Assessment of lead pollution in dogs around mining area and verification of effects of soil amendments on tissue lead concentrations and toxicity in mouse model

(鉱山地域のイヌにおける鉛汚染の評価と
土壌改良が及ぼすマウスの組織中鉛濃度
および毒性影響への効果検証**)**

Dissertation Summary

Nyein Chan Soe

Chapter 1: General Introduction

Lead (Pb) pollution has been a longstanding environmental concern, dating back centuries and documented in ancient civilizations. Its presence in the earth's crust and its release into the environment through natural processes like soil weathering and human activities such as mining and agriculture have led to widespread contamination of air, water, soil, and living organisms. Pb pollution affects various facets of human health, including neurological development, cardiovascular health, and kidney function. Children are particularly susceptible to Pb exposure, as their developing bodies absorb Pb more readily and are more sensitive to its toxic effects. Pb is listed in WHO's ten chemicals of major public health concern. Although the global removal of leaded gasoline has reduced human blood Pb levels, people still expose to Pb from different sources, especially in low-incomed countries and middle-income countries. The Republic of Zambia, a landlocked nation in southern Africa, is bordered by eight countries: Tanzania to the northeast, Malawi to the east, Mozambique to the southeast, Zimbadwe and Botswana to the south, Namibia to the southwest, Angola to the west, and the Democratic Republic of the Congo to the north. The economy of Zambia has remained heavily reliant on mining for over a century. The Pb-Zn mining at Kabwe, the capital of Central Province, began since 1904. It produced more than 1.8 million tons of Zn and 0.8 million tons of Pb, along with 7,816 tons of vanadium oxide, 80,000 kg of silver and 235,000 kg of Cd until 1994 when the mining ceased. Consequently, Pb pollution around mining areas in Kabwe has become a significant environmental and public health concern that arises from the extraction, processing, and disposal of Pb-containing ores. Understanding Pb speciation is essential because varying properties, behaviors, and toxicities of different Pb species can significantly influence its environmental fate, transport, and potential impacts on both human health and ecosystems. The significant amount of Pb produced from mining dumps causes severe pollution of the environment, especially in its vicinity. Therefore, there is an urgent need for cleaning up and rehabilitation of the contaminated soil. Remediation for Pb pollution concerns the application of plans to alleviate and minimize the existence of Pb contaminants in the environment, thereby reducing the risks to human health and ecosystems.

Therefore, the main objectives of this dissertation were;

1. To investigate the current accumulation of heavy metals in dogs' tissues around the Pb-Zn mine in Kawbe, Zambia.
2. To evaluate the in vivo efficacy of chicken manure amendment on Pb toxicity using mice.
3. To investigate the accumulation of different Pb species in mice model as Pb exists in different forms around the mining and smelting environment.
4. To investigate the effect of phosphate and liming amendments on soils spiked with different Pb species and its tissue accumulation in mice by simulating environmental exposure pathway.

Chapter 2: Metals and arsenic distribution in stray dogs' tissues around a lead–zinc mine in Kabwe, Zambia

Chapter 2 of the dissertation focused on assessing the current accumulation of toxic and trace metals in stray dogs' tissues in Kabwe, a town known for Pb pollution due to extensive lead and zinc mining activities, Zambia. Tissue samples collected from stray dogs near and distant from the mine site were analyzed for arsenic (As), cadmium (Cd), Pb, copper (Cu), and zinc (Zn) concentrations using inductively coupled plasma mass spectrometry (ICP-MS). Results showed stray dogs from Kabwe have higher Cd and Pb concentrations in kidneys and liver when compared with stray dogs from Naples, Italy where the minimal pollution. When concentration of As, Cd, Pb, Cu and Zn in tissues were compared between two locations (near the mine and far from the mine), the significant higher concentrations of Cd and Pb in tissues from dogs sampled from near the mine were observed. The highest levels of As, Cd, and Pb were distributed in the lungs, kidneys and bone, respectively. Additionally, higher Cu and Zn concentrations were observed in the liver across all samples. No sex and age of dogs were influenced on tissue distribution of metals. Associations were found among toxic (Pb and Cd) and trace metals (Cu and Zn) in various tissues, indicating complex interactions within biological systems. The metals accumulation in stray dogs from Kabwe, Zambia highlighted that toxicity may not be taken lightly. Because metal accumulation may be similar in human since dogs are sentinel animals. Moreover, environmental remediation is required for human, animals and environmental health concerns in Kabwe, Zambia.

Chapter 3: Effect of chicken manure amendment on lead burden in mice: exposure to lead-spiked soil

Chapter 3 of this dissertation aimed to investigate the efficacy of chicken manure on the Pb-contaminated soil via environmentally realistic exposure pathways in the laboratory setting by using mouse as a model. Because organic soil amendments like livestock manure are rich in OM content and improve soil structure. They can restrain Pb through different mechanisms (physical adsorption, precipitation and enhance microbial activities) and are relatively low cost, can facilitate the revegetation of contaminated soil. Moreover, it is the useful option to integrate organic waste management and disposal with soil remediation. When chicken manure was used as the immobilizer in Pb-spiked soil, Pb concentrations in the brain, kidneys, and lungs at day 30 and the brain, kidneys, liver, and bone also showed a similar reducing phenomenon on day 98. This result highlights that the use of chicken manure decreased Pb accumulation in tissues at 30 days and 98 days. This decline in Pb accumulation then eased the body's burden by triggering antioxidant genes in the brain, particularly HMOX1, SOD1, and CAT at 98 days.

Chapter 4: Assessing the Efficacy of Phosphate and Lime Amendments in Immobilizing various form of lead in Contaminated Soil: A vivo study on C57BL/6J Mice simulating realistic environmental exposure pathway

In chapter 4 of this dissertation, various Pb species ($2PbCO_3 \cdot Pb(OH)_2$, $PbSO_4$ and PbS) were used to investigate the accumulation of different Pb species in mice model as Pb exists in different forms around the mining and smelting environment. XANES analysis revealed that Pb-carbonate form is dominated in $2PbCO_3 \cdot Pb(OH)_2$-spiked soil and $PbSO_4$-spiked soil. It also revealed that Pb speciation was changed by the amendments added into different form of Pb-spiked soil. Mice exposed to $2PbCO_3 \cdot Pb(OH)_2$-spiked soil exhibited the highest Pb accumulation in the examined tissues except in the lungs where PbS-spiked soil exposed group accumulated more than others. In comparing the effectiveness of phosphate (TSP and PA) and liming (CD) amendments in various Pb-spiked soils under environmentally realistic exposure condition, TSP emerges as the most effective chemical immobilizer among the three

tested amendments. However, different amendments worked differently on various form of Pb.

Chapter 5: General Conclusion

Taken together, these findings highlighted the ongoing environmental pollution in Kabwe, Zambia and emphasizes the need for remediation efforts. Specifically, the use of organic soil amendments, such as chicken manure, was proposed to mitigate Pb mobility accompanied by activation of antioxidant genes in the brain of mouse model. The study also showed the difference in tissue accumulation when the specific Pb species existed in the environment alongside $2PbCO_3 \cdot Pb(OH)_2$ causes more burden in the mammalian body than other Pb species. The study evaluated different soil amendments' efficacy in the immobilization of Pb in different Pb-spiked soils, with TSP identified as the most effective chemical stabilizer while CD emerged as a promising candidate with $PbSO_4$-spiked soil. As we move forward, it is crucial for policymakers, businesses, and local communities work together to prioritize mitigating Pb pollution in these areas. By doing so, we can protect our environment, vulnerable communities and promote sustainable development in mining areas.