The Effect of Pb Sourced Application on the Growth and Yield of Vegetable Crops

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ABSTRACT
Pb input into agricultural land in addition to the natural geological activities, can also be produced from anthropogenic activities such as the application of organic matter, fertilizers and the use of pesticides. Although Pb is not an essential element for plants, plants can absorb it from the soil and accumulate it in plant parts in various concentrations. This study aims to determine the effect of the use of production inputs in the form of fertilizers and pesticides containing Pb on the growth and yield of vegetable crops. The research method used is Split Plot Design, as the main plot namely the source application of Pb (A) consists of 4 levels (A1: pesticide (99 mg Pbkg-1); A2 = fertilizer (21 mg Pb/kg); A3 = Pb(NO3)2 (50 mg kg-1) and A4 = control (without Pb input) and as a subplot namely type of vegetable (B) consists of two levels chinese cabbage(B1) and bean (B2). In general, the results show that the application of Pb sources from the use of fertilizers and pesticides has no significant effect on plant height, number of leaves, fresh weight, and total dry weight of the chinese cabbage and bean. Application of different Pb sources affects on the total chlorophyll of the chinese cabbage and bean vegetable plants. Chinese cabbage plants that applied Pb source from pesticides produced higher total chlorophyll (17.21 mg g-1), on the contrary, on the bean plants total chlorophyll that applied to the Pb source from pesticides produced the lowest total chlorophyll (4.66 mg g-1). The shoots of Chinese cabbage which applied Pb from pesticides accumulate Pb equal to 0.445 mg per dry weight (concentration of 38.63 mg Pb kg-1) higher than the accumulation of Pb on the control Chinese cabbage shoot. The Pb concentration that exceeds the safe limit for food from vegetables equal to 0.5 mg kg-1, needs to be a serious concern to be minimized.

Keywords: fertilizer, pesticide, Pb, vegetables

I. INTRODUCTION
Lead (Pb) has the potential to be deposited in agricultural soils, because in addition naturally can be produced from geological sources namely the result from weathering of host rock, its deposition can also be generated from anthropogenic activities such as the contamination of various industrial wastes around agricultural areas as well as the results of air pollutant distribution that containing lead (Pb) from the rest of the vehicle's fuel disposal which can reach the ground (deposited in the soil) and even spread. Pb input into agricultural soils can also be significantly derived from the application of organic matter and contaminants in fertilizers, so that the highest metal concentrations can generally be found in the surface layer of the soil profile [1].

The results of the initial survey in five villages in the agricultural fields of vegetable production centers in the subdistrict of Ponokusumo, Malang regency, indicates the presence of Pb metal content. The results of the Pb content analysis on soil samples taken in a composite manner in each village showed a total Pb range between 7.86-10.20 mg kg-1. Heavy metals contained in agricultural lands are generally relatively within the limit that is not dangerous but can be dangerous if heavy metals which becomes soil contamination, through its transfer from soil to plants and then enter the metabolic system of living things in amounts that exceed the threshold. Inclusion of metals can occur indirectly when foodstuffs such as vegetables are consumed. Although Pb is not an essential element for plants, plants can absorb it from the soil and accumulate it in edible parts in various concentrations. The amount absorbed in parts that can be eaten by humans is the most important thing for Pb soils that endanger humans. The accumulation of metal, toxicity and the presence of heavy metals in agricultural soils became an interesting topic to be studied extensively in soil science and the environment because activities in agricultural lands are very intensive in the use of fertilizers and pesticides[2]. The influence of the use of fertilizers and pesticides on the Pb concentration so far has been revealed, especially for agricultural lands and their absorption in plants through the soil, while the use of Pb sources in a foliar manner has not been widely revealed. For this reason, this experiment was carried out with the aim to find out how much influence the application of Pb sources in a foliar and through the soil to the accumulation of Pb in vegetable crops. Another consideration is that the
excess accumulation of heavy metals in the soil if absorbed by plants in high quantities can have toxic effects on humans and animals that eat them. So it is necessary to know how much the absorption potential level of Pb in vegetable crops, will determine whether vegetables are safe for consumption.

II. METHODS

Location and Time of Research

The plastic house experiment was carried out in the horticultural land of Wonorejo village, Poncokusumo subdistrict, Malang regency in July to December 2018. The research location is at an altitude of 717 m above sea level, with an average temperature of 23.5°C, average monthly rainfall ranges from 137 mm.

Treatment and experimental Design

The design used in this experiment is Split Plot Design, with details, as the main plot is the Pb application from several sources that contain Pb (A) consists of 4 levels: A1 = Application of Pesticides (Pb content of 99 mg kg⁻¹); A2 = Fertilizer Application (Pb content of 21 mg kg⁻¹); A3 = Application of Pb from Pb(NO₃)₂ (Pb content of 50 mg kg⁻¹); and A4 = Control (Without Pb applications from outside).

As a subplot is a type of vegetable plant (B) namely B1 = Leaf vegetables (Chinese cabbage) and B2 = Fruit vegetables (Beans). Each treatment combination was repeated three times, with plant samples for each treatment combination taken 10 plants. So that there are 240 units of sample experiments.

Data Analysis

To find out the effect of Pb contamination sources on growth, yield and Pb concentration in plant organs, conducted an analysis of variance (ANOVA) with the Anova table for Split Plot Design. If the F calculate is greater than the F table, it shows there is a real influence, and the average results then compared with the Tukey’s Honest Significant Difference at the level of 5%.

III. RESULTS AND DISCUSSION

The results of the analysis of variance showed that the application of different Pb sources did not show any significant effect on plant height and number of leaves (Table 1 and Table 2). Up to a certain concentration level, Pb does not endanger plant growth, so that plants are able to produce biomass normally.

Although it did not show any real effect, however based on Figure 1 and Figure 2, it was seen that the plants of Chinese cabbage and beans that were applied to higher Pb namely those from Pb(NO₃)₂ produced plant height and number of leaves lower than other treatments, while on the control plants (without the application of Pb from the outside) showed better growth than other treatments. According to Sala et al [7], some heavy metals such as Zn, Cu, Mn, Ni, Se, Co, Cr, and Mo are indeed involved in important biological functions, so they have positive biological effects on plant productivity. However, some metals are known to have no important metabolic functions such as Zr, Sb, As, Pb, Hg, and Cd so they can reduce plant productivity especially if the concentrations of these metals are in conditions of increasing until it reaches the suboptimal value 8,9,10,11.

The results of this study also showed that the application of Pb sources could reduce total plant chlorophyll (Table 3). In Chinese cabbage plants which were applied Pb from Pb(NO₃)₂ and phosphate fertilizer produced lower total chlorophyll compared to Chinese cabbage plants which were applied Pb from pesticides and controls. While in the bean plants that were applied to Pb from the source of Pb(NO₃)₂ and pesticides produced lower total chlorophyll compared to control. This is in line with what was revealed by Pourrut et al. [12] that exposure to heavy metals can reduce plant growth and biomass, and in extreme cases cause plant death. Heavy metals can also cause disorganization of grana structures, reduce root and shoot growth, reduce chlorophyll biosynthesis, disrupt respiration and photosynthesis.

| Pb Sources | Average Height of Plants on days after planting (dap) | Tukey’s HSD 5% | Vegetable Type |
|-------------|-----------------------------------------------------|----------------|----------------|
|             | 14  | 21  | 28  | 35  | 42  | 49  | 56  | 63  |            |               |               |
| A1 (pesticide) | 25.28 | 51.89 | 76.23 | 104.25 | 119.29 | 130.72 | 135.66 | 184.60 | a       | ns          | ns          |
| A2 (fertilizer) | 23.54 | 56.70 | 88.93 | 118.41 | 134.39 | 144.29 | 147.75 | 185.91 | a       | ns          | ns          |
| A3 (Pb(NO₃)₂) | 21.75 | 55.21 | 90.29 | 120.69 | 133.73 | 140.64 | 147.05 | 173.57 | a       | ns          | ns          |
| A4 (control)  | 25.80 | 60.64 | 92.14 | 122.92 | 135.69 | 142.54 | 150.18 | 182.17 | a       | ns          | ns          |
| Tukey’s HSD 5% | ns  | ns  | ns  | ns  | ns  | ns  | ns  | ns  |            | ns          | ns          |

Table 1. Average Plant Height (cm) in the provision of different sources of Pb and types of vegetable plants

Information: the numbers followed by the same letters in each of the same columns show no significant difference based on the test of Honest Significant Difference at the 5% level.
Figure 1. Comparison of plant height in Chinese cabbage and bean plants due to the application of Pb from several different Pb inputs. A1= Pesticide (99 mg Pb kg\(^{-1}\)); A2= fertilizer (21 mg Pb kg\(^{-1}\)); A3= Pb(NO\(_3\))\(_2\) (50 mg Pb kg\(^{-1}\)); A4= control (without Pb)

Figure 2. Comparison of number of leaves in Chinese cabbage and bean plants due to the application of Pb from several different Pb inputs. A1= Pesticide (99 mg Pb kg\(^{-1}\)); A2= fertilizer (21 mg Pb kg\(^{-1}\)); A3= Pb(NO\(_3\))\(_2\) (50 mg Pb kg\(^{-1}\)); A4= control (without Pb)

Table 2. Average Number of Leaves in the Provision of different Pb sources and types of vegetable plants

| Pb Sources       | Observation Average Number of Leaves on days after planting (dap) | 14 | 21 | 28 | 35 | 42 | 49 | 56 | 63 |
|------------------|-------------------------------------------------------------------|----|----|----|----|----|----|----|----|
| A1 (pesticide)   |                                                                   | 4.20 | a  | 8.03 | a  | 12.07 | a  | 15.94 | a  | 20.00 | a  | 22.32 | a  | 23.68 | a  | 24.60 | a  |
| A2 (fertilizer)  |                                                                   | 3.73 | a  | 7.43 | a  | 11.75 | a  | 16.73 | a  | 20.40 | a  | 22.78 | a  | 24.37 | a  | 26.93 | a  |
| A3 (Pb(NO\(_3\))\(_2\)) |                                                               | 3.90 | a  | 7.62 | a  | 11.80 | a  | 15.99 | a  | 20.25 | a  | 22.65 | a  | 24.36 | a  | 25.68 | a  |
| A4 (control)     |                                                                   | 3.85 | a  | 7.65 | a  | 12.20 | a  | 16.70 | a  | 21.68 | a  | 24.84 | a  | 26.25 | a  | 26.90 | a  |
| Tukey’s HSD      |                                                                   | ns  | ns  | ns  | ns  | ns  | ns  | ns  | ns  |
| Vegetable Type   |                                                                   | ns  | ns  | ns  | ns  | ns  | ns  | ns  | ns  |

The numbers followed by the same letters in each of the same columns show no significant difference based on the test of Honest Significant Difference at the 5% level
Table 3. Total chlorophyll of Chinese cabbage and bean plants due to Pb application from several different inputs.

| Treatment | Total Chlorophyll (mg g⁻¹) | Vegetable Type |
|-----------|----------------------------|----------------|
| Pb source application | B1 (Chinese cabbage) | B2 (bean) |
| A1 (Pesticide 99 mg Pb kg⁻¹) | 17,21 ± 4,68 b | 4,66 ± 0,31 a |
| A2 (Fertilizer 21 mg Pb kg⁻¹) | 7,42 ± 0,85 ab | 9,15 ± 4,48 ab |
| A3 (Pb(NO₃)₂ 50 mg kg⁻¹) | 7,50 ± 1,17 ab | 6,33 ± 0,20 ab |
| A4 (kontrol) | 12,23 ± 3,60 ab | 7,63 ± 4,81 ab |
| BNJ 5% | | 11,33 |

The data presented are means ± SD of three replications; the numbers accompanied by the same letters in the same row and column show no significant difference based on the test of Honest Significant Difference at the 5% level.

In general, the application of Pb provision from several Pb sources in this study also did not significantly affect crop yield or production. In Figure 4 biomass (dry weight) of Chinese cabbage and beans plants showed no significant difference between the control plants and plants which applied Pb sources from pesticides, fertilizers and Pb(NO₃)₂. Likewise, the average crop production shown in the fresh weight of canopy from Chinese bean plants and the fresh weight of fruit on green beans showed the same tendency, where the control plants and plants that were applied to Pb from some external inputs did not show any significant differences (Figure 5). This is presumably because the Pb metal concentration in the media is still in a condition that is not too extreme so it does not reduce plant productivity. Soil analysis results showed the average initial Pb concentration in soil media was in the range of 10 mg kg⁻¹. By contrast, if the concentration of Pb in the soil reaches a supraoptimal value, then it can reduce plant productivity [8].

Although Chinese cabbage and beans plants in this study can grow and produce well as control plants, however, the application of Pb from several inputs has a significant influence on the concentration and accumulation of Pb in each organ. Table 4 shows that in Chinese cabbage plants, Pb application from pesticides, fertilizers and Pb(NO₃)₂ accumulate Pb greater than control plants. Similarly, the bean plants that applied Pb sources from pesticides, fertilizers and Pb(NO₃)₂ accumulate Pb greater than the control plants. This is also made clear in Figure 6 and Figure 7 for the results of the accumulation of Pb and the Pb concentrations in each of the canopy of Chinese cabbage and bean as well as bean fruit. In plant growth, heavy metals begin to influence when they enter plant tissues as nutrients enter from the soil solution [13]. But before that, the ability of roots to absorb water and nutrients was known to decrease under conditions of metal stress [14]. Furthermore, heavy metals that enter the plant tissue will affect plant physiology, including photosynthesis, formation of chlorophyll, absorption of nutrients and the presence of oxidative stress [15]. However, according to Appenroth [16], the effects of heavy metals on photosynthesis still not very clear.

Heavy metals are very vulnerable contaminate agricultural lands, because soil is a basic and important part of an ecological system [17]. The entry of heavy metals from the soil into the roots of plants, indirectly begins by being absorbed by the roots followed by binding to the carboxyl uronic acid group around the roots, or directly to the polysaccharide sap from the surface of rhizodema cells [18]. Some of the processes involved in the absorption of metals by plants include absorption of metals from soil particles, transport of metals to plant roots, absorption of metals by roots and metal translocation to shoots [19].
Table 4. Pb accumulation per dry weight of Chinese cabbage and beans due to the application of Pb from several different inputs

| Treatment                      | Pb accumulation (mg per plant dry weight) | Vegetable Type |
|--------------------------------|-------------------------------------------|----------------|
| Pb source application          |                                           |                |
| A1 (Pesticide 99 mg Pb kg\(^{-1}\)) | 0,45±0,07 a                               | B1 (Chinese cabbage) |
| A2 (Fertilizer 21 mg Pb kg\(^{-1}\)) | 0,25±0,09 a                               | B2 (Bean)       |
| A3 (Pb(NO\(_3\))\(_2\) 50 mg kg\(^{-1}\)) | 0,42±0,19 a                               |                |
| A4 (control)                   | 0,18±0,03 a                               |                |
| Tukey’s HSD 5%                 | 0,48                                      |                |

The data presented are means ± SD of three replications; the numbers accompanied by the same letters in the same row and column show no significant difference based on test of Honest Significant Difference at the 5% level.

Figure 5. Comparison of fresh weight in the shoots of Chinese cabbage and bean, as well as bean pod as a result of the application of Pb from several different Pb inputs. A1= Pesticide (99 mg Pb kg\(^{-1}\)); A2= fertilizer (21 mg Pb kg\(^{-1}\)); A3= Pb(NO\(_3\))\(_2\) (50 mg Pb kg\(^{-1}\)); A4= control (without Pb).

Figure 6. Comparison of the accumulation of Pb in the shoots of Chinese cabbage and bean, as well as bean pod as a result of the application of Pb from several different Pb inputs. A1= Pesticide (99 mg Pb kg\(^{-1}\)); A2= fertilizer (21 mg Pb kg\(^{-1}\)); A3= Pb(NO\(_3\))\(_2\) (50 mg Pb kg\(^{-1}\)); A4= control (without Pb).

Figure 7. Comparison of the concentration of Pb in the shoots of Chinese cabbage and bean, as well as bean pod as a result of the application of Pb from several different Pb inputs. A1= Pesticide (99 mg Pb kg\(^{-1}\)); A2= fertilizer (21 mg Pb kg\(^{-1}\)); A3= Pb(NO\(_3\))\(_2\) (50 mg Pb kg\(^{-1}\)); A4= control (without Pb).

Heavy metals are generally transported apoplastisally in plant tissue, and their translation into the upper part of the plant via xylem vessels is most likely driven by transpiration of water through leaves [20]. To reach the xylem vessels from the roots, the metal must cross the endodermis and the Casparian band. As a result, a lot of metal absorption is carried out by younger roots where the Casparian band is not fully developed. After penetration into the central cylinder, the metal moves towards the top of the plant through the flow of water from the vascular tissue system [21], where water evaporates and metals accumulate. In xylem, metals such as Pb and Cd are bound to amino acids and form complexes [22-24].

The presence of metal translocation restriction, common in Pb compared to other metals. Limitation of heavy metal translocation to the top of the plant is related to accumulation in the plasma membrane [25], blocking by a casparian band, precipitation in an intercellular space [18], precipitation as insoluble metal salts in cell walls [26]-[30], or accumulation in corticol vacuoles and rhizoderm cells [31-32].

IV. CONCLUSION

The application of Pb sources from the use of fertilizers and pesticides has no significant effect on plant height, number of leaves, fresh weight, and total dry weight of the Chinese cabbage and bean. Application of different Pb sources effect on the total chlorophyll of the Chinese cabbage and bean vegetable plants. Chinese cabbage plants which applied Pb source from pesticides produced higher total chlorophyll (17,21 mg g\(^{-1}\)), on the contrary, on the bean plants total chlorophyll that applied to the Pb source from pesticides produced the lowest total chlorophyll (4,66 mg g\(^{-1}\)). The shoots of chinese cabbage which applied Pb from pesticides accumulate Pb equal to 0,445 mg per dry weight (concentration of 38,63 mg Pb kg\(^{-1}\)) higher than Pb concentration in the control chinese cabbage shoot. While beans from pesticide applications accumulate Pb of 1,17 mg per dry weight (concentration of 42,17 mg Pb kg\(^{-1}\)) higher than beans of control plants and Pb application from fertilizer. Pod beans as edible part of bean plant from Pb(NO\(_3\))\(_2\) application, accumulate 0,19 mg Pb per dry weight (concentration of 18,73 mg Pb kg\(^{-1}\)) higher than the others. The Pb concentration that exceeds the safe limit for food...
from vegetables equal to 0.5 mg kg⁻¹, needs to be a serious concern to be minimized.

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