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Rice plant tolerance response to the exposure of heavy metals and its Impact on Plant Overall Morphological and Histological Features

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ABSTRACT

Heavy metals bioaccumulation in the food supply chain is a critical environmental issue, required drastic action plans for health risk reduction. In this study, we aim to determine the effects of heavy metal exposure on the morphology and anatomy of paddy plant (*Oryza sativa* L.). Paddy plants were treated with single metal of Cd, Zn and Fe, while the combined metals were Cd+Zn, Cd+Fe and Zn+Fe, each at 5 different concentrations on 21st day of the germination. The results showed that all single and combined heavy metals upon solo and multiple exposures have a significant effect to the plant growth, where heavy metals influence root length, stem length, tree height, number of leaves, number of tillers, number of root hairs and total rice yield. The heavy metal content in paddy plants were in the order of root>stem>grain>leaf. CO_2 influences the growth of rice plants, but the effect of heavy metal exposure has influenced the rate of growth. A positive correlation was obtained between single exposure and double exposure in soil and paddy plant. The accumulation of single and combined heavy metals in greenhouses and CO_2 greenhouses alters the cell structure and tissues of rice plants. Our findings also suggested that paddy plant has the ability to regulate its internal structure and adapt to high accumulation of heavy metals in plant structure, seemed to have some degree of tolerance to heavy metals.

Key words: Paddy plant, Metal exposure, Soil pollution, Tolerance

Introduction

Soil pollution is determined by the presence of materials in the soil that really are detrimental to human health when their concentration levels exceed certain thresholds. Heavy metals remain in the soil for long periods of time even in small quantities and subsequently seep into plant roots, affecting the food chain Rice is one of the world's three main food crops, and it is consumed by about half of the global population. Paddy is a significant food crop for more than 3.5 billion people worldwide. Paddy is a staple food and a major source of nutrients for half of the world's population (Fukagawa and Ziska, 2019). Heavy metal pollution and increasing CO_2 concentrations in the environment have become

major concerns in the world as these two factors can affect the safety of the main food of the Malaysian population, namely rice (Turkyilmaz et al., 2019). In addition to the issue of heavy metal pollution, global climate change is a major concern now a days. The concentration of carbon dioxide (CO_2) in the atmosphere has recorded the highest record in 2019 which is 409.8 ± 0.1 ppm which shows an increase of 2.5 ± 0.1 ppm from 2018, (Arguez *et al.*, 2020). The concentration of carbon dioxide (CO₂) per capita in Malaysia has increased and is predicted to reach 15.5 tonnes per capita by 2030 which is a very worrying climate change. So, a comprehensive study related to heavy metals and global climate change needs to be given full attention for the sake of the sustainability of food production which in turn has an impact on food security.

Materials and Method

Muda Agriculture Development Authority (MADA) Kedah provided paddy seeds (Oryza sa*tiva*), Malaysia was cultivated in an Open CO₂ Greenhouse (Open Roof Ventilation System). Plant House Complex, University Kebangsaan Malaysia (UKM), under a covered structure and a transparent plastic roof. The planting method in this study consists of several steps namely soil preparation, sowing and germination of seeds, the step of transferring the seedlings into a temporary reservoir and then the step of transferring seedlings into containers. Heavy metal exposure to paddy plants was done once on the 21st day. Seeds were soaked in water for 24 hours before sowing, then drained and permitted to germinate before white root or radical emerged from the seeds. On the day 14th of planting, the single metal exposures are Cd, Zn and Fe while the combined metals are Cd+Zn, Cd+Fe and Zn+Fe. The metal treatment consisted of Cd as CdCl₂, Zn as $ZnSO_4$ and Fe as $FeSO_4$. Metal treatments are at 5 different concentrations namely for Cd (ppm): 5, 10, 15, 20 and 25, Zn (ppm): 150, 200, 250, 300 and 350 and Fe (ppm): 200, 250, 300, 350 and 400. Wet digestion method was used for the determination of heavy metals in rice plants. The ICP-MS (Inductive Coupled Plasma Mass Spectrometer) was used to calculate the concentration of heavy metals (ELAN 9000, Perkin Elemer SCIEX). The data obtained were collected, compiled and analyzed using Microsoft Excel and SPSS 15.0 software. The Duncan test (at the significance level of $\alpha = 0.05$) was used to study the differences in the effects of study metal exposure at different concentrations along with controls. P <0.05 was the relationship considered statistically significant.

Results and Discussion

The results in this study showed significant effects of single exposures and double exposures to the growth of paddy plants. Leaf chlorosis, decreases of roots, tiller number, number of leaf, number of root hair and shoot height of the paddy were detected after single and double exposures of heavy metals. The heavy metal content in paddy plants were in the order of root>stem>grain>leaf. A positive correlation was obtained in this study between single exposure and double exposure in soil and paddy plant. Bioaccumulation Factor (BF) grains/soil for combine metals are higher compare to single metal exposure in Open Roof CO₂Glass house. BF_{grains/soil} are in order: Fe>Zn>Cd (Figure 1). Translocation Factor (TF) for all metal study are higher in Open Roof CO, glass House compare to glass house and $\mathrm{TF}_{\mathrm{roots/soil}}$ for Cd is $>1TF_{shoot/roots}$ and $TF_{grainss/shoot}$ follow the order: Fe>Zn>Cd (Figure 2). The Bioaccumulation factor (FB) is the ratio of the concentration of heavy metals in plants to their concentration in soil. When FB <1 / FB = 1 means the plant absorbs heavy metals from the soil but does not accumulate it while, when FB> 1 means plants accumulate heavy metals. Translocation Factor (FT) refers to the mechanism and rate of absorption as well as the translocation of heavy metals from soil to plants. If the FT> 1 indicates that Oryza sativa is a plant capable of collecting metal absorbed from the soil. Figure 3 shows the FT for

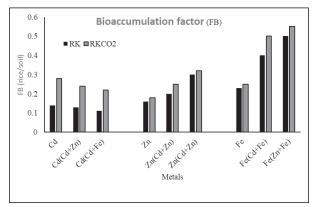


Fig. 1. Bioaccumulation factors (FB) in greenhouse (RK) and carbon dioxide (RK CO₂) glass house experiments

single and combined exposures. Data shows the FT root for metal single and combined Zn and Fe is less than 1 in greenhouses and Open Roof CO_2 glass House. For single metal and combined Cd, the value of FT root is FT = 1 /> 1 in the greenhouses and the Open Roof CO₂ glass House.

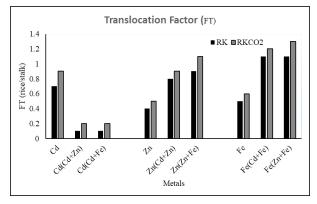


Fig. 2. Translocation Factor (FT) of single and combined heavy metals in greenhouse (RK) and carbon dioxide (RK CO₂) glass house.

The concentration of heavy metals in certain parts of the rice plant was high during the vegetative and reproductive stages while declining during the seeding stages for all study metals in greenhouses and CO, greenhouses. This study showed that the dispersion of Cd, Zn and Fe concentrations in rice plants decreased from root to leaf. The concentration of heavy metals in rice plants is in the order: roots> stems> rice> leaves. The rate of accumulation, absorption and transport of heavy metals in certain parts of the plant varies according to the type of plant species as well as on the same species. Fe metal concentrations were found to be high in the roots, stems, leaves and rice. The concentration of heavy metals in rice is an indicator of food safety because rice enters directly into the human body. The concentration of heavy metals in certain parts of the rice plant has a relationship with each other. The relationship of heavy metal concentrations is either antagonistic or synergistic. The findings of the study found that there is a synergistic relationship between the metals Cd with Zn and Cd with Fe. The concentration of Fe in the roots decreases as the exposure concentration of Cd metal increases. These results indicate that Cd is more easily utilized by crops than Zn and Fe. The concentration of heavy metals in the soil decreases after a certain period of time however, the concentration of Zn and Fe reaches its highest level when the rice crop is in the breeding phase. FB (rice/soil) refers to the content and accumulation of metals in tissues. Based on the results of the study, it was found that the FB (rice/ soil) for all study metals was less than 1 for the greenhouse and CO₂ greenhouse experiments. FB (rice/soil) for the combined metal was higher than the single metal for all study metals. for Fe and Zn are higher in root than Zn and Fe. These results indicate that soil is a reservoir of metal pollution and Cd is a metal that is more easily absorbed by rice plant roots. The FT for Zn and Fe for single and combined exposures were <1 and the influence of CO₂ influenced its increase in the root part. It can be concluded that the type of metal and the influence of CO₂ influence the rate of metal absorption in the root part. The presence and interaction of single metals (Cd, Zn and Fe) and combined metals (Cd+Zn, Cd+Fe and Zn+Fe) influence metal dispersion in soil, with Cd had higher absorption capacity than Zn and Fe. Rice/soil Bioaccumulation factor (FB) for Fe metal is higher than Zn and Cd, while for the combined metal it was higher than the single metal for all study metals. Root/soil Translocation Factor (FT) showed a value> 1 for Cd followed by Zn and Fe, while for stems/roots were in the order Fe> Zn> Cd. The accumulation of single and combined heavy metals in greenhouses and CO2 greenhouses alters the cell structure and tissues of rice plants. Cell wall thickening, cell destruction, increase in the number of elements in the cylindrical tube and fragility of aerenchyma cells occur in the root cells of rice plants.

Conclusion

Soil pollution is worsening day by day, leads to heavy metals bioaccumulation in the food supply chain, threatening human's health and required drastic action plans. Results from this study stipulated that paddy plant has the ability to regulate its internal structure and adapt to high accumulation of heavy metals in its plant. All single and combined heavy metals have a significant effect on rice plant growth.

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