COMBINED APPLICATION OF FERMENTED BAMBOO (*Bambusa spinosa*) AND MOLLUSK (*Achatina fulica*) LIQUID FERTILIZER CAN IMPROVED LETTUCE (*Lactuca sativa* var. CURLY GREEN) PRODUCTION

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ABSTRACT

Giant bamboos (*Bambusa spinosa*) are abundant along the Philippine countryside that contains carbohydrates, protein, and mineral. Meanwhile, the Japanese snail (*Achatina fulica*) is a mollusk pest found in most gardens in the Philippines. By increasing soil organic matter, organic farming can reinstate the damaged soil's natural fertility, which will improve crop productivity like in leafy vegetables such as lettuce (*Lactuca sativa* var. Curly Green). Four treatments (T0: commercially available liquid fermentation-based fertilizer, T1: fermented bamboo shoot-based liquid fertilizer, T2: fermented Japanese snail-based liquid fertilizer, and T3: Combination of both bamboo and Japanese snail-based liquid fertilizer) in three replications were conducted to determine their effects on the weekly height, leaf length and above and below ground fresh weight of lettuce. The recommended rate (10ml/L) for commercial liquid fertilizer was followed for all treatments. For the erectness of leaves (plant height) and length of leaves, the combined use of bamboo and Japanese snail-based liquid fertilizer is generally comparable to using commercially available liquid fermentation-based fertilizer. At termination, the combined application of Japanese snail and bamboo shoot liquid fertilizer was 13.88±0.51SE cm high and had leaves 15.78±0.78SE cm long. Lettuce applied with the commercial liquid fertilizer was 14.01±1.11SE cm high, with leaves measuring 15.75±0.86SE cm long. All treatments have approximately four fully opened leaves 20 days after transplanting. For above and below ground fresh weight, lettuce applied with fermented

bamboo shoot-based liquid fertilizer alone performed the least after four weeks at 80±4.47SE and 39.33±2.89SE g/plant, respectively. These suggest that the combined application of bamboo shoot and Japanese snail has the potential to be used as liquid fermented fertilizer for organic lettuce production.

Keywords: Bamboo shoot; fermentation-based fertilizer; Japanese snail; leafy vegetable; organic agriculture.

INTRODUCTION

Lettuce is widely cultivated across different countries due to its innumerable benefits, which includes development and regulation of the body brought about by its high vitamin and mineral contents [1].

Owing to their high market demand, mineral fertilization is usually resorted to increase volume production. Despite its advantages, mineral fertilization contributes to increasing soil salinity, decreasing long-term soil fertility, and decreasing soil organic matter and other negative effects on the ecosystems [2]. Organic fertilization contributes to greater availability of nutrients, especially nitrogen, phosphorus, sulfur, and micronutrients, increases the pH, and improves soil physical, chemical, and biological properties [3]. Several studies have demonstrated that organic farming, which strictly prohibits synthetic fertilizers, provides an alternative that has the potential to minimize the negative influence from by using chemical fertilization, and the products from the organic farming systems are generally endowed with improved nutritional properties [4].

Fermented animal and plant-based fertilizers are organic foliar fertilizers. Through foliar application, nutrients become promptly available to plants and are thus considered better than direct soil fertilization [5]. According to Gamuyao et al., [6], bamboo shoot extracts contain the growth regulator gibberellin, which has functions associated to cell division and elongation. Natural growth regulators are an easy alternative to chemical-based plant growth regulators and are relatively inexpensive and safe to use [7]. This also contains carbohydrates, protein, and mineral like Ca, Mg, P, K, Mn and Zn [8]. Furthermore, application of Fermented Swamp Cabbage Juice, a plant-based nutrient source, alone also improved the nutrient contents (N, P_2O_5 and K_2O) of the compost after four and eight weeks of composting [9]. On the other hand, the application of crushed golden snail amino acid together with fermented swamp cabbage juice increased soil microorganisms at 2.5 x 105 cfu/ml [10]. This can be inferred to an improved soil biological property. Its shell also has a chemical composition which includes proteins, carbohydrates, fats, and minerals such as iron, zinc, copper [11].

Considering the potential benefits of bamboo and Japanese snail as liquid organic fertilizer, the relative abundance of bamboo and that Japanese snail is considered an economically important mollusk pest, the study is hereby deemed necessary. Not to mention the need to transition towards organic agriculture, as there are many known ill effects of excessive inorganic fertilizers to the environment.

METHODOLOGY

Collection and Fermentation of Bamboo Shoot Fertilizer

Bamboo shoots were collected at Cebu Technological University Barili Campus farm vicinity. The shoots were washed, then chopped into small pieces for effective fermentation. The samples were rewashed three times to remove the dirt and were added with molasses with a ratio of 1:1. The study, used 5 kg of bamboo shoots and 5 liters of molasses, which was harvested after seven days of fermentation. The fermented bamboo shoot fertilizer was safely kept in a storage container.

Collection and Fermentation of Japanese Snail Fertilizer

The collected Japanese snails were washed three times and then crushed into pieces for the fermentation process. The samples were added with equal weight of molasses. The study used 5 kg of Japanese snail and 5 liters of molasses which

was harvested after seven days of fermentation following the procedure used by Pascual et al., [10]. The fermented Japanese snail fertilizer was safely kept in a storage container. These were compared against a commercial seaweed-based organic fertilizer which is described to also contain natural fermentation extracts of plants / herbs known to have natural insecticidal / fungicidal properties such as neem, jatropha, kakawati, hot pepper, garlic, onions, etc. that doesn't harm the plants, animals and farmers/consumers. The rate of application was based on the commercial liquid fertilizer recommended rate (product label).

Experimental Design and Treatments

The experiment was laid out in Completely Randomized Design with four treatments; commercially available liquid fermentation-based fertilizer, fermented bamboo shoot-based liquid fertilizer, fermented Japanese snail-based liquid fertilizer and Combination of both bamboo and Japanese snail-based liquid fertilizer. Each treatment was replicated three times.

Application of Treatments

All of the treatments were applied to the plant with a four-day interval after planting which was on the 4^{th} day, second at 8^{th} day, third at 12^{th} day, and fourth on 16^{th} day. The recommended rate (10ml/L) for commercial liquid fertilizer was followed.

Data Gathered

The following parameters were gathered in this study: plant height (cm) – measured using a ruler for 20 days with a five-day interval after planting; leaf length – measured using a ruler from the tip of the longest leaf at one end of the leaf to the other end; number of leaves – counted every five days in the whole plant; and above-ground fresh weight and below ground fresh weight – measured using a digital balance right after the plants were removed from the soil, then washed off any loose soil and was wiped off gently with the use of a soft paper towel to remove any free surface moisture.

Data Analysis

The data was subjected to analysis of variance (ANOVA) and further test was done using tukey's test at $p<0.05$ to test for differences between treatment means.

RESULTS AND DISCUSSION

For the whole duration of the study, the commercial liquid fermentation-based fertilizer produced the tallest plant at 14.01±1.11SE cm (Fig. 1). This however, is comparable to all other treatments wherein the combined Japanese snail and bamboo shoot produced 13.87±0.51SE tall plants. Sadikin and Manullang [12] reported that the interaction of the golden snail bio-activator with bamboo shoots significantly improved the average nitrogen content and increased the phosphorus content of the fermented fertilizer. On the other hand, it was also stated that with the combination of golden snail and bamboo shoot which was fermented with the use of molasses produced increased phosphorus than the optimum amount at 0.325. The high nitrogen content also influences the high content of phosphorus, the higher the nitrogen contained, the multiplication of microorganisms reacts with phosphorus thereby increasing phosphorus availability to plants [13]. It is known that nitrogen would significantly affect the growth of the plant. The study of Razaq et al. [14] indicated that both N and P applications significantly enhanced plant height, root collar diameter, chlorophyll content, and root morphology in Acer mono.

Table 1. Organic fertilizer treatments used for the growth of Lettuce (*Lactuca sativa*)

Treatment	
T0	**Commercial Liquid Fermentation-based Fertilizer (10 mL/L)**
T1	**Fermented Bamboo Shoot-Based Liquid Fertilizer (10 mL/L)**
T2	**Fermented Japanese Snail-Based Fertilizer (10 mL/L)**
T3	**$\frac{1}{2}$ Bamboo Shoot Fertilizer (5 mL/L) + $\frac{1}{2}$ Japanese Snail Fertilizer (5 mL/L)**

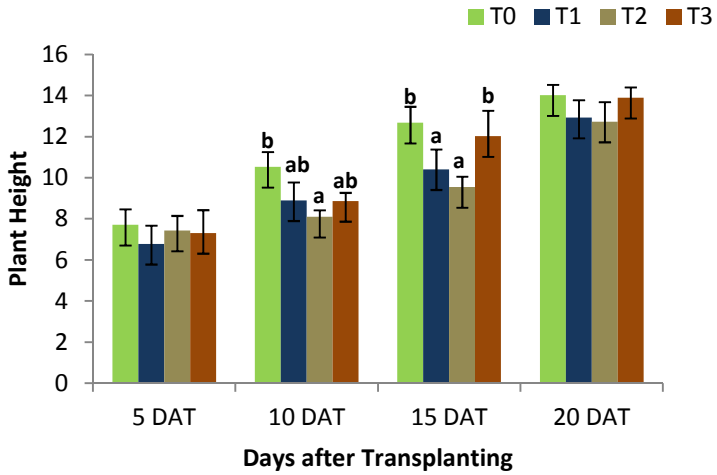

Fig. 1. Effects of bamboo and Japanese snail liquid fertilizer singly and in combination on the plant height of Lettuce (*Lactuca sativa* var. Curly Green). Different superscript letters indicate significant differences (Tukey HSD, α = 0.05)

The result clearly indicated that after 10 DAT, leaf length improved with the addition of combined bamboo and japanese snail-based fertilizer at 11.69±0.47SE cm. Generally, fermented mollusk is amino acid source like fish-based extracts. Ahuja et al., [15] evaluate the potential of production of fertilizers from captured fish which found to contain P and Ca and enhanced growth of radish. Next to nitrogen, phosphorus is a vital nutrient for plant growth and productivity [16]. In treatment three, this showed comparable results with the commercial liquid fermentation-based fertilizer as seen in Fig. 2. Because of the increase of nitrogen content on the combined bamboo and Japanese snail fertilizer, leaf length in lettuce was also increased. In the report of Khan et al., [17], the leaf area of maize was positively influenced by different levels of nitrogen. It was stated that the probable reason for the result might be that N increase vegetative growth. Phosphorus have also taken part in this improvement for in the study of Alahi et al., [18], leaf length increased after the application of the highest dosage of Phosphorus.

The result in Fig. 3 showed that all of the treatments are generally comparable to using the commercially available liquid fermentation-based fertilizer. At 15 DAT, application of fermented bamboo shoot-based liquid fertilizer alone produced 3.64±0.47SE leaves that is comparable to using the commercially available liquid fermentation-based fertilizer at 3.69±0.33SE. This was supported with the study of Ariana et al., [19] which states that gibberellins, commonly found in bamboo shoot, increase cell division and cell enlargement.

Above ground fresh weight improved greatly with the application of fermented japanese snail-based fertilizer at 167.67±4.63SE g/plant followed by the combination of bamboo and japanese snail fermented fertilizer and then the commercial liquid fermentation-based fertilizer at 119.67±4.26SE g/plant and 112.33±4.63SE g/plant, respectively (Fig. 4). The reason behind is that golden snail bio-activator with the addition of bamboo shoots increases the nitrogen level of the fertilizer [12]. According to the study of Chen et al., [20] above-ground biomass was increased by N fertilization in degraded grassland. It was reported that the initial increase in aboveground biomass may be because the added N alleviates ecosystem N limitation and improves the soil nutrient status. Furthermore, since fermented mollusk are possible source of amino acids, it was regarded by Teixeira et al., [21] that plants are capable of uptake of amino acids.

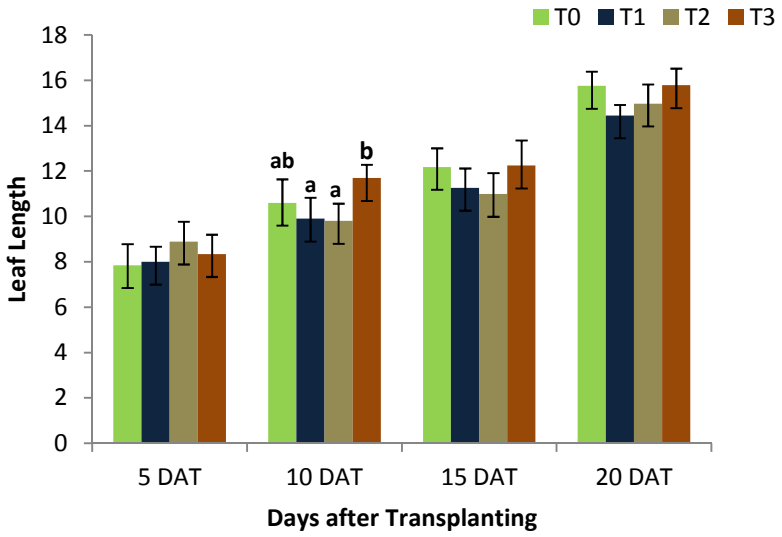

Fig. 2. Leaf length of lettuce (*Lactuca sativa*) as affected by bamboo and Japanese snail fertilizer singly and in combination. Different superscript letters indicate significant differences (Tukey HSD, α = 0.05)

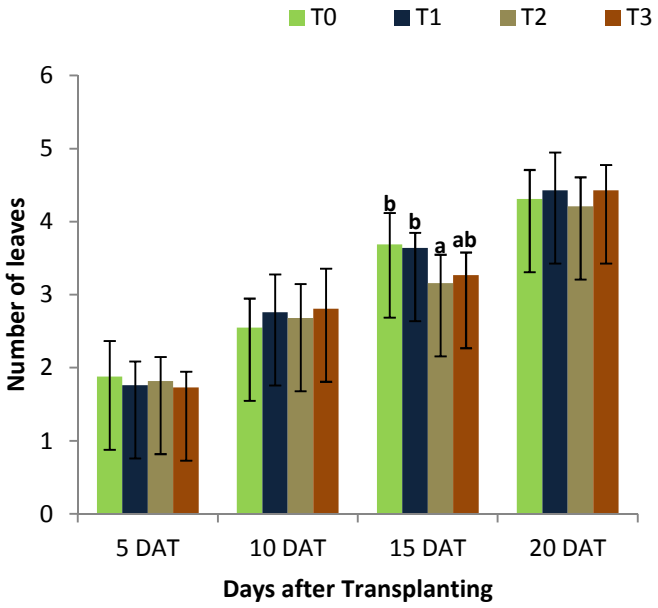

Fig. 3. Number of leaves of lettuce (*Lactuca sativa)* as affected by the application of bamboo and Japanese snail fermented fertilizer singly and in combination. Different superscript letters indicate significant differences (Tukey HSD, α = 0.05)

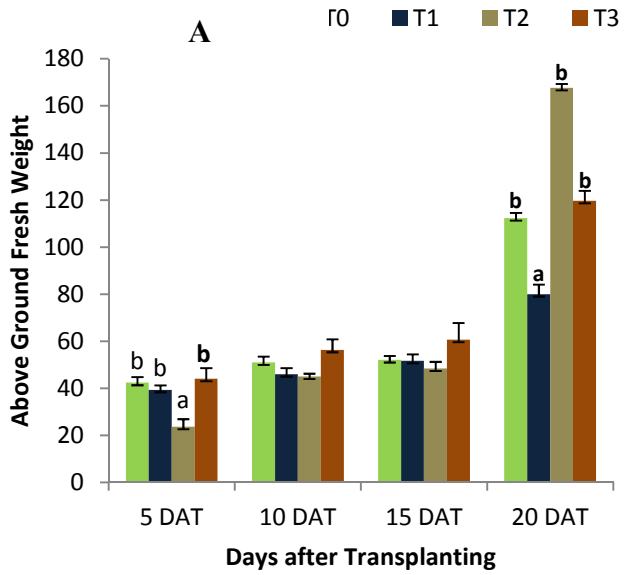

Fig. 4. Above ground fresh weight of Lettuce (*Lactuca sativa*) as affected by bamboo and Japanese snail fermented fertilizer singly and in combination (A-graph, B – appestise). Different small superscript letters indicate significant differences (Tukey HSD, α = 0.05)

As seen in Fig. 5, the combined use of bamboo and Japanese snail-based liquid fertilizer and the fermented japanese snail-based fertilizer alone greatly improved the below ground fresh weight of lettuce at 123.00±5.02SE cm and 117.33±3.91SE cm respectively. This result was supported by Chen et al., [22] where root length and root surface area have 36.06% increased under intermediate N levels. In the study of Kim and Li, [23], the total root length, root surface area, and root volume decreased by reduced P but tended to increase at 7 weeks as P concentration was also raised.

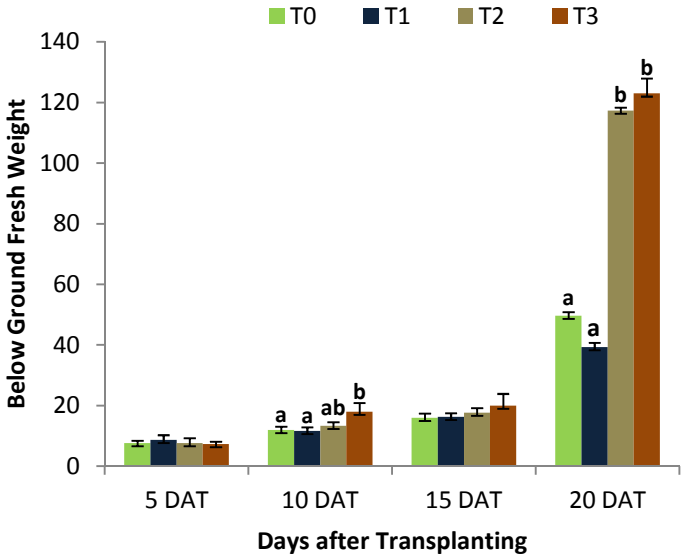

Fig. 5. Below ground fresh weight of Lettuce (*Lactuca sativa*) affected by bamboo and Japanese snail fermented fertilizer singly and in combination. Different small superscript letters indicate significant differences (Tukey HSD, α = 0.05)

CONCLUSION

Both bamboo shoot and Japanese snail-based fertilizer can be applied singly for lettuce production at 10ml/L. For better over-all performance, it should be applied in combination for this improved leaf length and below ground fresh weight. This treatment is also comparable with the commercial liquid fermentation-based fertilizer on the plant height.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Silva MRP, Pinheiro FC, De Paula MT, Prigol M. Parasitological evaluation of lettuce (*Lactuca sativa*) marketed in a municipality of western frontier, RS Brazil Tropical Pathology Magazine. 2015;44(2): 163-169. DOI:https://doi.org/10.5216/rpt.v44i2.36646.
2. Pedrotti A, Chagas RM, Ramos VC, Prata APN, Lucas AAT, Santos PB. Causes and consequences of the process of soil salinization. Electronic Magazine on Environmental Management, Education and Technology. Rev Eletr Gest Educ Technology Ambient. 2015;19:1308-1324. DOI:https://doi.org/10.5902/2236117016544
3. Jayamangkala N, Sutigoolabud P, Inthasan J, Sakhonwasee S. The effect of organic fertilizers on growth and yield of broccoli (*Brassica oleracea* L. var. italica Plenck cv. Top Green). Journal of Organic Systems. 2019;10(1).
4. Ye L, Zhao X, Bao E. et al. Bio-organic fertilizer with reduced rates of chemical fertilization improves soil fertility and enhances tomato yield and quality. Sci Rep. 2020;10:177. Available:https://doi.org/10.1038/s41598-019-56954-2

5. Alshaal T, El-Ramady H. Foliar application: From plant nutrition to biofortification. Environment, Biodiversity and Soil Security; 2017. DOI:10.21608/jenvbs.2017.1089.1006.
6. Gamuyao R, Nagai K, Ayano M, Mori Y, Minami A, Kojima M, Suzuki T, Sakakibara H, Higashiyama T, Ashikari M, Reuscher S. Hormone distribution and transcriptome profiles in bamboo shoots provide insights on bamboo stem emergence and growth. Plant and Cell Physiology. 2017;58(4):702–716. Available:https://doi.org/10.1093/pcp/pcx023
7. Rademacher W. Plant growth regulators: backgrounds and uses in plant production. Journal of Plant Growth Regulation. 2015;34:845-872. DOI: 10.1007/s00344-015-9541-6
8. Karanja PN, Kenji GM, Njoroge SM, Sila DN, Onyango CA, Koaze H, Baba N. Compositional characteristics of young shoots of selected bamboo species growing in kenya and their potential as food source. Journal of Food and Nutrition Research. 2015;3(9):607-612. DOI: 10.12691/jfnr-3-9-8
9. Pet Roey L. Pascual. Improved composting through fermented activators. JPAIR Multidisciplinary Research. 2013;13(1). August. DOI:https://doi.org/10.7719/jpair.v13i1.221
10. Pascual, P., Jarwar, A., and Nitural, P.S. (2013). Fertilizer, fermented activators, and EM utilization in pechay (*Brassica pekinensis* L.) production. Pak. J. Agri., Agril. Engg. Vet. Sci. 29(1):56-69. Available:https://agris.fao.org/agris-search/search.do?recordID=PK2014000384 (ISSN) 1023-1072
11. Jatto E, Imohimi A, Medjor W. Proximate and Mineral Composition of Different Species of Snail Shell. PJTS. 2010;11.
12. Sadikin RA, Manullang RR. Improving the quality of masters of bio- activators in golden apple snail with addition of bamboo shoot. December 2019 Edition. 2019;8(12). Retrieved from Available:http://www.ijstr.org/paper-references.php?ref=IJSTR-1219-27620 (ISSN) 2277-8616
13. Yuli A. Hidayati.et al. The quality of liquid fertilizers produced from Beef Cattle Feces using *Saccharomyces cereviceae*. Journal of Animal Science. 2011;11(2).
14. Razaq, Muhammad, Zhang, Peng, Shen, Hai-long, Salahuddin. Influence of nitrogen and phosphorous on the growth and root morphology of Acer mono. PLOS ONE. 2017;12:e0171321. DOI:10.1371/journal.pone.0171321.
15. Ahuja I, Dauksas E, Remme J, Richardsen R, Løes AK. Fish and fish waste-based fertilizers in organic farming – With status in Norway: A review. Waste Management. 2020;115:95-112. DOI:10.1016/j.wasman.2020.07.025.
16. Tang Z, Xu W, Zhou G, Bai Y, Li J, Tang X, Chen D, Liu Q, Wenhong M, Xiong G, He H, He N, Guo Y, Guo Q, Zhu J, Han W, Hu H, Fang J, Xie Z. Patterns of plant carbon, nitrogen, and phosphorus concentration in relation to productivity in China's terrestrial ecosystems. Proceedings of the National Academy of Sciences. 2018;115:4033-4038. 10.1073/pnas.1700295114.
17. Khan A, Munsif F, Akhtar K, Afridi M, Ahmad Z, Fahad S, Ullah R, Faheem A, Khan I, Din M, River Y. Response of fodder maize to various levels of nitrogen and phosphorus. American Journal of Plant Sciences. 2014;05. DOI: 10.4236/ajps.2014.515246
18. Alahi A, Hossain, Mohammad, Kabir, Khairul, Shah Jahan, Mohammad, Arefin SM, Anamul, Hosain Md. Effect of phosphorus and plant spacing on the growth and yield of lettuce. 2014;2:1-7. DOI:10.15192/PSCP.AAB.2014.2.1.17.
19. Ariana E, Wicaksono FY, Irwan AW, Nurmala T, Yuwariah Y. Pengaruh berbagai pengaturan jarak tanam dan konsentrasi gibberellin (GA3) terhadap pertumbuhan dan hasil tanaman gandum (*Triticum aestivum* L.) kultivar dewata di dataran medium Jatinangor [The Effect of Various Plant's Range and Gibberellin Concentration (GA), on The Growth and

Result of wheat (*Triticum aestivum* L.) dewata's cultivar in medium flatland Jatinangor] Journal of Agricultural Science. 2015;2(1):31–52
DOI: doi:10.1088/1755-1315/454/1/012170

20. Chen J, Dong C, Yao X, Wang W. Effects of nitrogen addition on plant biomass and tissue elemental content in different degradation stages of temperate steppe in northern China. Journal of Plant Ecology; 2017.
DOI: https://doi.org/10.1093/jpe/rtx035
21. Teixeira W, Fagan E, Soares L, Soares J, Reichardt K, Dourado-Neto D. Seed and foliar application of amino acids improve variables of nitrogen metabolism and productivity in soybean crop. Frontiers in Plant Science. 2018;9:396.
DOI:10.3389/fpls.2018.00396.
22. Chen J, Liu L, Wang Z, Zhang Y, Sun H, Song S, Bai Z, Lu Z, Li C. Nitrogen fertilization increases root growth and coordinates the root–shoot relationship in cotton. Frontiers in Plant Science. 2020;11:880.
DOI:10.3389/fpls.2020.00880.
23. Kim H, Li X. Effects of phosphorus on shoot and root growth, partitioning, and phosphorus utilization efficiency in lantana. Hort Science. 2016;51:1001-1009.
DOI:10.21273/HORTSCI.51.8.1001.