile, flora and fauna provide food to various different organisms. Relationships between all biota in an ecosystem can be described in the form of an ecological pyramid or trophic levels. Ecological pyramids refer to the community structure in an ecosystem, whereas trophic levels refer to how energy is moved throughout the system. Understanding how community structures occur in ecosystems is a fundamental goal in ecology. Community structure and function are formed based on interactions amongst organisms and the nature of these interactions is governed by the organisms involved (Trebilco et al. 2013). Simply put, trophic pyramids are composed of primary producers on the bottom level, herbivores in the middle, and carnivores on the top (Fath and Killian 2007). The sun is the primary source of energy for almost every ecosystem on Earth. Primary producers, typically referring to plants in terrestrial ecosystems, utilize energy from the sun to produce their own food. When primary producers are consumed by herbivores, energy flows up the pyramid one level at a time. There are several roles to be filled within ecological pyramids to ensure energy flows throughout the system; none of this can be achieved without adequate biodiversity.

Biodiversity is a broad unifying concept encompassing all forms, levels and combinations of natural variation, at all levels of biological organization. Put simply, biodiversity is the variety of life (Gaston and Spicer 2004, Gardner 2010). It can be measured on a variety of scales and is shaped by natural processes and increasingly by the influence of humans. The value of diversity can be classified into direct and indirect values. Direct values include consumptive use (i.e., global food supply) and productive use (i.e., commercial pesticides, pharmaceutical products). Whereas, the indirect values include: ethical, aesthetic, option, and environment service (Meerabai and Pullaiah 2016). For the purpose of this study, I would like to highlight the importance of biodiversity in plant communities.

Plants are a valuable resource for humanity; however, plant usage and importance are not limited to humanity. Plant life balances ecosystems, mitigates erosion, protects watersheds, moderates climate, and provides shelter for many animal species (Clark et al. 2018). Biodiversity is threatened by increasing human population, pollution, deforestation, and species extinction

(Pennsylvania State University 2020). Several species found on this Earth are indicator species give a signal by their presence, lack of abundance, or chemical composition about the distinctive aspects of the character or quality of an environment. A perfect example of an indicator species are moss species, sometimes referred to as bioindicators (Aboal et al. 2010, Angelovska et al. 2016, Berg and Steinnes 1997). The condition and health of moss can be used as an indication of ecosystem health.

Mosses are small green land plants found in a variety of habitats across the globe. Morphologically and anatomically, these plants are very simple (Godvindapyari et al. 2010, Schofield 1927). They lack a true root system allowing them to absorb heavy metals over their entire surface (Berg and Steinnes 1997, Degola et al. 2014, Stanković et al. 2018). Due to the lack of a cuticle layer, and a large surface-to-weight ratio, the cell walls of moss, with pronounced ion-exchange properties, are easily accessible to metal ions (Stanković et al. 2018). As a result of their simplicity, they react and reflect changes in heavy metal concentrations faster than most vascular plants making them an ideal organism for air pollution studies (Zvereva and Kozlov 2011). Moss species reproduce at a rapid rate, are totipotent, and have a high heavy metal accumulation capacity (Godvindapyari et al. 2010). Mosses have the capacity to absorb heavy metals from the atmosphere, their rooting substrate, or a combination of the two; as such, moss have recently become the most popular plant for biomonitoring studies (Aboal et al. 2010, Angelovska et al. 2016, Berg and Steinnes 1997). Moss lack specialized conducting tissue (Onianwa 2001) and have a slow growth rate (Chakrabortty and Paratkar 2006). Their growth segments can provide information about the exposure to heavy metals over longer periods of time, which can be very useful information in areas like mines where introduced heavy metals change rapidly (Stanković et al. 2018). The state of the moss species (whether it is living or dead) will affect the level of heavy metal contamination on the moss (Aboal et al. 2010, Berg and Steinnes 1997). Heavy metals have been classified as one of the most dangerous groups of anthropogenic pollutants due to their toxicity and persistence in the environment (Koz et al. 2012). Industrial processing and mining are two of the main sources of heavy metal contamination in the environment (Angelovska et al. 2016, Koz et al. 2012). Air pollution can result in a decrease in biodiversity and productivity in forests. In order to maintain a sustainable environment and minimize human expose to these strong pollutants, environmental policy must reflect desired outcomes.

There is a lack of environmental regulation, leading to a large input of toxic pollutants to the atmosphere, particularly from mining (Govindapyari et al. 2010). Biomonitoring is an excellent, cost-efficient method of monitoring air quality and could be used to test the impact mines are having on the environment surrounding their boundaries. Moss, as a bioindicator, is able to depict the quality of the environment on the basis of changes observed in the plant itself (Saxena and Harinder 2004). Air pollution problems can often arise in urban valleys due to high emissions of pollutants and limited ventilation resulting from inversions (Rendón et al. 2015). In complex terrain, such as in the Kamloops British Columbia region, topography limits the ventilation of pollution out of the valley and traps heat. With heavy metals being carried via air, this can result in the deposition of the particles in highly populated areas of the city, which will in turn have negative effects on human health and the vitality of plants species. Several elements pose a potential threat to human health in high levels including lead (Pb), cadmium (Cd), and arsenic (As). Copper (Cu) and Zinc (Zn) are essential to the human body for proper cellular function, regulation of the immune system, wound healing, and the synthesis of neutrotransmitters. Often elevated levels of Cu depress the absorption of Zn levels in the human body and vice versa (Hoffman et al. 1988). Zinc deficiencies can cause various symptoms such as, hair loss, weight loss, and problems with wound healing. Deficiencies of copper can lead to problems with connective tissue, muscle weakness, and neurological problems, however too much copper and zinc can be toxic (Uni. Of Roch. Med. Center 2020). Airborne emissions of these metals are of particular concern in terms of human health, because of the widespread dispersal capability and the inability of humans to avoid polluted air. Air quality in these areas also depends on the intensity of emissions, as well as atmospheric dispersion, chemical transformation conditions, and topographic location (Baumbach and Vogt 2003).

With environmental degradation becoming a prominent issue in today's society, there is growing concern for the environmental effects of mines. Studies have found high concentrations of heavy metals in the surrounding area of abandoned mines (Angelovska et al. 2016; Godvindapyari et al. 2010). Using atomic emission spectrometry with inductively coupled plasma, over 20 different elements were found in moss samples around a lead-zinc mine near Krivia Palanks town in Eastern Macedonia (Angelovska et al. 2016). Although heavy metals are naturally occurring, an excess amount can result in ultrastructural changes in chloroplasts and cell membranes as well as changes in the physiological processes and characteristics of moss (Choudhury and Panda 2005). Once heavy metals are introduced to the environment, they are often hard to remove and tend to accumulate in the tissues of plants and other organisms throughout the food chain (Lee and Von Lehmden, 1973, Maevskaya et al. 2001, Stanković et al. 2018.). Continuous monitoring is needed to ensure the heavy metals concentrations in the environment do not begin to negatively affect and influence ecosystems.

Highland Valley Copper Operations (HVC) is a copper and molybdenum operation located approximately 17km west of Logan Lake and about 50km southwest of Kamloops in British Columbia. Annual copper production is between 155,000 and 165,000 tonnes per year. This mine is one of the largest open pit copper mining and concentrating operations in the world (Teck Resources Limited 2019). Mining operations at Highland Valley began in 1962. HVC was created by the combination of three historic mining operations in 1986: Bethlehem, Lornex, and Highmont. The Bethlehem Copper operation was operating until 1982, meanwhile development began for the Lornex pit. Highmont Operating Corporation ran from 1979 until 1984 (BC Mine … n.d.). Over time, the mine increased in size and now occupies roughly 10,000 hectares (iMap BC 2020). Under the current mine plan, Highland Valley Copper Operations will be operating until 2027 (Teck Resources Limited 2019).

Prior to beginning their operations, HVC had to obtain a Project Approval Certificate at the mine. In order to do this, potential impacts on watersheds, fish and aquatic resources, wildlife resources, and land use was assessed through the use of models that facilitated projections of the effects under various management scenarios as required by the Environmental Code of Practice for Metal Mines. The code of practice was created with the objective of identifying and promoting the recommended best practices to encourage continual improvement in the environmental performance of mining through the mine life cycle (Gov. Canada 2013). A comprehensive mitigation and several monitoring plans were developed based on the results of the projections (Associated Environmental n.d.). My study took place outside the boundary of HVC to determine whether the environment outside of the mine boundary was being contaminated.

My study performed a chemical analysis on moss samples to determine heavy metal quantities in species located outside the boundaries of the Highland Valley Copper Mine. My overarching objective with this study was to collect new data and observations on the potential use of moss species as biomonitors within the low-elevation Douglas-fir forests in south-central British Columbia. More specifically, I sought to

1) **Determine whether moss outside the limits of the mine contain heavy metals: Lead (Pb), Zinc (Zn), Copper (Cu), and Nickel (Ni)**
2) **Analyze the potential use and effectiveness of biomonitoring to assess ecosystem response**

With these findings, further preventative measures can be put in place to ensure that mine companies are mitigating their impact on environmental pollution as much as possible.

STUDY SITES

Site determination was done using maps and forestry road exploration to determine access (Appendix A). My study sites were located approximately 20km west on the outskirts of Logan Lake, British Columbia adjacent to the boundary of the Highland Valley Copper Mine. Study sites were located within 1km, 2km, and 5km radii in the East, West, and North cardinal directions from the mine boundary (Appendix A). Due to laboratory restrictions as a result of COVID-19, the only samples that were analyzed for the purpose of this preliminary report include those collected at East 5km and West 2km.

All study sites were located within the Interior Douglas-fir (IDF) Biogeoclimatic (BEC) zone, dk1 (dry, cool) variant (Meidinger and Pojar 1991a and b). The West 2km site was located in a transition zone between the IDKdk1 zone and the MSxk (Montane Spruce, (very dry, cool) variant) zone. The IDFdk1 zone occurs in the lee of the Coast and Cascade mountains. It occupies lower to middle elevations of the southern Interior Plateau. Continental climate is characterized by warm, dry summers, a relatively long growing season, and cool winters. A rainshadow is the main factor controlling climate in this zone created in the lee of the Coast, Cascade, and Columbia mountains. Growing season moisture deficits are common as roughly 20-50% of all precipitation received falls as snow. The soils in this zone often have medium to rich nutrient status (Meidinger and Pojar 1991a). The Montane Spruce zone is found elevationally above the Interior Douglas-fir zone. The growing season is relatively warm and dry and moisture deficits can occur. This zone lacks many species that are characteristics of the IDF zone (Meidinger and Pojar 1991b).

The East 5km site (50.4740 -120.8943) was located at approximately 1200m in elevation (Figure 1). Forest cover consisted of mixed Interior Lodgepole pine (*Pinus contorta* var. *latifolia*) and Interior Douglas-fir (*Pseudotsuga menziesii* var. *glauca*) stand. Understory vegetation composition was limited to pinegrass (*Calamagrostis rubscens*), red-stem feathermoss (*Pleurozium schreberi*), and twinflower (*Linnea borealis*) (Table 4).

Figure 1. Photo taken on July 10th, 2020 at the starting point of the East 5km transect line.

Figure 2. Photo taken on July 10th, 2020 at the starting point of the West 2km transect line.

The West 2km site (50.5790 -121.200) was located in an area where forest harvesting commonly occurs (Figure 2). Vegetation on the site consisted mostly of young and mature Interior Douglas-fir (*Pseudotsuga menziesii var. glauca*) with minimal Interior Lodgepole pine the understory was comprised primarily of herbs and grasses. The understory of the East and West sites was very similar; however, percent cover of moss was higher at the West 2km site than the East 5km site (Table 4).

METHODS

FIELD DATA COLLECTION

Field data collection occurred on July 10th, 2020 during the peak of the growing season. Upon arrival to the sites, site classification information was collected along with vegetation information (Appendix B, Tables 1 and 2). A 10m transect line was laid in pre-determined study site locations deemed representative of the plant community. Along the transect line, a 30cm by 30cm grab sample of moss was taken every meter using a frame constructed out of white PVC piping. Species were identified to genus using a field identification guide written by Ryan, MW (n.d.) and a percentage estimate was taken for their coverage (Appendix C, Tables 3 and 4). In areas along the transect line where no moss was detected, the frame was moved up to 1.5m off the transect line in either direction creating a 1.5m buffer for moss detection.

CHEMICAL ANALYSIS

Chemical analysis was conducted by chemical biology undergraduate student, Abu Harera Nadeem and Zhi Chao Guo, a masters student in environmental chemistry, under the supervisor of Dr.Kinsley Donkor. Chemical analysis was conducted using methods developed by Rinne and Barclay (1980), to determine concentrations of Pb, Zn, Cu, and Ni measured in a microgram of the metal per gram of moss. Few modifications were made to the methods due to differing equipment available in the laboratory. Rather than boiling samples, a microwave digester was used and a pre-made program was created. The pre-made program began at 40°C then increased at 10mm/ss until it reached a temperature of 100°C. At this time, the digester held for 2 minutes and then proceeded to increase in temperature up to 185°C, once it cooled the samples were digested. Once digested, samples were filtered through a 0.45 micrometer nylon filter. Sample vials were rinsed twice with 18 MOhm water and diluted to 50mL in a volumetric flask using 18 MOhm water. For 48-72 hours the samples were stored in a fridge until the flame atomic absorption spectrometry (FAAS) analysis was conducted.

Statistical Analysis

Results were compiled using Microsoft Excel. Each sample was run through the FAAS three times; therefore, the results were averaged for each sample. A Two-Sample T-Test was conducted using MiniTab (Vers. 19) to compare whether the mean concentrations of Zn and Cu were equal at the East 5km site versus at the West 2km site.

Results

Site classification and vegetation information data were collected at a total of nine different study sites. A total of 90, 30cm by 30cm, grab samples of moss were collected. Results displayed are preliminary, based solely on the East 5km and West 2km locations. Due to COVID-19 restrictions throughout the university, the entire lab sample analysis will not be completed until December 2020. Majority of the samples collected at the East 5km site were identified as *Pleurozium spp.* (Appendix C, Table 5). The West 2km site showed more variation in the species found on site. The site was dominated by *Pleurozium spp.*, but *Dicranum spp., Pohlia spp.,* and *Brachythecium spp.* were also found and sampled (Appendix C, Table 6).

Chemical analysis was conducted to determine the concentrations of four main metals in the samples for the East 5km and West 2km sites. Both sites contained the same number of samples. The concentrations found for Pb and Ni thus far were not within the detectable limit of the FAAS, and therefore will not be discussed. When comparing the average concentration of Zinc measured in micrograms per gram of moss for the East 5km and West 2km sites using a two-sample t-test, it can be observed that the mean of the average concentrations is much higher at the East 5km site. The standard deviation and standard error mean are both greater values as well. The results determined the means to be significantly different with a p-value of 0.023 and a t-value of 2.49 (Table 1).

Table 1. Summary of two-sample t-test results. T-test compared average μg Zn/g of moss samples from each site (East 5km and West 2km). .

Sample	Sample Size	Mean	Standard Deviation	Standard Error Mean	T-Value	P-Value
East 5km	10	56.5	21.2	6.7	2.49	0.023
West 2km	10	34.2	18.8	5.9		

When comparing the average concentration of Copper in each one-gram sample, the mean for the East 5km site was again found to be greater than that of the West 2km site. The t-value was calculated to be 5.11 and the p-value was 0.000, respectively (Table 2).

Table 2. Summary of two-sample t-test results. T-test compared average µg Cu/g of moss sampled from each site (East 5km and West 2km).

Sample	Sample Size	Mean	Standard Deviation	Standard Error Mean	T-Value	P-Value
East 5km	10	133.0	54.9	17.0	5.11	0.000
West 2km	10	41.0	15.0	4.8		

Overall, mean concentrations of both Copper and Zinc were very high in the East 5km samples (133.0 µg Cu/g of moss and 56.47 µg Zn/g of moss) in comparison to the West 2km samples (40.97 µg Cu/g of moss and 31.17 µg Zn/g of moss) (Figure 3).

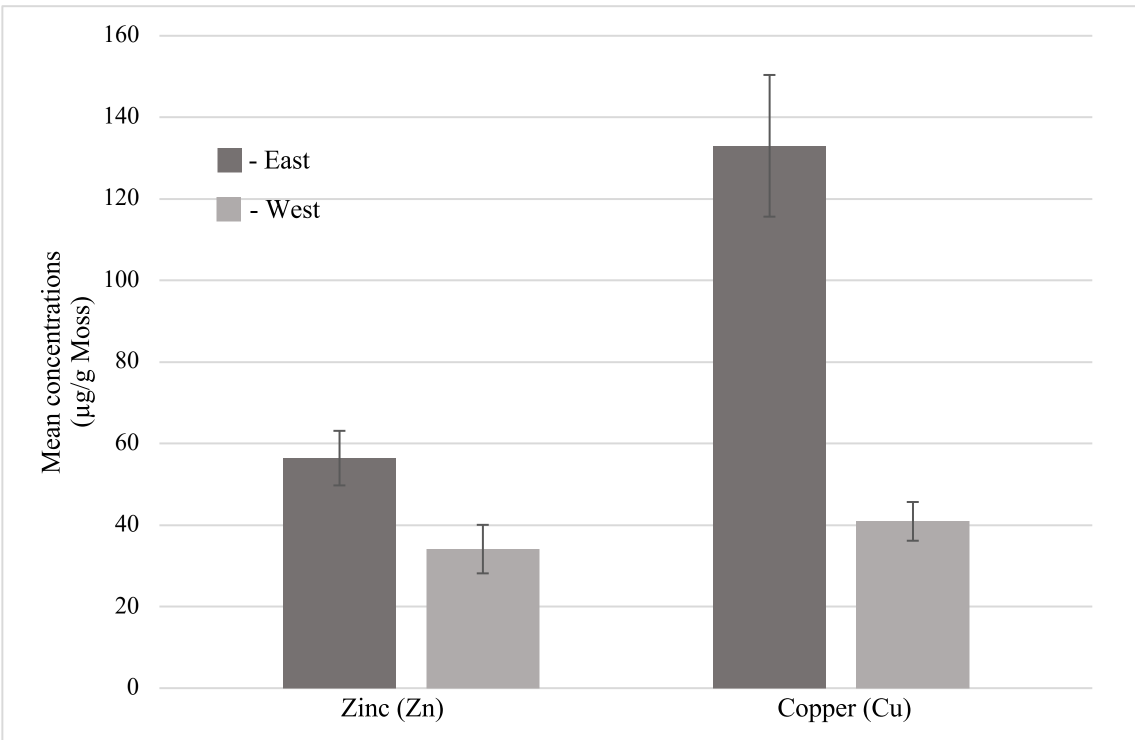

Figure 3. Average concentrations of Zn and Cu per gram of moss in samples collected from the East 5km and West 2km sites. Concentrations between the two sites were significantly different (Cu: p=0.023; Zn: p=0.000). Error bars represent 1 SE.

DISCUSSION

SITE CLASSIFICATION AND VEGETATION DATA

The West 2km site was more difficult to classify into a specific Biogeoclimatic zone based on the vegetation data and the site classification information collected. The site had plants from both the IDF zone and the MS zone. *Vaccinium scoparium* is a species more commonly found in the MS zone, whereas *Calamagrostis rubescens* is often found in the IDF zone. The MS zone generally occurs at higher elevations that the IDF zone, however upper elevations of the IDF zone are at 900 to 1450m. The MS zone ranges from 1100 to 1500m in elevation in wetter parts of the zone, and from 1250 to 1700m in drier areas. The West 2km site had an elevation of 1425m and showed visible signs of seepage and a water table close to the surface.

FACTORS AFFECTING HEAVY METALS CONCENTRATIONS

Results from the two-sample t-test comparing the two sites showed that mean concentrations were significantly different. Both t-values calculated (2.49 and 5.11) for the concentrations of Zn and Cu act as evidence that there is a significant difference between concentrations found at the East 5km site versus the West 2km site. The p-values are both very low (0.023 and 0.000) suggesting there is great difference in the concentrations collected. Mean concentrations of Zinc and Copper detected in the East 5km samples were much higher than those found in the West 2km samples. My original hypothesis was that samples collected further away from the mine boundary (i.e. 5km) would contain lower concentrations of heavy metals than those collected within one and two kilometers from the boundary.

All moss samples analyzed contained heavy metals, however none of the samples analyzed thus far contain Pb or Ni. Metals such as Zinc and Copper are essential to moss plants, therefore there may always be a background level of the element in the moss (Berg and Steinnes 1997). My study did not account for the base concentrations of metal elements that moss require. Copper and Zinc toxicity is relatively well studied in vascular plants (Elvira et al. 2020, Choudhury and Panda 2005), however much less is known about copper tolerance and resistance in bryophytes (Elvira et al. 2020). Various species of bryophytes such as the genera *Scopelophila* and *Mielichhoferia* are metallophilous, which means they require high metal levels to develop and therefore they uptake and concentrate metals in their tissues. Moss species that are native from contaminated soils often display higher tolerances to heavy metal contamination (Elvira et al. 2020). I struggled to find data in the literature describing acceptable tolerance levels of Zinc and Copper in moss species.

A large proportion of the pollutant load accumulates in mosses through wet deposition (Chakrabortty and Paratkar 2006). Copper is known to be an element that has a high uptake efficiency from wet deposition (Ross 1990). Accumulation and leaching are affected by the duration, intensity, and the amount of precipitation (Berg and Steinnes 1997). The East 5km site was characterized by a more arid climate, therefore the contribution of dry deposition would increase (Couto et al. 2004). Concentrations of heavy metals in the moss samples can be affected by various factors: stand throughfall, the nutrient status of the site, altitude, age of the moss, sampling technique, etc. (Chakrabortty and Paratkar 2006). Moss samples were collected using bare hands and samples were collected under the canopy and near other plants, which contradicts many sampling techniques found in the literature (Fernández et al. 2015).

I speculate that the moss samples collected at the East 5km site contain a greater concentration of heavy metals due to numerous factors. When looking at historical wind trends, it can be observed that prior to field data collection prevailing winds blew towards the East (Enviro. Canada 2020). Wind-blown dust may carry pollutants from the mine for several kilometers eventually resulting in deposition (Angelovska et al. 2016). Research conducted by Kim et al. (2012) concludes westerly winds were associated with enhanced concentrations of trace-elements in precipitation It is important to note that my study did not account for any soil dust that may have been deposited on the moss samples, therefore the results may be slightly skewed.

Natural sources may also contribute to the observed results: natural and cyclic processes, root uptake in higher plants and transfer to the moss, mineral particles releases to the air (i.e., from wind erosion to local soil), and uptake from the ground when the surface is saturated with water. The East 5km site is lower in elevation than the West 2km site, conceivably watershed processes could be impacting the deposition of metals as leaching occurs down the elevational gradient (Jarsjö et al. 2017). Perhaps there is an ideal distance that heavy metals can be suspended in the air for. After travelling 5km the particles may begin to deposit. There was no evidence found to support this claim, however it may be valuable to consider and study further.

Conclusions and Proposed Future Studies

Biomonitoring is a great way to assess ecosystem response moving forward. It is passive and provides a measure of integrated exposure over a period of time. This method is relatively inexpensive, and no long-term use of expensive sampling equipment is required. Heavy metal concentrations within the moss samples are higher than within the ecosystem as whole therefore, accuracy of measurements is improved. It is apparent that air pollution levels change rapidly over time. The use of moss as biomonitors has the capability to monitor and detect external conditions averaged over certain periods of time. Moss is a renowned and respected species to use a biomonitor as it is common to various areas across British Columbia, it is available for sampling during the field season, and it is relatively tolerant to pollutants (Berg and Steinnes 1997).

My primary objective was to provide insight into the environmental pollution caused by mines outside of their boundaries; I sought to compare data collected in two different sites, at two different distances from the mine boundary. The analysis of my results and the depth of my study was limited by time, my ability to access potential study sites, a small sample size, and ultimately restrictions imposed by the ongoing global pandemic, but overall, my data provides a foundation for a more detailed study on the environmental impacts of mines outside their boundaries. I feel biomonitoring studies lack a standardized protocol for sampling, sample preparation, and elemental analysis. Without a standardized method it is difficult to obtain comparable results. It would be interesting to conduct more studies combining the use of dustfall monitoring and biomonitoring. Soils often contain quantities of heavy metals as well and surely through soil dusting, soils may deposit on moss. In order to account for heavy metal deposition via soil dusting, biomonitoring in combination with soil analysis could be performed. It would be beneficial to further understand what mean levels of heavy metals moss samples contain to maintain themselves. As aforementioned, moss species require certain quantities of metal elements for growth, development, and reproduction (Berg and Steinnes 1997, DalCorso et al. 2014). With the knowledge of those quantities, calculations could account for them and subtract them from total concentration values.

Currently, the environmental impact of mines outside of their boundaries is poorly studied. The majority of the literature focuses on how moss can be used as biomonitors but does not delve deep into the importance of biomonitoring in Natural Resource Management. This research will contribute to bridging the knowledge gap of the air pollution effects outside mine boundaries, with emphasis on determining whether there are any heavy metals present in moss samples in those areas. It is known that bioindicators are a useful tool when assessing air quality; biomonitoring could be an effective and economical alternative to investigating trace-element atmospheric pollution. The findings of this project provide important information about the impact mines have on the environment surrounding their boundaries and will hopefully increase the frequency in which natural resource managers use moss as bioindicators. It is my hope that moving forward management strategies and policies will reflect the goal of decreasing air pollution caused by heavy metal mines.

LITERATURE CITED

Aboal JR, Fernández JA, Boquete T, Carballeira A. 2010. Is it possible to estimate atmospheric deposition of heavy metals by analysis of terrestrial mosses? Science of the Total Environment [Internet]. [cited 2020 Oct 2]. 408:6291–6297. Available from: https://www.sciencedirect.com /science/article/pii/S0048969710009745 doi: https://doi.org/10.1016/j.scitotenv.2010.09.013

Angelovska S, Stafilov T, Šajn R, Balabanova B. 2016. Geogenic and Anthropogenic Moss Responsiveness to Element Distribution Around Pb–Zn Mine, Toranica, Republic of Macedonia. Archives of Environmental Contamination and Toxicology [Internet]. [cited 2020 Oct 2]. 70: 487-505. Available from: https://link.springer.com/content/pdf /10.1007/s00244-015-0251-7.pdf doi: https://doi.org/10.1007/s00244-015-0251-7

Associated Environmental [Internet]. n.d. Environmental Impact Assessment, Highland Valley Copper Mine. [cited 2020 Nov 20]. Available from: https://www.ae.ca/projects/details/ environmental-impact-assessment-highland-valley-copper-mine

Baumbach, G. and Vogt, U. 2003. Influence of Inversion Layers on the Distribution of Air Pollutants in Urban Areas. Water, Air, & Soil Pollution: Focus. 3(5):67-78. doi: 10.1023/A:1026098305581

BC Mine Information [Internet]. n.d. Highland Valley Copper (HVC) Mine Overview. [cited 2020 Nov 20]. Available from: https://mines.nrs.gov.bc.ca/p/highland-valley-copper-hvc/overview

Berg T, Steinnes E. 1997. Uses of mosses (*Hylocomium splendens* and *Pleurozium schredberi*) as biomonitors of heavy metal depositions: from relative to absolute deposition values. Environmental Pollution [Internet]. [cited 2020 Oct 4]. Vol. 98, Issue 1, pg. 61-71. Available from: https://www.sciencedirect.com/science/article/pii/S0269749197001036 doi: https://doi.org/10.1016/S0269-7491(97)00103-6

Chakrabortty, S., Paratkar, G. T., 2006: Biomonitoring of trace element air pollution using mosses. Aerosol and Air Quality Research 6, 247–258.

Choudhury, S., Panda S. K., 2005: Toxic effects, oxidative stress and ultrastructural changes in moss *Taxithelium nepalense* (Schwaegr.) Broth. under chromium and lead phytotoxicity. Water, Air, Soil Pollution 167, 73–90

Clark, M., Douglas, M., Choi, J. [Internet]. 2018. Biology 2e. [cited 2020 Nov 29]. Houston, Texas. Available from: https://chromoscience.com/2020/07/25/biodiversity-of-plants/

Couto, J.A., Fernandez, J., Aboal, J.R., and Carballeira, A. (2004). Active Biomonitorng of Element Uptake with Terrestrial Mosses: A Comparison of Bulk and Dry Deposition. Sci. Total Environ. 324: 211-222.

Dalcorso, G., Manara, A., Piasentin, S., Furini, A. 2014. Nutrient metal elements in plants. Metallomics. 6(10): 1770-1788. Doi:https://doi.org/10.1039/C4MT00173G.

Degola, F. Benedictis, M., Petraglia, A., Massimi, A., Fattorini, L., Sorbo, S., Basile, A., Sanità di Toppi, L. 2014. A Cd/Fe/Znresponsive phytochelatin synthase is constitutively present in the ancient liverwort *Lunularia cruciata* (L.) Dumort. Plant and Cell Physiology 55, 1884–1891

Elvira, NJ., Medina, NG., Leo, M., Cala, V., Estébanez, B. 2020. Copper Content and Resistance Mechanisms in the Terrestrial Moss *Ptychostomum capillare*: A Case Study in an Abandoned Copper Mine in Central Spain. Archives of Environmental Contamination and Toxicology. 79: 49-59. Doi: https://doi.org/10.1007/s00244-020-00739-6

[Enviro. Canada] Environment Canada. 2020. Hourly Data Report for July 10, 2020. [cited 2020 Nov 24]. Available from: https://climate.weather.gc.ca/climate_data/hourly_data_e.html ?hlyRange=2011-05-15%7C2020-11-26&dlyRange=2011-05-03%7C2020-11-26& mly Range=% 7C&StationID=49408&Prov=BC&urlExtension=e.html&searchType=stn Name&optLimit=specDate&StartYear=1840&EndYear=2020&selRowPerPage=25&Lin e=0&searchMethod=contains&Month=7&Day=10&txtStationName=Merritt&timeframe =1&Year=2020

Fath, D. and Killian, M. 2007. The relevance of ecological pyramids in community assemblages. Eco.Model, 208(2-4):286-294.

Fernández, JA. Boquete, A., Carballeira, Aboal, JR. 2015. A critical review of protocols for moss biomonitoring of atmospheric deposition: Sampling and sample preparation. Science of The Total Environment. 517: 132-150. Doi: https://doi.org/10.1016 /j.scitotenv.2015.02.050.

Hoffman, H., Phyliky, R., Fleming, R. 1988. Zinc-Induced Copper Deficiency. Gastroenterology. 94: 508-12. Available from: https://www.gastrojournal.org/article/ 0016-5085(88)90445-3/pdf#:~:text=High%20dietary%20zinc%20intakes%20depress, copper%20can%20depress%20zinc%20absorption.

Gaston, K. J. and Spicer, J. I. 2004. Biodiversity: An Introduction, Blackwell Publishing, Oxford.

Gardner, T. 2010. Monitoring Forest Biodiversity: Improving Conservation Through Ecologically Responsible Management. Routledge.

[Gov. Canada] Government of Canada [Internet]. 2013. Environmental Code of Practice for Metal Mines. [cited 2020 Nov 26]. Available from: http://publications.gc.ca/site/fra /345934/publication.html

Govindapyari H, Leleeka M, Nivedita M, Uniyal PL. 2010. Bryophytes: indicators and monitoring agents of pollution. NeBIO [Internet]. [cited 2020 Oct 4]. 1(1): 35-41. Available from: https://www.anderson5.net/cms/lib02/SC01001931/Centricity/Domain /1901/byrophytes%20article.pdf iMap BC. 2020. B.C. Conservation Data Centre: CDC iMap [web application]. [cited 2020 Nov 23]. Victoria, British Columbia, Canada. Available from: http://maps.gov.bc.ca/ess/sv/cdc/

Jarsjö, J., Chalov, SR., Pietroń, J., Alekseenko, AV., Thorslund, J. 2017. Patterns of soil contamination, erosion and river loading of metals in a gold mining region of northern Mongolia. Regional Environmental Change. 17: 1991-2005. Doi: 10.1007/s10113-017-1169-6

Kim, JE., Han, YJ., Kim, PR., Holsen, T. 2012. Factors influencing atmospheric wet deposition of trace elements in rural Korea. 116: 185-194.

Koz, B., Cevik, U., Akbulut, S. 2012. Heavy Metal analysis around Murgul (Artvin) copper mining area of Turkey using moss and soil. Ecological Indicators, 20: 17-23. doi: https://doi.org/10.1016 /j.ecolind.2010.02.002

Lee Jr, R. E., Von Lehmden, D. J. 1973. Trace metal pollution in the environment. Journal of the Air Pollution Control Association 23, 853–857.

Maevskaya, S. et al. 2001. Absorption of cadmium and lead ions by gametophyte of the moss *Plagiomnium undulatum*. Russian Journal of Plant Physiology 48, 820–824.

Meerabai, G. and Pullaiah, T. 2016. Plant Biodiversity Conservation and Management. Daya Publishing House.

Meidinger, D. and Pojar, J. 1991a. Ecosystems of British Columbia, Chapter 10: Interior Douglas-fir Zone. British Columbia Ministry of forests. 330 pp. Available from: http://www.for.gov.bc.ca/hfd/pubs/Docs/Srs/SRseries. htm

Meidinger, D. and Pojar, J. 1991b. Ecosystems of British Columbia, Chapter 12: Montane Spruce Zone. British Columbia Ministry of forests. 330 pp. Available from: http://www.for.gov.bc.ca/hfd/pubs/Docs/Srs/SRseries. htm

Onianwa, P. C. 2001. Monitoring atmospheric metal pollution: a review of the use of mosses as indicators. Environmental Monitoring and Assessment 71, 13–50.

Pennsylvania State University [Internet]. 2020. Threats to Biodiversity. [cited 2020 Oct 30]. Available from: https://www.e-education.psu.edu/geog30/node/394

Rinne, JK., Barclay – Estrup, P. 1980. Heavy Metals in a Feather Moss, *Pleurozium* schreberi, and in Soils in NW Ontario, Canada. Oikos, 34(1): 59-67. Doi:https://doi.org/10.2307/3544550

Ross, H.B. 1990. On the Use of the Mosses *Hylocomium splendens* and *Pleurozium schreberi* for Estimating Atmospheric Trace Metal Deposition. Water, Air and Soil Poll. 50: 63-76.

Ryan MW. n.d. Mosses and Liverworts of the Sicamous Research Area.

Saxena DK, Harinder. 2004. Uses of Bryophytes. Resonance [Internet]. [cited 2020 Oct 4]. 9: 56-65. Available from: https://link.springer.com/content/pdf/10.1007/BF02839221.pdf doi: https://doi.org/10.1007/BF02839221

Schofield WB. 1927. Introduction to Bryology. New York (NY): Macmillan Publishing Company.

Stanković, J. et al. 2018. Bryophytes and heavy metals: a review. Acta Bot. Croat. 77 (2), 109–118. DOI: 10.2478/botcro-2018-0014

Teck Resources Limited [Internet]. 2019. Highland Valley Copper. Logan Lake, BC: Teck Resources Limited; [cited 2020 Oct 30]. Available from: https://www.teck.com/operations /canada/operations/highland-valley-copper/

Trebilco, R., Baum, JK., Salomon, AK., Dulvy, NK. 2013. [Internet]. Ecosystem ecology: size-based constraints on the pyramids of life. [cited 2020 Oct 30]. Available from: https://www.sciencedirect.com/science/article /pii/S016953471300 0888?casa_token=0sA6JS7tRcYAAAAA:StxothG8DhyxqiOwKrdK aTA5rEMMlmNT-sxQzUqSL76x6vgcPJXmYHZmLR83 wwu_lFtfXZ7ILg

[Uni. Of Roch. Med. Center] Univeristy of Rochester Medical Center. 2020. [Internet]. Total Copper (Blood). Health Encyclopedia. [cited 2020 Nov 27]. Available from: https://www.urmc.rochester.edu/encyclopedia/content.aspx?contenttypeid=167& contentid=total_copper_blood#:~:text=Copper%20deficiency%20can%20lead%20to,much%20copper%20can%20be%20toxic.

Zvereva, E. L., Kozlov, M. V., 2011: Impacts of industrial polluters on bryophytes: a meta-analysis of observational studies. Water, Air & Soil Pollution 218, 573–586.

Appendix A.

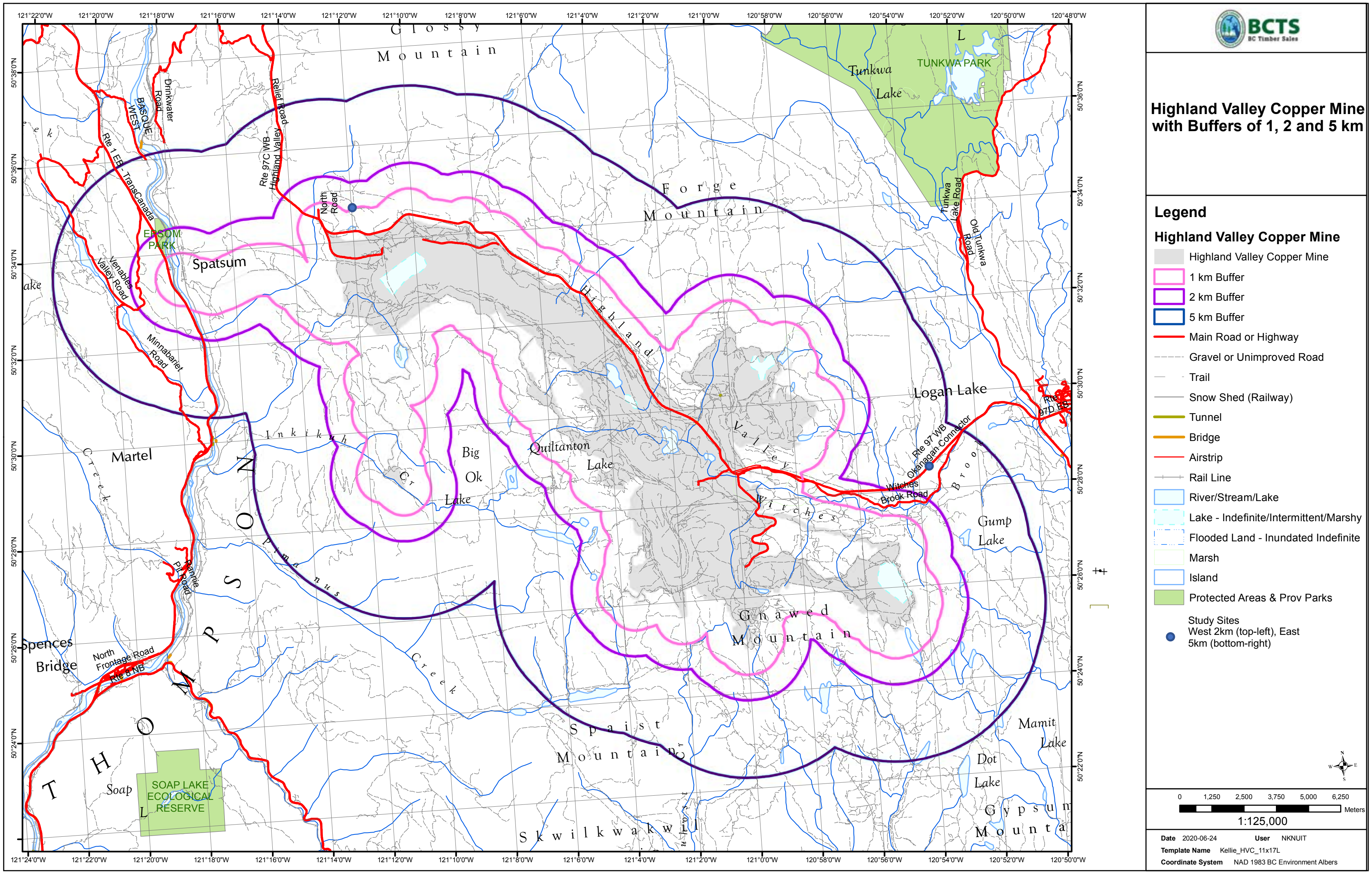

Figure 4. Map outlining the boundary of Highland Valley Copper Mine with 1km, 2km, and 5km radii overlaid. Potential access also outlined. The map was created and provided by Nikki Knuit, a GIS Technician with BC Timber Sales. Blue dots were added to display site locations.

APPENDIX B.

Table 3. Site classification information collected for the East 5km site, and the West 2km site.

General Features	**East 5 km**	**West 2 km**
BEC zone/subzone/variant	IDFdk1	IDFdk1/MSxk
Elevation (m)	1200	1425
Slope Gradient (%)	0	0
Aspect	-	-
Slope Position	Flat	Flat
Slope Shape	Straight	Straight
Soil Features		
Soil Texture	Loamy	Silty
Seepage or Ground Water Table	No	Yes
Gleyed Horizon	No	Unknown
Flooding	No	No
Soil Colour	Medium	Medium
A Horizon/Thickness (cm)	Ah/1cm	Ah/2cm
Humus Form/Thickness (cm)	Mor-Moder/5cm	Moder/3cm
Soil Order	Brunisol	Brunisol (Gleysol)

Table 4. Vegetation data collected for the East 5km site and the West 2km site. Data provided in percent cover.

Trees (% Cover)	**East 5 km**	**West 2 km**
Pinus contorta var. *latifolia*	25	5
Pseudotsuga menziesii var. *glauca*	2	30
Shrubs (% Cover)		
Arctostaphylos uva-ursi		4
Populus tremuloides	<1	
Rosa acicularis	3	1
Shepherdia canadensis	3	7
Vaccinium scoparium		1
Spiraea betulifolia		1
Juniperus communis		<1
Herbs/Grasses (% Cover)		
Calamagrostis rubescens	45	30
Arinca cordifolia	2	1
Fragaria virginiana		1
Aster consicuus		3
Epilobium angistifolium		1
Linnaea borealis	5	
Lathyrus nuttalli	1	
Mosses/Lichens (% Cover)		
Pleurozium shreberi	7	15

APPENDIX C.

Table 5. Summary of moss samples identified to Genus and percent coverage along a 10m transect within a 5km radius East of the mine. Data collected on July 10th, 2020.

Sample Name	Genus	Percent Cover (%)
E-5km-1m	*Pleurozium spp.*	80
E-5km-2m	*Brachythecium spp.*	12
E-5km-3m	*Pleurozium spp.*	80
E-5km-4m	*Pleurozium spp.*	20
E-5km-5m	*Pleurozium spp.*	30
E-5km-6m	*Pleurozium spp.*	50
E-5km-7m	*Pleurozium spp.*	80
E-5km-8m	*Pleurozium spp.*	80
E-5km-9m	*Pleurozium spp.*	80
E-5km-10m	*Pleurozium spp.*	70

Table 6. Summary of moss samples identified to Genus and percent coverage along a 10m transect within a 2km radius West of the mine. Data collected on July 10th, 2020.

Sample Name	Genus	Percent Cover (%)
W-2km-1m	*Pohlia spp.* and *Dicranum spp.*	80
W-2km -2m	*Brachythecium spp.*	12
W-2km -3m	*Pleurozium spp.*	80
W-2km -4m	*Pleurozium spp.*	20
W-2km -5m	*Pleurozium spp.* and *Dicranum spp.*	30
W-2km -6m	*Pleurozium spp.* and *Dicranum spp.*	50
W-2km -7m	*Brachythecium spp.*	80
W-2km -8m	*Pleurozium spp.*	80
W-2km -9m	*Pleurozium spp.*	80
W-2km -10m	*Pleurozium spp.* and *Dicranum spp.*	70