Western Honeybee and Honey as Biomonitor for Urban Metal Contamination:
with Case Study in Metro Vancouver, British Columbia, Canada

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Abstract

Due to the global trend of urbanization, environmental contamination has become a critical issue globally. Metal contamination of the soil-water system mainly results in disturbances in environmental health and biotic functions. Insect biomonitoring has become a valuable environmental assessment tool that quantifies the impacts of soil and water contamination. Western honeybee (*Apis mellifera*) is an ideal species commonly used in urban metal biomonitoring. Honey, the natural product, also serves as a food source for human consumption. High accessibility, global distribution, species diversity, and intense interactions with the environment are factors that contribute to the effectiveness of western honeybee and honey as the urban metal biomonitor. Metro Vancouver is selected as a case study of the urban system that encompasses a wide range of habitats. The concentrations of antimony (Sb), lead (Pb), copper (Cu), chromium (Cr), and rubidium (Rb) in honey and honeybee samples from a total of 28 study sites across Metro Vancouver are retrieved from the database of Smith et al. (2019) and Smith & Weis (2020). Enrichment factor (EF) and Honeybee Contamination Index (HCI) are two biomonitoring indicators used for metal contamination assessment. The results show that most study sites of Metro Vancouver are characterized by relatively insignificant anthropogenic metal deposition and high metal contamination. Based on EF values, significant metal enrichment only exists at 3 study sites (YVR Airport for Cr, E Cordova St, Downtown Eastside for Pb and Powell St, Downtown Eastside for Pb), with the enrichment of Cu and Cr lower than Sb and Pb. HCI values indicate that Cu is the least significant contaminant in Metro Vancouver in regards to the number of study sites with high metal contamination levels. Only two study sites (Alaksen National Wildlife Area and Terra Nova Rural Park) show low or intermediate metal contamination levels for all five metals. Overall, the anthropogenic metal input and contamination level of conservation and recreation land use are less significant than industrial and general urban land uses. The results of the case study are consistent with the outcomes of other relevant research. Metal concentrations in honey and honeybee samples are able to reveal the spatial variations of metal contamination in the surrounding environment. The indicators of EF and HCI are also with high applicability for extrapolation. Therefore, the method applied in this case study is found to be feasible among different regions, and further research is expected with more comprehensive spatial and temporal analysis.

1. Introduction

1.1 Urban System

The global trend of urbanization has led to rapid population growth and extensive changes in land uses and landscapes. As a result, a massive amount of people has immigrated from rural areas to cities. Currently, about 55% of the world population lives in urban environments, which has increased by 25% since 1950 (United Nations Department of Economic and Social Affairs, 2019). In terms of regional geography, the term urban system defines the region that is composed of cities, which was firstly used in 1960 for the establishment of metropolitan organizations in the United States (Simmons, 1981). Urban systems are usually characterized by the relatively large population size, which contains well-organized structures (also called functional regions) including tertiary services or laboursheds emerge in areas of various densities. Within an urban system, the interaction and specialization among different functional regions or cities are critical (Simmons, 1981). For instance, how intense different regions interact in an urban system, how does each city link with each other, as well as what is the structure for a specific functional region, can all become important subjects in the research of urban systems.

The transformation from natural ecosystems to urban systems can primarily modify the biotic and abiotic environmental factors. The land structure in urban systems is highly heterogeneous with different scales, and isolation of structures may happen spatially. Urban systems retain high degrees of anthropogenic impacts. Enhanced human disturbances can affect species composition and biodiversity by introducing invasive groups and causing endangered species. Meanwhile, management practices in urban systems have effectively lowered the frequency of natural disturbances. The changes in landscapes can also influence regional climates. The concept of heat island suggests higher city temperature in comparison with surrounding rural or terrestrial areas. Lastly, the construction of an urban system inevitably affects the community conditions for animal and plant growth, which may eventually alter the ecological successions (Elmqvist, Alfsen & Colding, 2008).

The urban system is associated with an extensive range of habitats, from the least disturbed habitats, such as lakes, mountains and Canadian national parks, to the most disturbed regions, including residential areas, roads and industrial centres. The habitats in urban systems are influenced by not only ecological but also socio-economic factors. Different types of landscapes and habitats within urban systems reveal the significance of ecological fragmentation, which may create challenges and risks for organisms, especially those with poor dispersion (Elmqvist, Alfsen & Colding, 2008).

1.2 Environmental contamination

Environmental contamination is a critical issue affecting people's goals for the achievement of sustainability. Along with global development and urbanization, anthropogenic disturbances have led to a range of environmental changes in many fields such as abiotic conditions and biological functions in soil and water contexts. Since past decades, environmental contamination has become a significant issue that raises concerns in ecological sustainability, human health and socio-economic values (Azam et al., 2015). Environmental contaminants can be released from various land uses from both point and non-point sources. Some chemical contaminants, such as hydrocarbons, radioactive elements, and metals, are likely to persist in ecosystems once entered and cause cumulative effects within the food webs. As a result, threats are posed to human health by contaminating the global food safety and potable water supply (Thompson & Darwish, 2019).

Anthropogenic contaminants are classified into two major categories, which are biodegradable and conservative substances (Conti, Mecozzi, Alimonti, Mattei & Iacobucci, 2008). Biodegradable contaminants are mainly substances containing organic matters that can be broken down by microbial decomposition. Compared with conservative substances, biodegradable contaminants can be more easily removed by bioremediation, as well as physical and chemical removal (Bahadori, 2020). Most of the biodegradable contaminants consist of high amounts of nitrogen, phosphorous, and carbon, which can be decomposed by either aerobic or anaerobic reactions, depending on the environmental conditions (Conti, Mecozzi, Alimonti, Mattei & Iacobucci, 2008). Conservative substances are contaminants that can hardly be decomposed, sustaining a significant residence period in the ecosystems. This type of contaminants includes metals, radioactive substances and chemicals like DDT (an insecticide), PCBs (an organic chlorine compound), and halogenated hydrocarbons (Conti, Mecozzi, Alimonti, Mattei & Iacobucci, 2008). In general, conservative substances cannot be treated by natural processes, and anthropogenic treatments are required (Bahadori, 2020). Some conservative substances, including metals and halogenated hydrocarbons, tend to leave long-lasting effects by accumulating in body tissues of species Conti, Mecozzi, Alimonti, Mattei & Iacobucci, 2008).

Contaminants with low concentration may leave insignificant impacts on the environment. However, almost every contaminant retains a threshold for hindering ecological functions and species development (Conti, Mecozzi, Alimonti, Mattei & Iacobucci, 2008). Examples of contaminants affecting the health of the environment include metals released from urbanization (e.g. Pb, Cd and Zn), chemical substances from the excess application of nutrients, pesticides in agriculture (e.g. N, P and DDT), as well as greenhouse gas emissions (e.g. CO_2 and CH_4) from the industry.

Metal Contaminants

Metal contamination is a concern all around the world, considering its high input levels and toxic effects around regions with significant industrial and urban activities. Metal contaminants can come from both natural inputs and anthropogenic disturbances, and the restoration of soils with metal contamination is difficult (Shakoor et al., 2013). The inputs of metal contaminants from anthropogenic activities are currently considered to be more significant than those natural processes, such as rock weathering and volcanic eruptions (Girgin, Kazanci, & Dügel, 2010). Metals such as lead, nickel, chromium, and cadmium are released during the cultivation of sugar cane due to the use of fertilizers (Corbi et al., 2010). Additionally, the disposal of industrial wastewater increases the concentrations of metal elements in the receiving water bodies. Urbanization, such as the construction of buildings, roads, and railways, also allows metal contamination in surrounding soil and water environments (Girgin, Kazanci, & Dügel, 2010).

Excessive metal concentrations in the environment can lead to toxic effects on environmental processes and living organisms. Metal contaminants in sediments can be transferred to higher trophic levels by absorption and ingestion (Corbi et al., 2010). Severe metal contamination can cause lethal effects to the soil and aquatic organisms by decreasing the population size, disturbing the spatial distributions, and lowering the intensity of biological activity. For instance, soil Pb contamination may interfere with some essential functions of plants, such as photosynthesis and cell division. Human exposure happens while Pb contaminated crops are consumed, considering the implication of the food chain. Pb does not serve as an essential nutrient, and human bodies do not metabolize it. By persisting in body tissues, Pb may induce health risks, such as immune suppression, malnutrition, and upper gastrointestinal cancer (Jiwan & Ajay, 2011).

Other Examples of Contaminants

In addition to urban metal inputs, agricultural and industrial productions can also cause the releasing of contaminants from the uses of fertilizer and pesticide, as well as the releasing of greenhouse gases. The application of fertilizers in agricultural activities increases the concentrations of nutrients (e.g. N, P and K) in soils, which enter water bodies by leaching and surface runoff processes. Excess nutrients in aquatic systems can cause eutrophication and disturb the essential functions of aquatic organisms. The utilization of pesticides may release persistent chemicals, such as DDT, into the soil and surrounding aquatic systems. Pesticide residuals are one of the major contaminants for soil, water and vegetation. Pesticides can cause acute and chronic toxicity for non-target species such as beneficial insects and aquatic species. The toxicity varies depending on the physical and chemical properties of pesticides applied, as well as the species of exposure (Aktar, Sengupta & Chowdhury, 2009). Industrial activities can result in significant emissions of greenhouse gases such as nitrous oxide, carbon dioxide and ozone, resulting in major impacts like climate change. The atmospheric temperature may increase as high greenhouse gas concentrations intensify the absorption and trapping of solar radiation, called the “greenhouse effect” (Earth Science Communications Team, 2020).

1.3 Biomonitoring

Biomonitoring is a valuable and accessible tool to assess the environmental contamination level and biological impacts. It monitors the early diagnosis, toxic influences, as well as synergetic and antagonistic impacts of different contaminants on organisms (e.g. plants, animals, and humans) (Parmar, Rawtani & Agrawal, 2016). Biomonitoring consists of three major categories, which are environmental monitoring, ecological monitoring and biodiversity monitoring (McGeoch, 2007). Environmental monitoring relies on the species whose responses to environmental changes are measurable and predictable. Ecological monitoring mainly quantifies the impacts of environmental disturbances on the biosphere and biological communities. Instead of specific species, biodiversity monitoring utilizes taxa or functional groups to track the diversity and the health status of habitats. Biomonitoring systems utilize the biological characteristics of specific organisms to provide quantitative information by measuring the response of an organism to the exposure to environmental changes (Ponikvar, Šnajder & Sedej, 2005).

Biomonitor is the taxa applied to detect environmental changes due to contaminant inputs and indicates the impacts on the presence and biodiversity of the community. The biomonitoring implication of different species varies depending on their ecological variations and environmental resistance (Parmar, Rawtani & Agrawal, 2016). The selection of species that are suitable for biomonitoring should refer to several factors, including abundance and distribution, sensitivity and responding ability, well-studied characteristics, as well as ecological and commercial values (Holt & Miller, 2010). Species applied as biomonitor should have high abundance and wide distribution within the research scope, and the abundance and distribution should be relatively stable under variation of climate and environmental conditions. Biomonitor should be sensitive to changes in the surrounding environment without lethal or accumulating effects from contaminants. The response should be measured based on the level of disturbances, and the reaction of the biomonitor should be consistent with that of the entire community. The history and biological characteristics of biomonitoring species should be well-documented with relatively consistent taxonomic. Species that are inexpensive and simple for study are optimal. Species that generate significant ecological and commercial values, such as being applied for producing activities, are often with high public awareness (Holt & Miller, 2010).

2. Background: Insect Biomonitoring

During the decades of biomonitoring development, precedent studies have found insect to be one of the most effective biomonitoring groups for environmental conservation. Insects are largely applied as biomonitor in enormous research, such as soil pollution and water quality, because of their abundance and species diversity, as well as the essential functions they provide within the ecosystem (McGeoch, 2007).

2.1 Abundance and Biodiversity

Class Insecta (insects) is the largest group of Arthropoda phylum with extensively high richness and diversity (Rocha et al., 2010). So far, approximately 1,000,000 insect species have been identified and described, which constitutes more than 70% of the total known animal species (Wigglesworth, 2020). Even so, there still exist millions of insect species globally that have not been discovered. The large number and diversity of insects mainly result from their high reproductive rates and remarkable adaptability to habitats, which lead to higher accessibility in target ecosystems (Meyer, 2007). The high diversity of insect species also contributes to measurable demographic or behavioural variations. Based on their biological niches, different insect species can be utilized to study specific habitats or the degree of environmental disturbances (Rocha et al., 2010). Insects appear in almost all types of habitats, which range from terrestrial, desert, urban ecosystems to surface water, glaciers, and brackish environments (Meyer, 2007). The small size of insects also makes transportation and sample processing easier (Rocha et al., 2010).

2.2 Ecological Values

Insects work as the indispensable component in many environmental processes in almost all ecosystems, which generate significant ecological and economic values. The understanding of the correlation between insects and ecosystems is necessary for the biomonitoring of future environmental management and sustainability (Rocha et al., 2010). Here, decomposition and pollination are introduced as two representative environmental services of insects.

Decomposition

Insects are found to have significant impacts on ecological nutrient dynamics by adding biomass directly and contributing to the microbial decomposition. Yang & Gratton (2014) suggests that the total biomass of insect accounts for a significant proportion of the total biomass inputs into the ecological nutrient pool in many systems. For example, the biomass input of 13 and 17-year periodical cicadas in North America forest systems is considerable, which is beneficial to ecological functions like microbial distribution and nitrogen mineralization (Yang & Gratton, 2014). In addition, insects also play essential roles in soil decomposition processes by affecting both biotic and abiotic factors. Insects' living activities and interaction with other species help arrange the distribution of microorganisms such as bacteria. Sites with higher microbial abundance are likely to retain a higher decomposition rate. Meanwhile, insect mobilization and nesting underground will increase soil porosity by forming pores and channels. This soil structure is associated with a better-exchanging capacity of air and higher moisture retention (Meyer, 2007). The changes in abiotic factors can impact nutrient cycling by altering the microbial decomposition rate.

Pollination

Insects' pollination is an essential ecological service in many terrestrial systems as flowering plants serve as significant primary producers. The majority of flowering plants are not capable of asexual reproduction but mainly depend on pollination (Gill et al., 2016). In this case, insects work as the intermediate to support the pollen exchange between flowering plants so that the symbiotic relationship forms. Different plant species may exhibit unique colours, odours, or shapes to attract specific insect groups. In addition to butterflies and bees, insects like moths and beetles are also significant pollinators in terrestrial environments (Wigglesworth, 2020). Ecologically, insect pollination helps maintain the genetic diversity of flowering plants by transferring pollens among different species. It also contributes to good-quality fruit production and increased seed fertility (Gill et al., 2016). Meanwhile, insect pollination is a critical component in agriculture. In total, approximately 75% of crop production relies on insect pollination. Fruit and vegetable production may decrease by 40% and 16% without insect pollination (Gill et al., 2016). Insect pollination also contributes to essential lipids and micronutrients in diets. In future, higher insect pollination will be required due to the increasing demands of food security, resulting from population growth and environmental changes (Gill et al., 2016).

2.3 Aquatic & Terrestrial Insects

Compared with other taxa, invertebrates are associated with quicker and clearer responses under the changes in environmental conditions. Insects, the major group of invertebrates, also retain a short responding period in comparison to other animal species under stress posed by other species or human activities (Rocha et al., 2010). As the increasing urbanization and resource development cause increased amounts of contaminants into the environments, aquatic and terrestrial insects have been developed and studied as effective biomonitors in the research of soil and water contexts (Bargańska, Ślebioda & Namieśnik, 2016).

Aquatic Insects

Aquatic insects are the most sensitive and vulnerable to water contamination (Girgin, Kazanci & Dügel 2010). Aquatic communities were considered as a component for monitoring the approach of water quality since the 1900s (Bonada et al., 2006). Metal contamination in surface water systems has become a critical issue, as anthropogenic contaminant inputs have currently become the dominant metal disposal (Girgin, Kazanci & Dügel 2010). Metal contaminant accumulation in freshwater can be further intensified by the accumulation in sediment particles, which allows more metals transferred into higher trophic levels of the aquatic community by adsorption and direct consumption (Corbi et al. 2010).

Aquatic insects are a commonly used insect group for current freshwater biomonitoring systems. While chemical and physical testing can only assess the contamination level at the sampling time, the use of aquatic insect monitors the cumulative effects of contamination, which contributes to the temporal discussion (Holt & Miller, 2010). Meanwhile, the capacity of aquatic insects and benthic invertebrates to reveal the contaminating sources also allows people to identify the thresholds of ecological restoration and the limit of environmental stress (Bonada et al., 2006). Ephemeroptera, Plecoptera, and Trichoptera are three insect groups that are found to be the most abundant and effective in revealing water quality. Many insect species, such as *Platycnemis pennipes* (Trichopteran) and *Baetis fuscatus* (Ephemeropteran), retain a positive correlation with metal concentrations in water (Girgin, Kazanci & Dügel, 2010).

The development of aquatic insect biomonitoring has allowed monitoring methods to be differentiated depending on regional environmental conditions and specific monitoring requirements. For instance, each type of disturbance may release different contaminants into the ecosystems. Different management objectives can also have specific requirements for biomonitoring, such as the precision of results and the frequency of monitoring. Besides, the method should be modified to adapt to the structure of ecosystems. Ecosystems with more complex structures may maintain different species interactions from ecosystems with simple structures (Bonada et al., 2006).

Terrestrial Insects

In addition to aquatic insects, the terrestrial insect is another significant invertebrate group that is applied in contamination biomonitoring. Terrestrial ecosystems support a larger number of insect species than aquatic ecosystems with various abundance and diversity. The biomonitoring system used in the terrestrial environment is more complex and diverse so that the contamination is more difficult to be quantified (McGeoch, 1998). Gerlach, Samways & Pryke (2013) reviewed a range of terrestrial taxonomic groups that can be used as biomonitor, depending on their sensitivity, biological characteristics and geographical distributions. In biomonitoring, however, one taxonomic group may not be sufficient, and it is more reliable to include several insect taxa. The fluctuations of insect abundance because of life histories and biological interactions should be distinguished against population size changes due to environmental disturbances. Also, to achieve the most accurate and precise results, the correlation between insect response and environmental changes should be identified quantitively (Gerlach, Samways & Pryke, 2013). In the following, the beetle is briefly introduced as a representative species used for terrestrial biomonitoring.

The beetle serves as the arthropod species that are more commonly used in terrestrial biomonitoring systems. Beetles are found to maintain a wide distribution in many types of habitats in the terrestrial environment. Similar to other biomonitoring species, beetles are very sensitive to environmental changes and disturbances. Also, the sampling method for beetle analysis is relatively easy and inexpensive. Therefore, beetles are considered an effective biomonitor in the monitoring of a variety of ecological parameters (Ghannem, Touaylia & Boumaiza, 2018).

In the research of Rainio & Niemelä (2003), carabid beetles are examined as a reliable beetle species to detect environmental contaminations. The studies of carabid beetles mainly concentrate on temperate regions. It suggests that carabid beetles are widespread; they distribute among almost all types of habitats besides environments with extremely low moisture levels. Carabid beetles have been successfully applied for environmental monitoring, such as forest fragmentation, insecticide contamination and management approaches. In general, the population size and distribution of carabid beetles can be affected by vegetation variation, moisture content, and soil fragmentation. However, the effectiveness of using carabid beetles for biodiversity monitoring remains uncertain, and further research is expected (Rainio & Niemelä, 2003).

Beetles are also valuable biomonitors for metal contamination. Metal contamination tends to reduce the richness and diversity of beetle species. The beetle may be more vulnerable to other disturbances with exposure to toxic metals, and the reproduction rate will also decrease. Specific metal contaminants (e.g. Pb, Zn) can influence the physiology of beetles, such as body size and wing length. However, the responses of each beetle species may vary under environmental changes. The suitability of different beetle species may vary based on specific ecological parameters and managing principles (Ghannem, Touaylia & Boumaiza, 2018).

Although many insect species are determined to be good biomonitor, the western honeybee (*Apis mellifera*) has been documented to be particularly effective in the biomonitoring of urban metal contamination. A. *mellifera* occurs widely geographically with high biodiversity (Han, Wallberg & Webster, 2012). Honeybees form colonies and maintains active biological activities throughout the year (Seeley, 2019). The interactions between western honeybees and the environment make it closely related to anthropogenic disturbances. The intensified metal contamination in the environment tends to increase the metal contaminant level in not only the honeybee but also honey, a food source for human consumption (Bargańska, Ślebioda & Namieśnik, 2016). As a result, the honeybee and honey are selected as the major focus in this research for metal contamination biomonitoring.

3. Objective

This research aims to discuss the implication of the western honeybee and honey as an urban metal biomonitor by explaining their biological characteristics and geographical distributions that contributes to the biomonitoring effectiveness. This report includes Metro Vancouver, British Columbia, Canada as a case study. The anthropogenic metal inputs and metal contamination pattern of antimony (Sb), lead (Pb), copper (Cu), chromium (Cr), and rubidium (Rb) in Metro Vancouver are assessed with metal concentrations in honey and honeybee tissue using enrichment factor (EF) and Honeybee Contamination Index (HCI) as indicators.

4. Method

Literature was reviewed comprehensively to introduce western honeybee as the urban metal biomonitor. The year-round biological activeness, global distribution and high biodiversity, as well as the interaction between honeybee and the environment, were reviewed and discussed as major characteristics that contributed to effective biomonitoring.

Metro Vancouver was selected as the case study that analyzes the metal biomonitoring honey and the honeybee with Sb, Pb, Cu, Cr, and Rb as the metal of focus. The metal concentration in honey and the honeybee samples were retrieved from the research of Smith et al. (2019) and Smith & Weis (2020). Samples of honey were collected from a total of 28 study sites across Metro Vancouver (Figure 1). Honeybee tissue was only sampled from 13 of the 28 sites. Sample collection and processing procedures can refer to Smith et al. (2019) and Smith & Weis (2020) for more details. The data of metal concentration was not available due to the lack of soil survey for Metro Vancouver. Soil metal contamination, in this case, was assessed using regional land uses in Metro Vancouver. It assumed that areas with more significant anthropogenic and industrial activities tended to have more urban metal inputs and higher degrees of environmental disturbances.

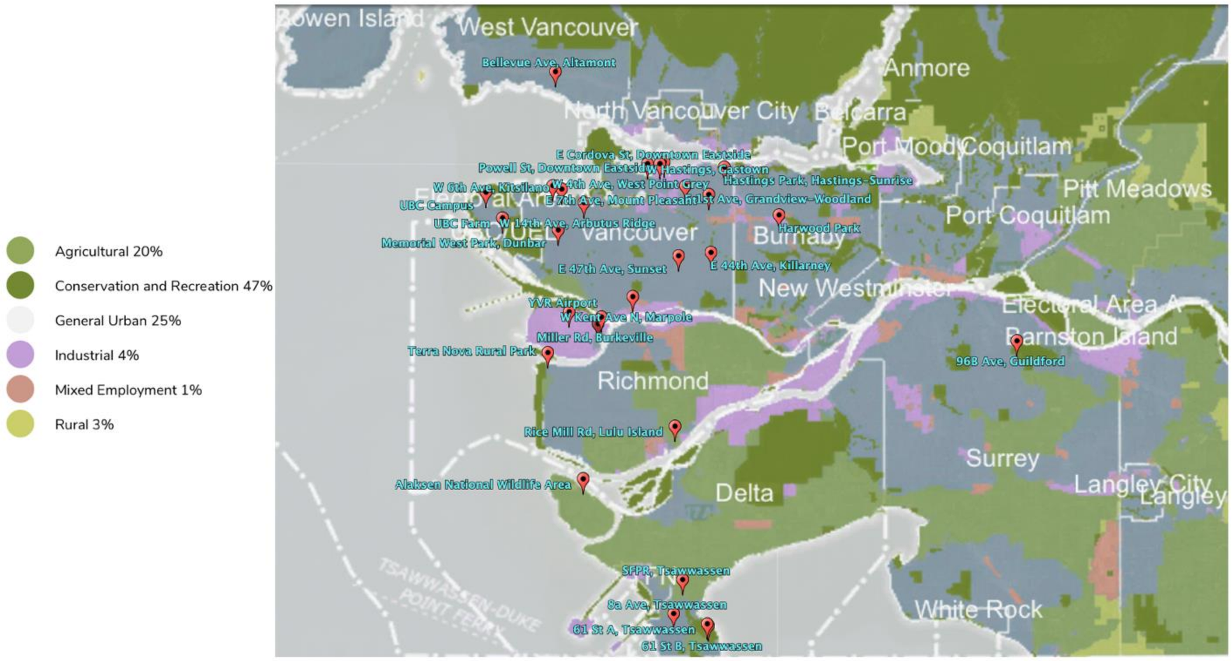

Figure 1. Map of land use distribution in Metro Vancouver retrieved from Metro Vancouver (2017). Study sites that honeybee and honey samples collected from are labelled with a red placemark. Locations are retrieved from the dataset of Smith & Weis (2020).

4.1 Field Description

Metro Vancouver is the metropolitan district located along the southwest coast of British Columbia, Canada, including a number of major cities, such as Vancouver, Richmond, Delta, and Langley. Metro Vancouver usually refers to the area of Vancouver and its surrounding urban and suburban regions, which are governed by the Metro Vancouver Regional District. Geographically, Metro Vancouver belongs to the lower mainland of British Columbia, and it contains a total of 21 municipalities (e.g. Langley, Delta, Burnaby, and Vancouver), one treaty First Nation (Tsawwassen) and one electoral area (Electoral Area A) (Metro Vancouver, 2019).

The land use pattern in Metro Vancouver is presented in Figure 1. The map is retrieved from Metro Vancouver (2017), and it indicates the regional land uses of different municipalities of Metro Vancouver. Metro Vancouver is composed of six types of habitats, which are conservation and recreation, general urban, agricultural, industrial, mixed employment, and rural regions (Metro Vancouver, 2017). Overall, Metro Vancouver is dominated by conservation and recreation areas with a 47% land cover out of the total land area. However, most of the conservation and recreation areas are composed of mountains, watersheds and natural parks, which are located in the northern part of Metro Vancouver with a low population size. Most of the municipalities are covered with a high proportion of urban land use, especially Vancouver and Burnaby. Industrial land uses scatter among all municipalities, leading to many potential industrial hotspots, but few single and large industrial area exists. Agricultural land uses account for approximately 20% of the total Metro Vancouver land area, which is mainly located within Delta, Langley, and Pitt Meadows. Other municipalities, including West Vancouver, North Vancouver, Coquitlam, Richmond, Surrey and Maple Ridge maintains a mixture of different land uses.

4.2 Choice of Elements

Antimony (Sb)

Sb is a metalloid element that was initially discovered from sulphide ore as the natural sources, which can be used in the production of alloy, synthetic rubbers and painting materials (Cooper & Harrison, 2009). Sb was firstly applied for gold purifying in the 18th century, and anthropogenic utilization had become the dominant Sb source compared to natural fluxes. By 2000, the total Sb production worldwide has reached around 140,000 tons by mining industries annually (Filella, Belzile & Chen, 2002). The release of Sb can lead to potential urban contamination in many regions due to its increasing application around the world (Li et al., 2018). Antimony contaminants in urban systems can pose risks to public health, and the impacts may vary depending on the exposure mode, duration, and quantity, as well as the presence of other chemicals (Cooper & Harrison, 2009). In urban systems, Sb mainly comes from industrial and agricultural activities (Li et al., 2018). One of the major Sb exposures is industrial production, such as Sb alloys in the manufacturing of lead batteries, flame retardant, castings, and pipes. Sb can also enter the soil and water contents from mining activities and the processing of sulphide ores. The Sb concentration in soils is usually less than one ppm if only natural fluxes are considered. In highly contaminated regions, such as waste sites, the soil Sb concentration can reach more than 2000 ppm in maximum (Cooper & Harrison, 2009). Once Sb is absorbed by animals or humans, it can combine with sulfhydryl, a functional group, and lead to the dysfunction of enzymes and ion balance. Eventually, nervous systems and metabolic reactions are damaged (Li et al., 2018).

Lead (Pb)

City, industry and traffic are three major sources of urban metal inputs, and Pb is one of the best-studied heavy metal in metal contamination analysis, which usually exists in soils along urban roads. It has been investigated that Pb accumulation in soils is correlated to the traffic intensity and distances to traffic since the 1970s (Wang & Zhang, 2018). More recent studies also determine that roadside soil Pb contamination is positively related to vehicle emission and negatively related to distances from roads (Zhang et al., 2015). In the 1960s, Pb contamination attracted public concerns due to lead poisoning among children with the diagnosis of nervous system damage (Mahaffey, 1983). Higher concentrations of Pb are found in the atmosphere and soils in regions with higher degrees of social activities. Pb contaminants can enter the human body by direct consumption of contaminated food and water, as well as the uptake of Pb particulates from the atmosphere and soil surfaces (Mahaffey, 1983). The potential lead exposure can result from lead paint, mining and smelting and other industrial implications, such as pipes, water distribution operations and battery recycling. Leaded gasoline was a major source of Pb contamination, but its use has been banned in many countries (World Health Organization, 2019). The Pb contamination in specific regions may vary significantly because of the nature of emission and sink pools, which brings more challenges to the precise detection of the lead hotspot. In future, geochemical mapping can be expected as an effective technique with the application of remote sensing (Filippelli et al., 2018).

Copper (Cu)

Cu serves as the conductive materials mainly used in cables as its low electrical resistance allows current to flow through quickly with low energy loss. The Cu concentration in city soils is positively related to the duration of urbanization. The study in Beijing, China, discovered that roadside soils are hotspots for metal contamination, including Cu accumulation. Levels of Cu contamination are positively correlated to the intensity of anthropogenic activity, such as traffic, but negatively correlated to the distances of soils from roads (Wang & Zhang, 2018). Copper contamination in cities mainly comes from vehicle transportation as Cu plays essential roles in the production of a brake pad to allow smooth braking. Cu was increasingly applied in brake pads since the mid-1980s. Even though Cu content can vary based on different vehicle makes and models, on average, Cu content can reach up to 10 – 15% in brake pads produced in North America and Europe regions (Hwang, Fiala, Park & Wade, 2016). During braking, friction is made between the brake pad and the disk rotor. A small amount of brake pad materials is released while heated so that Cu particles are released to the road surface and roadside soils. In general, the number of Cu contaminants released from the brake pad can account for a significant proportion of the total Cu release into the environment (Hwang, Fiala, Park & Wade, 2016). However, the total Cu loads may be different spatially under the impacts of environmental factors, such as soil pH, wind conditions, organic matter content and vegetation distributions (Wang & Zhang, 2018).

Chromium (Cr)

Cr is a transition metal element associated with the harmful potential to the environment, and its anthropogenic sources mainly consist of industrial and transportation disposals (Byrne, Taylor, Hudson-Edwards & Barrett, 2017). Cr is widely applied in the industry of metallurgy and ferrochromium alloys, such as the production of steels, nonferrous alloys, as well as the production of materials including mortars, refractory bricks and coating components (Canadian Council of Ministers of the Environment, 1999). Meanwhile, the utilization of car braking and tyres in traffic also constitutes a significant Cr releasing source (Byrne, Taylor, Hudson-Edwards & Barrett, 2017). The major exposure pathways of Cr are through water, soil, aerosols and other particulate matters. Its influences on public health are highly dependent on its chemical speciation and mobility (Byrne, Taylor, Hudson-Edwards & Barrett, 2017). Cr usually exists in natural environments in the form of compounds with oxygen and other metal elements, including iron and lead. In total, Cr retains nine oxidation states in the environments, and the most common ones are trivalent and hexavalent chromium. Trivalent chromium is characterized by thermodynamic stability, while hexavalent chromium is with high oxidizing capacity (Canadian Council of Ministers of the Environment, 1999). Trivalent Cr exists as the dominant form under an anoxic environment while hexavalent Cr dominates under oxic conditions with more significant toxicity. The mobility of Cr contaminants can be affected by various environmental factors, such as climate and urban dynamics (Byrne, Taylor, Hudson-Edwards & Barrett, 2017).

Rubidium (Rb)

Rb is an alkaline metal existing with high abundance in the earth's crust, and it can form oxide in minerals, such as leucite, lepidolite and pollucite. Rb is a soft and conductive metal that can appear in liquid form under room temperature, and it reacts spontaneously with air and water. Rb is associated with relatively low ionization energy and Rb ions form while it is heated. The ionizing capacity of Rb is applied in many fields of industrial production, including thermoelectric generator and ion engine for space vehicles. The electronic conductivity of Rb is also utilized in the manufacturing of vacuum tubes and photoelectric cells (Los Alamos National Security, 2016). As a lithogenic element, rubidium is geochemically stable for the evaluation of soil metal contamination (Boës, Rydberg, Martinez-Cortizas, Bindler & Renberg, 2011). In contrast with elements with significant anthropogenic inputs, lithogenic elements mostly inherit from the geographic components, including the weathering of parent materials and bedrocks (Yunginger et al., 2018). Lithogenic elements are with low mobility or abundance, and organisms rarely utilize them. Rb forms resistant minerals and is conservative in most environments. Along with other lithogenic elements, such as titanium and zirconium, Rb can be applied in sediment metal contamination investigation as the reference element.

4.3 Biomonitoring Indicators

Enrichment Factor (EF)

The metal concentrations measured in honey samples are analyzed by calculating the enrichment factor for each metal of focus. EF is an indicator that assesses the level of anthropogenic metal inputs, including non-point and point sources, with the normalization of metals based on the presence of reference elements. It is able to reveal a background reference across the geographical area and give the baseline to measure the fluctuation of other metal abundance (Boës, Rydberg, Martinez-Cortizas, Bindler & Renberg, 2011). Rb is selected as the reference metal in this study since it is a lithogenic metal with high geographic inheritance and stability. The enrichment factor of each metal is calculated using the equation:

$EF = ([M] / [Rb]_{sampled}) / ([M]_{background} / [Rb]_{background})$, where

$[M]$ = metal concentration of honey sampled at each location;

$[Rb]_{sampled}$ = Rb concentration of honey sampled at each location;

$[M]_{background}$ = background metal concentration;

$[Rb]_{background}$ = background concentration of Rb.

The mean metal concentrations of Sb, Pb, Cu and Cr at each sampling site are used to calculate the enrichment factor. Conventionally, the regional upper crust concentrations of metals and the reference element should be applied as background values (Boës, Rydberg, Martinez-Cortizas, Bindler & Renberg, 2011). However, the upper crustal concentrations may be less significant in the study of biological matrices than soil assessments. In this case, the background values of metal and reference elements are assumed to be the median value of metal concentrations in honey sampled from the background land types (agricultural and rural land uses in Smith & Weis, 2020).

Metals with an EF less than 2 is considered to have insignificant anthropogenic contaminant deposition. EFs that are between 2 and 5 suggests a moderate enrichment of metal contamination. The EF of 5 – 20 indicates significant anthropogenic inputs. EFs that are greater than 20 reveals a dramatically high metal enrichment (Barbieri, 2016).

Honeybee Contamination Index (HCI)

The metal contaminant data of honeybee tissue samples are analyzed using Honeybee Contamination Index. HCI is an effective and reliable tool that uses western honeybees as a biomonitor to indicate the contamination level in terrestrial environments for different metal contaminants. Each Honeybee Contamination Index is calculated based on data of one type of metal (Goretti et al., 2020). The background metal concentration may be various spatially due to different natural metal sources (e.g. the composition of parent materials). The application of HCI generalizes the metal contamination quantification based on the concept of bioaccumulation (Goretti et al., 2020). Therefore, the impacts of background metal concentration are minimized, and metal contamination results become comparable on a broader scale. HCI also displays the pattern of environmental health within regions, and management practices can be suggested for locations with relatively higher contamination levels. HCI_1 and HCI_2 are calculated for each metal of focus using the equation:

$HCI = \log([M]_{honeybee} / [M]_{threshold})$, where

$[M]_{honeybee}$ = metal concentration of honeybee tissue sampled at each location;

$[M]_{threshold}$ = environmental threshold for metal contaminant (the high threshold is used for HCI_1 while the low threshold is used for HCI_2).

Ideally, the regional thresholds of metal contamination should be applied (Goretti et al., 2020). As specific reference values are not available for metal elements in Metro Vancouver, similar to the approach of EF calculation, metal concentrations measured at the background land use are assumed to be background thresholds. The 75% quartile of metal concentrations in honeybee tissue sampled from the agricultural and rural land use is applied as the high threshold, and the 25% quartile of metal concentrations is applied as the low threshold.

An illustration of the Honeybee Contamination Index is shown in Figure 2. Environments with negative HCI_2 values are considered to have relatively low metal contamination levels, and environments with positive HCI_1 values have relatively high contamination levels. Intermediate contamination level is determined with negative HCI_1 values and positive HCI_2 values (Goretti et al., 2020).

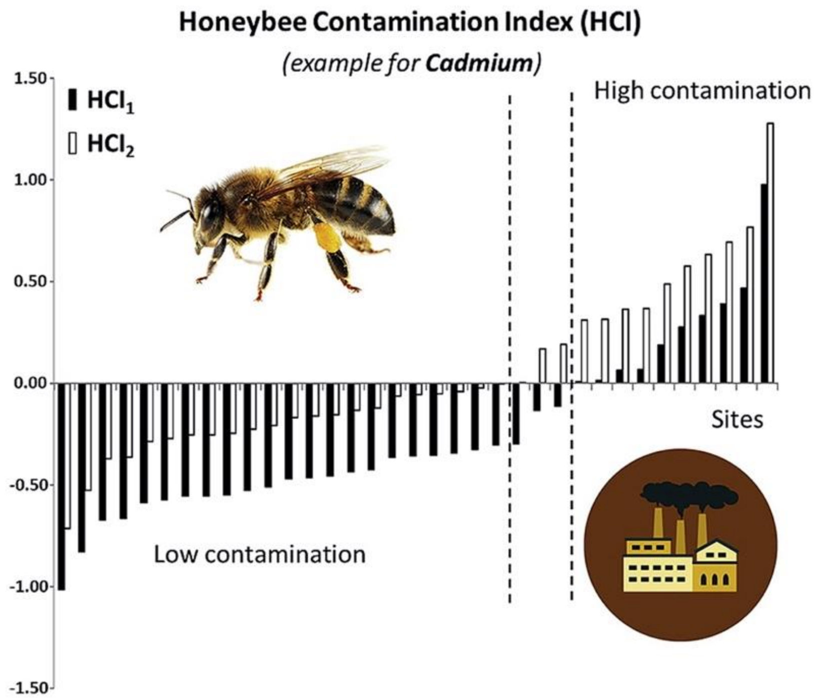

Figure 2. Honeybee Contamination Index with Cadmium data as an example, retrieved from Goretti et al. (2020).

5. Result & Discussion

5.1 Western Honeybee (*Apis mellifera*)

Western honeybee (*Apis mellifera*), in particular, is an ideal species that monitor the metal contamination level in urban systems, considering its year-round accessibility, high diversity and wide distribution, as well as intensive interaction with local environments.

Apis mellifera is a social insect that lives in a group of families. Western honeybees form colonies, and they are involved in the well-organized social structure with strict labour division (Bargańska, Ślebioda & Namieśnik, 2016). Each bee colony contains three categories of honeybees: the queen, drones, and workers. The queen honeybee usually has a lifespan of two to five years and is the only female bee in a colony that produces a maximum of 1500 eggs per day (Mackean, 2004). The drone bees within a colony have a much shorter lifespan of approximately four weeks, and they are male bees responsible for fertilizing the queen (Mackean, 2004). Worker bees are female bees without their reproductive system fully developed, which forage pollen and nectar and bring them back to the combs from the surrounding environment. (Mackean, 2004).

Accessibility

The honeybee is with high accessibility throughout the year as a biomonitor. While other social bees, such as bumblebees, tend to have the population size decrease significantly during cold seasons, honeybees are able to retain the active biological activities and functions in colonies because of their annual cycle. For most of the social bee groups, only the queen is remained for cold seasons by hibernation (Seeley, 2019). The honeybee is good at temperature control, resulting from the well-insulated structure they form, and the heat produced by their muscle contraction. This temperature control ability allows the honeybee to keep a warm habitat even though the background temperature is as low as -30 °C. In winter, the social structure of a honeybee colony maintains to be intact with more than 10,000 worker bees and one queen bee (Seeley, 2019). The brood-rearing of honeybees usually happens after winter finishes as the comb temperature is raised (Seeley, 2019). The work of honeybees resumes since early spring when the climate gets warmer with the temperature around 15 °C. During this period, canopies and vegetations are within the blooming period, which leads to higher availability of pollens and nectars. The honeybee development also renews the bee group with a high growth rate. A recover period occurs from late autumn to early winter, and the number of bees in the colony decreases, especially for old bees with low accessibility to food resources (Bargańska, Ślebioda & Namieśnik, 2016).

Diversity and Distribution

The *Apis* genus consists of 10 honeybee species. Based on the identification of nuclear DNA and mitochondrial DNA, these species are categorized into three clades, which are cavity-nesting bees, giant bees, and dwarf bees. *Apis mellifera* belongs to the cavity-nest bees (Han, Wallberg & Webster, 2012). Figure 3 shows the categorization of honeybee species in cavity-nesting, giant and dwarf groups. Western honeybee has 29 subspecies based on its morphometric characteristics. These subspecies are composed of 4 honeybee groups in total, which are M group, A group, C group and O group, with their native ranges across Europe, Africa and the Middle East. Other species of the *Apis* genus originate from Asia regions.

However, the honeybee is further introduced into other regions by human activities such as beekeeping, and currently, the honeybee population has dispersed in almost all continents worldwide (Han, Wallberg & Webster, 2012). As mentioned above, the temperature control ability also helps honeybee adapt to the colder climate, which allows the expansion of cavity-nesting honeybees.

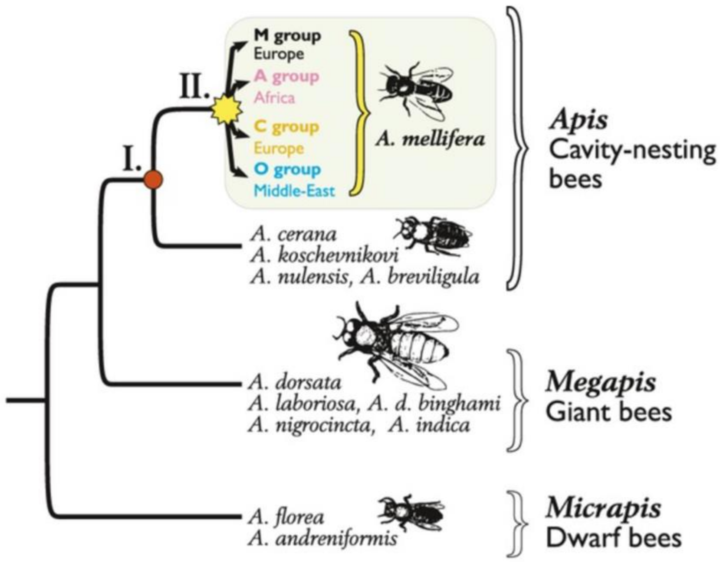

Figure 3. Categorization of different honeybee species in cavity-nesting bees, giant bees and dwarf bees, retrieved from Han, Wallberg & Webster, 2012. (I) The split of Apis mellifera and other bees in the Apis genus. (II) Common ancestors of Apis mellifera.

Environmental Interaction

The honeybee is associated with intensive interaction with the environment during their daily foraging activity (Figure 4). Honeybee collect nectar and pollen from flowering plants, so that contaminant elements tend to be absorbed and bioaccumulate in honeybees during this process (Smith et al., 2019). The intensity of pollen foraging highly depends on the colony pollen demand, which is regulated by quantities of pollen stored and the amount of brood in combs. The amount of brood in combs act as the positively affecting factors for pollen foraging, while the foraging activity is negatively correlated to the abundance of pollen (Dreller & Tarpy, 2000) The direct consumption of water also leads to the uptake of contaminants by a honeybee in the water content. Meanwhile, during the flight and movement on soils and vegetations, contaminant particles will be adsorbed to its body and carried by the honeybee. The honeybee has an average working radius of 3 km, depending on the availability of flowering vegetation and the density of beehives (Smith et al., 2019). As a result, it can provide the composite sample for environmental contamination within the working region of the honeybee (Van der Steen, Kraker & Grotenhuis, 2012). As environmental contaminants are picked up by honeybees, these elements can be eventually transferred into bee products, including honey, royal jelly and beeswax. Honey is considered a stable product, and its composition can be affected by elevated concentrations of environmental contaminants, such as lead, zinc, and copper on a relatively small scale (Smith et al., 2019). The exposure of honeybee to contamination from vegetation, soils and water are significant from spring to autumn with the most intensive foraging activity (Bargańska, Ślebioda & Namieśnik, 2016).

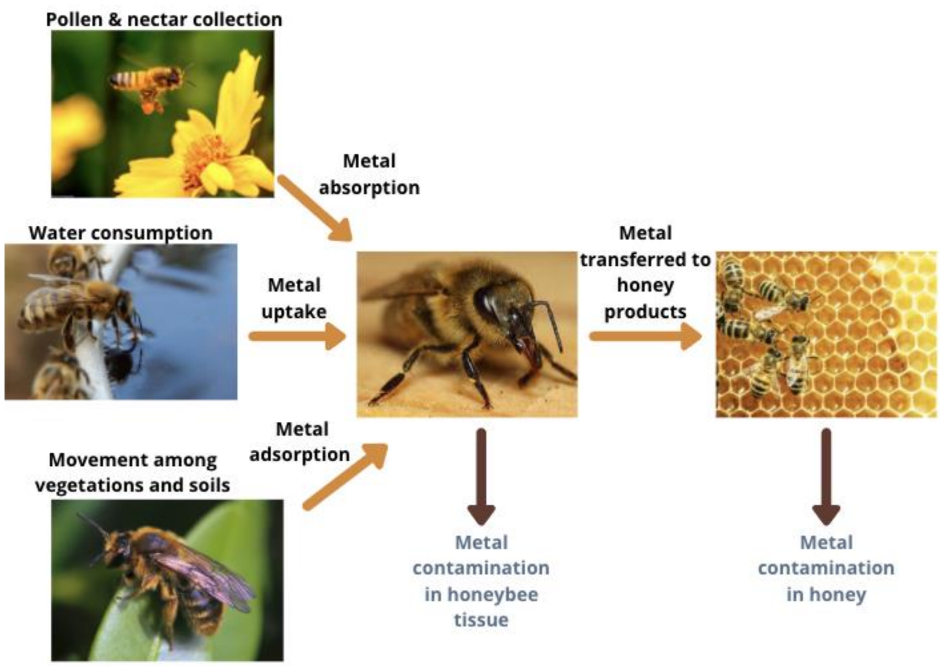

Figure 4. Major interactions between honeybee and the environment, as well as the processes that metal contaminants are transferred into honeybee and honey products,

The honeybee and honey can be used as a biomonitor for metal contaminants released from both point sources and non-point sources. In urban systems, point sources usually refer to specific locations, such as mining spots or shipping ports, while non-point sources are environments with diffusive metal inputs including areas with heavy transportation (Smith et al., 2019). Honeybee and its products have been used to monitor metal contamination since 1935 (Van der Steen, Kraker & Grotenhuis, 2012). As honeybees are able to visit as more as thousands of flowers within one day, the application of honeybee as a contamination monitor is much less expensive than establishing localized monitoring equipment. Honeybee has also been applied for air quality assessment at Frankfurt Airport in Germany since 2006 (Kaufman, 2017). In previous urban studies, honeybee, honey and wax were used to determine the presence and quantity of metals, and metals such as lead, zinc and cadmium are frequently studied (Kaufman, 2017).

Nevertheless, using honeybee and honey as a biomonitor has several drawbacks. Even though honeybee and its products are effective in assessing urban trace metals with high concentrations or extensive inputs, they are not capable of tracking the contaminating sources. Contaminants detected from honeybee and honey often serve as the integrated samples of all sources within the honeybee working radius (Smith et al., 2019). The optimal conditions for bee foraging are with relatively low moisture content and a warm temperature of 20 – 25 °C. The mobility of honeybee can be disturbed under extreme weather (Bargańska, Ślebioda & Namieśnik, 2016).

5.2 Case Study: Metro Vancouver, British Columbia, Canada

EF values are summarized in Table 1, and the result is shown in Figure 5. In Metro Vancouver, most study sites maintain insignificant enrichment (EF < 2) of metal deposition. The EFs of Sb also show insignificant enrichment in many of the study sites. However, 8 study sites show a moderate level of Sb anthropogenic inputs. Cu maintains the enrichment factor lower than 2 in most of the study sites with three exceptions of Powell St, Downtown Eastside (EF = 2.02), Alaksen National Wildlife Area (EF = 2.03) and SFPR, Tsawwassen (EF = 2.25). The natural input contributes to the major sources of Cu deposition. Similarly, most EFs of Cr is less than 2, and the anthropogenic deposition of Cr is insignificant in the majority of regions in Metro Vancouver. Moderate anthropogenic Cr deposition is identified at 61 St B, Tsawwassen (EF = 2.37) and Powell St, Downtown Eastside (EF = 4.03). The enrichment of Cr is significant at YVR Airport (EF = 5.04). In a total of 8 study sites, including UBC Campus (EF = 2.54), W Hastings, Gastown (EF = 2.16), Bellevue Ave, Altamont (EF = 3.44), 8a Ave, Tsawwassen (EF = 2.38) , W 6th Ave, Kitsilano (EF = 3.46), W 14th Ave, Arbutus Ridge (EF = 3.61), W Kent Ave N, Marpole (EF = 4.94) and Memorial West Park, Dunbar (EF = 2.96), indicates the moderate intensity of anthropogenic Pb deposition. E Cordova St, Downtown Eastside (EF = 5.22) is associated with significant Pb enrichment. Additionally, Powell St, Downtown Eastside retains an extremely high EF of 22.21, which shows a very high anthropogenic input of Pb.

Table 1. Summary table of EF values for Sb, Pb, Cu and Cr at 28 study sites in Metro Vancouver.

Region	Location	EF Sb	EF Pb	EF Cu	EF Cr
UBC	UBC Campus	1.10963774	2.54135677	0.44161422	0.92306137
	UBC Farm	0.54836367	1.38241309	0.28997134	0.60214015
Downtown	Powell St, Downtown Eastside	3.45269405	22.2127886	2.02068957	4.03557256
	E Cordova St, Downtown Eastside	2.60564833	5.21583734	0.48301336	1.607258
	W Hastings, Gastown	2.10888615	2.1624858	0.40912151	1.18971404
Vancouver Eastside	E 1st Ave, Grandview-Woodland	0.72834337	0.92368736	0.23856842	0.39527962
	E 7th Ave, Mount Pleasant	0.55281087	0.61816961	0.39406831	0.3338726
	Hastings Park, Hastings-Sunrise	0.86751694	1.57114768	0.36567178	0.65299304
	E 44th Ave, Killarney	0.92378639	1.14325158	0.31551227	0.61046157
	E 47th Ave, Sunset	0.73987343	0.99442616	0.4067532	0.49963079
West Van	Bellevue Ave, Altamont	1.7648681	3.44226481	0.47852372	0.84468634
Delta	61 St A, Tsawwassen	0.69900121	1.17141017	0.63599471	0.42262528
	61 St B, Tsawwassen	2.55590098	1.58955372	0.74465389	2.37380043
	Alaksen National Wildlife Area	1.20165674	1.03000243	2.03381931	1.72091883
	SFPR, Tsawwassen	1.54346289	1.06702708	2.25261084	1.50212069
	8a Ave, Tsawwassen	1.39024598	2.38091139	0.36534849	1.54164013
Burnaby	Harwood Park	0.95560484	0.92263764	0.28293057	0.77539665
Vancouver Westside	W 6th Ave, Kitsilano	2.25059268	3.46487362	0.41821495	1.15668235
	W 14th Ave, Arbutus Ridge	2.31028876	3.60982712	0.38696144	1.34313633
	W Kent Ave N, Marpole	2.83467037	4.93841107	0.4445939	1.62242708
	Memorial West Park, Dunbar	1.31884598	2.95614618	0.33916293	1.13903949
	W 4th Ave, West Point Grey	1.02963662	1.05886366	0.42587745	0.78089614
Richmond	YVR Airport	0.5935292	1.90083019	1.84095361	5.04136142
	Wellington Crescent, Burkeville	0.73668956	0.93489783	0.41936194	0.48615139
	Miller Rd, Burkeville	1.94220774	0.93675321	0.33112274	0.38655725
	Terra Nova Rural Park	0.1529622	0.30262656	0.8007685	0.44478859
	Rice Mill Rd, Lulu Island	2.07965527	1.52490028	1.54948278	1.69176048
Surrey	96B Ave, Guildford	0.18133802	0.33026661	0.10885545	0.12672452

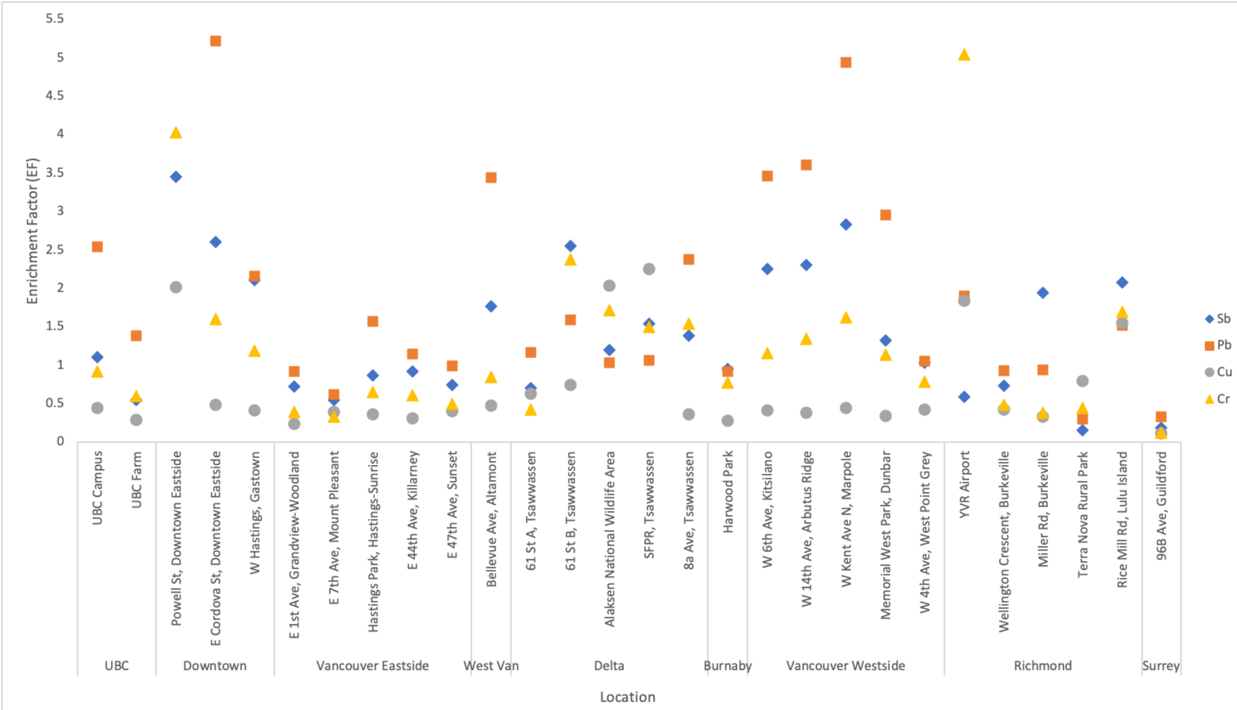

Figure 5. Enrichment factors of Sb, Pb, Cu and Cu in honey sampled from different study locations in Metro Vancouver. The enrichment factor of Pb at Powell St, Downtown Eastside (EF = 22.21) is dramatically higher than EF at other locations, which is not included in this figure.

According to this approach using EF, Pb constitutes the most significant anthropogenic metal input within Metro Vancouver, followed by Sb and then Cr. The deposition of Cu from anthropogenic activities and land uses are the least significant based on the results in this study. Improved management practices are expected in the future, especially in those regions with moderate enrichment levels. Three study sites are indicating significant anthropogenic metal deposition (YVR Airport for Cr, E Cordova St, Downtown Eastside for Pb and Powell St, Downtown Eastside for Pb), especially the extremely high Pb enrichment in Powell St, Downtown Eastside. More effective contaminant controlling and land restoring approaches may be required in these regions. Overall, the anthropogenic deposition is the lowest in Surrey for all metals of focus. However, as only one study site was sampled in Surrey, a larger sample size is recommended for further study to test the statistical significance. Metal enrichment is relatively higher in Vancouver Westside, Downtown, and Delta. Vancouver Westside and Downtown regions are dominated by the general urban land use, while Delta is dominated by agricultural uses. Several potential industrial hotspots distribute across these three regions, which may also contribute to the anthropogenic metal inputs.

The HCI of metals of focus is shown in Figure 6. Please refer to HCI Summary Table in Appendix for the full HCI values. For Sb, Pb, Cr and Rb, the majority of study sites in Metro Vancouver are found to have a relatively high level of urban metal contamination. According to the HCI values of Sb, Alaksen National Wildlife Area is the only region that has low contamination. Three sites (Terra Nova Rural Park, Rice Mill Rd, Lulu Island and SFPR, Tsawwassen) show intermediate contamination, and the remaining nine sites are with high metal contamination. Bellevue Ave, Altamont and Terra Nova Rural Park are two sites having low Cu contamination. Six study sites of Cu show intermediate contamination, and five of them reveal high Cu contamination. None of the study sites is identified with a low contamination level in consideration of Pb, Cr or Rb. Four study sites of Pb and Rb investigation show intermediate contamination level while three study sites are considered intermediately contaminated regions for Cr. All other study sites are with high contamination levels for Pb, Cr and Rb. Study sites of Alaksen National Wildlife Area, Terra Nova Rural Park, Rice Mill Rd, Lulu Island and SFPR, Tsawwassen shows relatively less significant metal contamination for all metals of focus in this case. E Cordova St, Downtown Eastside, E 44th Ave, Killarney and Hastings Park, Hastings-Sunrise are three sites have higher metal contamination level with respect to all metal contaminants.

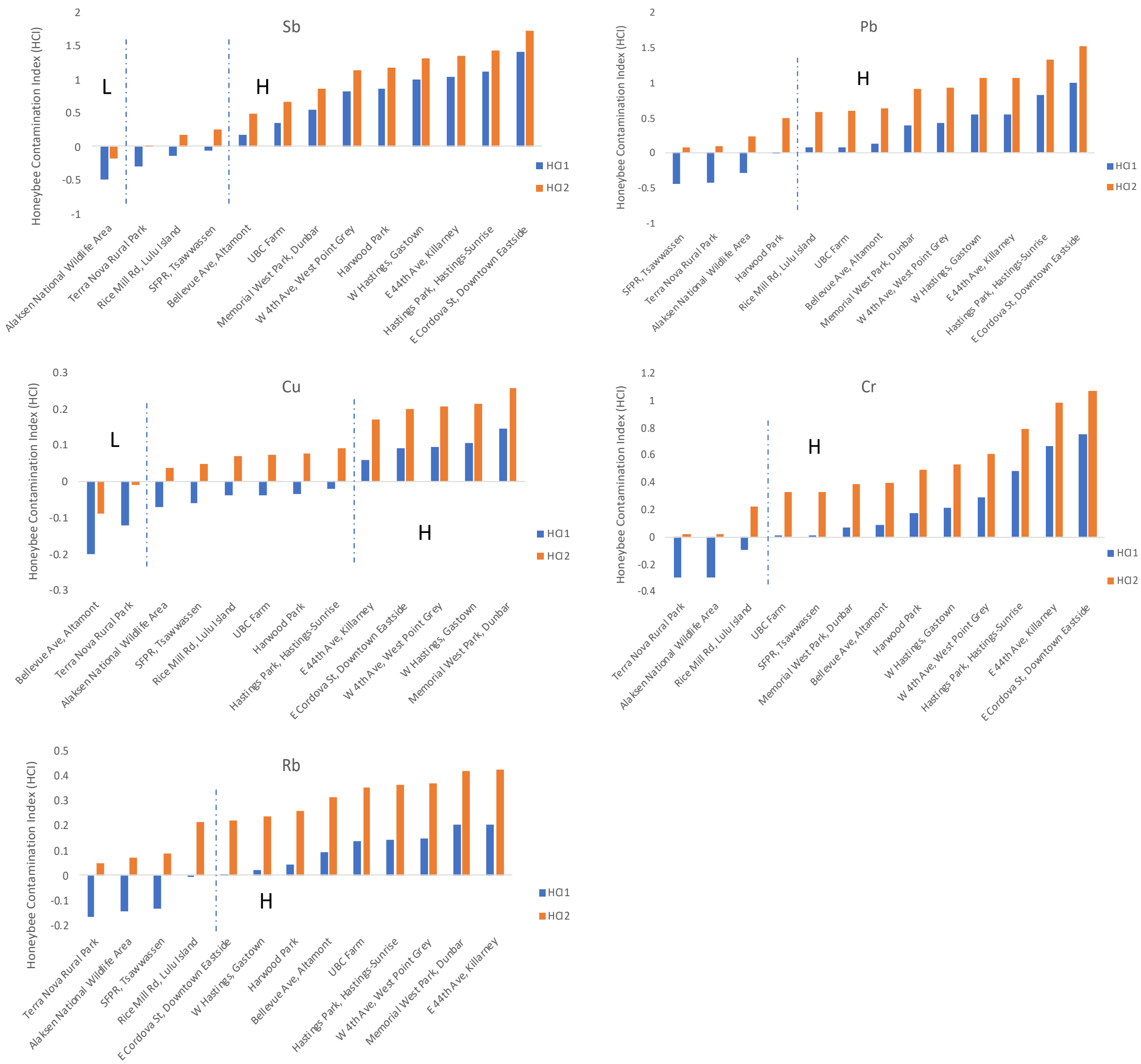

Figure 6. Honeybee Contamination Index of Sb, Pb, Cu, Cr and Rb in honeybee tissue sampled from Metro Vancouver. Study sites with high metal contamination are labelled with 'H', and study sites with low metal contamination are labelled with 'L'.

The HCI results show that many regions within Metro Vancouver are associated with a high level of metal contamination. Among five metals of focus, Cu is the least significant contaminant in Metro Vancouver, followed by Sb, regarding the number of study sites that indicate low and intermediate contamination levels. Except for Alaksen National Wildlife Area and Terra Nova Rural Park, the remaining study sites all present high contamination of at least one metal element. As none of these study sites is detected with low contamination levels for all metals of focus, management and restoration practices are suggested for the entire range of Metro Vancouver.

However, while comparing the results between the calculated enrichment factor and Honeybee Contamination Index, no significant correlation is discovered between these two indicators. Regions with low anthropogenic metal deposition do not necessarily have low metal contamination levels, and vice versa.

Overall, the results of EF and HCI are relatively consistent with the land use distribution in Metro Vancouver. The EF shows that industrial land uses tend to result in higher anthropogenic metal deposition than other land uses. Conservation and recreation regions are usually associated with lower metal enrichment. For instance, E Cordova St, Downtown Eastside, Powell St, Downtown Eastside, W Kent Ave N, Marpole and YVR Airport are found to have significantly higher anthropogenic metal inputs, and all of them located within potential industrial hotspots of Metro Vancouver. 96B Ave, Guildford and Terra Nova Rural Park are two study sites within conservation and recreation land use, and their EFs both indicate insignificant enrichment for all four metals of focus. Nevertheless, anthropogenic metal deposition is identified to be spatially various for urban land uses. E 7th Ave, Mount Pleasant and Bellevue Ave, Altamont both located in the general urban land uses, but the metal enrichment of E 7th Ave, Mount Pleasant is significantly lower than that of Bellevue Ave, Altamont.

The HCI results indicate that conservation and recreation, as well as agricultural land, uses retain relatively lower metal contamination levels than general urban and industrial land uses. For instance, Alaksen National Wildlife Area and Terra Nova Rural Park are two regions with either low or intermediate contamination levels among all study sites of honeybee tissue sampling. While Alaksen National Wildlife Area is located within agricultural land use, Terra Nova Rural Park belongs to conservation and recreation land use. The contamination levels of urban and industrial land use tend to vary depending on the metal of focus. For example, E Cordova St, Downtown Eastside, the industrial study site, is characterized by more intense contamination levels for Sb, Pb and Cr. However, the Rb contamination level at E Cordova St, Downtown Eastside is less significant with a lower HCI_1 value. Memorial West Park, Dunbar, as an example of urban land use, is associated with high contamination for Cu and Rb, but it is less significantly contaminated by Sb, Pb and Cr.

Van der Steen, Kraker & Grotenhuis (2012) have conducted comprehensive research that tests the metal concentrations of the honeybee in both temporal and spatial manners. The research includes the results for a total of 18 metals, which are Cd, Cr, Cu, Pd, Zn, Al, Sb, As, Co, Li, Mn, Mo, Ni, Se, Sr, Sn, Ti and V, at three locations (Maastricht, Buggenum and Hoek van Holland) of Netherlands. Within a research period of 3 months, 18 samples are taken from each location bi-weekly. The worker bee is applied as the major subject of this research. During the activity of pollen and nectar collection, a worker bee retains the most significant interactions with environmental contamination, resulting in more reliable results. The metal concentrations in honeybee samples are determined with inductive coupled plasma–atomic emission spectrometry technique (Van der Steen, Kraker & Grotenhuis, 2012). The concentrations of 9 metals (Cd, Cr, Cu, Al, Co, Mn, Sr, Ti and V) in honeybees are found to fluctuate significantly throughout the 3-month period for at least one location. As a result, the exposure of honeybee to metal contaminant sources at study locations are considered to vary significantly temporally. Meanwhile, the mean metal concentrations in honeybee are found to be significantly different among three locations for Co, Sr and V. The spatial variation may result from the specific intensity of industrial activities. Compared to temporal trends, the spatial pattern of metal contamination in the Netherlands is limited due to its small area and concentrated population (Van der Steen, Kraker & Grotenhuis, 2012).

Honey is a natural product of honeybee, with its composition largely affected by the metal contamination in the foraging range of honeybee. The research of Bosancic, Zabic, Mihajlovic, Samardzic & Mirjanic (2020) investigates the effectiveness of honey for metal biomonitoring by comparing the metal concentrations in honey produced from three types of environmental systems, which are apiaries near the thermal power plant, certified organic apiaries, and conventional apiaries that are semi-controlled. In total, 27 honey samples are collected from randomly selected beehives in three systems, and the concentrations of Pb, Cd, Ni, and Zn in honey are discussed. The results show that the concentrations of Pb, Cd, and Zn are significantly higher in honey sampled from areas near the thermal power plant, and the concentrations of Pb and Cd are the lowest in honey from certified organic apiaries (Bosancic, Zabic, Mihajlovic, Samardzic & Mirjanic, 2020). However, the concentration of Ni is found to be lower in honey from apiaries near the thermal power plant and conventional apiaries than that from certifies organic apiaries, which may result from the types of flower used (e.g. variations between polyfloral and monofloral honey) (Oroian et al., 2016).

By integrating the results from the case study in Metro Vancouver and previous research on honeybee and honey as a metal biomonitor, it concludes that metal concentrations in honeybee and honey can vary significantly in both spatial and temporal perspectives. Metal contaminants are traceable in honeybee tissue and honey as metal contaminants are picked up by honeybee and transferred to bee products. As a result, honeybee and honey can both perform as the urban metal biomonitor. EF and HCI, as biomonitoring indicators, are applicable in different regions for metal contamination assessment. The calculation of EF and HCI uses background and reference values from the study region, which provides generalized results that are useful for making inferences among various regions (Boës, Rydberg, Martinez-Cortizas, Bindler & Renberg, 2011; Goretti et al., 2020).

6. Limitations

This research provides a general explanation for honeybee as an effective biomonitor by explaining their biological activeness, species diversity and global distribution and intense interactions with the surrounding environments. Due to the outbreak of COVID-19 in Canada, the major method used for this section is a literature review, and primary sampling or testing is avoided for safety. Also, the impacts of local environmental conditions (e.g. climate, geographic features) on the effectiveness of honeybee biomonitoring among different regions (e.g. continents) are not included in this case.

Metro Vancouver is used as a case study in this research. Thus, the scope of the study region is relatively small. The results from the case study may be limited in extrapolating to a larger geographical region, or an entire country or other regions around the world. Ideally, the metal concentration of the upper crust or deeper soil layer should be used. However, due to the lack of information, local measurements in the background land use (agricultural and rural land use) are applied as background and reference values in calculations. The results in the case study only show spatial variations of Metro Vancouver with different types of land uses without the consideration of temporal variation.

In further studies, the effectiveness of insect biomonitor is recommended to be evaluated by incorporating variations of local environmental conditions. For example, whether the local climate will impact the responding efficiency of the biomonitor of focus. As a consequence, the results can be more comprehensive and significant, and more accurate guidelines can be developed to choose the best biomonitoring species based on the local conditions (e.g. land use, climate, etc.). For the case study in Metro Vancouver, more samples are expected to be tested so that more land uses and regions across Metro Vancouver can be covered. With honey and honeybee tissue sampled at different stages of the honeybee annual cycle, the temporal variation of urban metal contamination is recommended to be analyzed in the future.

7. Conclusion

Along with future urbanization, biomonitoring will turn to be a valuable and reliable assessing method for environmental contamination. Biomonitoring can help determine the impacts of environmental disturbances and contamination elements quantitatively. Insects are widely applied as an effective biomonitor for environmental contamination because of its large biodiversity and population size, global geographic distribution and responding efficiency. Honey and honeybee are ideal biomonitors for metal contamination in urban systems, and its effectiveness in biomonitoring is discussed here based on the high accessibility, distribution, diversity and environmental interactions. In this research, EF and HCI are calculated for different regions and land uses within Metro Vancouver using metal concentrations in honey and honeybee tissue samples, indicating the anthropogenic metal enrichment and regional metal contamination level. The results not only show the metal enrichment and contamination levels among different locations but also determine the correlation between urban metal contamination and types of land uses. Along with previous relevant research, the spatial variation of metal contamination in Metro Vancouver reveals the effectiveness of honey and honeybee as biomonitor and the applicability of EF and HCI as useful indicators. The case study can serve as the background research for further study in Metro Vancouver, and the results can also be used to compare with metal contamination levels in other regions. The biomonitoring system is also expected to be applied more extensively in the future with contributions to ecological conservation and public health.

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Appendix

Honeybee Contamination Index (HCI) Summary Table

Sb		
Location	**HCI1**	**HCI2**
Alaksen National Wildlife Area	-0.49792	-0.18468
Terra Nova Rural Park	-0.29173	0.021502
Rice Mill Rd, Lulu Island	-0.13822	0.175016
SFPR, Tsawwassen	-0.05934	0.253895
Bellevue Ave, Altamont	0.172946	0.486179
UBC Farm	0.357374	0.670606
Memorial West Park, Dunbar	0.549141	0.862374
W 4th Ave, West Point Grey	0.819943	1.133175
Harwood Park	0.865814	1.179046
W Hastings, Gastown	1.009668	1.3229
E 44th Ave, Killarney	1.042246	1.355479
Hastings Park, Hastings-Sunrise	1.122109	1.435342
E Cordova St, Downtown Eastside	1.405765	1.718998

Pb		
Location	**HCI1**	**HCI2**
SFPR, Tsawwassen	-0.43756	0.079402
Terra Nova Rural Park	-0.42109	0.095874
Alaksen National Wildlife Area	-0.28213	0.234834
Harwood Park	-0.01514	0.501822
Rice Mill Rd, Lulu Island	0.07304	0.590004
UBC Farm	0.084432	0.601396
Bellevue Ave, Altamont	0.124374	0.641338
Memorial West Park, Dunbar	0.391726	0.90869
W 4th Ave, West Point Grey	0.419304	0.936268
W Hastings, Gastown	0.552005	1.068969
E 44th Ave, Killarney	0.556656	1.07362
Hastings Park, Hastings-Sunrise	0.822495	1.339459
E Cordova St, Downtown Eastside	1.007635	1.524599

Cu		
Location	**HCI1**	**HCI2**
Bellevue Ave, Altamont	-0.20056	-0.08999
Terra Nova Rural Park	-0.11971	-0.00914
Alaksen National Wildlife Area	-0.07179	0.03878
SFPR, Tsawwassen	-0.06035	0.050213
Rice Mill Rd, Lulu Island	-0.03937	0.071191
UBC Farm	-0.03848	0.072086
Harwood Park	-0.0334	0.077166
Hastings Park, Hastings-Sunrise	-0.01851	0.092058
E 44th Ave, Killarney	0.059451	0.170017
E Cordova St, Downtown Eastside	0.090907	0.201473
W 4th Ave, West Point Grey	0.096185	0.206751
W Hastings, Gastown	0.104596	0.215162
Memorial West Park, Dunbar	0.14546	0.256026

Cr		
Location	**HCI1**	**HCI2**
Terra Nova Rural Park	-0.29831	0.016599
Alaksen National Wildlife Area	-0.2956	0.019305
Rice Mill Rd, Lulu Island	-0.09227	0.222638
UBC Farm	0.008118	0.323024
SFPR, Tsawwassen	0.013447	0.328353
Memorial West Park, Dunbar	0.070444	0.385351
Bellevue Ave, Altamont	0.08416	0.399067
Harwood Park	0.171515	0.486421
W Hastings, Gastown	0.214688	0.529595
W 4th Ave, West Point Grey	0.289965	0.604871
Hastings Park, Hastings-Sunrise	0.478031	0.792937
E 44th Ave, Killarney	0.664299	0.979205
E Cordova St, Downtown Eastside	0.752845	1.067752

Rb		
Location	**HCl1**	**HCl2**
Terra Nova Rural Park	-0.16726	0.049364
Alaksen National Wildlife Area	-0.14352	0.073102
SFPR, Tsawwassen	-0.13072	0.085906
Rice Mill Rd, Lulu Island	-0.00438	0.212244
E Cordova St, Downtown Eastside	0.003925	0.220548
W Hastings, Gastown	0.020335	0.236959
Harwood Park	0.040959	0.257582
Bellevue Ave, Altamont	0.094872	0.311495
UBC Farm	0.13681	0.353434
Hastings Park, Hastings-Sunrise	0.145208	0.361832
W 4th Ave, West Point Grey	0.150522	0.367146
Memorial West Park, Dunbar	0.20063	0.417254
E 44th Ave, Killarney	0.203497	0.420121