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The Effect of Eco-enzymes and Husk Charcoal on the Growth of Vanilla Seeds

H. Haryuni[1], M.N. Wahid[1], S. Priyadi[1], A.F. Aziez[1], I.E. Maryanti[2]

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ABSTRACT

Background: The successful growth and development of the vanilla plant are supported by the availability of soil nutrients that are absorbed by the plant roots. Natural fertilizers derived from plant waste easily decompose in the soil so that they are more quickly absorbed by roots and stored in plant cells. The purpose of this study was to determine the effect of eco-enzymes and husk charcoal on the growth of vanilla seedlings.

Methods: This study was designed using a complete randomized block design. The first factor was eco-enzyme (without eco-enzyme and with eco-enzyme 15 ml/plant), namely E0 and E1, the second factor was the husk charcoal dose (0, 5, 10, 15) g/plant, P0, P1, P2, P3. Data were analyzed using ANOVA analysis of variance with Duncan's multiple range test (DMRT) with a significant difference of 5%. The results showed that the best vanilla growth was obtained by treating it with 15 ml/polybag eco-enzyme and 15 g/polybag coconut shell charcoal (E1P3).

Result: The plant height reaches 79.67 cm, the number of leaves reaches 12.67 strands and the width reaches 405.42 mm. The fresh weight of the plants reached 71.33 g, the dry weight of the plants reached 28 g, the fresh weight of the roots reached 8.6 g, and the dry weight of the roots reached 1.27 g. Microscopi observation showed that eco enzymes and coconut shell charcoal infected the roots and filled the tissues, then played a role in increasing the growth of the vanilla seedlings.

Key words: Eco-enzyme, Husk charcoal, Vanilla.

INTRODUCTION

Vanilla is called green gold because of its quite fantastic price, in November 2022 per kg of dry vanilla is worth 1.5 million, compared to other plantation products such as arabica coffee (green beans), nutmeg (dried peeled beans), and cocoa (fermented) respectively. The price is only IDR 64,160, IDR. 65,000 and IDR. 30,000. Starting from 2020, the area of vanilla smallholder plantations is 184 ha, increasing in 2022 to 206 ha as well as vanilla production from 23 tons to 24 tons (Directorate General of Plantations Ministry of Agriculture Republic of Indonesia, 2022).

Vanilla grows epiphytically (Frenkel and Belanger, 2018), the soil media needed for initial growth is sufficient air, crumb/loose and contains nutrients that can be absorbed (Kartikawati and Rosman, 2018). Fertilization during vanilla growth affects the success of cultivation, the addition of organic fertilizer helps improve soil quality and soil aggregate stability and supports agricultural sustainability (Hayati *et al.*, 2021; Kartikawati and Rosman, 2018). Eco enzyme and husk charcoal are agricultural wastes that can be recycled into environmentally friendly organic fertilizers.

Eco enzyme made from citrus waste is a complex solution Li *et al.* (2013) results in calming as a substrate with sugar, has a fresh aroma and high acidity (Vama and Cherekar, 2020; Mavani *et al.*, 2020), is an organic solution that acts as a plant fertilizer Hasanah, (2021), contains 203 mg L^{-1} potassium, 21.79 mg L^{-1} phosphorus Hasanah *et al.* (2022), stabilizing effect of industrial waste activated sludge (Arun and Sivashanmugam, 2015). The effect of eco-enzymes increases the growth of *Brassica juncea* L Lumbanraja *et al.* (2021) and does not harm the environment (Vama and Cherekar, 2020).

Agricultural husk charcoal waste can be used to increase plant fertility, containing lignin, cellulose, and hemicellulose with ash and silica content of 87-97%, 1% nitrogen, 2% potassium. The function of potassium for plants is essential, playing a role in the function of the chemical structure of plants, biochemical regulation and physiological processes, which affect plant growth and development. The use of potassium for other nutrients for plants increases productivity, quality, against biotic and abiotic disturbances, especially during drought stress conditions (Dizaji *et al.*, 2019; Imas, 2013). The husk charcoal affected the proline content, body height, fresh weight and dry weight of vanilla plants induced by the BNR fungus Haryuni *et al.* (2022).

The purpose of this study was to determine the effect

[1]Faculty of Agriculture, Tunas Pembangunan University, Surakarta Central of Java, 57139, Indonesia.
[2]Faculty of Economic and Business, Tunas Pembangunan University, Surakarta Central of Java, 57139, Indonesia.

Corresponding Author: H. Haryuni, Faculty of Agriculture, Tunas Pembangunan University, Surakarta Central of Java, 57139, Indonesia. Email: haryuni@lecture.utp.ac.id

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of eco-enzymes and husk charcoal on the growth of vanilla beans. This study provides information on the dosage of husk charcoal and eco-enzymes and their effect on the growth of vanilla seedlings and the microscopic appearance of stem parts.

MATERIALS AND METHODS

Preparation of vanilla and soil media using the method from Haryuni *et al.* (2020). Preparation of eco-enzym, takes 3 kg of clean orange peel waste and 10 L of clean water. Put the orange peel waste into a large bucket filled with water and add 1 kg of brown sugar. Let it be stored for 3 months. After 1 and 2 weeks, uncover the bucket for 1 minute and stir well to release any gas that builds up. Before using eco-enzyme, it is important to filter it first. The rice husk charcoal is cleaned of other materials which are mixed and then collected together and the center is equipped with a tubular chimney to drain the resulting smoke, the section is given a zinc mat so that the fire does not go out and stays clean, the rice husk is burned together with charcoal and paper, the rice husk is Burn it upside down so that everything is exposed to the fire evenly and turns black, leave it for 24 hours until it cools down and is ready to be used as a medium for treating vanilla plants Haryuni *et al.* (2022). This research was conducted in a greenhouse with a temperature of around 30°C from May to November 2022 at the Department of Agronomy, Faculty of Agriculture, Tunas Pembangunan University, Surakarta, Central Java, Indonesia, using Andosol soil type. This study used a completely randomized group design, with the first factor without eco-enzyme and with eco-enzyme 15 ml/polybag as E0, E1 and the second factor was rice husk charcoal dose treatment (0, 5, 10, 15, 20) g /polybag as P0, P1, P2, P3, P4.

Observation of vanilla bean growth was carried out four weeks after the first treatment of husk charcoal and eco-enzymes, including plant height (measured from the base of the stem to the top of the stem), leaf width, plant fresh weight, plant dry weight, root fresh weight, root dry weight. Furthermore, the dry weight of the leaves and all roots was observed by drying the leaves and roots in an oven at 105°C for 2 hours and continuously at 80°C until constant weight Huang *et al.* (2019). Analysis of variance (ANOVA) was performed in this study. If there is a difference between the treatments, then a follow-up test is performed using the Duncan multiple range test (DMRT) at a significance level of 5% (Gomez and Gomez, 1995).

RESULTS AND DISCUSSION

Eco-enzymes

Data in Table 1 shows the average effective dose of the effect of husk carbon and eco-enzymes on the growth of vanilla beans analyzed using Duncan's multiple range test (DMRT) at a 5% level in the experimental and control activities and shows the differences in eco-enzyme treatment on various aspects of plant growth, such as height, leaf width, plant dry weight and root fresh weight. However, no significant differences were observed in terms of number of leaves, plant fresh weight and root dry weight.

Eco-enzymes are organic substances that help break down soil nutrients into components that can be used for plant growth. This process occurs through the process of photosynthesis which then causes an increase in the number and size of cells so that the plants become taller, the leaves are wider, the dry weight of the plants is greater and the weight of the roots is heavier (Harman *et al*., 2021; Novianto, 2022). Photosynthesis also plays an important role in increasing the growth rate of plants (Rahmawan *et al*., 2019). According to Nurhayati, (2021), photosynthesis is responsible for ATP synthesis, the production of photosynthetic enzymes (*eg* RuBP carboxylase), absorption of CO_2 through leaf stomata, and maintaining electrical balance during the photophosphorylation process in chloroplasts, all of which are influenced by nutrients absorbed by plants. Organic molecules such as proteins, carbohydrates and lipids can be broken down by enzymes such as proteases, amylase and lipases, which are then used for plant metabolism (Arun and Sivashanmugam, 2015).

The use of eco-enzyme chicory orange as much as 15 ml L^{-1} on the dry weight of Lokananta tubers increased by 20.47% compared to the control on the Sanren variety Hasanah *et al.* (2022), with significant effects on root length, stem circumference and dry weight of lettuce plants Yuliandewi *et al.* (2018), supported by research from Novianto, (2022) which showed that eco-enzymes increased root length and number of shallots, as well as the growth of Sacha inchi *Plukenetia volubilis* L Rosnina *et al*. (2022) and Turi (*Sesbania grandiflora*) Ginting *et al*. (2021).

Husk charcoal

Table 2 shows how different doses of husk charcoal affect plant growth. Parameters of leaf width, plant fresh weight, and root dry weight were significantly different at P4, while plant height and number of leaves were not significantly different. The husk charcoal contains potassium, an essential macronutrient for plant transport and assimilation. It also plays a role in soil improvement, silica extraction, and supports plant growth in extreme soil conditions and saline soils (Perdanatika *et al.,* 2018). The decomposition of husk charcoal affects various factors such as seed germination, root growth, seedling emergence, number of shoots, nutrient availability and plant productivity (Kumar *et al*., 2022). Potassium is very important for wet stem-type plants to maintain the balance between the vegetative and reproductive phases (Sardans and Peñuelas, 2021).

In that study, it was found that giving rice husk charcoal at a dose of 20 g/plant to vanilla seedlings increased leaf area, plant fresh weight, plant dry weight, fresh root weight, and root dry weight (Table 2). Whereas in *Lycopersicum esculentum* Mill., plant growth requires a dose of 50 g/plant (Kiswondo, 2011). In another study, a dose of 5 g/plant on *Lactuca sativa* plants increased fresh weight, and the addition of fresh weight also occurred on Arabica coffee and shallot plants (Bismantara *et al.*, 2022; Ngindi *et al.*, 2022). In soybeans, this dose can also increase seed weight Perdanatika *et al.* (2018). In addition, in *Vigna sinensis* L long bean plants, this dose also increased plant height, fresh weight of long bean plants, and fruit weight per plot (Walianggen, 2022).

Combination of eco-enzymes and rice husk charcoal

Data in Table 3 shows that the combination of eco-enzymes and rice husk charcoal did not have a significant effect on plant height, live area, plant fresh weight, plant dry weight, and vanilla root dry weight. However, there was a significant difference in the fresh weight of vanilla tubers. The only significant difference found was in the treatment of fresh root weight. This is because each treatment has a different effect and they do not interact significantly, except for the treatment of fresh root weight which showed a significant effect, indicating an interrelation between the two treatments. When eco-enzymes and husk charcoal enter the plant tissue, they are stored and fill in the gaps in the tissue.

Eco-enzyme which comes from orange peel has advantages because oranges contain various substances such as phenols, amino acids, essential oils, pectin, carotenoids, flavonoids and vitamin C (Epifano *et al.* 2014; Benny *et al.*, 2023), hormones, organic acids, enzymes and mineral salts that accelerate plant biochemical reactions (Fadlilla *et al.*, 2023). One of the advantages of eco-enzyme is its ability to dissolve organic compounds in inside. what was previously insoluble becomes a soluble form. The protease, amylase, and lipase enzymes in eco-enzymes can help break down proteins, carbohydrates, and fats (Verma *et al.*, 2019) and have antifungal, antibacterial and insecticidal properties (Hemalatha and Visantini, 2020). Giving eco-enzymes enhances the growth of chili plants (Darusman *et al.*, 2023), *Brassica chinensis* L (Nugraha and Sa'diyah, 2023), *Allium cepa* (Panataria *et al.*, 2023).

Microscopic observation

Vanilla stems filled with ecological enzymes (A) and husk charcoal (B) fill the transport packets at the base of the stem (Fig 1). This shows that the treatment of ecological enzymes and rice husk charcoal enters the plant tissue and is useful and influences plant growth and development (Tables 1, 2 and 3). Ecological enzymes present in vanilla tissue include functional enzymes amylase, lipase, protease, and cellulase (Gallage and Møller 2015; Raveendran *et al.*, 2018), as well as secondary metabolites such as flavonoids, quinones, saponins, alkaloids, and cardio glycosides (Vama and Cherekar, 2020), thereby reducing environmental toxicity. in agriculture and as liquid organic fertilizer (Hemalatha and Visanti, 2020). Supported by research conducted by Nasar

Table 1: The effect eco enzyme application on height of plant, wide of life, fresh weight of the plant, dry weight of plant, fresh weight of root and dry weight of root on vanilla.

Treatments	Parameters						
	Height of plant (cm)	Amount of leaf (sheet)	Wide of leaf (mm)	Fresh weight of plant (g)	Dry weight of plant (g)	Fresh weight of root (g)	Dry weight of root (g)
Eco enzymes application (E)							
E0	59.67[a]	10.8	323.83[a]	51.73	19.27[a]	5.67[a]	0.86
E1	67.93[b]	11.33	336.74[b]	58.07	23.33[b]	6.87[b]	0.98

Note: E0= Without eco-enzyme, E1= Eco-enzyme 15 ml/polybag.

Numbers in the same column followed by the same letter are not significantly different according to Duncan's multiple range test (DMRT) at a 5% level.

Table 2: The effect doses husk charcoal application on height of the plant, wide of leaf, fresh weight of plant, dry weight of the plant, fresh weight of the root, dry weight of the root on vanilla.

Treatments	Parameters						
	Hight of plant (cm)	Amount of leaf (sheet)	Wide of leaf (mm)	Fresh weight of plant (g)	Dry weight of plant (g)	Fresh weight of root (g)	Dry weight of root (g)
Eco enzymes application (E)							
P0	57.33	10.67	297.83[a]	43.00[a]	18.50[ab]	5.50[a]	0.65[a]
P1	64.33	10.67	350.88[ab]	52.17[a]	21.83[bc]	5.33[a]	0.84[a]
P2	65.00	10.5	304.08[a]	46.17[a]	18.33[a]	5.00[a]	0.77[a]
P3	68.00	11.83	395.54[bc]	65.50[b]	23.83[c]	7.67[b]	1.14[b]
P4	64.33	11.67	428.12[c]	67.67[b]	24.00[c]	7.83[b]	1.19[b]

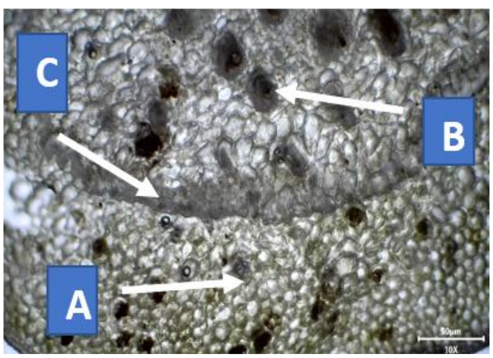
Note: P0= Doses of huck charcoal 0 g/polybag; P1= Doses of huck charcoal 0 g/polybag; P2= Doses of huck charcoal 5 g/polybag; P3= Doses of huck charcoal 10 g/polybag, P3= Doses of huck charcoal 15g/polybag; P4= Doses of huck charcoal 20 g/polybag.

Numbers in the same column followed by the same letter are not significantly different according to Duncan's multiple range test (DMRT) at a 5% level.

Table 3: The effect doses husk charcoal and eco-enzyme application on height of plant, amount of leaf, wide of life, fresh weight of plant, dry weight of plant, fresh weight of root, dry weight of root on vanilla.

Treatments	Parameters						
	Hight of plant (cm)	Amount of leaf (sheet)	Wide of leaf(mm)	Fresh weight of plant (g)	Dry weight of plant (g)	Fresh weight of root (g)	Dry weight of root (g)
Interaction of eco-enzymes and husk charcoal application							
E0P0	55.00	10.67	253.33	40.33	18.33	5.67^{bc}	0.61
E0P1	64.67	10.33	325.08	49.33	21.00	6.00^{bcd}	0.77
E0P2	61.33	10.67	260.75	46.67	16.33	2.33^{a}	0.79
E0P3	56.33	11.00	385.67	59.67	19.67	6.67^{bcde}	1.01
E0P4	61.00	11.33	394.33	62.67	21.00	7.67^{cde}	1.10
E1P0	59.67	10.67	342.33	45.67	18.67	5.33^{b}	0.69
E1P1	64.00	11.00	376.67	55.00	22.67	4.67^{b}	0.91
E1P2	68.67	10.33	347.40	45.67	20.33	7.67^{cde}	0.75
E1P3	79.67	12.67	405.42	71.33	28.00	8.67^{e}	1.27
E1P4	67.67	12.00	461.91	72.67	27.00	8.00^{de}	1.29

Note: P0= Doses of huck charcoal 0 g/polybag P1= Doses of huck charcoal 0 g/polybag, P2= Doses of huck charcoal 5 g/polybag, P3= Doses of huck charcoal 10 g/polybag, P3= Doses of huck charcoal 15 g/polybag, P4= Doses of huck charcoal 20 g/polybag.
E0= Without eco-enzyme, E1= Eco-enzyme 15 ml/polybag.



[A: Eco-enzymes, B: Husk charcoal, C: Endodermis]

Fig 1: Cross-section of a stem treated with eco-enzyme and rice husk with a magnification of 10 × 50 µm.

et al. (2019) that charcoal and compost treatment play a role in maintaining soil fertility including soil physio-chemical properties and increasing yields consisting of (number of tillers per square meter, plant height, number of grains per spike and 1000 grain weight) in wheat plants (*Triticum aestivum* L), in chickpea (*Cicer arietinum* L.) by Anjali *et al.* (2021). The husk charcoal in the soil is porous, light, not dirty, and can store air, then enters the plant through the roots to fill the plant tissue Mishra *et al.* (2017). Rice husk charcoal functions as a biological fertilizer Maftuah *et al.* (2020) and biopesticide Sala *et al.* (2020). The effect of rice husk ash in improving soil dryness is the same as using lime to adjust acidic soil pH in the Kuttanad area (Mini and Lekshmi, 2021).

CONCLUSION

The effect of eco-enzyme doses had an effect on increasing growth and was significantly different on plant height, leaf width, plant dry weight and root fresh weight, husk charcoal had an effect and was significantly different on leaf width, plant fresh weight, plant dry weight, root fresh weight, weight dry roots. Meanwhile, the interaction of eco-enzymes and rice husk charcoal had an effect and was significantly different on the fresh weight of the roots 8.6 g (E1P3). Dosing of eco-enzymes had an effect on increasing growth and was very significantly different on plant height, leaf width, plant dry weight and fresh root weight, husk charcoal had an effect and was significantly different on leaf width, plant fresh weight, plant dry weight and plant weight. fresh root, fresh weight of root. Meanwhile, the interaction of eco-enzymes and husk charcoal had an effect and was significantly different on fresh root weight of 8.6 g (E1P3).

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Conflict of interest

Authors report no conflict of interest.

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