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Planta Tropika focuses related to various themes, topics and aspects including (but not limited) to the following topics Agro-Biotechnology, Plant Breeding, Agriculture Waste Management, Plant Protection, Soil Science, Post Harvest Science and Technology, Horticulture. Planta Tropika published two times a year (February and August) by Universitas Muhammadiyah Yogyakarta in collaboration with Indonesian Association of Agrotechnology / Agroecotechnology (PAGI). The subscriptions for one year : IDR 350.000.

Editorial Address

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Editorial

Journal of Planta Tropika ISSN 0216-499X published by Study Program of Agrotechnology, Faculty of Agriculture, Universitas Muhammadiyah Yogyakarta, is journal presenting scientific articles of agricultural science (Journal of Agro Science). With full sense of gratitude to the Almighty Allah, Volume 10 Number 2 for the year of 2022 has been published.
In this edition, Journal of Planta Tropika presents seven research articles in the field of Agro sciences comprising post harvest physiology, crop cultivation system, weeds management, tissue culture, land management, and climate. The scientific articles discuss about:

(1) Litter Production of Cocoa-Based Agroforestry in West Sumatera, Indonesia, (2) Analysis of Soil Penetration Resistance in Coffee Plantation Agroecosystems in Bangelan, Malang, East Java, (3) Application of Streptomyces sp. and Trichoderma sp. for Promoting Generative Plants Growth of Cherry Tomato (Lycopersicum cerasiformae Mill.), (4) Magnesium Fertilizer Increased Growth, Rhizome Yield, and Essential Oil Content of Ginger (*Zingiber officinale*), (5) Increasing Growth and Yield of Shallot Using Nano Zeolite and Nano Crab Shell Encapsulated NK Fertilizer in Entisols and Inceptisols, (6) Seed Bio-Priming to Enhance Seed Germination and Seed Vigor of Rice Using Rhizobacteria from The Northern Coast of Pemalang, Central Java, Indonesia, (7) Application of Empty Fruit Bunches of Oil Palm and *Indigofera zollingeriana* for Conservation of Oil Palm Plantation, (8) Utilization of Several Agricultural Wastes Into Briquette as Renewable Energy Source, (9) Effects of Foliar Application of Oil Palm Empty Fruit Bunch Ash Nanoparticles on Stomatal Anatomy of Potato Leaf Plants (*Solanum tuberosum* L.), (10) Effects of Mycorrhiza Doses and Manure Types on Growth and Yield of Cassava in Gunungkidul, (11) Fertilizers for Improving the Growth Characteristics and N Uptake of Wild *Rorippa indica* L. Hiern in Different Soil, and (12) Inoculation of Merapi Indigenous Rhizobacteria as A Substitute Compost for Application in Rice Cultivation on Coastal Sandy Under Drought Stress.

The editors would like to thank the authors, reviewers, executive editors, leaders and LRI UMY for their participation and cooperation. Our hope, this journal can be useful for readers or be a reference for other researchers and useful for the advancement of the agriculture.

Editors

GUIDE FOR AUTHORS

TYPE OF PAPERS

PLANTA TROPIKA receives manuscripts in the form of research papers in Bahasa Indonesia or English. The manuscript submitted is a research paper that has never been published in a journal or other publication.

SUBMISSION

The submission of the manuscript is done through our journal website http://journal.umy.ac.id/index.php/pt/index. If you need information regarding the process and procedure for sending the manuscript, you can send it via email at plantatropika@umy.ac.id. Editor's address: Program Studi Agroteknologi, Fakultas Pertanian, Universitas Muhammadiyah Yogyakarta, Jl. Ring Road Selatan, Tamantirto, Kasihan, Bantul, Telp (0274) 387646 psw 224, ISSN: 2528-7079.

ARTICLE STRUCTURE

The submitted manuscripts should consist of 15-20 pages of A4 size paper with 12-point Times New Roman fonts, 1.5 spacing with left-right margin and top-bottom of the paper is 2.5 cm each. All manuscript pages including images, tables and references should be page-numbered. Each table or picture should be numbered and titled.

The systematic of the manuscript writing is as follows:

TITLE : The title should be brief and informative and written bold. Only the first letter of the words is written in uppercase. Maximum length should be 14 words.

AUTHOR NAMES : The author names should be written in lowercase letters (only the first letter of the words is written in uppercase) and should be written from the first author and followed by the others along with the marker of each author's affiliation.

AUTHOR AFFILIATIONS : The author affiliation should be written in lowercase letters (only the first letter of the words is written in uppercase) and it is written according to the order of the number marker of each author's affiliation.

EMAIL : Please list one of authors' email address used for paper's correspondence.

ABSTRAK : Abstrak is written in Bahasa Indonesia using single space in a paragraph with maximum length of 200 words. It should contain background, objective, method, results, and conclusion followed by keywords containing maximum of 5 words.

ABSTRACT : Abstract is written in English using single space in a paragraph with maximum length of 200 words. It should contain background, objective, method, results, and conclusion followed by keywords containing maximum of 5 words.

INTRODUCTION : Introduction contains background, hypothesis or problem outline, and the objective of the research.

MATERIALS AND METHOD : Explaining in detail about materials and method used in the research as well as the data collection and analysis.

RESULT AND DISCUSSION : The results of the research should be clear. State the results collected according to analyzed data. Discussion should include the significance of the results.

CONCLUSION : Authors are expected to give brief conclusion and to answer the objective of the research.

ACKNOWLEDGEMENT : If necessary.

REFERENCES : Single space, according to the authors' guide of Planta Tropika.

EXAMPLES ON HOW TO WRITE REFERENCES

References are written in alphabetical order according to the rules below:

REFERENCE TO A BOOK

Gardner, F.P., R.B. Pearce, and R.L. Mitchell. 1991. Fisiologi Tanaman Budidaya (Translated by Herawati Susilo). UI Press. Jakarta.

REFERENCE TO A JOURNAL PUBLICATION

Parwata, I.G.M.A., D. Indradewa, P.Yudono dan B.Dj. Kertonegoro. 2010. Pengelompokan genotipe jarak pagar berdasarkan ketahanannya terhadap kekeringan pada fase pembibitan di lahan pasir pantai. J. Agron. Indonesia 38:156-162.

REFERENCE TO A THESIS/DISSERTATION

Churiah. 2006. Protein bioaktif dari bagian tanaman dan akar transgenic Cucurbitaceae serta aktivitas antiproliferasi galur sel kanker *in vitro*. Disertasi. Sekolah Pascasarjana. Institut Pertanian Bogor. Bogor.

REFERENCE TO AN ARTICLE IN PROCEEDING

Widaryanto dan Damanhuri. 1990. Pengaruh cara pengendalian gulma dan pemberian mulsa jerami terhadap pertumbuhan dan produksi bawang putih (Allium sativum L.). Prosiding Konferensi Nasional X HIGI hal. 376-384.

FIGURE FORMATTING

Title should be given **below each figure**. Additional information (notes) should be written in lowercase letters except the first letter in each sentence. All figures need to be numbered respectively. Figures should be placed close to explanation/ discussion about the figure.

Examples :

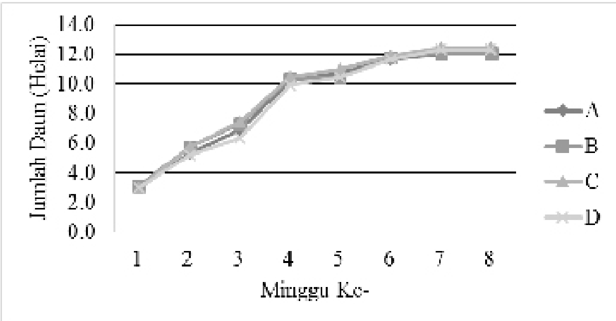

Figure 1. Number of leaves of corn plant

Notes:
A = 250 kg KCl/ha + 0 kg KJP/ha
B = 125 kg KCl/ha + 273,89 kg KJP/ha
C = 62,5 kg KCl/ha + 410,84 kg KJP/ha
D = 0 kg KCl/ha + 547,79 kg KJP/ha

Fig. 1., Fig. 2., and so on. The title of the figure is written with lowercase letters (use uppercase letter at the beginning of the title only) and without full stop (.). Additional information (notes) is placed below the figure.

TABLE FORMATTING

The title of the table should be written **above the table** started from the left (left alignment). Additional information related to the table (notes) is placed below the table. The information is written in uppercase letters at the beginning only as well as the titles inside the table. Table is placed close to the discussion of the table.

Examples :

Table 1. Fruit compost analysis

Variable	Jatropha before composted	Jatropha after composted	SNI (National standard) for compost	Category
Water content	22,49 %	45,79 %	≤ 50 %	Qualified
pH	7,05	8,02	4-8	Qualified
C-Organic content	10,01	5,11	9,8-32 %	Not qualified
Organic matter	17,42 %	8,81 %	27-58	Not qualified
N-Total	0,97 %	2,69 %	< 6 %	Qualified
C/N Ratio	10,44	1.90	≤ 20	Qualified
Potassium	-	9,06 %	< 6 %**	Qualified

Notes: **) Certain materials originated from natural organic matters are allowed to contain P_2O_5 dan K_2O level > 6% (proved with the results of laboratory analysis).

Litter Production of Cocoa-Based Agroforestry in West Sumatera, Indonesia

10.18196/pt.v10i2.11092

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ABSTRACT

Litter is a fragment of aboveground carbon stocks, a vital bridge to the belowground carbon cycle. Land conversion to agricultural purposes will affect litter production. This study aimed to compare the litter production of natural forests with cocoa-based agroforestry systems (AFS) in West Sumatra. Litter production was measured in five different types of ecosystems, namely natural forest (NF), cocoa-rubber-based AFS (CR), multistrata cocoa-based AFS (CM), cocoa-coconut-based AFS (CC), and cocoa monoculture (M). This study is quantitative research with the collection method. The difference in litter production between the five ecosystems observed was tested using ANOVA parametric statistical method. Litter was collected monthly for one year in which litter traps were evenly distributed in each research plot. Ecosystems of NF and M produced the highest annual litter (6.04 Mg ha^{-1} and 4.65 Mg ha^{-1} respectively), while CR produced the lowest one (2.52 Mg ha^{-1}). Although this study did not perform comprehensive modeling of decomposition dynamics, the measurement of annual litter production can provide a further understanding of the dynamics of ecosystem carbon, especially in cocoa-based agroforestry.

Keywords: Agroforestry, Carbon stock, Cocoa, Litter

ABSTRAK

Serasah adalah bagian dari stok karbon di atas permukaan tanah yang merupakan penghubung penting pada siklus karbon di bawah permukaan tanah. Konversi lahan untuk kepentingan pertanian bisa mempengaruhi produksi serasah. Penelitian ini bertujuan untuk membandingkan produksi serasah dari hutan alami dengan sistem agroforestri (SAF) berbasis kakao di Sumatera Barat. Produksi serasah diukur pada lima tipe ekosistem yang berbeda, antara lain hutan alami (H), SAF berbasis kakao-karet (KK), SAF kakao multistrata, (KM), SAF berbasis kakao kelapa (KKel) dan ekosistem monokultur kakao (M). Penelitian ini merupakan penelitian kuantitatif dengan metode koleksi. Perbedaan produksi serasah di antara lima ekosistem diuji dengan pendekatan statistik ANOVA parametrik. Serasah dikoleksi per bulan selama satu tahun menggunakan perangkap serasah yang didistribusikan secara merata di setiap petak penelitian. Ekosistem H dan M memproduksi serasah tahunan tertinggi (6.04 Mg ha^{-1} dan 4.65 Mg ha^{-1} berturut-turut), sementara KK terendah (2.52 Mg ha^{-1}). Meskipun penelitian ini tidak menyediakan pemodelan dinamika dekomposisi yang komprehensif, pengukuran produksi serasah tahunan dapat menambah pengetahuan untuk lebih memahami dinamika karbon ekosistem, terutama pada sistem agroforestri berbasis kakao.

Kata Kunci: Agroforestri, Stok karbon, Kakao, Serasah

INTRODUCTION

Among the various components of the soil-plant system, nutrient cycling is related directly to aboveground system productivity. Litter is one of the aboveground system fragments, which is a vital bridge to the belowground carbon cycle. The cycle of carbon and nutrient is the main ecosystem process driven by plant litter decomposition (Bradford et al., 2017; Giweta, 2020). Therefore, apart in addition to the data on the vegetation biomass, the National Inventory Report and the Kyoto Protocol Report under the United Nations Framework Convention on Climate Change (UNFCCC) require separated measurements of litter and wood debris biomass (The United Nations Framework Convention on Climate Change [UNFCC], 2015).

Based on the structural parameters of vegeta-

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tion, such as species abundance and diversity, litter production provides key information about the functioning of a balanced ecosystem (Petraglia et al., 2019). Litter production patterns between ecosystems vary depending on altitude, latitude, soil fertility, standing structure, climate, and tree species composition (Apriyanto et al., 2021; Primo et al., 2021). Apart from these factors, land management of various types of human activities also provides dynamics to litter production and decomposition patterns. Regarding the effects of human activities on terrestrial ecosystems, land use categories and histories are the key factors in determining the level of carbon stock balance in the soil (Sleeter et al., 2018). Conversion of forest land to agricultural land reduces soil carbon stock because it will affect litter production (Auliyani et al., 2019 ; Yue et al., 2020). Thus, research is needed to compare litter carbon stocks in natural forests and agricultural land.

The Agroforestry System (AFS) is one of the best approaches to reduce pressure on natural forest while still meeting local economic needs. In Indonesia, one of the most common crops grown by AFS approach is cocoa (*Theobroma cacao* L.), which originally grows in tropical rain forests. This study aimed to compare the litter production of natural forest with cocoa-based agroforestry systems (AFS) and cocoa monoculture in West Sumatera. Cocoa-based agroforestry and other types of agroforestry can be awarded credit for its services in storing carbon (Roziaty & Pristiwi, 2020) . Carbon stocks of AFS with perennial mixtures such as cocoa and coffee vary between 12 and 228 MgC per hectare and have the potential to mitigate climate change (Madountsap et al., 2018; Santhyami et al., 2018; Besar et al., 2020; Batsi et al., 2021).

Estimates of annual litter production are a prerequisite for forest soil carbon stocks modeling and their associated changes in biodiversity, decomposition dynamics, and even energy cycles (Krishna & Mohan, 2017). Although this study did not carry out comprehensive modeling of decomposition dynamics, the measurement of annual litter production could provide further understanding of ecosystem carbon dynamics, especially cocoa-based agroforestry.

MATERIALS AND METHODS

Study Area

This study is quantitative research with the collection method. The study was conducted from March 2017 to March 2018. The research was conducted in cocoa-rubber based AFS (CR), multistrata cocoa-based AFS (CM), and natural ecosystem (NF) located in Nagari Simpang, Simpati Subdistrict, Pasaman Region. Meanwhile, the cocoa-based AFS grown under coconut (C. nucifera) (CC) and cocoa monoculture (M) are located in Sungai Geringging, Padang Pariaman Region, West Sumatera (Figure 1). Tree community composition and structure of each cocoa-based agroforestry has been studied by Santhyami et al., (2020). Natural forest was represented by Bukit Badindiang in Pasaman, a traditionally protected and managed by local community (Santhyami et al., 2021).

The location of the NF is at an altitude of 500 meters above sea level with hilly topography. The location of the CM and CR is at an altitude of 250 meters above sea level with a flat topography. The location of the CC and M is at an altitude of 180 meters above sea level with a flat topography. The soil in Simpati Subdistrict is classified as red and yellow litosol and podzolic (Badan Pusat Statistik [BPS] Pasaman, 2018). The soil in Sungai Geringging District is alluvial, podzolic, and peat (Pemerintah Kabupaten Padang Pariaman 2013). These two districts are classified in wet areas with type A rainfall (Schmidt & Ferguson, 1951).

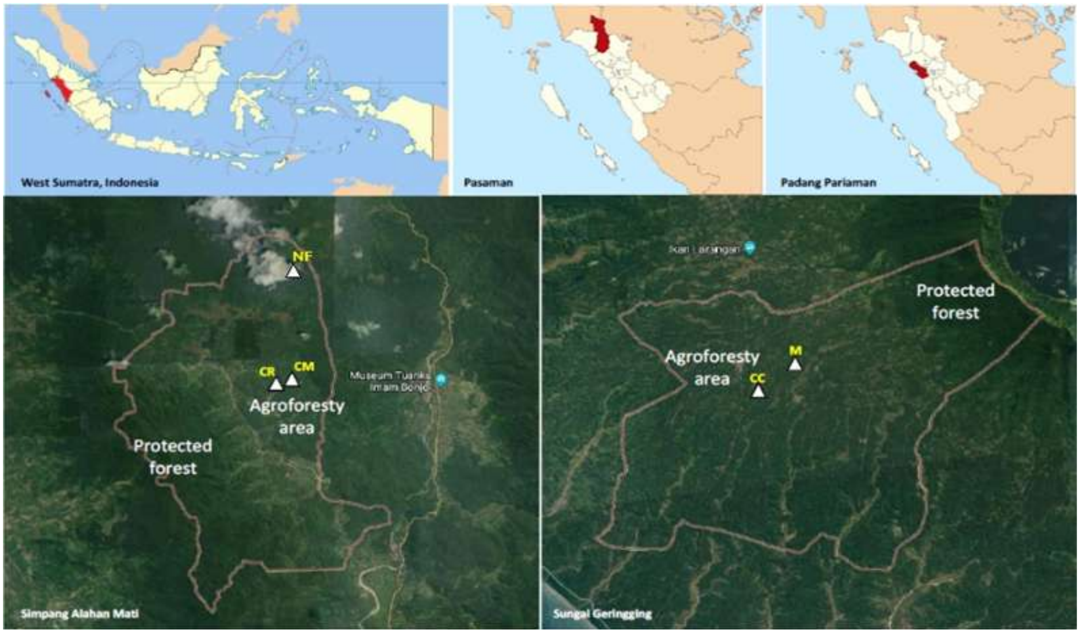

Figure 1. Research location in West Sumatera, Indonesia: CM (Multi-strata Cocoa), CR (Cocoa-Rubber) and NF (Natural Forest) in Nagari Simpang Alahan Mati, Kabupaten Pasaman; CC (Cocoa-Coconut) and M (Monoculture) in Sungai Geringging, Padang Pariaman

Data Collection

The litter was collected monthly for one year with a litter trap, every beginning of the month. The traps were spread inside plots. The minimal area of plots for a natural forest in Indonesia is 1 ha (Rosalina et al., 2014), while for plantation land as agroforestry is ¼ ha (ForestWorks ISC, 2014). On this basis, 25 plots were designed in the forest and six plots on each agroforestry and monoculture practices with a size of 400 m2 for each plot (Badan Standar Nasional Indonesia [BSNI], 2011).

The litter trap is an open wooden frame with a size of 50x50 cm and a height of 30 cm. This wooden frame was covered with 1-mm nylon cloth material. Each plot consisted four litter traps, randomly distributed within the plot. Trap positions were changed monthly (Dawoe et al., 2010). Each litter trap was raised 10 cm above the ground surface to prevent decomposition (Figure 2). The collected litter was then dried until it reached a constant weight. Litter production is expressed in Mg ha^{-1}.

Figure 2. Litter trap design

The point intercept method (Mueller-Dombois and Ellenberg, 1974; Nunes et al., 2015; Thacker et al., 2015) was used to calculate the percentage of land canopy cover. A plot of 400m2 with a size of 20 x 20m was divided into 100 square frames and mapped on a piece of graph paper. This point interception method has the principle of reducing each small square to a midpoint and observing and calculating the cut point as a percentage of the tree canopy. The interception was measured by a simple periscope using a tube with a mirrored base to see if the canopy was closed.

Data Analysis

The difference in litter production between the five ecosystems observed was tested using the ANOVA parametric statistical method with a 95% confidence level for normally distributed and homogeneous data or the Kruskal Wallis non-parametric statistical method with a 95% confidence level for normally undistributed and non-homogeneous data. The post hoc tests were performed, namely Tukey's Honestly Significant Different (Tukey's HSD) for ANOVA and the Mann Whitney U test for Kruskal Wallis.

RESULTS AND DISCUSSION

Monthly litter production was measured from March 2017 to February 2018 in five different ecosystem types, namely natural forest (NF), cacao-rubber-based AFS (CR), multistrata cocoa-based AFS (CM), cacao-coconut-based AFS (CC), and cacao monoculture (M). Table 1 shows the comparison of annual litter production in four cocoa-based AFS and natural forest. This table also shows the comparison of the stand basal area (Santhyami et al., 2018) and the percentage of canopy cover (%) as the basis for analysis. Forest and monoculture ecosystems were the groups that produce the highest annual total litter, which was 6.04 Mg ha^{-1} and 4.65 Mg ha^{-1}, respectively, while the lowest was produced by CR (2.52 Mg ha^{-1}).

This study shows that litter production is related to the value of the basal area and the percentage of canopy cover (Table 1). Basal area is reflected by tree size, stand volume, and biomass (Torres & Lovett 2013), so forests have a higher litter production. Huang et al., (2018) reported that litter production in natural forests was strongly influenced by the stand basal area, age structure, stem volume, altitude, and seasonal and climatic factors. The stand basal area of cocoa-based AFS in this study was lower than in forests, therefore the litter production was also lower than in forests. This result is in line with the research of Owusu-Sekyere et al., (2006) and Triadiati et al., (2011), reporting that the annual litter production of primary or secondary forests is greater than that of cocoa plantations. The natural forest in this study had a litter production of 6.04 Mg ha^{-1} year^{-1}. The forest in this study was customary land in a protected area. The most dominant tree species in this forest was Tarok tree (Campnospera auriculata). This species has thickly leathery broad leaf blades. However, the natural forest litter production in this study was smaller than in the primary forest in Lore Lindu National Park, Central Sulawesi (13.67 Mg ha^{-1} year^{-1}) (Triadiati et al., 2011) and Ghana (8 Mg ha^{-1} year^{-1}) (Owusu-Sekyere et al., 2006).

Table 1. Litter carbon stock of natural forest and four cocoa-based AFS

Type of land use	Stand Basal Area (SBA) (m^2 ha^{-1})	Canopy cover percentages (%)	Annual litter production (Mg ha^{-1})
Cocoa – Rubber (CR)	22.27	79.50	2.52^a
Multistrata Cocoa (CM)	34.42	92.67	2.92^a
Cocoa Coconut (CC)	29.15	82.17	3.96^b
Cocoa Monoculture (M)	9.74	61.67	4.65^{bc}
Natural Forest (NF)	43.34	93.24	6.04^c

Remarks: Values followed by the same letters in the same column are not significantly different based on Tukey's HSD test

According to the results of the one-way ANOVA

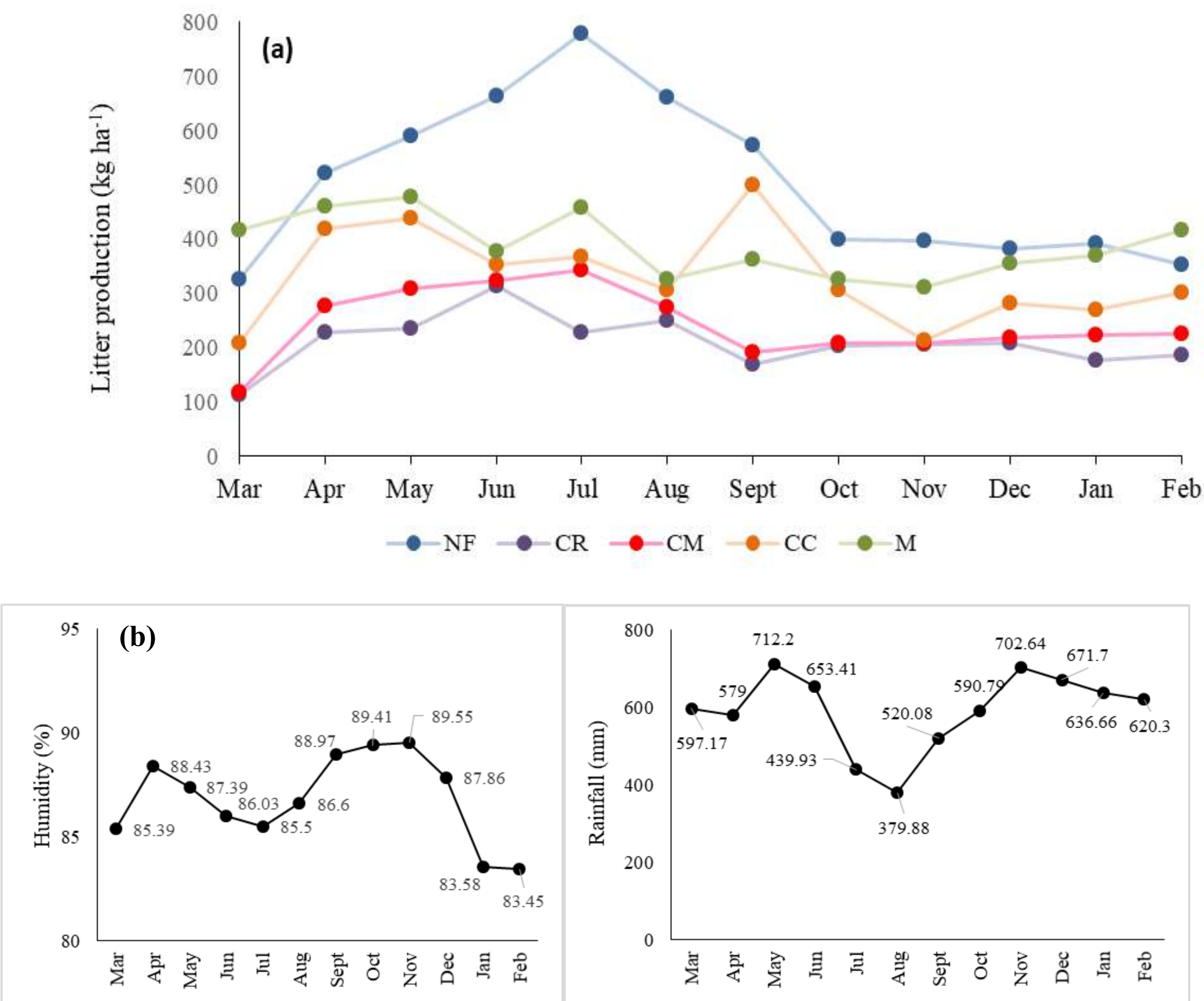

Figure 3. (a). Monthly litter production (March 2017 – February 2018), (b). Climatic condition of West Sumatera (March 2017 – February 2018) (Source: BMKG 2018)

test, the annual litter production of these five ecosystems was significantly different. Ecosystems based on the level of litter production from the lowest to the highest were grouped into three groups, namely CR - CM, CC - M, and M - NF. Statistically, the production of litter in cocoa monocultures was higher than in agroforestry practices and forests. In monoculture farming, farmers performed more intensive care than those in agroforestry. The action of pruning was done periodically. Some litter from this pruning process were netted into the trap.

Apart from the pruning factor, the high annual litter production in monoculture practices is also influenced by environmental pressures. Miyaji et al., (1997) mention that cocoa leaves have a shorter lifespan and easily fall when planted in an environment with high sunlight exposure. This theory is in line with the data of canopy cover percentages measured by the point intercept method. The monoculture cocoa ecosystem had the lowest canopy cover percentage, causing high exposure to sunlight (Table 1). Exposure to full sunlight can result in the stomata closure to reduce water loss so that photosynthetic activity and growth are slowed down. This sensitive response is related to the nature of cocoa as understory species of the forest. T. cacao is a C3 plant species that adapts to semi-shade on the forest floor. Full sunshine can be a growth stress factor rather than a stimulant factor. Photosynthesis in cocoa is saturated at a photon flux density of 400 µmol m^{-2} s^{-1} that is equivalent to 25% full light. The litter production data in this study fit the description of this theory. The production rate of monoculture litter is much higher than that of shaded cocoa indicating shorter leaves age as a form of pressure response to drought and high sunlight radiation (Kunikullaya et al., 2018).

The cacao - coconut and cacao monocultures produced higher litter production than the cacao-rubber and multistrata cocoa-based AFS. Cocoa has relatively broad leaves. Kuruppuarachchi et al., (2013) reported that forests dominated by broad-leaf trees were able to contribute higher litter as a nutrient source compared to forests dominated by narrow-leaf trees. This explains why the production of litter in cocoa monoculture is relatively high.

Litter production in cocoa-based AFS in West Sumatra is smaller than that at two other locations in Indonesia, namely in Central Sulawesi and Lampung. In Central Sulawesi, the type of cocoa-based AFS varied between cocoa planted under intentionally planted shade trees (*Glyricidium sepium*), cocoa planted between local trees, and cocoa planted under heavily shaded forests (rustic cacao agroforestry). The annual litter production ranged from 4.98 to 8.23 Mg ha^{-1} $year^{-1}$ (Triadiati et al., 2011). In Lampung, most cocoa-based AFS was dominated by durian and coffee trees as mixture plant. This AFS produced 11.56 Mg ha^{-1} $year^{-1}$ litter (Indriyanto, 2009). Otherwise, tree stand densities in CR, CM, and CC in West Sumatra were higher than that in cocoa agroforestry in Lampung and Sulawesi (Santhyami et al., 2020). Vegetation standing on fertile soils results in a higher rate of litter production because biomass input from litter contributes back to soil fertility (Dawoe et al., 2010). On this basis, agroforestry land in West Sumatra is likely to be less fertile compared to that in Lampung and Central Sulawesi, given the acidic soil conditions and relatively low nutrient content in the study sites (Santhyami et al., 2018).

Litter production fluctuates every month (Triadiati et al., 2011; Kitayama et al., 2020). This study supports this theory. Figure 3a shows the variation in monthly litter production in five ecosystem groups compared to variations in climatic conditions (air humidity and monthly average rainfall) in Figure 3b.

Forest (NF) and multistrata cocoa-based AFS (CM) produced the highest litter in July - September 2017 (0.66 - 0.78 Mg ha^{-1}). Other ecosystem types did not show a dominant trend of monthly litter production during certain seasons. Litter production in natural forest and multistrata cocoa-based AFS was influenced by the interaction of monthly climatic factors. In these two ecosystems, high litter production coincides with periods of low humidity and low precipitation. July - September of 2017 were the driest months, showing the lowest precipitation rate throughout the year in West Sumatra (Badan Meteorologi, Klimatologi dan Geofisika [BMKG], 2018). Seasonal patterns of litter production in primary forests and cocoa agroforestry in Ghana, which increase in the dry season, indicate a physiological response to drought/reduced humidity playing a major role in this process (Dawoe et al., 2010). This factor, along with the low night time temperature in the dry season, stimulates the synthesis of abscisic acid in leaves. Abscisic acid enhances the leaf fall (Yang et al., 2003). Most studies on litter production in tropical forests show a strong association between seasonal litter production and dry season as the peak litter production (Seta & Zerihun, 2018; Giweta, 2020; Primo et al., 2021). The litter season pattern generally depends on factors related to leaf shedding (Lian & Zhang, 1998).

The pattern of litter production in forest and multistrata cocoa-based AFS in this study is in line with the pattern of litter production in other tropical natural forest. On the other hand, the cacao - rubber, cocoa - coconut and cacao monoculture did not show any peak of litter production pattern. This contradictory finding was also reported in forests without dry seasons, such as the tropical rain forest of Atlantis in Brazil where the peak of litter production occurs during the rainy season. This indicates that the litter loss is due to mechanical factors (de Moraes et al., 1999). The mechanical factors referred to in the three groups in this study (CR, CC and M) were all anthropogenic factors, such as maintenance, pruning, and harvesting that trigger leaf fall. Fertilization was done routinely by Pariaman farmers once a year, while the cocoa-based AFS in Pasaman was generally not fertilized. To keep the soil moisture, especially during the period before flowering and fruiting, farmers carry out pruning practice. Cocoa farmers will at least carry out the maintenance process three to four times a year. The pruning rejuvenates cocoa trees and increases higher cacao yields. By pruning, the trees have been re-grown to optimal crown shape and height (Rouse et al., 2017). The pruning allows the efficiency of cultivation management and harvest. The open canopy allows the shift of a full-sun plantation to agroforestry. The light can penetrate the land floor, thereby facilitating the growth of planted tree seedlings and other crops (Riedel et al., 2019).

CONCLUSION

Litter is one of the aboveground system fragments, which is a vital bridge to the belowground carbon cycle. Natural forest and cocoa monoculture ecosystems produced the highest annual total litter of 6.04 Mg ha^{-1} and 4.65 Mg ha^{-1}, respectively, while the lowest production was in CR (2.52 Mg ha^{-1}). Ecosystems based on the level of litter production from the lowest to the highest were grouped into three, namely Cocoa-Rubber – Multistrata Cocoa, Cocoa-Coconut – Cocoa Monoculture, and Cocoa Monoculture – Natural Forest. Litter production in cocoa monocultures was higher than in agroforestry practices and natural forests. Cocoa-based monoculture farmers performed more intensive care than those in agroforestry one. The pruning increased the litter production trapped into the net. Litter production fluctuated every month. The NF and CM ecosystems produced the highest litter during the dry season, around 0.66 - 0.78 Mg ha^{-1}. Other ecosystem groups did not show a dominant trend of litter production during certain seasons.

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Analysis of Soil Penetration Resistance in Coffee Plantation Agroecosystems in Bangelan, Malang, East Java

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ABSTRACT

Agriculture land shows soil compaction problems due to long-term agricultural cultivation activities. Soil compaction indicator can be seen from the value of soil penetration resistance at different soil depths (0 - 60 cm). This research aimed to determine soil penetration resistance at different coffee plantation ages with different soil depths and to analyze the relationship between soil penetration resistance with soil physical characteristics and coffee productivity. The survey activities include observation of minipits, measuring soil penetration resistance at soil depths of 0-20 cm, 20-40 cm, and 40-60 cm using a hand penetrometer, and soil sampling. The results showed that the soil penetration resistance at each LU and soil depth suggested variation were categorized into moderate and high soil penetration resistance classes (1.34 MPa - 3.35 MPa). Soil characteristics, such as soil aggregate stability, water content, bulk density, porosity, silt content, and clay content, significantly correlate with soil penetration resistance. However, soil penetration resistance has a negative correlation with coffee productivity. The value of soil penetration resistance (at a depth of 0-60 cm) has a significant negative correlation with the average productivity of coffee plantations (r=-0.5936**). Therefore, increased soil penetration resistance decreased root growth, decreasing plant productivity.

Keywords: Coffee plantation, Penetration resistance, Soil depth

ABSTRAK

*Lahan pertanian menunjukkan masalah pemadatan tanah karena kegiatan budidaya pertanian jangka panjang. Indikator pemadatan tanah dapat dilihat dari nilai ketahanan penetrasi tanah pada kedalaman tanah yang berbeda (0 - 60 cm). Penelitian bertujuan untuk mengetahui ketahanan penetrasi tanah pada berbagai umur tanaman kopi dengan kedalaman tanah yang berbeda dan untuk menganalisis hubungan ketahanan penetrasi tanah dengan karakteristik fisik tanah, serta produktivitas kopi. Kegiatan survey meliputi observasi minipit, pengukuran ketahanan penetrasi tanah pada kedalaman 0-20 cm, 20 - 40 cm, dan 40 - 60 cm dengan menggunakan hand penetrometer; dan pengambilan sampel tanah. Hasil penelitian menunjukkan bahwa ketahanan penetrasi tanah pada setiap LU dan kedalaman tanah menunjukkan adanya variasi, dan dikategorikan ke dalam kelas ketahanan penetrasi tanah sedang dan tinggi (1,34 MPa - 3,35 MPa). Karakteristik tanah seperti kestabilan agregat tanah, kadar air, berat isi tanah, porositas tanah, kadar lanau, dan kadar lempung menunjukkan korelasi yang signifikan dengan ketahanan penetrasi tanah. Namun ketahanan penetrasi tanah menunjukkan korelasi negatif dengan produktivitas kopi. Nilai ketahanan penetrasi tanah (pada kedalaman 0-60 cm) memiliki korelasi negatif yang signifikan dengan rata-rata produktivitas tanaman kopi (r=-0,5936**). Oleh karena itu, peningkatan ketahanan penetrasi tanah dapat menyebabkan penurunan pertumbuhan akar, sehingga produktivitas tanaman juga menurun.*

Kata kunci: *Tanaman Kopi, Ketahanan Penetrasi, Kedalaman tanah*

INTRODUCTION

Indonesian coffee production dominates the global coffee market, in addition to Vietnam, Brazil, and Colombia (Atmadji et al., 2019). Coffee plantations are dominated by robusta coffee, which reaches 90% of total coffee plantations (Rahardjo, 2017). National coffee production in 2016-2018 gradually decreased, and the National productivity in 2018 was 775 kg/ha. Provinces with the highest coffee productivity in Indonesia are North Sumatra (1,081 kg/ha), Riau (949 kg/ha), Jambi and South Sumatra (878 kg/ha), and East Java (809 kg/ha) (BPS, 2020). Although coffee productivity in East Java Province is above the national average, it is still lower than coffee productivity in North Sumatra,

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Riau, Jambi, and South Sumatra. Judging from its geographical conditions, East Java Province has the possibility to increase coffee production. Thus, plantations in East Java Province are expected to meet the increasing demand for coffee, which in 2017 increased by 43% compared to 2010, which was 36.5%.

In order to increase coffee productivity, one of the obstacles faced by coffee farmers is soil quality in the root zone of coffee plants, especially soil penetration (Nzeyimana et al., 2013) , in which a more suitable level of soil penetration is important for the growth and development of coffee plant roots (Silva et al., 2019). The roots of coffee plants are divided into horizontal and vertical roots, and the growth is influenced by plant factors, growing environment, and soil characteristics. The development of roots that spread into the soil layer vertically and horizontally impacts increasing soil macroporosity. The destruction of soil aggregates that enters the soil layer along with the flow of water causes blockage of soil pores so that soil penetration resistance increases and macroporosity decreases.

Soil penetration is an important indicator in soil management systems (Girardello et al., 2014) and soil quality in agricultural evaluation that can directly affect root growth and production (Bengough et al., 2011). Soil penetration is influenced by soil texture, bulk density, and porosity. The negative impact of increased soil penetration is related to decreased aeration, availability of water and nutrients, reduced root growth (Beutler et al., 2004), soil ability to hold water, and water movement in the soil. In addition, soil depth and soil moisture can also affect the growth of coffee roots (Kufa & Burkhardt, 2013; Silva et al., 2016)

Coffee plants are suitable for planting in soils that are not compact with loam and clay loam textures (Yadessa et al., 2008; Tarigan et al., 2015; Nzeyimana et al., 2017). However, the existence of coffee plantation management activities and the maintenance of coffee plants, especially in the surface layer, has decreased soil quality, such as compaction. The management of coffee plantations and the maintenance of coffee plants include machinery and human foot stamping in the long term (Alakukku et al., 2003; Miranda, et al., 2003; Araujo-Junior et al., 2008; Tracy et al., 2011; Martins et al., 2012; Hundera et al., 2013; Utomo et al., 2015; Sitania et al., 2018). Soil compaction affects the growth and production of the coffee plant because it is difficult for plant roots to penetrate the soil to meet water and nutrient requirements (Chancellor, 1971; Allmaras et al., 1988). In addition, plant roots will be stunted because they face a fairly high resistance to soil penetration (Shierlaw & Alston, 1984; Taylor & Brar, 1991; Unger & Kaspar, 1994; Kirby & Bengough, 2002; Clark et al., 2003; Masaka & Khumbula, 2007; Place et al., 2008; Masulili et al., 2014).

Soil compaction indicators can be seen from its penetration resistance and soil physical characteristics (Carter, 1990; Assouline et al., 1997; Richard et al., 2001). This soil penetration resistance reflects the ease or difficulty of soil penetration by plant roots. High soil penetration resistance can inhibit root penetration through the soil mass (Bengough & Mullins, 1990; Chen & Weil, 2010; Andrade et al., 2018). Barriers to root penetration can cause a decrease in plant growth and production (Ishaq et al., 2001; Lipiec & Hatano, 2003; DaMatta et al., 2007; Siqueira et al., 2013). Soil compaction in coffee cultivation lands inhibits coffee plant growth due to the difficulty of infiltrating water into the soil and hindering the growth of plant roots, and it can reduce coffee productivity (Fernandes et al., 2012).

The age of the coffee plant is thought to affect the plant roots penetration level. Coffee and tamarind (shade plants) contribute a lot of litter along with increasing plant age and plant biomass, which increases the activity of soil microorganisms, thereby increasing soil organic carbon content, nutrients, soil moisture, and other physical properties (Araujo et al., 2008; Hansel et al., 2008). Braun et al., (2009) showed that the greater the added organic matter, the greater the infiltration, water retention, aeration, temperature, and soil penetration (Oliveira et al., 2009).

The need for information on the importance of root penetration distribution for better management of coffee plantation agroecosystems requires research on this matter. Therefore, this study was conducted to analyze the soil penetration resistance of the root zone at several ages of coffee plantations and determine the distribution of soil penetration resistance and its relationship with soil physical characteristics and coffee plantation productivity.

MATERIALS AND METHODS

Place and sample selection

This research was carried out from November 2019 to February 2020. Rainfall during the research reached 9215 mm in January 2020. Besides, the soil water content in the five Land Units Map was 21-41%. The research location was in the Robusta coffee plantation owned by PTPN XII, Bangelan, Wonosari, Malang. PTPN XII Bangelan has located at 8°05'38.3" South Latitude and 8°05'38.3" East Longitude. Soil types in Bangelan are classified as Alfisols and Inceptisols, but the soil type in the study area is Inceptisols. The height of the plantation from sea level ranges from 450 - 680 m asl. Topographic points of flat land are classified based on the slope of 0 - 8% covering an area of 707.20 ha (80%), 8 - 15% covering an area of 93.05 ha (11%), and 15 - 40% covering an area of 82.95 ha (9%). The soil's physical characteristics were analyzed at the Soil Physics Laboratory, Faculty of Agriculture, Universitas Brawijaya.

Data collection

This research applied a survey method, which was divided into three stages: pre-survey, survey, and post-survey. The pre-survey activity is in the form of determining observation points using a purposive sampling method with criteria of land being planted by robusta coffee plants and based on LU maps (Land Units Map), which were made using ArcGIS 10.2.2 software. In this study, there were 5 LU with three replications based on the age of the plant. There are 15 points of soil sampling and measurement of soil penetration resistance. Survey activities include making minipits and measuring soil penetration resistance in coffee plantations aged 7 to 78 years at a depth of 0-20 cm, 20-40 cm, and 40-60 cm (from the soil surface to the optimal limit of coffee root growth). The plant age class interval was divided into five classes with an interval of 16 years. At LU 1, LU 2, LU 3, LU 4, and LU 5, the ages of the coffee plant were 78 years, 56 years, 45 years, 30 years, and 7 years. The measurement of soil penetration resistance using a hand penetrometer.

As supporting data, sampling of whole/ring or composite soil samples is required. The soil physical characteristics observed include soil structure (aggregate stability), bulk density, particle density, actual water content, total soil pores, particle density, and soil texture, which were analyzed using the wet sieve method, cylinder method, pycnometer method, gravimetric method, the calculation of density, and pipette method, respectively. The average productivity of coffee plants was obtained from secondary data from PTPN XII Bangelan.

Post-survey activities include processing data from soil sample analysis in the laboratory, including statistical and spatial data analysis. Classification of soil penetration resistance is presented in Table 1.

Table 1. Soil Penetration Resistance Classes

Class	Penetration Resistance (MPa)
Extremely low	< 0.01
Very low	0.01 – 0.1
Low	0.1 – 1.0
Moderate	1.0 – 2.0
High	2.0 – 4.0
Very high	4.0 – 8.0
Extremely low	> 8.0

Source: USDA (1993)

Data Analysis

The data was analyzed using Microsoft Excel and Genstat Twelfth Edition software. Statistical data analysis performed includes t-test, correlation test, and regression test. The t-test of two unpaired samples was used to analyze the difference in soil penetration resistance between LU at each depth.

RESULTS AND DISCUSSION

Soil Penetration Resistance

Soil penetration resistance in the field was measured at 0-20 cm, 20-40 cm, and 40-60 cm at each LU (Land Unit: coffee plant age). This measurement produced different soil penetration resistance values. LU 1, with an average plant age of 78 years, had the highest penetration resistance value of 2.71 MPa and the lowest value of 2.24 MPa. Overall, the value of soil penetration resistance in LU1 is classified in the high class, which decreases with soil depth. Table 2 showed several physical characteristics of the soil, such as decreasing soil density, soil porosity, and increasing clay fraction content at each LU 1. According to Silalahi & Nelvia (2017), the factors that affect soil penetration resistance are soil density and total pore space (soil porosity). In addition, soil texture (sand, silt, clay fraction content) also affects soil penetration resistance (Landsberg et al., 2003).

The high value of soil penetration resistance (>2.0 MPa) indicates inhibition of plant root growth, especially in the top layer of soil (0-20 cm). The results are in accordance with Silva et al., (2000); Bergamin et al., (2010); Martins et al., (2012); Palma et al., (2013); and Andrade et al., (2018), reporting that the critical range of soil penetration resistance for plant root growth is 2-3 MPa, soil penetration resistance that does not inhibit plant root growth is <2 MPa, and soil penetration resistance that cannot be penetrated by roots of annual plants and roots of annual plants is >3 MPa.

Table 2. Soil physical properties at each LU

LU	Coffee plant age	Soil Depth (cm)	AS (mm)	BD ($g.cm^{-3}$)	PD ($g.cm^{-3}$)	Por. (%)	WC ($g.g^{-1}$)	Sand (%)	Silt (%)	Clay (%)	Texture
1	78 years	0-20	5.02	1.32	2.16	38.83	0.32	13.52	43.93	42.55	SC
		20-40	4.62	1.21	2.21	45.32	0.38	12.55	47.68	39.76	SCL
		40-60	4.76	1.11	2.07	46.30	0.35	16.13	44.49	39.38	SCL
2	56 years	0-20	5.14	1.26	2.07	39.14	0.30	17.47	43.59	38.93	SCL
		20-40	4.78	1.23	2.20	44.24	0.33	16.49	45.73	37.77	SCL
		40-60	4.25	1.25	2.17	42.14	0.27	15.05	49.35	35.60	SCL
3	45 years	0-20	4.81	1.13	2.00	42.89	0.40	14.94	45.81	39.25	SCL
		20-40	4.86	1.20	2.00	39.74	0.41	14.38	42.77	42.85	SC
		40-60	4.51	1.16	1.90	39.27	0.37	14.02	42.95	43.04	SC
4	30 years	0-20	5.11	1.43	2.09	31.21	0.33	17.72	45.73	36.55	SCL
		20-40	4.79	1.33	1.99	32.98	0.31	17.26	43.41	39.33	SCL
		40-60	5.06	1.31	2.11	37.57	0.28	15.15	44.00	40.85	SC
5	7 years	0-20	5.02	1.37	2.04	32.73	0.22	11.19	38.66	50.15	C
		20-40	4.99	1.14	2.00	42.73	0.21	18.37	42.80	38.83	SCL
		40-60	4.80	1.25	2.01	37.78	0.24	20.92	46.22	32.86	CL

Remarks: AS: Aggregate Stability; BD: Bulk density; PD: Particle density; Por. : Porosity; WC: Water Content; C: Clay; SC: Silty Clay; SCL: Silty Clay Loam; CL: Clay Loam

Soil penetration resistance at LU2 with an average plant age of 56 years is classified in moderate to high class. The highest penetration resistance value was 2.25 MPa, and the lowest was 1.42 MPa. The decrease in soil penetration resistance occurred at a depth of 20-40 cm compared to at a depth of 0-20 cm. This is caused by several physical characteristics of the soil in the form of soil density, porosity, and water content (Table 2). The soil at a depth of 20-40 cm illustrates the soil condition with more pore space so that the penetrometer more easily penetrates it. This is in accordance with Silalahi & Nelvia (2017), mentioning that the density and total soil pore space can affect the value of soil penetration resistance. In addition, soil water content also affects soil penetration resistance. According to Azzuhra et al., (2019), when the soil is dry or the soil moisture content is low, it is more difficult for plant roots to penetrate the soil because the bond (cohesion force) between soil particles is very strong.

LU3 is land with an average coffee plant age of 45 years. The highest soil penetration resistance value was 2.18 MPa, and the lowest was 1.34 MPa, categorized into the Moderate to High penetration resistance class. At LU 3, the value of soil penetration resistance at a depth of 20-40 cm was higher than at other depths. This condition can occur allegedly due to aggregate stability and soil density (Table 2). According to Landsberg et al., (2003), the penetration resistance is influenced by the density of the soil and the stability of the soil structure (aggregate).

The average age of coffee plants in LU 4 is 30 years, and the value of soil penetration resistance is classified in the high class. The highest soil penetration resistance value was 2.48 MPa, and the lowest was 2.08 MPa. Soil penetration resistance was lower at a depth of 20-60 cm compared to that at a depth of 0-20 cm. The difference in the value of soil penetration resistance is caused by the decreased soil density (Table 2). This soil condition is in accordance with the results of research by Silalahi & Nelvia (2017), stating that the factor that affects soil penetration resistance is density. The higher value of soil density in the 0-20 cm layer indicates the effect of soil compaction due to coffee plantation management activities that take place on the soil surface.

In LU5, with the youngest plant age, which is about 7 years, the soil penetration resistance value is classified in the high class, which is thought to be due to the influence of the aggregate stability value and soil texture in the form of sand, dust, and clay (Table 2). The highest soil penetration resistance value at LU5 was 3.35 MPa, and the lowest was 2.43 MPa. The high value of soil penetration resistance at a depth of 0-20 cm is thought to be due to the high clay fraction content (50.15%) and high aggregate stability. In addition, the effect of soil compaction also occurs due to the influence of coffee plantation management activities (Sitania et al., 2018).

The results of the T-test of two unpaired samples showed a significant difference in soil penetration resistance between LU2 and LU5 at a depth of 20-40 cm and 40-60 cm, and between LU3 and LU5 at a depth of 0-20 cm (Table 3). This difference is thought to have something to do with the age of the coffee plantation. Routine plantation management activities carried out every year can cause compaction of the topsoil (0-20 cm). In addition, older plants (coffee and shade trees) have more root systems, which directly and indirectly affect the physical characteristics of the soil (soil aggregation, soil porosity).

Soil penetration resistance obtained from each LU and depth in the field also did not always increase but also decreased with increasing soil depth. According to Oduma et al., (2017), the increase

Tabel 3. T-Test of two unpaired samples

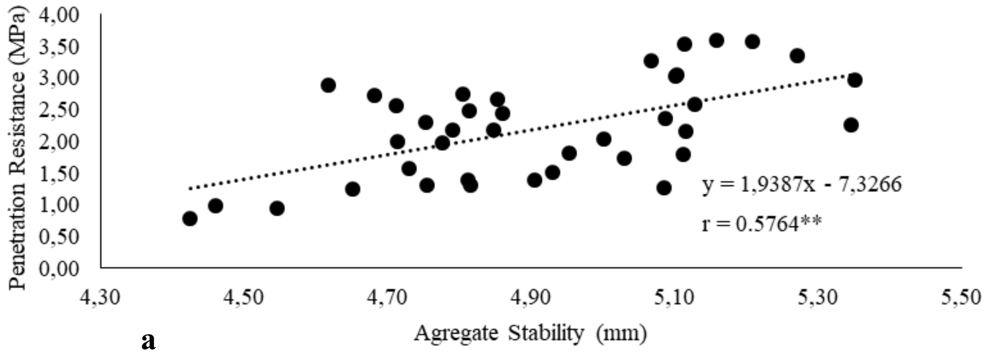
LU	Soil Depth (cm)	T Stat	T-table	T-test result
1 : 2	0-20	0.7384		NS
	20-40	2.3665	2.7764	NS
	40-60	1.5184		NS
1 : 3	0-20	1.6396		NS
	20-40	0.2653	2.7764	NS
	40-60	1.3856		NS
1 : 4	0-20	0.3195		NS
	20-40	0.4651	2.7764	NS
	40-60	0.2254		NS
1 : 5	0-20	-1.3115		NS
	20-40	-0.5341	2.7764	NS
	40-60	-0.4060		NS
2 : 3	0-20	0.9628		NS
	20-40	-1.5666	2.7764	NS
	40-60	0.3364		NS
2 : 4	0-20	-0.3457		NS
	20-40	-1.3970	2.7764	NS
	40-60	-0.8629		NS
2 : 5	0-20	-2.6005		NS
	20-40	-3.0215	2.7764	S
	40-60	-3.0738		S
3 : 4	0-20	-1.1692		NS
	20-40	0.1658	2.7764	NS
	40-60	-0.9406		NS
3 : 5	0-20	-4.0952		S
	20-40	-0.6921	2.7764	NS
	40-60	-2.0435		NS
4 : 5	0-20	-1.5840		NS
	20-40	-0.9045	2.7764	NS
	40-60	-0.5597		NS

Remarks: NS: No Significant; S: Significant

and decrease in the value of soil penetration resistance can occur due to soil and plant management activities, such as land sanitation, plant care, such as pruning, fertilization, weed control, pest and disease control and harvesting of produce, which are carried out annually. Meanwhile, according to Landsberg et al., (2003), several soil characteristics that affect penetration resistance are bulk density, soil structure, soil texture (content of sand, silt, clay fraction), and soil organic matter content.

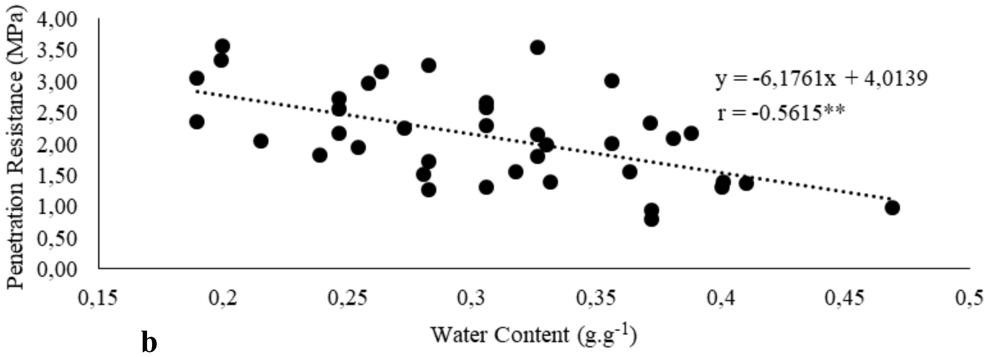
The Relationship between Soil Penetration Resistance and Soil Physical Characteristics

The correlation between penetration resistance and aggregate stability shows a value of r= 0.5764** (Figure 1a), which means an increase in soil aggregate stability results in an increase in soil penetration resistance. In soils with a high clay fraction, the stability of the aggregate is related to the adhesive function of clay particles in the soil aggregation process (Brady & Weil, 2009). Increasing the stability of the aggregate means the



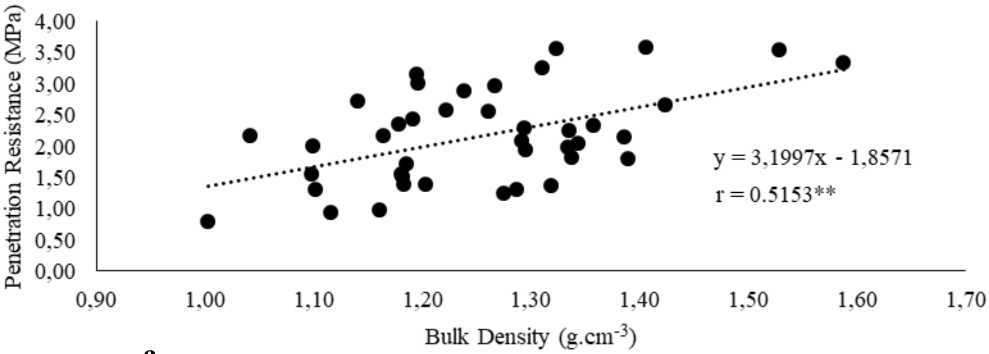
Penetration Resistance (MPa)
4,00
3,50
3,00
2,50
2,00
1,50
1,00
0,50
0,00
4,30
4,50
4,70
4,90
5,10
5,30
5,50
Agregate Stability (mm)
y = 1,9387x - 7,3266
r = 0.5764**

a



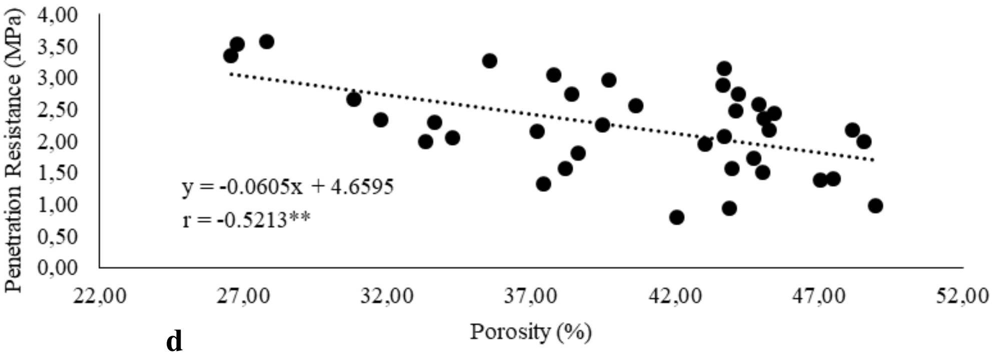
Penetration Resistance (MPa)
4,00
3,50
3,00
2,50
2,00
1,50
1,00
0,50
0,00
0,15
0,2
0,25
0,3
0,35
0,4
0,45
0,5
Water Content (g.g-1)
y = -6,1761x + 4,0139
r = -0.5615**

b



Penetration Resistance (MPa)
4,00
3,50
3,00
2,50
2,00
1,50
1,00
0,50
0,00
0,90
1,00
1,10
1,20
1,30
1,40
1,50
1,60
1,70
Bulk Density (g.cm-3)
y = 3,1997x - 1,8571
r = 0.5153**

c



Penetration Resistance (MPa)
4,00
3,50
3,00
2,50
2,00
1,50
1,00
0,50
0,00
22,00
27,00
32,00
37,00
42,00
47,00
52,00
Porosity (%)
y = -0.0605x + 4.6595
r = -0.5213**

d

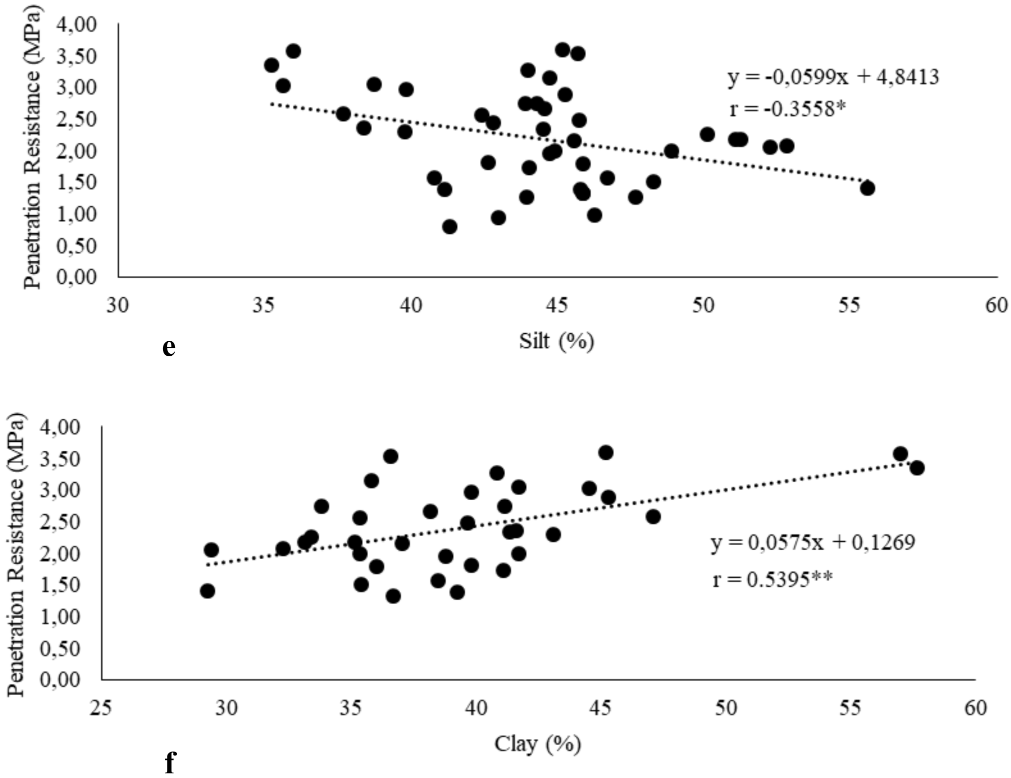


Figure 1. The relationship between soil penetration resistance with soil physical characteristics: a. Aggregate stability, b. Water content, c. Bulk density, d. Porosity, e. Silt, f. clay

greater the bond strength between soil particles so that the soil is more difficult to penetrate by roots or by penetrometers. According to Haridjaja et al., (2010), the value of soil penetration resistance increases when soil compaction occurs. Meanwhile, the results of the study by Catania et al., (2018) showed that tillage to overcome soil compaction was related to soil penetration resistance and soil aggregate stability.

The results of the correlation test between penetration resistance and water content at depths of 0-20 cm, 20-40 cm, and 40-60 cm showed a negative relationship with the calculated r-value of -0.5615 (Figure 1b). The negative direction in the correlation test results means that any increase in water content will decrease soil penetration resistance. The decrease in soil penetration resistance is thought to be due to an increase in the number of water particles in the soil so that the density decreases, and the distance between soil particles increases, which causes a decrease in the attractive force between soil particles, causing the soil to become less hard, making it easier for plant roots to penetrate. According to Azzuhra et al., (2019), plant roots will find it difficult to penetrate the soil when the soil water content is low because the soil has a strong particle bond that makes the soil hard, whereas if the soil water content is high, the soil will be slippery, thereby making it easier for roots to penetrate the soil.

The correlation test results between penetration resistance and soil density at a depth of 0-20 cm, 20-40 cm, and 40-60 cm showed a positive relationship with the r-value of 0.5153 (Figure 1c). These results mean that any increase in the soil's density will increase the soil's penetration resistance. Prasetyo et al., (2014) reported a negative relationship between soil density and plant roots with an r value of -0.728, which means that an increase in soil density will cause the total length of plant roots to decrease because plant roots are difficult to penetrate. Panayiotopoulos et al., (1994) also showed a positive relationship between soil penetration resistance and soil density (r = 0.64).

The correlation test between penetration resistance and soil porosity at depths of 0-20 cm, 20-40 cm, and 40-60 cm resulted in an r-value of -0.5213 (Figure 1d), which means that the relationship between penetration resistance and soil porosity has the same direction. The direction of the negative relationship means that any increase in soil porosity will decrease soil penetration resistance. According to Colombi & Walter (2016), macro pores and meso pores will disappear when soil compaction causes a decrease in soil porosity (Cannell, 1977). Furthermore, the denser the soil, the higher the soil penetration resistance and the more difficult it is for plant roots to penetrate the soil (Refliaty & Endriani, 2018).

The results of the correlation test between penetration resistance and dust content at depths of 0-20 cm, 20-40 cm, and 40-60 cm showed a negative relationship with the calculated r-value of -0.3558 (Figure 1e). These results mean that any dust content increase will decrease the soil's penetration resistance. According to Zhang et al., (2017), dust positively correlates with macroporosity with an R-value of 0.709. High macroporosity conditions make soil penetration resistance decrease, which causes the soil to be more easily penetrated by plant roots.

The correlation test results between soil penetration resistance and clay fraction content at a depth of 0-20 cm, 20-40 cm, and 40-60 cm resulted in a value of r=0.5395** (Figure 1f). This means that an increase in the content of the clay fraction results in an increase in the penetration resistance of the soil. The results of Suprayogo et al., (2004) showed that an increase in the content of the clay fraction resulted in a decrease in soil macro-porosity caused by the blockage of soil pores by clay particles of small size and resulted in increased soil penetration resistance. This is in line with the results of research by Wahyunie et al., (2012), reporting that high clay fraction content will reduce soil macroporosity and can have an impact on increasing soil penetration resistance due to blockage of macro soil pores.

Relationship of Plant Productivity with Soil Penetration Resistance

The productivity of the coffee plant is influenced by one of the physical characteristics of the soil, namely soil penetration resistance. The average productivity in LU1, LU2, LU3, LU4, and LU5 was 2535, 1617, 5232, 10433, and 2498 kg/ha, respectively. Thus, it is necessary to do a correlation test to determine the relationship between coffee plant productivity and soil penetration resistance. The correlation test between soil penetration resistance at a depth of 0-20 cm, 20-40 cm, and 40-60 cm with the productivity of coffee plants in 2019 resulted in a value of r = -0.5936** (Figure 2). This means that increasing soil penetration resistance can reduce the productivity of coffee plants. Increased soil penetration resistance can cause decreased root growth, thereby decreasing plant productivity.

The relationship between penetration resistance and productivity is inversely proportional. There is a decrease in plant productivity with an increase in soil penetration resistance (Colombi & Walter, 2016). Increased soil penetration resistance is related to the effect of soil compaction, resulting in disturbances in plant root growth, thereby decreasing plant productivity (Carmi et al., 1983; Bartzen et al., 2019).

The difference in the value of soil penetration resistance between LUs is related to three things: the age of the coffee plantation, the technology of coffee plantation management, and the soil characteristics. According to Mechram et al., (2013),

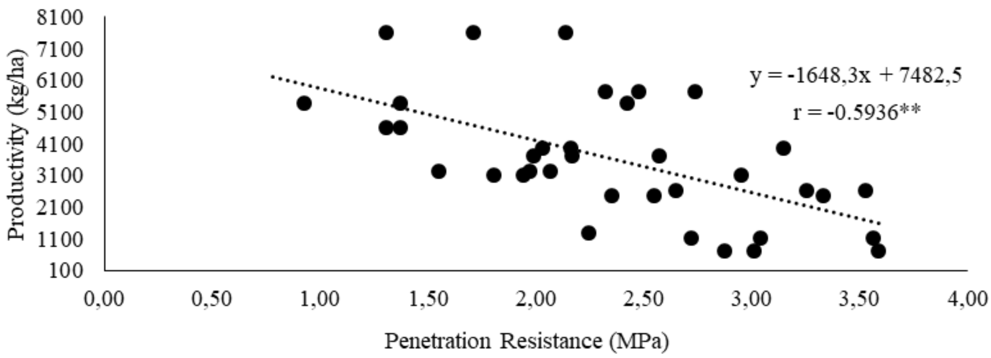


Figure 2. Relationship between coffee productivity in 2019 and penetration resistance

the value of soil penetration resistance increases or decreases with soil depth, presumably due to the soil compaction resulting from coffee plantation management activities. This compaction effect is more pronounced in the topsoil (0-20cm). Soil physical characteristics that can affect the value of soil penetration resistance include aggregate stability, water content, bulk density, porosity, dust fraction content, and clay fraction content (Assouline et al., 1997).

Aggregate stability is closely related to soil penetration resistance value, and every 0.1 mm increase in aggregate stability will increase soil penetration resistance by 0.19 MPa. This is presumably because the Robusta coffee area, Bangelan Plantation, has dominant clay soil, so the attractive force between soil particles (cohesion) becomes strong. The value of soil aggregate stability in Robusta coffee land, Bangelan Plantations, is classified into a very stable class and causes the value of soil penetration resistance to be high because the soil is difficult to destroy. This is supported by Serosero et al., (2016), stating that clay is a particle that can form a bond, so soils containing a lot of clay can form stable aggregates.

Soil water content has a significant negative relationship with soil penetration resistance, and an increase in water content results in a decrease in soil penetration resistance. This has something to do with the stability of the aggregate and the strength of the attractive forces between soil particles. When the soil water content is low, the soil has strong cohesion between particles and makes the soil dense and hard, whereas if the soil moisture content is high, the cohesion force between soil particles becomes weaker, and the penetration resistance is lower (Azzuhra et al., 2019).

Soil density and penetration resistance have a significant positive correlation, where an increase in soil density results in an increase in soil penetration resistance. This happens because the bulk density of the soil is an illustration of the solid composition and pore space of the soil. According to Panayiotopoulos et al., (1994), soil penetration resistance and soil density have a positive relationship, meaning that when the soil density is high, the soil penetration resistance value will also be high.

Soil porosity has a significant negative correlation with soil penetration resistance. A large number of pore spaces in the soil makes the soil less dense, and the penetration resistance of the soil is lower. This is in accordance with the results of research by Colombi & Walter (2016), reporting that a number of soil pores disappear when soil compaction occurs and soil porosity decreases. Soil compaction like this has an impact on decreasing soil porosity and increasing soil penetration resistance (Kooistra & Trovey, 1994; Carducci et al., 2014).

The content of dust fraction has a negative effect on the value of soil penetration resistance. This is because the dust particles have a larger size than clay so in the process of soil aggregation, it produces meso and macro pores, and the penetration resistance of the soil becomes lower. This is supported by Serosero et al., (2016), stating that dust particle has a size of 0.05 mm to 0.002 mm, but the surface of dust particles is not electrically charged, so it cannot form bonds and does not act as an adhesive in the aggregation process (Kemper, & Rosenau, 1986; Amezketa, 1999; Bronick & Lal, 2005).

The content of clay fraction has a significant positive relationship with soil penetration resistance. This has something to do with the very small size of clay particles, and clay can act as an adhesive in the soil aggregation process. The more clay particles, the more stable and stronger the soil aggregates, and the pores formed are mostly micro pores, so the penetration resistance of the soil becomes greater. The results of Suprayogo et al., (2004) showed that the increase in clay fraction content was followed by a decrease in soil macropores and an increase in micropores, which resulted in increased soil penetration resistance. Other factors that may affect the value of penetration resistance are soil organic matter content, aeration pores, and soil aggregation (Day et al., 1995; Carducci et al., 2015).

Penetration resistance and soil compaction have a significant relationship with crop productivity. If the value of soil penetration resistance is high, then plant root growth and development will be disrupted, which can inhibit plant growth and decrease plant productivity (Gilman et al., 1987; Bengough & Mullins, 1990; Ehlers et al., 1983; Kozlowski, 1999; Masaka & Khumbula, 2007). This is also in accordance with the results of research by Colombi & Walter (2016), reporting that increased soil penetration resistance causes plant root growth to decrease, thereby reducing plant productivity. The results also showed a decrease in productivity by 27%, along with an increase in soil penetration resistance from T0 (0.32 MPa) to T4 (1.83 MPa).

Soil penetration resistance in the root zone of coffee plants is, directly and indirectly, related to the age of the coffee plantation and its management (dos Santos et al., 2009; Martins et al., 2012; Refliaty & Endriani, 2018). Information related to the distribution of soil penetration resistance at various ages of coffee plantations is very important to support efforts to manage coffee plantations more sustainably.

CONCLUSION

Soil penetration is an essential soil quality indicator in agricultural evaluation that directly affects root growth and coffee production. The research on soil penetration resistance conducted at various ages of coffee plantations (7-78 years) and soil depth (0-60 cm) showed a reasonably significant variation, but overall, it was classified into the "Moderate" to "High" soil penetration resistance class (1.34 MPa to 3.35 MPa). Differences in plant age cause this difference in soil penetration resistance. Age differences cause additional soil compaction depending on plant growth conditions. Older tree plants have more roots and are more actively growing, which indirectly affects the density of the soil. Soil physical characteristics that have a significant correlation with soil penetration resistance are aggregate stability, water content, bulk density, soil porosity, dust fraction content, and clay fraction content. The value of soil penetration resistance (at a depth of 0-60 cm) has a significant negative correlation with the average productivity of coffee plantations (r=-0.5936**). Therefore, increased soil penetration resistance can cause decreased root growth, thereby reducing plant productivity.

Soil penetration resistance has a close relationship with plant productivity and has an effect of 35.24% (R2 = 0.3524) with the equation of y = -1648.3x + 7482.5. This equation means that every 1 MPa increase in soil penetration resistance will reduce plant productivity by 1.64 tons/ha. If the value of soil penetration resistance is high, then plant roots will be disturbed in their growth and development, which causes plant productivity to decrease.

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Application of *Streptomyces* sp. and *Trichoderma* sp. for Promoting Generative Plants Growth of Cherry Tomato (*Lycopersicum cerasiformae* Mill.)

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ABSTRACT

Production of cherry tomatoes in Indonesia is still low, which might be due to the inappropriate planting and maintenance processes. This research applied biological agent microorganisms *Streptomyces* sp. and *Trichoderma* sp. as Plant Growth Promoting Microorganisms (PGPM) in sustainable agricultural systems. This study aimed to determine the effect of the concentration of microorganisms *Streptomyces* sp. and *Trichoderma* sp. on the growth and production of cherry tomato plants on the polybag scale. The study was arranged with different concentrations of microorganisms *Streptomyces* sp. and *Trichoderma* sp. These concentration applied consisted of 1 : 0 ; 0 : 1 ; 2: 2 ; 3 ; 1 and without PGPM, each repeated four times. The results showed that the treatment of PGPM *Streptomyces* sp. and without *Trichoderma* sp. (1:0) resulted in the shortest flowering period (33.99 days after planting). Meanwhile, the treatment without *Streptomyces* sp. and *Trichoderma* sp. (0 : 1) produced the highest solid weight fruit (69.82 grams/plant).

Keywords: Biological, Growth, Microorganisms, Production

ABSTRAK

Produksi tanaman tomat cherry di Indonesia masih rendah hal ini dapat terjadi proses penanaman dan pemeliharaan yang kurang tepat. Penelitian ini menggunakan mikroorganisme agen hayati Streptomyces sp. dan Trichoderma sp. sebagai Plants Growth Promoting Microorganism (PGPM) dalam sistim pertanian berkelanjutan. Tujuan penelitian ini untuk mengetahui pengaruh konsentrasi mikroorganisme Streptomyces sp. dan Trichoderma sp. terhadap pertumbuhan dan produksi tanaman tomat cherry. Penelitian disusun dengan Rancangan Acak Kelompok (RAK) dengan faktor konsentrasi mikroorganisme Streptomyces sp. dan Trichoderma sp. Faktor perlakuan tersebut yaitu Kosentrasi 1 : 0 ; 0 : 1 ; 2 : 2 ; 3; 1, dan tanpa pemberian PGPM masing masing diulang sebanyak empat kali. Hasil penelitian menunjukkan awal bunga muncul terpendek 33,99 hari setelah tanam pada perlakuan pemberian PGPM Streptomyces sp. dan tanpa Trichoderma sp. (1:0). Perlakuan pemberian PGPM Konsentrasi tanpa Streptomyces sp.: dengan Trichoderma sp. (0:1) menghasilkan berat buah pertanaman tertinggi dengan nilai rata rata 69,82 gram.

Kata Kunci: *Hayati, Pertumbuhan, Mikroorganisme, Produksi*

INTRODUCTION

Indonesian people know the Cherry tomato plant for its nutritional content and good benefits for health. Cherry tomato plants belong to annual plants that can be harvested many times in one year. Cherry tomato crop production reached 962,849 tons in 2017 (Kementerian Pertanian, 2017). Tomato crop production in 2014 and 2015 decreased by 7.74% and 4.17%, respectively, which had not met national demand, so 11 tons of tomato imports were needed (Direktorat Jenderal Hortikultura, 2015). The declining tomato crop production can occur due to the decreasing agricultural land (Pusat Data dan Sistem Informasi Pertanian, 2014). Therefore, an effort is needed to change the strategy to get optimal results.

In addition to paying attention to the environmental conditions, the cultivation of cherry tomato plants also needs to consider the needs of nutrients in plants, as well as providing fertilizers containing micronutrients and macro nutrients

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that function as activators for various enzymes and help the plant growth and development (Yanti et al., 2013). One of the efforts that can be made to improve the quality and quantity of cherry tomato plants is to add microorganisms that act as PGPM and biological agents. The application of these microorganisms is considered the most promising technology for sustainable agriculture. However, it requires effective adoption and standardization of bio formulations for applications in the field. PGPM also acts as a biological agent and is very promising for successful implementation in sustainable agriculture (Verma et al., 2019).

Trichoderma sp. is an antagonistic fungus that has a role as a biological agent and decomposing organism of organic matter. Trichoderma sp. use can increase shallot plants' growth and control diseases that attack plants (Yanti et al., 2019). The filtrate from *Trichoderma viridae* VKF3 derived from mangrove soils can produce a fairly high IAA and suppress pathogens' development (Kumar et al., 2017). Putri et al., (2018) reported that using *Streptomyces* sp. as a growth booster could increase plant height, the number of productive branches, and the roots volume by 27.3%, 24.3%, and 20.7%, respectively. According to Tamreihao et al., (2016), in addition to acting as a biocontrol, the biological agency *Streptomyces* sp. has a role in increasing plant growth. Using biological agents *Streptomyces* sp. as a growth promoter can increase plant height, the number of productive branches, and root volume.

The use of bacteria *Streptomyces* sp. as biological agents has a role in reducing the application of inorganic fertilizers. The isolates of *Streptomyces* sp. are the biological agents of the fruit fly Bactrocera sp., which are potential as a PGPB for tomato and chili plants and can increase plant height, number of flowers, and number of fruits (Suryaminarsih et al., 2019). *Streptomyces griseorubens*, *Gliocladium virens* and *Trichoderma harzianum* are compatible, and the association of these microorganisms can precipitate fusarium wilt disease (Suryaminarsih et al., 2015). *Streptomyces* sp. is a bacterium that has a role as a biological agent.

The use of microorganisms as biological agents and PGPM for plants can reduce the use of inorganic fertilizers because using microorganisms with good habitat management will be able to decompose organic matter into nutrients available for plants. These microorganisms as decomposers will also be available at all times, mainly used in sustainable agricultural systems so that it is expected that the needs of nutrients in plants can be met. The application of biological agents *Streptomyces* sp. and *Trichoderma* sp. at the proper doses and concentrations are expected to spur the growth and production of cherry tomato plants (*Lycopersicum cerasiformae* Mill.). Thus, this study aimed to determine the concentration of *Streptomyces* sp. and *Trichoderma,* which can promote the growth of cherry tomato plants in a sustainable and environmentally friendly farming system.

MATERIALS AND METHODS

The research was conducted in the trial field of the Faculty of Agriculture, Universitas Pembangunan Nasional Veteran East Java, from November 2020 to March 2021. This study used cherry tomato plants cv. Juliet and Plants Growth Promoting Microorganism *Streptomyces* sp. and *Trichoderma* sp. obtained from the Plant Health Laboratory of the Faculty of Agriculture, Universitas Pembangunan Nasional Veteran East Java. This study was arranged in a Randomized Block Design (RBD) with one treatment factor, namely the PGPM formula consisting of several concentrations and combinations of Streptomyces sp. and Trichoderma sp., namely without the administration of PGPM (Control), PGPM 1 (ST 1:0) containing *Streptomyces* sp., PGPM 2 (ST 0:1) containing *Trichoderma* sp., PGPM 3 (ST 2:2) containing *Streptomyces* sp. and *Trichoderma* sp. with a concentration of 2 : 2 and PGPM 4 (ST 3: 1) containing *Streptomyces* sp. and *Trichoderma* sp. with a concentration of 3 : 1.

Production Media and PGPM

The production media were made of Sugar Potato Extract (SPE). The composition of the SPE media was 250 grams of potatoes, 22.5 grams of sugar, and 1 liter of sterile distilled water. The manufacture of PGPM used the ratio of *Streptomyces* sp. and *Trichoderma* sp., which were propagated in the production media. A colony of *Streptomyces* sp. and *Trichoderma* sp. isolates were cut using a cork borer with a diameter of 0.5 cm. In the treatments of single biological agents, four colonies of *Streptomyces* sp. (PGPM1) and *Trichoderma* sp. (PGPM2) were added in 150 mL of SPE media. In the combination treatment of PGPM 3, two colonies of *Streptomyces* sp. and *Trichoderma* sp. were added in 150 mL of SPE media (ST 2:2). In the combination treatment of PGMP 4, the colonies of *Streptomyces* sp. and a colony of *Trichoderma* sp. were added in 150 mL of SPE media (TS 1:3). Meanwhile, in control treatment was prepared without addition of bioagents (K). Each treatment was shaken using IKA Yellow line RS 10 for 14 days.

Planting and Maintenance

Seeds of cherry tomato plants were sown on a soil medium and composted in a ratio of 1:2 using tray pots for 21-28 days. Afterward, the seedlings were transplanted to polybags measuring 35 cm x 35 cm containing garden soil planting media and compost (Ramdani et al., 2018). Transplanting was carried out after the seedlings were 30 days old, with one cherry tomato seedling/polybag. Fertilizing was carried out using 2 grams NPK fertilizer in each polybag seven days after planting (DAP) and 5 grams on 15, 30, 45, and 60 DAP.

The Application of PGPM Streptomyces sp. and Trichoderma sp.

PGPM *Streptomyces* sp. and *Trichoderma* sp. applied had been dissolved at 20 mL of PGPM in 980 mL of distilled water. PGPM solution was given at the time of transplanting by casting (200 mL/plant) and spraying (100 mL/plant) according to treatment at 7, 21, and 35 DAP.

Observation and Data Analysis

The variables observed in the effect of biological agents on the generative growth of cherry tomato plants include the flowering period, the number of flowers per plant, the number of fruits, and the weight of fruits per plant. The data were analyzed using ANOVA (software, type, year) and followed by an HSD test at 5% (Rochiman, 2008).

RESULTS AND DISCUSSION

Number of Flowers, Number of Fruits, and Weight of Fruits per Plant Period 1

Based on the ANOVA results, there was no significant effect of the treatment of PGPM (Plants Growth Promoting Microorganism) *Streptomyces* sp. and *Trichoderma* sp. on the number of flowers, the number of fruits per plant, the number of fruits per plant, and the weight of the total fruits per plant. However, the final observation on the number of flowers and fruit weight in cherry tomato plants treated with PGPM *Streptomyces* sp. showed larger average values than those without the application of PGPM and with the application of other PGPM concentration formulas. (Table 1).

The highest number of flowers (40.32) was obtained in the treatment of PGPM *Streptomyces* sp., while the lowest number of flowers (38.24) was in the treatment without PGPM. The highest number fruits per plant (5.37) was observed in the combination treatment of PGPM *Streptomyces* sp. and *Trichoderma* sp. (3: 1), while the lowest number

Table 1. The average flowering period and weight of fruits per plant at period 2

Concentration of PGPM *Streptomyces: Trichoderma*	Parameter	
	Flowering period (35 days after planting)	Weight of fruits per plant (gram)
Without PGPM	35.16ab	43.08a
ST (1:0)	33.99a	54.93ab
ST (0:1)	37.58b	69.82b
ST (2:2)	37.53b	65.88b
ST (3:1)	34.31a	60.31 ab
BNJ 5%	2.88	17.55

Remarks: Means followed by the same letters are not significantly different based on HSD test at 5%, S = *Streptomyces* sp., T = *Trichoderma* sp.

(4.41) was in the treatment without PGPM. The highest total number of fruits (34.08) was obtained in the treatment of PGPM *Streptomyces* sp., while the lowest total number of fruits (31.74) was in the treatment without PGPM. Meanwhile, the average total weight of fruits was the highest (447.96) in the treatment of PGPM *Streptomyces* sp. and the lowest (388.92) in the treatment without PGPM.

The administration of PGPM (Plants Growth Promoting Microorganism) *Streptomyces* sp. and *Trichoderma* sp. at several different concentrations with a dose of 200 mL/plant by casting and 100 mL/plant by spraying has not been able to increase the number of flowers, the number of fruits per plant, the total number of fruits, and the weight of fruits. This can be because the dose and concentration of PGPM is less than optimal and less effective in increasing plant growth and production. This follows the opinion of Ardiyanto et al., (2017), stating that the frequency of administration and the concentration used are related to the process of plant growth and production. The results of the study that have been registered as a simple patent IPR Application Number: S00202005990 claim that the application of multi-antagonist *Streptomyces narbonensis* and *Trichoderma harzianum* (3:1) on tomato, melon, and chili plants is effective with application dose of 200 mL/plant and 300 mL/plant in vertisol (Suryaminarsih et al., 2019).

Flowering Period and Weight of Fruits per Plant Period 2

Based on the results of ANOVA, there was a significant effect of biological agents *Streptomyces* sp. and *Trichoderma* sp. on the flowering period and weight of fruits period 2. The average values of flowering period and weight of fruits due to biological agent treatment (PGPM) is presented in Table 2.

The latest flowering period (37.58 DAP) was obtained in the treatment of PGPM *Trichoderma* sp., while the earliest (33.99 DAP) was in the treatment of PGPM *Streptomyces* sp. The application of biological agents *Streptomyces* sp. and *Trichoderma* sp. significantly affected the weight of fruits for period 2, which was the highest (69.82 grams) in the treatment of *Trichoderma* sp. and the lowest (43.08 grams) in the control treatment. This follows Putri et al., (2018), stating that the use of 20 ml of PGPM influences the weight of fruits. *Trichoderma* sp. is a fungus that has a role as a biological agent that can increase the growth of shallot plants due to the application carried out directly to the planting media (Yanti et al., 2019). According to Kumar et al., (2017), the filtrate / soluble substance from *Trichoderma viridae* VKF3 derived from mangrove soils can produce a fairly high IAA and can suppress the development of pathogens.

The cherry tomato plants in this study were not attacked by *Fusarium* sp. because at the begin-

Table 2. The average plant height, number of leaves, number of flowers, number of fruits/plant, total fruits/plant, and weight of fruits/plant

Concentration of PGPM *Streptomyces: Trichoderma*	Parameters					
	Plant height	Number of leaves	Number of flowers	Number of fruits/ plant	Total fruits/plant	Weight of fruits/plant
Without PGPM	116,33	15,66	38,24	4,41	31,74	388,92
ST (1:0)	126,91	17,24	40,32	4,58	34,08	447,96
ST (0:1)	120,24	15,58	39,58	4,74	32,2	435,03
ST (2:2)	123,45	15,87	39,49	4,95	32,7	417,55
ST (3:1)	188,62	15,24	39,33	5,37	32,45	424,87
SD	6,44	1,14	1,76	0,78	1,46	22,19

Remarks: SD = Standard deviation S = *Streptomyces* sp., T = *Trichoderma* sp.

ning of planting, biological agents of *Streptomyces* sp. and *Trichoderma* sp. were added to the soil in polybags. *Trichoderma* sp. is also a fungus that can degrade organic matter. Streptomyces sp. is a gram-positive bacterium functioning as a biofertilizer, bioremediation, and biological control agants that effectively controls plant disease pests. *Streptomyces* sp. and *Trichoderma* sp. are biological agents combined to be used as Plants Growth Promoting Microorganism (PGPM) to determine the increase in plant growth when the Plants Growth Promoting Microorganism (PGPM) has been given to cherry tomato plants. This follows Sutarman (2016), stating that *Trichoderma* sp. is a parasite fungus taking nutrients from other fungi. Widodo (2016) states that using PGPM can protect plants from pathogen infections. According to Keliat & Iftari (2017), the fungus *Trichoderma* sp. is a fungus found in all types of soil. *Trichoderma* sp. can be used as biological agents because of their ability to control pathogens that attack plants to reduce the presence of pests and plant diseases. According to Dendang (2015), *Trichoderma* sp. produces the enzyme ß – (1-3) glucanase and chitinase, which can cause exolysis able destroy the cell walls of the fungus *Fusarium*. Trichoderma sp. use can increase shallot plants' growth and control diseases that attack plants (Yanti et al., 2019). Suryaminarsih et al., (2015) found that multi antagonist *Trichoderma* sp. can be applied to horticultural crops because it can help increase crop production. *Trichoderma* sp. can function as a fertilizer that is usually packaged in the form of compost as P and K solvents, increasing plant root growth and height. The *Streptomyces* sp. application at a dose of 30 ml, 20 ml, and 10 ml showed that the height, number of productive branches, root volume, diameter, and weight of fruits harvested in chili peppers tended to be better than control plants or without treatment. Plant height, the number of productive branches and the volume of roots increased by 27.3%, 24.3%, and 20.7%, respectively, with the application of *Streptomyces* sp. (Putri et al., 2018). The biological agent *Streptomyces* sp. is an actinobacterium that can produce bioactive compounds containing antibiotics, antiparasitics, and antifungals (Ekundayo et al., 2014). According to Purnomo et al., (2017),*Streptomyces* sp. can interfere with cell membrane function and synthesis of proteins and nucleic acids so that it can inhibit the growth of pathogenic fungi.

CONCLUSION

The application of *Streptomyces* sp. and *Trichoderma* sp. at a dose of 200 mL/plant by casting and 100 mL/plant by spraying with several different concentrations has not been able to promote the generative growth of cherry tomato plants. The application of *Trichoderma* sp. resulted in the latest flowering period (37.58 DAP), while the application of *Streptomyces* sp. produced the lowest one (33.99 DAP). The administration of PGPM has an influence on the weight of fruits which has the highest weight of fruits (69.82 grams) was obtained in the treatment of *Trichoderma* sp., while the lowest (43.08 grams) was in the control treatment.

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Magnesium Fertilizer Increased Growth, Rhizome Yield, and Essential Oil Content of Ginger (*Zingiber officinale*) in Organic Field

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ABSTRACT

Ginger (*Zingiber officinale*) is the main biopharmaceutical export commodity of Indonesia. However, its productivity and quality are low because it is not cultivated using optimal techniques. This study aimed to examine the effect of magnesium (Mg) fertilizer on the growth, rhizome yield, and essential oil content of two ginger varieties in the organic field. The two factors tested were the rate of Mg fertilizer application (0, 50, 100, and 150 kg MgO ha^{-1}) and the variety of ginger (elephant ginger [*Zingiber officinale* var. *officinarum*] and red ginger [*Zingiber officinale* var. *rubrum*]). The variables measured were plant height, number of leaves, number of tillers, rhizome weight, and essential oil content. Mg fertilizer application rate and ginger variety significantly affected growth, yield, and essential oil content. No interaction effects were found between the two factors. Mg fertilizer applied at 150 kg MgO ha^{-1} resulted in the highest rhizome yield and essential oil content, with an increase of 21.74% and 15.38%, respectively, compared to the control (0 kg MgO ha^{-1}). The yield of elephant ginger was 29.41% higher than that of red ginger, whereas the essential oil content of the red ginger was 16.67% higher than that of the elephant ginger.

Keywords: Optimal Cultivation, Productivity, Quality, Variety

ABSTRAK

Jahe (Zingiber officinale) merupakan komoditas ekspor biofarmasi utama Indonesia. Namun, produktivitas dan kualitasnya rendah karena tidak dibudidayakan dengan teknik yang optimal. Penelitian ini bertujuan untuk mengetahui pengaruh pemupukan magnesium (Mg) terhadap pertumbuhan, hasil rimpang, dan kandungan minyak atsiri dua varietas jahe pada lahan organik. Dua faktor yang diuji adalah takaran pemupukan Mg (0, 50, 100, dan 150 kg MgO ha-1) dan varietas jahe (jahe gajah [Zingiber officinale var. officinarum] dan jahe merah [Zingiber officinale var. rubrum.]). Variabel yang diukur adalah tinggi tanaman, jumlah daun, jumlah anakan, bobot rimpang, dan kandungan minyak atsiri. Pemberian pupuk Mg dan varietas jahe berpengaruh nyata terhadap pertumbuhan, hasil, dan kandungan minyak atsiri. Tidak ada efek interaksi yang ditemukan antara kedua faktor. Pemberian pupuk Mg pada 150 kg MgO ha-1 menghasilkan hasil rimpang dan kandungan minyak atsiri tertinggi, masing-masing meningkat 21,74% dan 15,38% dibandingkan kontrol (0 kg MgO ha-1). Hasil jahe gajah lebih tinggi 29,41% dibandingkan jahe merah, sedangkan kandungan minyak atsiri jahe merah 16,67% lebih tinggi dibandingkan jahe gajah.

Kata kunci: *Budidaya Optimal, Produktivitas, Kualitas, Varietas*

INTRODUCTION

Ginger (*Zingiber officinale*) is a high-value crop that is widely used, especially for its medicinal and flavoring potential. This rhizomatous plant has the highest harvest area in Indonesia, amounting to 10,205.03 hectares in 2018 (BPS, 2018). Although exports of ginger exceed those of other biopharmaceutical crops, it is not grown using optimal cultivation techniques, resulting in low productivity and quality (Bermawie, 2002). Several factors contribute to a crop's chemical composition, including plant genotype, growing conditions, and crop management to modify edible organs to improve the quality of the final products (Akula & Ravishankar, 2011; Dordas, 2009; Stagnari et al., 2018). In this regard, fertilization has an important role because plant metabolite accumulation is closely

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related to the mineral elements available in the growing substrate (Botella et al., 2017; Michalska et al., 2016). Magnesium (Mg) fertilization can be used to improve the yield and quality of crops (D'Egidio et al., 2019).

Magnesium is a macronutrient essential for plant growth (Cakmak & Yazici, 2010). It is a major element of chlorophyll and is needed for harvesting solar energy; it also plays a crucial role in phloem-loading and photo-assimilate transport to sink organs, such as fruits, roots, and seeds (Cakmak & Kirkby, 2008). Mg is also pivotal in synthesizing oils, and together with sulfur, it increases oil levels in various plants. Therefore, soil amendment with Mg is crucial to increase the levels of essential oils in ginger plants (Marschner, 2012). A lack of Mg will cause the suppression of plant growth as it impacts photosynthesis (Cakmak, 2013; Verbruggen & Hermans, 2013). The deficiency of Mg will reduce the dry matter partitioning between roots and shoots, increase the accumulation of starch, sugar, and amino acid in the leaves, damage the chlorophyll molecules, lead to the over-reduction of the electron transport chain in photosynthesis, and generate highly reactive oxygen species (ROS) (Cakmak & Kirkby, 2008; Verbruggen & Hermans, 2013). Therefore, enhancing the level of Mg nutrition is necessary to maintain the high yield quality. Magnesium sulfate is a mineral that is allowed to be given in limited doses in organic farming (BSN, 2013)

Several studies have investigated the application of Mg to improve soil fertility, crop production, and oil content (Senbayram et al., 2015; Wang et al., 2020). Mg fertilizers generally promote the yields of most crops, essential oil yields (Dordas, 2009), and oil palm (Tang et al., 2001). However, information on the critical Mg^{2+} values for ginger is scanty. The concentration of critical leaf Mg^{2+} in a majority of plants is 2–4 mg g^{1} DW (Bergmann, 1992). Visual symptoms determine the critical value without yielding a response.

Moreover, studies have reported that a number of crops had a deficiency of Mg that impacted yield, despite no vegetative signs of deficiency and despite the adequate range of Mg concentrations (Prasad et al., 2008). Since Mg is highly needed during the reproductive stage of ginger, the application of Mg can possibly lead to high yield productivity and increase the essential oil content. This study aimed to examine the effect of magnesium (Mg) fertilizer on growth, rhizome yield, and essential oil content of two ginger varieties on organic field. This research is important in order to increase the yield and quality of ginger on organic field and to find out which varieties of ginger give better yields and quality.

MATERIALS AND METHODS

Study area

The study was conducted in the village of Tegallalang, Gianyar Regency, Bali Province, Indonesia. The experimental field was located 750 m above sea level at 8°19'40"S and 115°15'18"E in Bali's major ginger-producing area. The climate is tropical, with mean annual temperatures ranging from 21–31 °C; the mean annual precipitation is 1848 mm. The soil type of the experimental field is Alfisol. The laboratories analysis results of soil prior to the experiment showed that the soil pH was 6.07 (neutral), the organic carbon content was 2.18% (moderate), the total nitrogen content was 0.14% (low), the phosphorus available was 44.25 ppm (high), the available potassium was 118.18 ppm (moderate), the available Mg was 0.52% (low), soil moisture content at field capacity was 39.55 %, and the soil texture was silty loam. The experiment was carried out in organic fields. Organic certification was applied for the fields after August 2015, but the result was not received. Farmers use composted

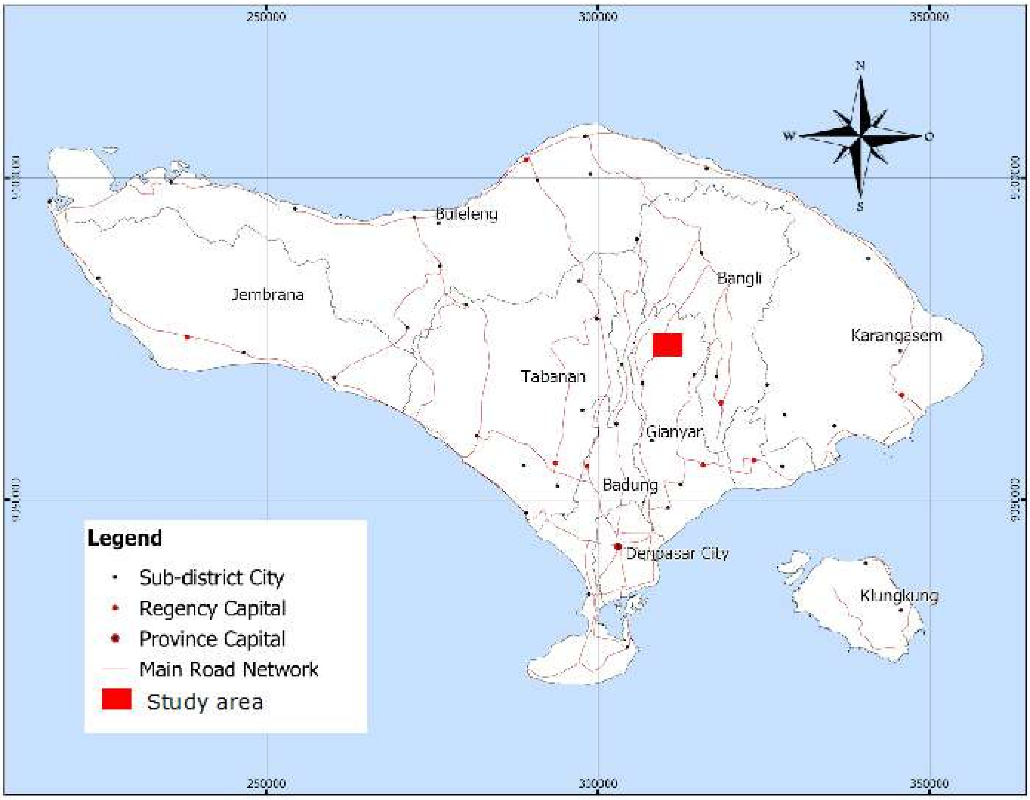

Figure 1. The location of the study site on Bali Island

cow manure for organic ginger production at a rate of 10 tons ha^{-1}. This organic fertilizer's composition of dry matter nutrients includes 17.36% carbon, 1.16% nitrogen, 0.53% phosphorus, and 0.14% potassium.

Experimental design

A randomized block design with two treatment factors was employed in the experiment. The first factor was the rate of Mg fertilizer application with four levels (0, 50, 100, and 150 kg MgO ha^{-1}). The dose of Mg fertilizer refers to the research of Wang et al., (2020), who reported that the agronomic efficiency of Mg fertilizers was correlated with application levels of Mg, at levels (50–120 kg ha^{-1}). The second factor was a variety of ginger, including red ginger (*Zingiber officinale* var. *rubrum*) and elephant ginger (*Zingiber officinale* var. *officinarum*). Both varieties are the dominant varieties cultivated by farmers in this area. These treatments were randomized and replicated four times. Mg was applied in the form of kieserite, a secondary mineral forming solid crystals with the chemical formula $MgSO_4.H_2O$ and containing 26% MgO. Kieserite is used as a fertilizer and is easily soluble in water. The experiment was carried out from April to November 2018 (eight months).

The experiment comprised 32 plots, each 3.5 × 1.5 m in size. Compost was administered at 10 t ha^{-1} one week before planting. In the treatments with fertilizer, MgO was administered two times, one day before planting and 30 days after planting (DAP) at a rate of 50, 100, or 150 kg ha^{-1}. In each plot, three seeds were planted in each of several holes with a dimension of 60 × 40 cm to maintain one healthy seedling per hole or 30 plants per plot (a population of 45,000 plants ha^{-1}). The seedlings were thinned at 14 days after planting (DAP). The plantation was spray-watered two times daily, especially during the beginning of plant growth and harvesting. The plots were weeded once at 14 days after planting (DAP).

Plant height, number of leaves, number of tillers, fresh rhizome weight, and essential oil content were obtained after harvest. Fresh rhizome weight was measured from a 1.26 m^2 quadrant with 12 plants in each plot, then analyzed and converted to fresh weight per hectare. The essential oil content was measured using the Stahl distillation method (*SNI* 06-2385-2006). A ginger rhizome weighing 150 g was chopped into pieces, placed in a round-bottom flask with 300 ml of distilled water, and then boiled. The water vapor condensed in the condenser (the cooling device), consisting of a mixture of oil and water, was collected and transferred to a separating funnel to which Na_2SO_4 was added. The water and oil will separate after being left for some time, depending on their specific gravity. The essential oil and water can then be partitioned in a separating funnel (Taufiq, 2007). The essential oil was identified using thin layer chromatography (TLC). The TLC plate was dried in the oven ± 3 minutes. Then, the lower and upper border was marked with a distance of 10 cm. The mobile phase in the TLC chamber was benzene and ethyl acetate at a ratio of 90:10. The TLC plate was spotted at the lower boundary line with the essential oil obtained and placed in the chamber with the mobile phase. The TLC plate was dried once the solvent had reached the upper border, and the essential oils were visualized under UV light to calculate Rf.

Statistical analysis

The data were analyzed statistically with ANOVA using Costat and MstatC software. Means comparison was performed using Least Significant Difference analysis with statistical significance at 5 % level (Gomez & Gomez, 2007). Pearson correlation coefficients were calculated between Mg fertilizer application rate and growth, rhizome yield, and essential oil content of ginger. Data transformation was done if necessary.

RESULTS AND DISCUSSION

Plant growth

Higher rates of application of Mg fertilizer, from 50 to 150 kg MgO ha^{-1}, significantly increased the height of ginger plants by 14.76% ($p < 0.05$) compared to the control (0 kg MgO ha^{-1}). However, this effect was not significantly different between 150 kg MgO ha^{-1} and 100 kg MgO ha^{-1} (Table 1). Elephant ginger was taller than red ginger. The application of Mg at 100 and 150 kg MgO ha^{-1} significantly increased ($p < 0.05$) the number of leaves by 24.02% and 8.06%, respectively, compared to the control and the 50 kg MgO ha^{-1} rate (Table 1). Elephant ginger produced 16.75% more leaves

Table 1. The effects of magnesium mineral fertilizer (MgO) application rate on the growth, including plant height, number of leaves, and number of tillers per plant of two ginger varieties

Treatment	Plant height (cm)	Number of leaves $plant^{-1}$	Number of tillers $plant^{-1}$
MgO rate (kg ha^{-1})			
0	47.90 ^{b}	56.75 ^{c}	8.88 ^{b}
50	49.85 ^{b}	61.02 ^{bc}	10.5 ^{a}
100	50.87 ^{ab}	65.13 ^{b}	10.5 ^{a}
150	54.97 ^{a}	70.38 ^{a}	10.75 ^{a}
5% LSD	16.571	1.595	0.574
Ginger variety			
Elephant ginger	65.2 ^{d}	72.75 ^{e}	10.97 ^{c}
Red ginger	52.56 ^{c}	62.31 ^{d}	9.06 ^{c}
5% LSD	16.571	1.595	0.574

Notes: Means followed by the same letters in the same column for each factor are not significantly different based on DMRT at α=5%.

Table 2. The effects of four rates of magnesium (Mg) fertilizer application on rhizome weight of two ginger varieties

Treatment	Rhizome weight (g crop-1)	Rhizome weight (t ha-1)
MgO rate (kg ha^{-1})		
0	139.37^{b}	6.27^{b}
50	143.12^{b}	6.89^{b}
100	158.43^{ab}	7.12^{ab}
150	169.67^{a}	7.63^{a}
5% LSD	4.262	0.12
Ginger variety		
Elephant ginger	166.34^{d}	7.49^{d}
Red ginger	128.53^{c}	5.78^{c}
5% LSD	4.262	0.12

Notes: The same letters in the same column for each treatment indicate no significant difference at the 5% level of the LSD test.

than red ginger.

Application of Mg at the rate of 150 kg MgO ha^{-1} significantly increased (p < 0.05) the number of tillers by 21.05% compared to the control. Although elephant ginger had more tillers (2.08%) than red ginger (Table 1), the difference was not statistically significant (p > 0.05).

There was a significant (p <0.05) effect of Magnesium fertilizer application on several plant growth variables. Plant height, number of leaves, and number of tillers significantly increased by 12.5%, 10.6%, and 9.4%, respectively, in the 150 MgO ha^{-1} treatment compared to the control. These growth variables were higher by 12.5%, 10.6%, and 9.4%, respectively, in elephant ginger compared to red ginger. There was no interaction between ginger variety and Mg fertilizer treatment. Ginger responds to Mg application when the soil is deficient in the element (Marschner, 2012), as shown by soil analysis, which indicated a low level (0.42%). These results show that Mg application impacts ginger's growth and productivity, especially when it is planted in soils with low magnesium levels. Under Mg deficiency, the chlorophyll content declines, which may be due to chlorophyll degradation or inhibition of chlorophyll biosynthesis because of a deficiency of Mg and carbohydrates (Marschner, 2012). In addition, a decline in chlorophyll results from interveinal chlorosis of older leaves and the formation of ROS and photooxidation caused by a lack of Mg (Marschner, 2012). Similarly, it was reported that the application of Mg could increase the number of stems in peach and plum (Alcaraz-López et al., 2004). Plants with a low level of Mg become shorter and produce less total biomass than plants with sufficient Mg (Cakmak & Kirkby, 2008; Marschner, 2012). In this study, Mg increased the number of tillers per plant. This may be due to the effect of Mg on carbohydrate transport, which impact components of yield in many plants (Cakmak & Kirkby, 2008; Marschner, 2012). The leaf plays a vital role in photosynthesis by rapidly taking up CO_2, and photosynthetic products can be used to establish rhizomes (Gardner & Pearce, 1991).

Crop yield

Higher rates of application of magnesium fertilizer, which are 100 and 150 kg MgO ha^{-1}, significantly (p <0.05) increased fresh rhizome weight per crop by 21.74% and 10.81%, respectively, compared to the control and the rate of 50 kg ha^{-1} (Table 2). The fresh weight of elephant ginger was 22.72% higher (p < 0.05) than that of red ginger (Table 2). The rhizome yield of ginger had increased significantly due to the application of Mg fertilizer. The rate of 100 kg MgO ha^{-1} increased rhizome yield by 15.12% (or 0.79 t ha^{-1}) compared to the control (Table 2). Elephant ginger had a 29.41% higher

Table 3. The correlation coefficients of the relationships between Mg fertilizer application rate with growth, rhizome yield, and essential oil content of ginger.

	Mg rate	Plant height	Number of leaves	Number of tillers	Rhizome yield
Mg rate	-				
Plant height	0,96	-			
Number of leaves	0.96*	0.99**	-		
Number of tillers	0.96*	0.99**	0,.9**	-	
Rhizome yield	0.99**	0.96*	0.96*	0.96*	-
Essential oil content	0.99**	0.93*	0.93*	0.93*	0.98**

Notes: *p <0.05; **p <0.01

rhizome yield (7.49 t ha^{-1}) than red ginger (Table 2).

The effect of Mg on yield can be attributed to the effects on rhizome weight. This result shows the importance of Mg in increasing the yield of ginger. Mg is essential for ginger; the reproductive phase has higher Mg requirements, and Mg application can directly impact on yield (Cakmak & Kirkby, 2008).

Essential oil content in rhizomes

The Mg fertilizer application could also increase the essential oil content in rhizomes. The 150 kg MgO ha^{-1} rate gave the highest essential oil content (0.42%), an increase of 15.38% compared to the control (Figure 2). The essential oil content of red ginger was 16.67% higher than that of elephant ginger (Figure 2).

Increased Mg fertilizer application led to significantly higher (p <0.05) essential oil content in ginger. The control (without Mg) resulted in the lowest essential oil content, 0.28%, compared to 0.42% (an increase of 15.38%) in the 150 kg MgO ha^{-1} treatment. Insufficient Mg results in a low yield of essential oils because, together with sulfur, Mg increases the synthesis of oils in various plant species. Mg takes part in enzymatic processes, the formation of chlorophyll, and the metabolism of carbohydrates and proteins. All of which can enhance the process of photosynthesis. Photosynthetic carbohydrates are used as a substrate for forming essential oils through glycolysis. Glycolysis produces pyruvic acid, which undergoes a number of reactions to produce geranyl pyrophosphate, a precursor in the formation of essential oils in the

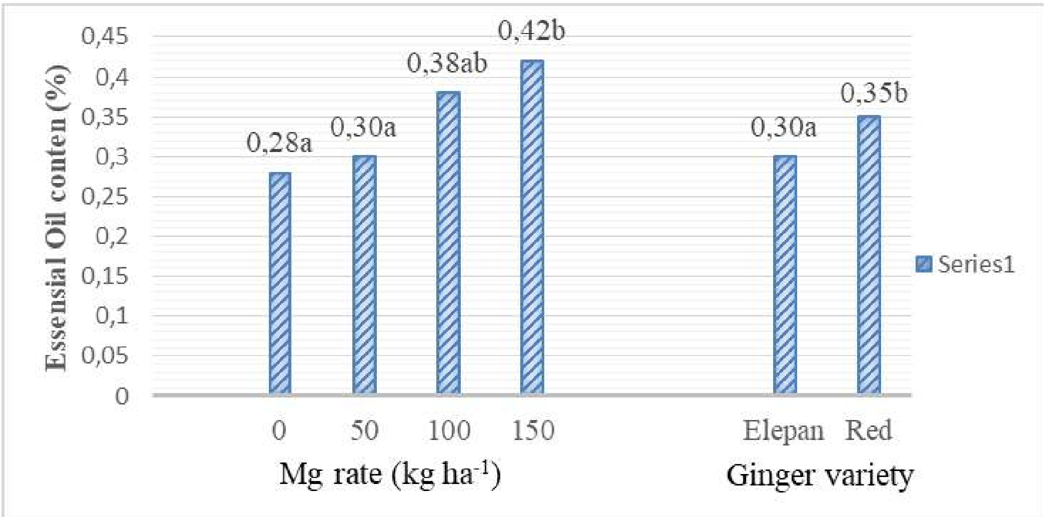

Figure 2. Effects of Mg fertilizers rate and ginger varieties on the essential oil content of ginger plants. The same letters in the same column for each treatment indicate no significant difference at the 5% level of the LSD test.

terpenoid group. The essential oil of ginger includes sesquiterpenes. Sesquiterpene biosynthesis involves photosynthesis. The increased availability of Mg increases the metabolic process of plants, which can further increase the levels of secondary plant metabolites, including essential oil. The effects of Mg fertilizer application in this study are similar to those observed in previous research on other plant species (Prasad et al., 2008). Red ginger produced 16.67% higher essential oil content than elephant ginger. These results may be due to the genetic differences between the two varieties (Rizqullah et al., 2018). It is in line with Jyotsna et al., (2012), who found that different ginger varieties differed significantly in quality.

Correlations

There was a positive correlation between Magnesium concentration with plant height, number of tillers per plant, rhizome weight, and essential oil content (Table 3). Plant height was correlated positively with number of tillers per plant, rhizome weight, and essential oil content (Table 3). A positive correlation was also found between the total number of leaves and number of tillers per plant, rhizome weight, and essential oil content. There was a positive correlation between number of tillers per plant with rhizome weight and essential oil content. Rhizome weight was positively correlated with essential oil content.

CONCLUSION

The growth, yield, and essential oil content of the two ginger varieties significantly increased after the application of Mg mineral fertilizer. No interaction effects were found between the two factors. The Mg fertilizer application rate of 150 kg MgO ha^{-1} resulted in the highest rhizome yield and essential oil content, with an increase of 21.74% and 15.38%, respectively, if compared to the control. The elephant ginger yield was 29.41% higher than that of red ginger. Conversely, the essential oil content of red ginger was 16.67% higher than elephant ginger. We recommend farmers apply Mg fertilizer at the rate of 150 MgO ha^{-1} or plant a red ginger variety to obtain high essential oil yields. Further research on organic fertilizer applications should be undertaken to evaluate the potential benefits for rhizome yield and essential oil production.

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Increasing Growth and Yield of Shallot Using Nano Zeolite and Nano Crab Shell Encapsulated NK Fertilizer in Entisols and Inceptisols

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ABSTRACT

Nanotechnology can be used to produce slow-release fertilizers. Zeolite and crab shells are materials that can be used as fertilizer encapsulation. This study aimed to compare the effects of nano zeolite and crab shells for encapsulation of nitrogen- potassium fertilizers tested on Entisols and Inceptisols soil on the growth and yield of shallots. The research method used a completely randomized design with three factors. The factors were soil type (Entisol and Inceptisol), coating materials (nano-zeolite and nano-crab shell), and NK fertilizer doses (125:50, 250:100, 375:150, and 500:200). The variables observed include initial soil physical and chemical properties, nanoparticle characterization, growth and yield, and agronomic efficiency. Nanoparticles were characterized using SEM and analyzed using ImageJ. The data collected were tested by ANOVA and Tukey. The ball milling method succeeded in producing 91.41% zeolite and 97.50% nano-sized crab shells. Plant height showed that using crab shells as fertilizer encapsulation with a dose of 125:50 gave better results. The yield of crab shells as encapsulation with a dose of 250:100 in inceptisols was better than that in entisols, but the highest agronomic efficiency (EA) was obtained in zeolite treatment as fertilizer encapsulation with a dose of 125:50.

Keywords: Entisol, Inceptisol, Nano crab shell, Nano zeolite, Slow-release.

ABSTRAK

Nanoteknologi dapat digunakan untuk membuat pupuk slow release. Zeolit dan cangkang kepiting merupakan bahan yang dapat dijadikan enkapsulasi. Penelitian ini bertujuan membandingkan pengaruh nano zeolite dan cangkang kepiting sebagai enkapsulasi pupuk nitrogen dan kalium yang diujikan pada tanah Entisols dan Inceptisols terhadap pertumbuhan dan hasil bawang merah. Metode Penelitian menggunakan Rancangan Acak Lengkap 3 Faktor. Faktor 1: Jenis tanah (Entisol dan Inceptisol), faktor 2: Bahan pelapis (nano-zeolit dan nano-cangkang kepiting), faktor 3: dosis NK dengan rasio 125:50, 250:100, 375:150, 500:200. Parameter yang diamati adalah sifat fisika-kimia tanah awal, karakterisasi partikel nano, pertumbuhan dan hasil tanaman serta efisiensi agronomi. Parikel nano dikarakterisasi menggunakan SEM dan dilanjutkan dengan analisis menggunakan ImageJ. Data diuji dengan ANOVA dan dilanjutkan Tukey. Metode ballmilling berhasil menjadikan 91.41% zeolit dan 97.50% cangkang kepiting berukuran nano. Tinggi tanaman menunjukkan bahwa penggunaan cangkang kepiting sebagai enkapsulasi pupuk dengan dosis 125:50 memberikan hasil yang lebih baik dibandingkan perlakuan lain. Hasil tanaman pada inceptisol menggunakan cangkang kepiting sebagai enkapsulasi dengan dosis 250:100 lebih baik dari entisol, namun nilai efisiensi agronomi (EA) paling tinggi diperoleh pada zeolit sebagai enkapsulasi pupuk dengan dosis 125:50.

Kata kunci: *Entisol, Inseptisol, Nano-cangkang kepiting, Nano-zeolit, Pupuk lepas lambat*

INTRODUCTION

Entisols and Inceptisols have the potential to be used as agricultural land. Entisol has an area of 3.80 million ha, and Inceptisol has an area of about 40.88 million ha in Indonesia. Entisols are soils that have little or no clear horizon development. Sandy land is one of the soils classified as Entisol. According to Tuhuteru et al., (2019), Entisol is one of the soils with low productivity due to low water-holding and storage capacity, high infiltration and evaporation, low fertility and organic matter, and low water use efficiency. Entisol also has a low cation exchange capacity and very low nutrient content, especially nitrogen. Inceptisols are soils modified from their parent material through a soil formation process that can be distinguished from entisols. However, they are not sufficient to

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form horizons needed to form other soil types. The total N content of inceptisol is low (0.15-0.42%), the cation exchange capacity is relatively moderate (14.1-17.3 cmol (+)/kg), the base saturation is relatively low (24-29%), and it has a relatively acidic pH (Safitri et al., 2018).

Shallot is one of the commodities with a fairly high economic value and an increasing consumption level (Burhan & Proyogo, 2019). The level of consumption of shallots during 2002-2021 is relatively fluctuating but tends to increase. Shallot consumption in 2017 and 2018 was 2,570 and 2,764 kg/capita/year, respectively. Meanwhile, shallot consumption in 2019 increased to 2,796 kg/capita/year. In 2020 and 2021, the level of shallot consumption was also predicted to increase by 1.18% and 1.28%, respectively. Shallots can grow in the lowlands and highlands with an altitude between 0-900 m asl. It indicates that shallots have the potential to be planted in Entisol and Inceptisol soils. However, shallots require soils with good drainage and aeration, organic matter, and slightly acidic to normal pH. Thus, it is necessary to improve the properties of Entisols and Inceptisols (Syawal et al., 2019). Increasing the efficiency of fertilization can be done, among others, by improving fertilization application techniques and improving the physical and chemical properties of fertilizers through changes in the nutrient solubility system, shape and size of fertilizers, and formulations of fertilizer nutrient levels.

Nano-fertilizers that are very small (1 nm = 10^{-9} µm) have more reactive properties, which can directly hit the target, and their use only requires small amounts. The ball milling method is a method that is often used to grind the powder to a nanometer scale. Its working principle depends on the energy released due to friction between the ball, the powder, and the operating time. The longer the friction occurs, the finer the particles are produced (Piras et al., 2019). In addition, one way to increase the efficiency of fertilization is to modify the fertilizer into a slow-release fertilizer so that the nutrients contained in the fertilizer can be released gradually according to the time needed by the plant. Coating nanomaterials in fertilizers can slow the release of nutrients in the fertilizer.

Rugayah et al., (2018) mentioned that slow-release fertilizer (SRF) was a mechanism for releasing nutrients gradually, following the pattern of nutrient absorption by plants, thereby leading to optimal fertilizer absorption by plants. One way to modify fertilizers into slow release is to mix fertilizers and materials with a high cation exchange capacity, such as zeolites (Dubey & Mailapalli, 2019). Zeolite is a hollow silicate mineral with a high cation exchange capacity so that it can exchange cations. Crab shells can also manufacture slow-release fertilizers because they have pores that can hold nutrients. Zeolite and crab shells can be made nano-sized and then encapsulate fertilizers to become slow-released. According to Noviyanita (2018), increasing the dose of inorganic fertilizers can increase the production of shallots. Hartatik et al., (2020) stated that zeolites could play a role in the gradual release of nutrients. Zeolite that has undergone a change in size to nano is able to increase the efficiency and effectiveness of fertilization on plants because it releases nutrients slowly and is able to reduce fertilizer doses (Lateef et al., 2016). The chitin content in crab shells can be used as an adsorbent to adsorb phosphate, so that crab shells are expected to be able to absorb nutrients in fertilizers, such as nitrogen and potassium, which can then be rereleased for plants.

This research was carried out by converting zeolite and crab shells into nano size with a ball mill, which were then used to encapsulate NK fertilizers to improve their effectiveness. Fertilizer application was given to shallots in Entisol and

Inceptisol soils. Urea and KCL are water-soluble fertilizers, so they will easily leach out from the plant root zone when applied to sandy soil. Those causes the low agronomic efficiency of these two types of fertilizers. Therefore, encapsulation technology is needed to engineer the two fertilizers into slow-release fertilizers. Materials such as zeolite and crab shells are widely available and inexpensive, so they are prospective enough to be used as nano-materials. This study aimed to compare the agronomic effectiveness of NK-SRF encapsulated with nano-zeolite and nano-crab on Shallots in Entisols and Inceptisols.

MATERIALS AND METHODS

Experimental Design

This research was conducted in the experimental field and Soil Laboratory from September 2020 to February 2021. This research was arranged in a completely randomized design with three treatment factors. The first factor was the types of soils, consisting of Entisol and Inceptisol, the second was the types of nano material, including nano zeolite and nano crab shell, and the third was the doses of NK, which were125:50, 250:100, 375:150, and 500:200. Each treatment was replicated three times. Soil preparation was carried out by taking Entisol soil from Kulon Progo (7.9^{0}67.3'34.9"S 110.18^{0}18.2'60.7"E) and Inceptisol from Gunung Kidul (7^{0}52'20.6"S 110^{0}31'35.1"E) in the tillage layer at a depth of 1-20 cm.

Research Methods

Material preparation was carried out by preparing urea-KCl fertilizer, zeolite, and crab shells. Zeolite and crab shells were cleaned and then mashed to a size of 100 mesh. Fertilizer was manufactured by making zeolite and crab shells into nano size using a ball mill, with a ratio of steel balls, zeolite/crab shells, and water of 500 g: 100 g: 60 ml, respectively. The milling process was carried out for six hours. The principle of milling is grinding the materials on the surface of balls due to colliding with other balls. The manufacture of nanomaterials is successful if 70% or more of the particles have a size of 1-100 nm (Khan et al., 2019). The formulation was carried out by mixing urea and KCl with nano zeolite and nano crab shells according to treatment in a ratio of 6:1 (Kottegoda et al., 2017). Mixing was done conventionally using centrifugal force.

The planting media was prepared by taking samples of Entisol soil from Kulon Progo beach sand and Inceptisol soil from the tillage layer at a depth of 1-20 cm. The soil was air-dried, sifted, cleaned from dirt and weeds, and then put into polybags according to the volume of polybags 28.260 dm^3/0.28 L. Entisol soil was obtained from Kulon Progo beach sand, and Inceptisol soil was obtained from Karangsari, Gunung Kidul. Planting was done by immersing all shallot seeds in the soil. The seeds used have previously gone through a shelf life of ±3 months and then cut 1/3 of the ends to break dormancy so that the growth is uniform. The treatments were applied after basic fertilization (cow manure 20 tons/ha and SP-36 90 kg/ha incubated for 7 days). Harvesting was done when the shallot plants had fallen 50% and turned yellow (55 days after planting).

Variables Observation

The variables observed were soil physical and chemical properties, characterization of nanoparticles, plant height, number of leaves, bulb diameter, shoot fresh and dry weight, root fresh and dry weight, and bulb fresh and dry weight. Soil physical and chemical properties include soil texture, pH, organic C, organic matter, total N, available P, avaible K, CEC, Ca, Mg, Na, and base saturation. Soil physical and chemical properties of incubated soils were analyzed in the laboratory. Plant height and diameter of bulbs were measured every week, while shoot fresh and dry weight, root fresh and dry weight, and bulb fresh and dry weight were measured after the harvest. Fresh weight was the weight after harvest. Dry weight was obtained after the samples were dried in the oven at a temperature of 65^{0}C for 3-4 days.

Data Analysis

The data collected were analyzed using ANOVA and continued with Tukey's test (HSD) using SAS program to find out the significant differences between treatments. Data from the characterization of nano particles were analyzed using Scanning Electron Microscopy (SEM) followed by analysis using image-J to find out the size of nano particles and the element content of nano particles. The agronomic efficiency was calculated by the formula as follows:

$$AE\ (\%) = \frac{(Y\text{-}Y0)x100\%}{F} \quad (1)$$

Where, Y = yield of harvested portion of crop with nutrient applied; Y0 = yield without nutrient application; F = amount of nutrient applied.

RESULTS AND DISCUSSION

Soil Physical and Chemical Properties

Based on Table 1, the texture of entisol soil before incubation is categorized as sand with a slightly alkaline pH. The organic C, total N, and available P contents are low, extremely low, and low, respectively. Soils are poor in P minerals, and the management level is still not intensive. Cation exchange capacity (CEC) is low, as well as the levels of Ca, Mg, and Na are categorized as very low to low ratings. Gunawan et al., (2019) stated that the soil cation exchange capacity (CEC) affects the availability of cations such as Ca, Mg and Na in the soil, if the soil CEC is low, the availability of these cations is also low. This is because base saturation is often directly proportional to cation exchange capacity (CEC) because it illustrates the level of cations present in the soil. The analysis results follow Sutardi (2017), stating that entisols have a sand texture, granular soil structure, loose consistency, and are very porous so they have low water and fertilizer buffering capacity. The content of organic matter and total N is also relatively low. The available K contained in the entisol is relatively high, related to the soil pH. According to Gunawan et al., (2019), an acidic pH can cause an increase in potassium fixation, resulting in a decrease in the availability of K in the soil.

Based on Table 1, Inceptisol has a silt loam texture and an acidic pH. The organic C, total N, and available P contents are low, very high, and moderate, respectively. Meanwhile, the cation exchange capacity (CEC) is moderate and directly proportional to cations such as Ca, Na, Mg, and K, which are classified as extremely low, medium, medium, and low, respectively. The cation exchange capacity (CEC) and the low number of base cations make the base saturation of the Inceptisol extremely low. The analysis results were supported by Sudirja et al., (2017), stating that in general, Inceptisols have an acidic pH and high clay content, and the surface layer is easily washed so that it is easy to lose nutrients. The Inceptisol soil of Karangsari, Gunung Kidul is dominated by the silt fraction and has a very high available N content.

Nano Material Characteristics

Table 2 presents the particle size distribution of the zeolite obtained after pounding with steel balls for 6 hours. The proportions of particle sizes are as follows: 1-10, 10-20, 20-100, 100-250, 250-1000, and > 1000 nm, each of which has a percentage of 35.61, 24.09, 31.71, 6 .19, 2.40 and

Table 1. Physical and chemical properties of Entisols and Inceptisols

No.	Parameter	Entisol	Inceptisol
1	Texture (USDA)	Sand	Loam
	Sand (%)	91.94[S]	33.43[SL]
	Silt (%)	4.48[S]	57.63[SL]
	Clay (%)	3.58[S]	8.94[SL]
2	pH-H_2O (1:5)	7.95[SA]	5.2[A]
3	pH-KCl (1:5)	7.70	5.1
4	Organic C (%)	0.33[L]	1.27[L]
5	Organic matter (%)	0.56	2.19
6	Total N (%)	0.91[EH]	1.18[EH]
7	Available P (mg kg^{-1})	7.07[L]	10.92[M]
8	Available K (cmol(+)kg^{-1})	0.95[H]	0.21[L]
9	CEC (cmol(-)kg^{-1})	12.43[L]	19.95[M]
10	Available Ca(cmol(+)kg^{-1})	0.62[EL]	1.13[EL]
11	Available Mg (cmol(+)kg^{-1})	0.11[EL]	1.98[M]
12	Available Na (cmol(+)kg^{-1})	0.32[L]	0.40[M]
13	Base Saturation (%)	16.04[EL]	18.68[EL]

Remarks: S=sand, SL=silt loam, SA=slightly alkaline, A=acid, L=low, EL=extremely low, M=medium, H=high, EH=extremely high

0.02%. The total percentage of zeolite size that can be categorized as nano particles (<100 nm) is 91.41%. While the particle size distribution of crab shells are: 10-20, 20-100, 100-250, 250-1000, and > 1000 nm with each percentage 52.85, 44.72, 1.68%, 0.71 and 0.02%. The percentage of total particle size of crab shells that can be categorized as nanoparticles (<100 nm) is 97.50%. The best size of materials for plants is 1-100 nm. The splitting of the particles into nano size is thought to be caused by the collision between the particles and the steel ball for 6 hours. This is in line with Subramanian et al., (2015), mentioning that the synthesis of nanoparticles using a physical approach, especially milling using high-energy ball milling can make the particle size into nano. Milling for 1, 2, 4, and 6 hours reduced the particle size to 1078, 475, 398, and 203 respectively. The reduction in size resulted in an increase in the surface area to 41, 55, 72, 83, and 110 $m^2g^{1,}$ respectively.

The highest elemental content in nano zeolite is O, with 56.82%, and Si, with 30.71%, while the lowest element content is Mg, which is 0.62%. This is in line with Estiaty (2015) research results, reporting that the main composition of zeolite is dominated by Si 72.3% and Al 10.68%. In addition, there are also cations such as Na, K, Ca, and Mg, which function as a counterweight to the negative charge originating from Si and Al filling the center of the tetrahedron of four oxygen atoms. The highest elemental content in nano crab shells is O, 46.02%, and Ca, 28.01%. Meanwhile, the lowest element content is K, which is 0.06%. This is in line that Ca is the highest elemental content found in crab shells, which is 14.96%. The main content in crab shells are calcium and magnesium carbonate, chitin, and some proteins. Handayani et al., (2019) stated that crab shells contained high Ca, which could be identified early by the hard-shell shape.

Table 2. Particle size of Nano Materials

Diameter (nm)	Nano Zeolite		Nano Crab shell	
	Total	%	Total	%
1-10	1669	35.61	0	0
10-20	1129	24.09	2448	52.86
20-100	1486	31.71	2071	44.72
100-250	290	6.19	78	1.68
250-1000	112	2.40	33	0.71
>1000	1	0.02	1	0.02
	Mean = 39.78 nm		Mean = 30.84 nm	

Table 3. Effect of treatments on agronomic traits

Treatments	Plant Height (cm)	Leaf Number	Bulp Number	Bulp Diameter (cm)	Shoot Fresh Weight (g)	Shoot Dry Weight (g)	Root Fresh Weight (g)	Root Dry Weight (g)	Bulb Fresh Weight (g)	Bulb Dry Weight (g)	Yield (kg/ha)
Soil Types											
Entisol	21.75 b	15.47 b	8.45 b	11.21 b	6.29 b	0.47 b	1.70 b	0.12 b	11.97 b	1.44 b	1685 b
Inceptisol	28.37 a	24.93 a	12.97 a	17.35 a	20.27 a	1.42 a	3.09 a	0.41 a	38.70 a	1.80 a	5450 a
Nano Materials											
Zeolite	24.98 p	22.20 p	10.97 p	14.44 p	13.72 p	0.99 p	2.13 x	0.22 x	24.80 p	1.56 p	3490 p
Crab Shell	25.14 p	18.20 q	10.45 p	14.12 p	12.69 p	0.90 p	2.66 x	0.31 x	25.90 p	1.67 p	3636 p
NK doses											
0 (blanko)	24.55 x	17.58 x	8.97 x	10.69 y	8.35 x	0.68 x	1.28 x	0.15 x	17.80 y	1.31 x	2506 y
125:50	27.67 x	18.00 x	10.75 x	15.81 x	14.08 x	0.96 xy	2.26 x	0.21 x	28.73 x	1.61 x	4046 x
250:100	24.32 x	20.58 x	12 x	14.85 x	15.78 x	1.14 x	3.155 x	0.35 x	30.57 x	1.81 x	4305
375:150	24.75 x	22.67 x	10.67 x	15.15 x	13.96 x	0.09 xy	2.70 x	0.34 x	23 xy	1.67 x	3238xy
500:200	24.01 x	22.17 x	11.17 x	14.90 x	13.87 x	1.03 xy	2.59 x	0.28 x	26.60 xy	1.68x	3745xy
Soil> <Nano> <NK dose	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)	(-)
CV (%)	21.24	36.66	31.12	14.87	24.58	37.37	30.78	14.73	34.45	43.99	34.46

Plant Height (cm)

Based on Figure 1, there was an increase in plant height from the first week to the seventh week. The decrease in plant height at week 8 was caused by the plant starting to fall. Shallot plants ready to be harvested have the characteristics of a yellowing crown, and the plant begins to fall. The combination of treatments with the highest average plant height was the treatment of shallots grown in Inceptisol soil using nano crab shell coated fertilizer with a dose of 125:50 (Table 1). The best shallots height reached 35 cm, according to the height of shallots in general between 1-50 cm depending on the variety. This treatment had a better plant height than the conventional treatment, producing a plant height of less than 30 cm. This is thought to be caused by nano fertilizer with the right dose, which can increase Inceptisol's ability to increase the growth of shallot plants. Khan et al., (2021) we propose macronutrients incorporated slow-release based nano-fertilizer using nanozeolite as a carrier. A simple chemical approach was used

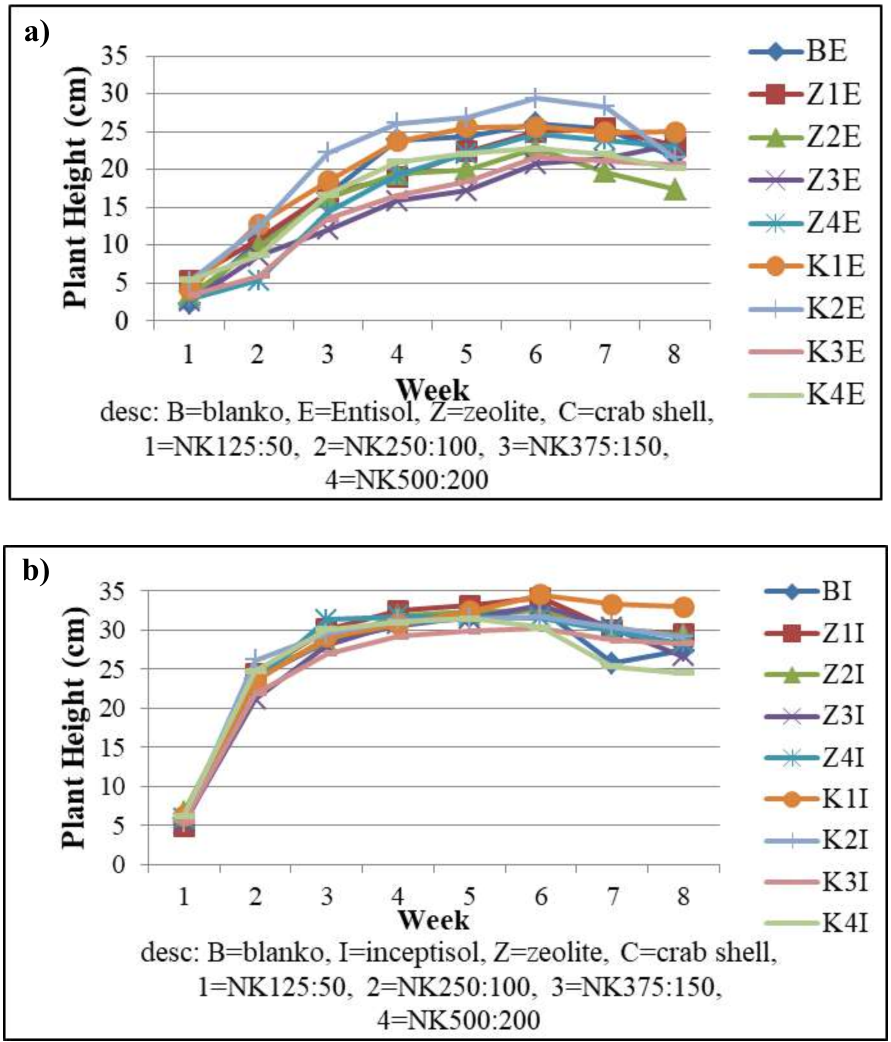

Figure 1. Plant Height as affected by NK-Z and NK-C in (a) Entisols and (b) Inceptisols

to synthesis the proposed nanozeolite composite fertilizer (NZCF) stated that nano fertilizer was a nano-sized fertilizer containing nanoparticles and nutrient encapsulation, capable of releasing micro and macronutrients targeted at plants. Nanomaterials can be used to hold nutrients for plants for a long time.

Number of Leaves

Based on Table 1, each treatment did not show significant results. Based on Figure 2, the combination of Inceptisol soil treatment with nano zeolite coated fertilizer at a dose of 125:50 had the highest average number of leaves. The number of leaves for each treatment increased every week. However, there was a decrease in the number of leaves in the last week. The shallot that is ready to be harvested begins to fall, and the leaves turn yellow until they are almost wilted. The highest number of leaves was observed in the combination of Inceptisol treatment and zeolite coated fertilizer at a dose of 125:50 (Table 1), proving that nano fertilizers can increase plant growth by controlling the release of nutrients so that they are available according to to plant needs. The highest number of leaves reached 45 strands; this treatment was better than the conventional treatment, producing less than 30 leaves. In general, nano zeolite has advantages compared

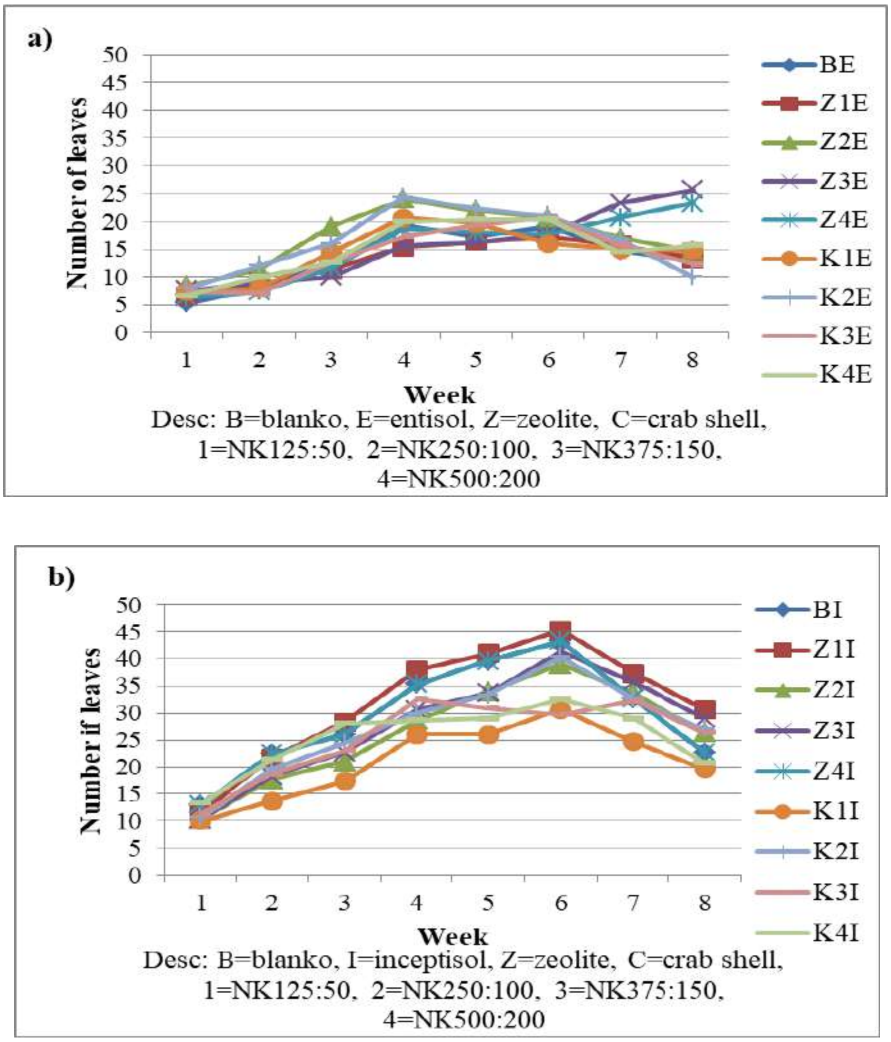

Figure 2. Number of leaves as affected by NK-Z and NK-C in (a) Entisols and (b) Inceptisols

to zeolite. Zeolite modified to nano size has a high surface area, mesoporous structure, and higher nutrient loading capacity. Using nanomaterials in slow-release fertilizers can increase the nutrient retention capacity of the soil (Khan et al., 2021).

Number of Bulbs

Based on Table 1, the highest average number of bulbs was found in the combination of Inceptisol treatment and nano zeolite coated fertilizer at a dose of 250:100. The highest number of bulbs reached 15 bulbs, and this treatment was better compared to the conventional treatment, which produced less than 6-7 bulbs. This proves that the use of nano zeolite as a coating can affect the yield of shallots. Coatings using nanomaterials can control nutrient output so that nutrients remain available according to plant needs during the planting period. The research results of Khan et al., (2021) also showed that using nano zeolite fertilizers could improve the soil's physical, chemical, and biological properties to increase plant growth and yield. Nano zeolite can release nutrients for a longer period when compared to conventional fertilizers, thereby reducing nutrient leaching (Lateef et al., 2016).

The average number of bulbs in Inceptisol was better than in Entisol, proving that Inceptisol had more favorable physical and chemical properties

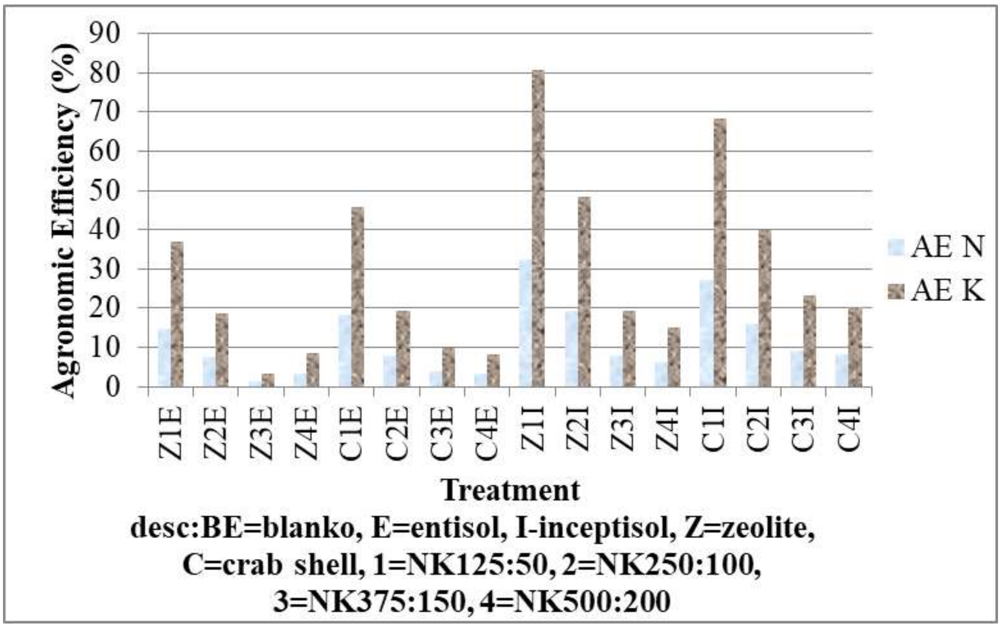

Figure 3. Agronomic Efficiency of NK-Z and NK-C

for plants. Fertilization at the appropriate dose can improve the physical and chemical properties of the soil. Napitulu & Winarto (2010) stated that the use of N fertilizer at a dose of 250 kg/ha and K fertilizer at 100 kg/ha was estimated to be able to increase the quantity and quality of shallot crop yields.

Bulb Diameter (mm)

Based on Table 1, the highest value of bulb diameter was found in the combination of Inceptisol treatment and nano zeolite coated fertilizer at a dose of 125:50. The highest value of bulb diameter reached 19 mm, and this treatment was better compared to the conventional treatment, which produced bulb diameter of less than 6 mm. This result shows that nano zeolite can control the nutrient output according to the plant's needs during the growing period. Yuvaraj & Subramanian (2018) mentioned that modification of fertilizer with nano zeolite could reduce fertilizer loss due to evaporation and leaching. Zeolites could retain nutrients in the root zone, which could be released according to plant needs.

Shoot Fresh Weight (g)

The combination of Inceptisol treatment and nano zeolite coated fertilizer at a dose of 250:100 had the highest average fresh weight of shoots compared to other treatments (Table 1). The highest shoot fresh weight reached 29 g, which was better than the conventional treatment, producing a fresh shoot weight of less than 6 g. Nano zeolite can control the release of nutrients properly to fit the needs of plants. Based on the analysis of the physical and chemical properties of the soil, Inceptisol has more favorable properties for plant growth and development than Entisol. The change of particles to nano size can increase the surface area and the number of pores to increase the ability to hold nutrients. The surface area of zeolite can be increased by ball milling. Ball milling is a top-down approach to reduce the size of the zeolite so as to increase absorption. Cations such as NH^{4+} and K^{+} can be adsorbed and desorbed slowly according to plant needs. Nano zeolite has a negative charge that is able to absorb cations and then release them regularly and stably. This process will produce nano fertilizer formulations that help regulate the release of nutrients, increase fertilization efficiency, and prevent environmental harm (Yuvaraj and Subramanian, 2018).

The elements N and K are very important in plant growth and yield. The research results by Napitulu & Winarto (2010) showed that applying N fertilizer at a dose of 250 kg/ha and K fertilizer at a dose of 100 kg/ha could increase the growth and yield of shallots.

Shoot Dry Weight (g)

The combination of Inceptisol treatment and nano zeolite coated fertilizer at a dose of 250:100 had a higher average shoot dry weight compared to other treatments (Table 1). The highest shoot dry weight reached 1.82 g, and this treatment was better compared to the conventional treatment, which produced shoot dry weight of less than 0.45 g. The shoot dry weight is related to the fresh shoot weight, in which the dry shoot weight shows the absolute weight of the plant canopy. In harmony with the fresh shoot weight, the average shoot dry weight in the treatment combination proved that the nutrients were well absorbed, thereby increasing the yield of shallots. The use of nano zeolite as a fertilizer coating was proven to be able to provide plant nutrient needs during the growing period.

Root Fresh Weight (g)

The highest fresh root weight was found in the combination of Inceptisol treatment using nano crab shell coated fertilizer at a dose of 375:150. The highest root fresh weight reached 5.56 g, which was better than the conventional treatment, producing a fresh root weight of less than 1.20 g. This is thought to be caused by the influence of crab shells as a coating, which is able to hold nutrients so they are not released directly into the environment. In addition, crab shells can chelate metals in the soil to prevent interactions between metals and nutrients, such as P and K, so that nutrients can be available to plants. The higher the dose of fertilizer given to plants, the more nano crab shells were given. This then increases the availability of nutrients in the soil and increases nutrient uptake by plant roots (Rais et al., 2017).

Root Dry Weight (g)

The combination of Inceptisol treatment and nano crab shell coated fertilizer at a dose of 375:150 had maximum root dry weight compared to other treatments (Table 1). The highest root dry weight reached 0.88 g, and this treatment was better compared to the conventional treatment, which produced root dry weight of less than 0.12 g. In accordance with the fresh weight of the roots, the combination of treatments with the highest dry weight of roots proved that the highest accumulation of organic compounds in the roots was found in the combination of Inceptisol and nano crab shell coated fertilizer at a dose of 375:150. Modifying crab shells into nano size can also increase the surface area and pore density so that it can hold nutrients and is not easily released into the environment. In addition, based on the analysis of the elemental content contained in crab shells, it shows that crab shells contain elements such as Ca, C, and Na, which can increase plant growth and yield.

Bulb Fresh Weight (g)

The highest value of fresh bulb weight was found in the combination of Inceptisol and nano zeolite coated fertilizer at a dose of 250:100 (Table 1). The maximum bulb fresh weight reached 49.78 g, and this treatment was better compared to the conventional treatment, which produced bulb fresh weight of less than 9.31 g. In addition to affecting plant growth, the use of nano zeolite coated fertilizer also affects the yield of shallots.

The quality of the bulb is related to the formation of the canopy. N element helps in the formation of the canopy, and the K element helps the process of translocation of photosynthate products to be better so that the quality of the bulb produced is also better (Prasetya et al., 2015). Zeolite formulation into nano fertilizer can increase the efficiency of N and K fertilization in plants. The slow release of nutrients is able to provide nutrients during the growing period according to the period needed by the plant.

Bulb Dry Weight (g)

Based on Table 1, the combination of Inceptisol and nano zeolite coated fertilizer at a dose of 250:100 had higher bulb dry weight compared to other treatments. The highest bulb dry weight reached 8.95 g, and this treatment was better compared to the conventional treatment, which produced bulb dry weight of less than 1.36 g. Bulb dry weight is the absolute weight of the tubers, which is also related to the bulb fresh weight. The combination of treatments with the highest bulb dry weight was in line with the combination of treatments with the highest bulb fresh weight (Table 1). Fertilization can improve the quality of Inceptisol's physical and chemical properties, which are beneficial for plants. Modification of zeolite into nano size was also proven to be able to increase the ability of zeolite to hold nutrients better than the previous size so as to control the release of nutrients better as well.

Agronomic Efficiency (%)

Based on Figure 3, the best agronomic effectivity value was found in the combination of Inceptisol and fertilizer coated with nano zeolite at a dose of 125:50. This result proves that nano zeolite is able to hold nutrients well and release it according to plant needs during the growing period. Crop yields influence agronomic efficiency. Inceptisols were proven to have more favorable soil physical and chemical properties for the growth and yield of shallots compared to Entisols. Application of fertilizer that has been coated using zeolite allows nutrient output to be well controlled. Yuvaraj & Subramanian (2020) stated that fertilizers coated with nanomaterials could increase nutrient absorption, increase soil fertility, and reduce fertilizer toxicity. Nano zeolite is able to carry nitrogen, phosphorus, potassium, and micronutrients in fertilizers so as to increase plant productivity. The use of zeolite in nano-tech fertilizers can provide nutrients to plants for 50 days. This shows that nano zeolite coated fertilizer is able to provide nutrients longer than conventional fertilizers, which can provide nutrients only for 10-12 days. The best result, which is at a dose of 125:50, is in line with the research of Napitulu & Winarto (2010), stating that N at a dose pf 250 kg/ha and K at a dose of 100 kg/ha gave the best results on uncoated shallots, thus allowing the use of smaller doses of fertilizers with coatings.

CONCLUSIONS

Treatments of soil types, encapsulation materials, and doses of NK fertilizer did not significantly affect the agronomic traits of shallot plants. These plants grew well and produced better yields in inceptisols than in entisols. The types of encapsulation material and NK doses had no significant effect. Although not statistically significant, the shallot yield produced by nano crab shell coated NK treatment was better than that produced by nano zeolite coated NK. NK doses with a ratio of 125:50 and 250:100 obtained the highest yields, but the highest agronomic efficiency (EA) was obtained in nano zeolite coated NK at a dose of 125:50.

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Seed Bio-Priming to Enhance Seed Germination and Seed Vigor of Rice Using Rhizobacteria from The Northern Coast of Pemalang, Central Java, Indonesia

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ABSTRACT

The growth and yield of plants are strongly influenced by the early growth ability of the plants. Similar germination and good seed vigor will greatly support plant growth and increase production. Increasing the germination and vigor of seeds can be done through biopriming. The application of biopriming using rhizobacteria is developing environmentally friendly agricultural technology. This study aimed to determine the effect of inoculation of rhizobacteria from the north coast of Pemalang on rice plants' germination and vigor index. The study was arranged in a Randomized Block Design, consisting of 10 treatments with three replications. Ten rhizobacteria isolates were isolated from the North Coast of Pemalang, Central Java, consisting of Ju1, Jn3, Jn1, J, J12, J5, Kn1, A3, Jn, and K3. The biopriming with rhizobacteria isolated from the rice rhizosphere of the Northern Coast of Pemalang increased the seed germination rate, seed vigor index, and early vegetative growth of rice seedlings. Inoculation with isolate J12 produced the highest vigor index of 8280.01. The results of this study imply that the application of rhizobacteria from saline soil has the potential to increase the vigor of rice seedlings to impact better seedling growth in saline conditions.

Keywords: Biopriming, Germination, Rhizobacteria, Rice, Vigor

ABSTRAK

Pertumbuhan dan hasil tanaman sangat dipengaruhi oleh kemampuan tumbuh awal tanaman. Daya kecambah yang seragam dan vigor benih yang baik sangat mendukung untuk dapat tumbuh dengan baik dan mendukung peningkatan produksi. Upaya peningkatan daya kecambah dan vigor benih dapat dilakukan dengan perlakuan biopriming. Penerapan biopriming menggunakan rhizobakteri merupakan pengembangan teknologi pertanian yang ramah lingkungan. Penelitian ini bertujuan untuk menguji pengaruh inokulasi rhizobakteri dari tanah salin di pantai utara Pemalang terhadap daya berkecambah dan indeks vigor tanaman padi. Penelitian disusun menggunakan Rancangan Acak Kelompok, dengan tiga ulangan. Sebagai perlakuan, 10 isolat rhizobakteri diisolasi dari Pantai Utara Pemalang Jawa Tengah yakni Ju1, Jn3, Jn1, J, J12, J5, Kn1, A3, Jn, dan K3. Perlakuan biopriming dengan isolat rhizobakteri yang berasal dari rizosfer padi asal Pantai Utara Pemalang mampu meningkatkan kecepatan perkecambahan benih, indeks vigor benih dan pertumbuhan vegetatif awal benih padi. Inokulasi dengan isolat J12 mampu menghasilkan indeks vigor tertinggi sebesar 8280,01. Implikasi dari hasil penelitian ini adalah bahwa aplikasi rhizobcateri yang berasal dari lahan salin berpotensi untuk meningkatkan vigor bibit tanaman padi sehingga akan memberikan dampak terhadap pertumbuhan bibit yang lebih baik pada kondisi saline.

Kata kunci: *Biopriming, Perkecambahan, Rhizobakteria, Padi, Vigor*

INTRODUCTION

Rice is the staple food of the Indonesian people, and the consumption pattern of the people in urban areas is almost the same as that in rural areas (Saliem et al., 2019). On the other hand, Indonesian rice consumption has been quite high since 1996. However, there is a downward trend wherein 2020, and it has reached 78.42 kg per capita per year (Anggraeni, 2020). The trend of decreasing rice consumption is a positive thing. Nevertheless, national rice production must continue to be increased in terms of quality and food safety (Saliem et al., 2019; Anggraeni, 2020).

Increased agricultural production is strongly influenced by the interaction between environmental genetics and plant management. Good plant growth will start with good quality plant seeds in

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terms of seed germination and seed vigor (Ayalew et al., 2018). Seeds that can germinate quickly and have uniform seedling growth are essential in crop production (Hélnia et al., 2021). Seed vigor is a very important index of seed quality. It is a physiological marker of commercial seed lots mostly those with similar germination percentages, aiming to identify lots with a higher probability of performing well after sowing and/or during storage (Wen et al., 2018). Hao et al., (2020) stated that high seed vigor would determine the potential for rapid and uniform seed emergence and increase yields by up to 20 percent.

Various studies have reported that priming treatment was able to increase germination and seed vigor by using various materials, such as Polyethylene glycol, Calcium chloride, Calcium aluminum silicate, gibberellic acid (GA), salicylic acid, citric acid (CA), sodium chloride (NaCl), potassium chloride (KCl), zinc (Zn) and iron (Fe) (Nouri & Haddioui, 2021). The development of environmentally friendly priming technology is urgently needed. The use of beneficial microorganisms to increase seedling vigor is environmentally friendly, which positively affects plants and the soil environment. Beneficial microorganisms, such as Plant Growth Promotion Rhizobacteria (PGPR), have an important role in stimulating plant growth through N2 fixing mechanisms, suppressing ethylene levels, induction of resistance to pathogens, solubilizing nutrient, production of siderophores, and phytohormones (dos Santo et al., 2020). Bacterial inoculation methods to promote plant growth have been developed, among others, through seed coating, foliar application, direct application through the soil, and seed priming, by immersing the seeds in a bacterial suspension before the physiological process of the seed begins in the seed. At the same time, the radicle and plumule emergence is prevented (Mahmood et al., 2016). Madyasari et al., (2017) reported that seed priming using rhizobacteria increased the vigor of chili seeds after being stored for 24 weeks. Furthermore, Roslan et al., (2020) reported that *Enterobacter* spp. increased the vigor index 19.6% higher than without *Enterobacter* spp. inoculation promotes the initial vegetative growth of okra plants and increases leaf area and greenness.

Various researchers have reported that beneficial microorganisms can be utilized to increase the vigor index of seeds. *Pseudomonas fluorecence*to could increase the germination and vigor of the East Indian Sandalwood (*Santalum album* L) (Chitra & Jijeesh, 2021), Enterobacter spp. could increase the vigor of okra seeds (Roslan et al., 2020), and *Azospirillum, Azotobacter, and Bacillus* could increase the germination and vigor index of sorghum plants (Widawati & Suliasih, 2018).This condition opens up opportunities for using rhizobacteria originating from a saline environment to stimulate germination and early vegetative growth of rice plants. Saline soils in Indonesia are still very large, reaching 12,020 million ha or 6.20% of the total land area of Indonesia, and 9 million ha is potential for rice cultivation (Karolinoerita & Yusuf, 2020). Several PGPR isolates isolated from the rhizosphere of rice plants in saline rice fields can produce growth regulators of the auxin group and fix N. These isolates have the potential to stimulate growth, and in saline conditions, are expected to increase the vigor of rice seedlings. The effectiveness of Rhizobacteria derived from saline soils needs to be tested to determine their potential to improve the vigor index and early vegetative growth of rice plants. This study aimed to examine the effects of rhizobacteria inoculation from saline soils on the Northern Coast of Pemalang on the germination and vigor index of rice plants.

MATERIALS AND METHODS

The Seed Material

The rice seed used in this study was Inpari Unsoed 79 Agritan Rice Variety collection from the Laboratory of Plant Breeding and Biotechnology, Faculty of Agriculture, Jenderal Soedirman University, Purwokerto. The Inpari Unsoed 79 Agritan variety is a rice variety that is resistant to salinity stress.

Bacterial Culture Preparation

A total of 10 rhizobacteria isolates were prepared by cultivating them in a Nutrient Broth (Himedia) media. A total of 1 ose of bacterial colonies were inoculated on 250 ml of Nutrient Broth media, then incubated with a shaker at a speed of 120 rpm for 24 hours at room temperature to reach a population density of 10^7 CFU/mg.

Bacterial Inoculation

Each treatment consisted of 100 grains of rice seeds. Before being inoculated, the rice seeds were sterilized using sodium hypochlorite 0.02% for two minutes (Widawati & Suliasih, 2018) and washed with sterile distilled water three times. Sterile rice seeds were put in a petridish and then soaked in 20 ml of bacterial culture for 30 minutes. The inoculated rice seeds were then planted in a seed box with sterile sand media and maintained in a greenhouse until the age of 25 days after planting.

Experimental Design

The research was carried out in the Laboratory of Agronomy and Horticulture, Faculty of Agriculture, Jenderal Sudirman University, Purwokerto, Central Java, Indonesia. The study was conducted for two months, starting from September to October 2021. The study was arranged using a Randomized Block Design, consisting of 10 treatments with three replications. As treatments, 10 rhizobacteria were isolated from the North Coast of Pemalang, Central Java, consisting of Ju1, Jn3, Jn1, J, J12, J5, Kn1, A3, Jn, and K3.

Observed Variables

The seeds were planted in trays containing sterile sand, with each treatment comprising of 100 seeds. Germinated seeds were recorded every time they germinated from the total number of seeds sown. Based on the germination data, the percentage of germination was calculated according to the formula of Polaiah et al., (2020), and the germination rate was calculated by the formula of Chitra & Jijeesh, (2021) as follows :

Germination (%) =

$$\frac{Number\ of\ seeds\ that\ germinated}{Total\ number\ of\ seeds} x100\% \qquad (1)$$

Germination rate =

$$\frac{G1}{T1} + \frac{G2}{T2} + \frac{G3}{T3} + \ldots\ldots + \frac{Gn}{Tn} \qquad (2)$$

Remarks: G1, G2, G3, and Gn are % seeds germinate at T1, T2, T3, and Tn, respectively, and T1, T2, T3, and Tn are the first, second, third, and n day counting from sowing, respectively.

Variables of early vegetative growth of rice seedlings included plant height (cm), total root length measured by the intersection method (Bohm, 1979), leaf greenness measured by chlorophyll meter (Konica Minolta Chlorophyll Meter SPAD-502Plus), and biomass. The seed Vigor index was calculated based on the following formula:

Seed Vigor Index =

(shoot length + root length) x germination (%) (3)

Statistical analysis

The data obtained from this study were analyzed by ANOVA using SAS 9.1 software followed by DMRT at α=5%.

RESULTS AND DISCUSSION

Seed germination and germination rate

The observations found that the biopriming of rice seeds with various rhizobacteria isolates did not show any effect on rice seed germination. The percentage of rice seed germination was still high, ranging from 93.33% to 100.00 percent (Table 1). The high percentage of germination in all treatments was caused by the condition of the seeds where the seeds used were rice seeds that had just been harvested for about two months so that the seeds were still in good condition and had not deteriorated. The germination rate showed the impact of biopriming ($p<0.05$). The germination rate of rice seeds in different biopriming treatments varied between 32.89 – 24.99. The highest germination rate was obtained in the treatment of rhizobacteria of J5 isolate, while the lowest germination rate was obtained in isolate K3 (Table 1). The germination rate in treatment J5 isolate was not significantly different from that in control, J12, J, Ju1, and Jn (Table 1). Germination rate indicates the speed at which sprouts appear, and the ability of sprouts to emerge is strongly related to the energy for germination.

The results of this study indicated that biopriming with rhizobacteria could enhance seed vigor and early vegetative growth of rice seedlings. Biopriming treatment did not significantly affect the seed germination percentage, which was seen from the percentage of germination showing an insignificant difference between control and other treatments, ranging from 93.33 percent to 100 percent. This illustrates that the physiological quality of the seeds is still good. These results are in line with the results of Madyasari et al., (2017), where the seed biopriming treatment did not significantly affect seed germination because each seed had high vigor. The seed germination rate in this study showed a higher increase in the J5 isolate treatment of 32.89 seeds/day (Table 1). The increase in seed germination rate in the rhizobacteria inoculation treatment is closely related to the presence of plant growth substances that are capable of being synthesized by bacteria from the auxin, cytokinin and gibberellin groups, which trigger the activity of specific enzymes that promote faster germination, such as -amylase which helps starch assimilation (Nezarat & Gholami, 2009). Starch assimilation in the seed germination process will also increase the energy available for the germination process, which will cause an increase in the germination rate (Chitra & Jijeesh, 2021). According to Mitra et al., (2021), living microorganisms have different multifunctional capabilities, such as the production of plant growth regulators like auxins, cytokines, abscisic acid and gibberellins, which are produced as secretions of effector molecules and secondary metabolites through modulation of various pathways, which are the most suitable for the biopriming method. Murunde & Wainwright (2018) reported that biopriming treatment using *Bacillus subtilis* and *Serratia nematodiphila* increased the germination of onion seeds.

Seedling growth and biomass

Seed priming treatment in this study positively affected seedling growth and biomass. Seed biopriming with rhizobacteria had a significant effect on the variables of plant height ($p=0.0340$), root length ($p=0.0191$), leaf greenness ($p=0.0030$), and plant biomass variables. The treatment of rhizobacteria inoculation strongly influenced the root length of rice seedlings. Overall total root length increased by 83.41 percent compared to the control. The inoculation treatment of Kn1 isolate reached the highest plant height much higher than the control, although inoculation treatments of rhizobacteria isolates were not significantly different (Table 2). Biopriming treatment using rhizobacteria was able to increase plant height by 17.61 percent.

Table 1. The effect of rhizobacteria inoculation on seeds germination and germination rate

Treatments	Germination (%)	Germination Rate (germination/day)
Control	98.67 a	31.09 ab
Ju1	100.00 a	29.91 abc
Jn3	99.00 a	27.94 bcd
Jn1	98.67 a	28.67 bc
J	97.00 a	29.48 abc
J12	98.33 a	30.67 ab
J5	97.67 a	32.89 a
Kn1	98.33 a	28.78 bc
A3	97.67 a	26.48 cd
Jn	100.00 a	29.28abc
K3	93.33 a	24.99 d

Remarks: Means followed by same letters in the same column are not significantly different according to DMRT 5%.

Table 2. The effect of rhizobacteria inoculation on vegetative growth of rice seedling

Treatments	Plant Height (cm)	Roots Length (cm)	Leaf Greennes (SPAD unit)	Biomass (mg)
Control	24.72 b	26.66 b	17.84 c	32.67 c
Ju1	29.15 a	47.76 a	19.38 bc	44.67 a
Jn3	30.03 a	43.23 a	23.59 a	46.67 a
Jn1	28.85 a	42.72 a	19.27 bc	44.67 a
J	29.25 a	47.66 a	18.17 c	42.67 ab
J12	29.58 a	54.60 a	19.63 bc	42.67 ab
J5	29.24 a	51.67 a	19.73 bc	44.00 ab
Kn1	29.85 a	49.51 a	20.97 b	41.33 ab
A3	29.65 a	52.91 a	20.08 bc	36.67 bc
Jn	26.85 ab	51.46 a	20.34 bc	47.33 a
K3	28.28 a	47.45 a	19.55 bc	42.67 ab

Remarks: Means followed by same letters in the same column are not significantly different according to DMRT 5%.

The results indicated an increase in the greenness of the leaves. The greenness of the leaves reflects the total chlorophyll content in the plant leaves. The biopriming treatment with rhizobacteria isolates had a significant effect (p=0.0030) on increasing the greenness of the leaves, with an average value of 20.07 units.

The effect of biopriming treatment is clearly visible in the variable biomass of rice seedlings. Plant biomass in the biopriming treatment, on average, was able to produce biomass of 43.33 mg, which was greater than the control. The highest biomass of rice seedlings was achieved in the inoculation treatment of Jn isolate (Table 2). It can be seen that all rhizobacteria isolates were able to increase biomass production by 32.64 percent.

In general, biopriming treatment using rhizobacteria isolates increased the growth of rice seedlings. The application of rhizobacteria enhanced vegetative growth, which was triggered by the ability of rhizobacteria to produce auxins, especially indole acetic acid (IAA) (Chitra & Jijeesh, 2021; Chauhan et al., 2021). The ability of rhizobacteria to produce IAA will stimulate root elongation so that the

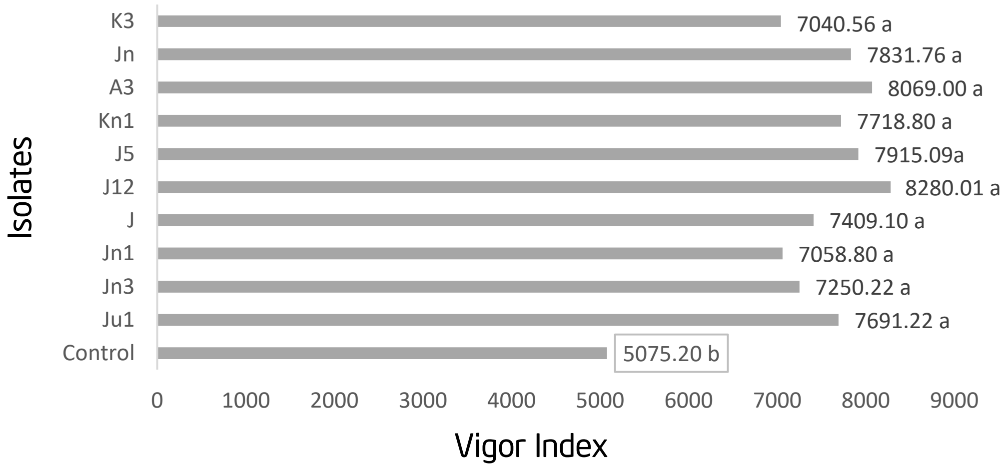

Figure 1. The effect of rhizobacteria isolates on seed vigor index

Table 3. Pearson Correlation Coefficient

	Plant Height	Germination	Root Length	Seed Vigor	Germination Rate	Leaf greenness	Biomass
Plant Height	1.00000						
Germination	-0.02904	1.00000					
Root Length	0.68563*	-0.17992	1.00000				
Seed Vigor	0.76862*	0.12582	0.94482*	1.0000			
Germination Rate	-0.18291	0.51039*	-0.20473	-0.06713	1.0000		
Leaf Greenness	0.22703	0.16298	0.00342	0.09224	-0.01063	1.0000	
Biomass	0.383882*	0.01838	0.43825*	0.45448*	-0.07033	0.29876	1.00000

root surface area that interacts with soil colloids increases and results in increased nutrient and water uptake (Purwanto et al., 2017; Purwanto et al., 2019; Rahma et al., 2019). Rahma et al., (2019) stated that the increase in root growth through the expansion of the root system was stimulated by hormones, thereby increasing nutrient uptake caused by the ability of rhizobacteria to dissolve nutrients such as P. Rhizobacteria can increase the availability of nutrients in the soil (N,P, K) so that nutrient uptake (N, P, K) increases, thereby increasing photosynthetic pigment and activity (Chauhan et al., 2021). Inoculation of rhizobacteria isolates can increase plant height and root length of rice seedlings through the ability to provide and mobilize the absorption of various nutrients in the soil through the ability to enhance capacity in synthesizing and modifying the concentration of numerous phytohormones, dissolving P elements, and producing the Indole Acetic Acid hormone (Rahma et al., 2019). The results of this study also showed that the biopriming treatment with rhizobacteria isolates was able to increase the biomass of rice seedlings. This result is in line with Moeinzadeh et al., (2010), stating that biopriming of sunflower seeds with *Pseudomonas fluorescens* significantly improved the growth of seedling height, root length, and biomass compared to control.

Seed vigor

The effect of biopriming rice seeds with rhizobacteria isolates was significant on the vigor of the seeds. The variance analysis showed that the rhizobacteria isolates' treatment significantly affected rice seedlings' vigor (p=0.0182). The observations found that the highest seed vigor was achieved in the J12 isolate treatment, and the lowest was in control (Figure 1).

Biopriming of rice seeds with rhizobacteria isolates significantly increased the vigor index. It can be seen that in all rhizobacteria isolate treatments, and the vigor index value increased compared to the treatment without biopriming (control). Seed biopriming increased rice seed vigor by 50.27 percent compared to control. The highest vigor index was achieved in biopriming with J12 isolate, where the vigor index value increased by 63.15 percent compared to the control. The germination percentage influences the increase in the vigor index. Still, it is also strongly influenced by the initial growth of rice seedlings, especially root growth and plant height. The results showed a significant correlation between plant height and vigor index (r=0.76862), as well as between root length and the vigor index variable (r=0.94482) (Table 3). The effect of biopriming on seed vigor index is induced by the ability of rhizobacteria to synthesize cytokines. This hormone stimulates cell division, and the effect of auxin as a hormone stimulates cell elongation (Agbodjato et al., 2016). Roslan et al., (2020) reported that inoculation of okra seeds with *Enterobacter* sp. increased the initial growth of okra seedlings compared to inoculation based on hypocotyl length, radicle, number of lateral roots and vigor index.

CONCLUSIONS

In general, it can be concluded that the biopriming treatment with rhizobacteria isolates derived from the rice rhizosphere from the Northern Coast of Pemalang increased the seed germination rate, seed vigor index, and early vegetative growth of rice seedlings. Inoculation with J12 isolates produced a higher vigor index than the control but was not significantly different from other isolates. The implication of the results of this study is that the application of rhizobacteria from saline soil has the potential to increase the vigor of rice seedlings so that it will have an impact on better seedling growth in saline conditions.

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Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Application of Empty Fruit Bunches of Oil Palm and *Indigofera zollingeriana* for Conservation of Oil Palm Plantation

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ABSTRACT

Oil palm empty fruit bunches are materials used as organic fertilizers that can be applied to oil palm plantations, thereby reducing the use of inorganic fertilizers. *Indigofera zollingeriana* is an appropriate alternative as an interplant because of its high branch and leaf development. Functions as a ground cover and a supplier of carbon stocks naturally plays a role in water and soil conservation. This study aims to determine the effect of oil palm empty fruit bunches and I. *zollingeriana* on land improvement to support oil palm growth and production. Variables observed included changes in soil water content, soil microorganism activity, and carbon stock. The results showed that the soil planted with I. *zollingeriana* and given the empty fruit bunches of oil palm had a higher soil moisture content. The highest soil carbon stock, oil palm carbon stock, and vegetation carbon stock were 81.6 t ha^{-1}, 36.60 t ha^{-1}, and 1.89 t ha^{-1}, respectively. The population and activity of microorganisms varies. The highest total microorganisms were treated with I. *zollingeriana* and oil palm EFB 105 (10^5CFU g^{-1}), while the lowest was 60 (10^5CFU g^{-1}). Planting I. *zollingeriana* and providing oil palm empty fruit bunches increased groundwater reserves by 36.71%.

Keywords: Carbon stock, Indigofera zollingeriana, Microorganisms

ABSTRAK

Tandan kosong kelapa sawit merupakan bahan yang digunakan sebagai pupuk organik yang dapat diaplikasikan pada perkebunan kelapa sawit, sehingga dapat mengurangi penggunaan pupuk anorganik. Indigofera zollingeriana merupakan alternatif yang tepat sebagai tanaman interplant karena pertumbuhan cabang dan daunnya yang tinggi. Fungsinya sebagai penutup tanah dan pemasok stok karbon secara alami berperan dalam konservasi air dan tanah. Penelitian ini bertujuan untuk mengetahui pengaruh tandan kosong kelapa sawit dan I. zollingeriana terhadap perbaikan lahan untuk mendukung pertumbuhan dan produksi kelapa sawit. Variabel yang diamati meliputi perubahan kadar air tanah, aktivitas mikroorganisme tanah, dan stok karbon. Hasil penelitian menunjukkan bahwa tanah yang ditanami I. zollingeriana dan diberi tandan kosong kelapa sawit memiliki kadar air tanah yang lebih tinggi. Stok karbon tanah, stok karbon kelapa sawit, dan stok karbon vegetasi tertinggi berturut-turut adalah 81,6 t ha^{-1}, 36,60 t ha^{-1}, dan 1,89 t ha^{-1}. Populasi dan aktivitas mikroorganisme bervariasi. Total mikroorganisme tertinggi pada perlakuan I. zollingeriana dan TKKS kelapa sawit 105 (10^5CFU g^{-1}), sedangkan terendah 60 (10^5CFU g^{-1}). Penanaman I. zollingeriana dan penyediaan tandan kosong kelapa sawit meningkatkan cadangan air tanah sebesar 36,71%.

Kata kunci: *Stok karbon, Indigofera zollingeriana, Mikroorganisme*

INTRODUCTION

Oil palm plant grows, develops, and produces optimally if the availability of groundwater is sufficient all years, with a rainfall of 2000-2.500 mm in the first year and a dry season of less than one month or no dry season (Henson et al., 2005; Kallarackal et al., 2004; Umana & Chinchille, 1991). The oil palm industry has grown exponentially over the past years. This case causes an increase in the number of waste products from the oil palm industry, especially empty fruit bunch (EFB) of palm oil. EFB is produced in large quantities in the local area. Recycling EFB through conversion into a usable product is the most appropriate way to reduce raw waste materials. There are several

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potential solutions for EFB to be used as compost material (Danmanhuri, 1998), and EFB can also be used as ground cover on the plantation (Mohammad et al., 2012), becoming an effective solution and inexpensive waste utilization.

Agricultural land conservation aims to create minimum soil mechanical disturbance, make nurseries without tillage, use organic ground cover, and diversify plants. Benefit potential includes higher productivity and income, climate change adaptation and susceptibility to erratic rainfall distribution, and reduction of greenhouse gas emissions (Kassam et al., 2012). Soil and water conservation through the agroecosystem approach can increase the benefit of farming and improve food security and land productivity (Robert, 2001). Other efforts include simultaneously applying three principles of soil and water conservation, namely minimum tillage, use of permanent ground cover, and plant rotation. A recent study has shown that minimum tillage combined with the cover plant has the potential to offer better soil conservation in cropping systems in tropical mountain areas, as well as to facilitate stability and increase harvest production several times, without the main weaknesses found with the hedge contour (Hobbs, 2007; Shafi et al., 2007). Pansak et al., (2008) found that planting ground cover and legumes showed a positive response and helped control lost nutrients in corn on the moderate slope in northeast Thailand, making this type of soil conservation a proper alternative to tropical mountain areas.

Indigofera zollingeriana is a type of shrub-shaped legume plant that has many leaves. This plant has an important benefit in developing sustainable oil palm plantations because the leaves can be used as a ground cover plant, increasing the source of organic material and carbon stock (Hassen et al., 2006). *Indigofera zollingeriana* can adapt highly to diverse environments and has a variety of important morphological and agronomic characteristics to be used as a forage and ground cover plant (Hassen et al., 2006). *Indigofera zollingeriana* can be used as a ground cover plant to prevent the transport of organic matter and nutrient loss on the soil's surface (Hassen et al., 2006). Utilization of oil palm empty bunches of waste applies the traditional composting method for several months or years to achieve complete decomposition. The high C: N ratio and polymer, such as cellulose and lignin in EFB, act as a natural inhibitor of natural biodegradation (Gaind & Nain, 2007). Using organic waste as organic fertilizer can increase plant productivity, improve soil health, and reduce the waste problem (Gaind & Nain, 2007). This study aimed to determine the effects of *Indigofera zollingeriana* and EFB as well as other treatments on improving the growing environment to support the growth and production of oil palm.

MATERIALS AND METHODS

The materials used were 5-month-*Indigofera zollingeriana* seedlings, 5-year-old-oil palm plants with a shade range of 33-50%, and EFB. Meanwhile, the equipment used included analytical and digital scales (Shimadzu ATX224), digital camera (Sony), Oven (Memmer), multimeter (Tofuda DT830B), and binocular microscope **(Olympus)**. The research was carried out for 12 months from November 2019 to October 2020, at the Oil Palm Plantation at an elevation of 115 m asl. Analysis of soil, fertilizer, and plant net was carried out at the Soil Fertility Laboratory of IPB Bogor.

Treatments Application

The research was arranged in a single factor Randomized Complete Block Design, consisting of five treatments, namely experimental plot overgrown with natural vegetation, experimental plot without natural vegetation, experiment plot

Figure 1. EFB Application

Figure 2. 3-month-*Indigofera zollingeriana* plants

planted with *Indigofera zollingeriana*, experiment plot treated with empty fruit bunch (EFB) of palms oil, and experimental plot planted with *Indigofera zollingeriana* and treated with EFB. Each treatment was replicated three times, resulting in 15 experimental units. The data collected were analyzed statistically using ANOVA at a 5% significance level (Steel and Torrie, 1993)

The first step was preparing the experimental field design by making a test plot with a size of 45 m x 8 m. The number of oil palm plant samples observed during the study was three plants for each treatment plot, so there were 45 plant samples. The image of the application of empty palm bunches can be seen in Figure 1, and the performance of 3-month- *Indigofera zollingeriana* plants can be seen in Figure 2.

The variables observed in this study were groundwater reserves, activities of soil microorganisms, and carbon stocks.

Soil moisture content

Measurement of soil moisture content was carried out in an experimental plot using the planted sensor in the soil at a depth of 10 cm, 20 cm, 30 cm, 40 cm, 50 cm, and 60 cm (Asbur & Ariyanti, 2017), which was then measured by a multimeter (Tofuda DT830B). Measurement was made only once at a determined time in the morning. The measured value was the soil conductivity value. The value of soil conductivity has a certain correlation function with soil moisture content. The correlation function was obtained from calibration (Asbur & Ariyanti, 2017).

The activity of soil microorganisms

The activities of soil microorganisms observed include the total number of microorganisms and soil microorganisms' respiration. The content of organic C, total N, available P, and total K was determined using the Walkley and Black method (Kjeldahl), Bray method and 25% HCL extract by spectrophotometer, and 25% HCL compound with f flame-photometer, respectively. The soil sample was taken from each plot at 0-20 cm depth (Asbur & Ariyanti, 2017)

Carbon stock

Soil carbon stock was calculated by the formula of **C-k n = C-conc × BD × d × CFst.** Oil palm carbon stock was calculated by formula of AGB = 0.0976^{h}total + 0.0706. Meanwhile, the carbon stock of natural vegetation was measured by making a sample plot (1 m x 1 m), and all vegetation was taken and then dried at a temperature of 80°C to constant weight. The dry weight of the biomass obtained was converted to kg ha^{-1} to determine biomass weight in the experimental plot. Then, the

Figure 3. Multi Meter

Figure 4. Measuring SMC

carbon stock was calculated by the formula of C = biomass (kg ha^{-1}) x vegetation C content. (Hairiah et al., 2011)

Data analysis

The formula for calculating soil moisture content is

$$w = \frac{W2\ W3}{W3\text{-}W1} x\,100\% \quad (1)$$

Soil biological activity is calculated by the formula

$$\mathbf{r} = \frac{(a-b)xtx120}{n} \quad (2)$$

while the formula for calculating carbon stock in oil palm plantations is Y = 0,002382 .D2,3385 . H0,9411. Statistical analysis design, using minitab Software version 19 (Sihombing & Arsani, 2022).

RESULTS AND DISCUSSION

The study area is located at an altitude of ± 115 m above sea level, with a relatively flat topography. Climatic conditions show rainfall ranging from 100-489 mm with an average temperature of 26-30°C. Humidity ranges from 78% to 80%, indicating that external environmental conditions require action to improve the growing environment by planting *Indigofera zollingeriana* and giving empty bunches of oil palm. *Indigofera zollingeriana* can be used as a ground cover plant and water storage (Hassen et al., 2006). It also functions as green fertilizer that can have a symbiotic relationship with *Rhizobium* sp. so that it can fix N from the air. Besides, *Indigofera zollingeriana* plants are plants adapted to the shade intensity of 40% (Saijo et al., 2018), so they are planted under oil palm stand 3-5 years after planting. Goh & Hardter (2003) state that the provision of nitrogen can increase leaf area, number of leaves, and average assimilation level in oil palm plants. In this study, the availability of water reserves in the soil was influenced by the planting of *Indigofera zollingeriana* and the treatment of empty oil palm bunches. Figure 3 shows a multimeter tool used to measure soil moisture content, while the documentation when measuring soil moisture content is shown in Figure 4.

The lowest soil moisture content was observed in July (28.78%), while in November, the soil moisture content was 60%. However, the deficit of soil moisture content tended to decrease by treating EFB and *Indigofera zollingeriana* as ground cover. At almost all depths (0-60 cm), the effects of *Indigofera zollingeriana* could reduce the deficit of soil moisture content. Water tends to be available below the soil with *Indigofera zollingeriana* root system (Hassen et al., 2006). In this case, the role of *Indigofera zollingeriana* plant was more significant, especially in dry months, where soil moisture content in the plot planted with *Indigofera zollingeriana*

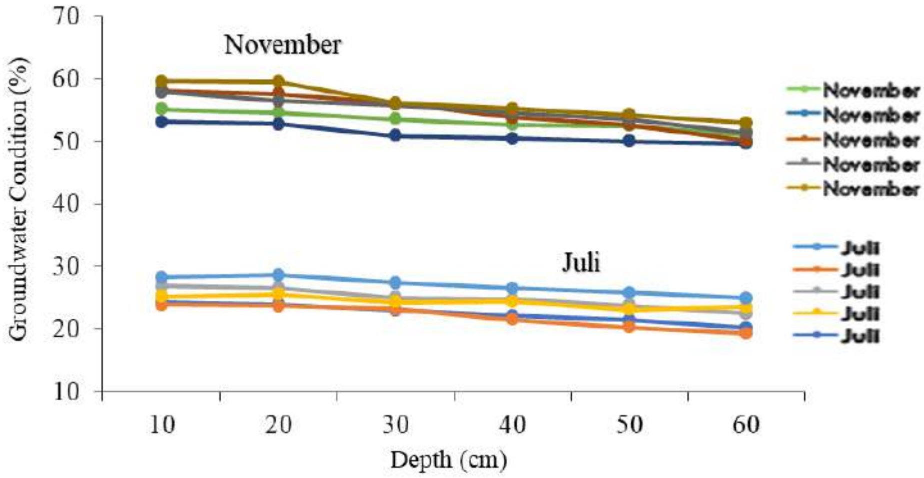

Figure 5. Effects of treatments on the soil moisture content in an extremely dry month (July) and wet month (November)

tended to be better than in the plot without plants. The extreme conditions of soil moisture content in the dry month (July) and wet month (November) can be shown in Figure 5.

To retain soil moisture content in the dry season, it is recommended to provide shade plants (above 80%) and cover the soil with litter (100%) (Saijo et al., 2018). The ground cover with the application of *Asystasia gangetica* can significantly increase the soil moisture content to 33%-66% (Saijo et al., 2018). From October-February, there was an increase in the average daily soil moisture content due to the high rainfall that occurred during these months. The effects of *Indigofera zollingeriana* + EFB started to appear in January, especially at the soil depth of 10-20 cm. The average daily soil moisture content increased in the plot planted with *Indigofera zollingeriana* plants as ground cover. In the rainy season, *Indigofera zollingeriana* + EFB in retaining soil moisture was effective only at a soil depth of 30 cm. This result is due to the effective growth and spread of *Indigofera zollingeriana* roots at a soil depth of 30 cm, thereby allowing rainwater to be retained in the zone.

Meanwhile, in November, these plants could retain higher soil moisture levels, up to a soil depth of 60 cm, because there was water surplus at depths of 10 cm to 60 cm. Water that enters the soil mostly flows as air percolation so that it is not trapped in the soil profile. The roots of *Indigofera zollingeriana* plants can reduce the occurrence of greater percolation, which is indicated by a lower air deficit at a soil depth of 20 cm.

The activity and population of soil microorganisms varied between treatments. The highest respiration was in the experimental plot planted with *Indigofera zollingeriana* + EFB (72.00 CO_2-C 100^{-1} g soil of day^{-1}), and the lowest was in control (61.71 CO_2-C 100^{-1} g soil of day^{-1}). Meanwhile, the highest total microorganism was also found in the plot planted with *Indigofera zollingeriana* + EFB, which was 105 (10^5 CFU g^{-1}), while the lowest total microorganisms were in the control plot (60 (10^5 CFU g^{-1})). The high level of respiration and the large number of total microorganisms on the plots treated with *Indigofera zollingeriana* and empty bunches of palms are thought to be due to the presence of litter sourced from *Indigofera zollingeriana* leaves and empty bunches of palms that contain a lot of organic matter, thereby automatically increas-

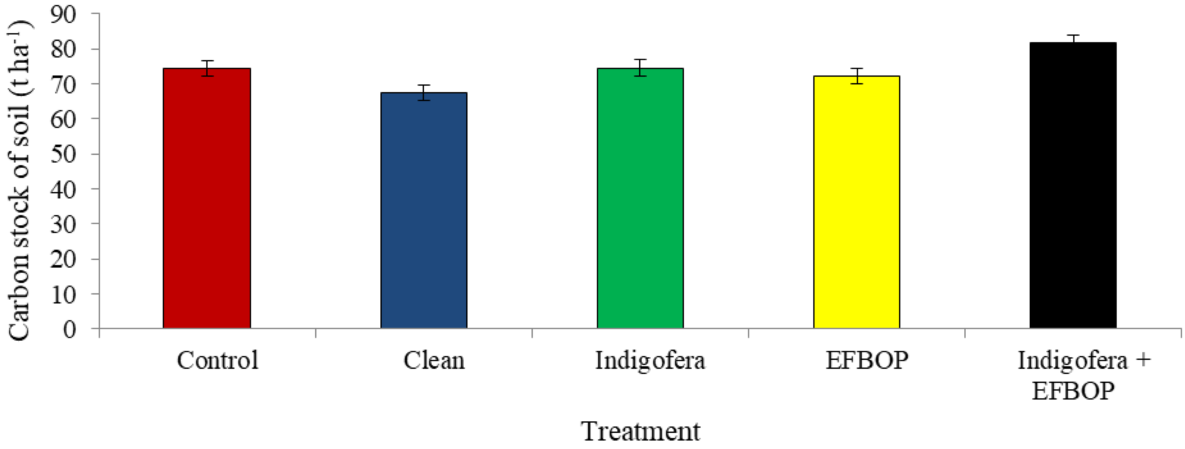

Figure 6. Effects of the treatments on carbon stocks of soil (t ha^{-1})

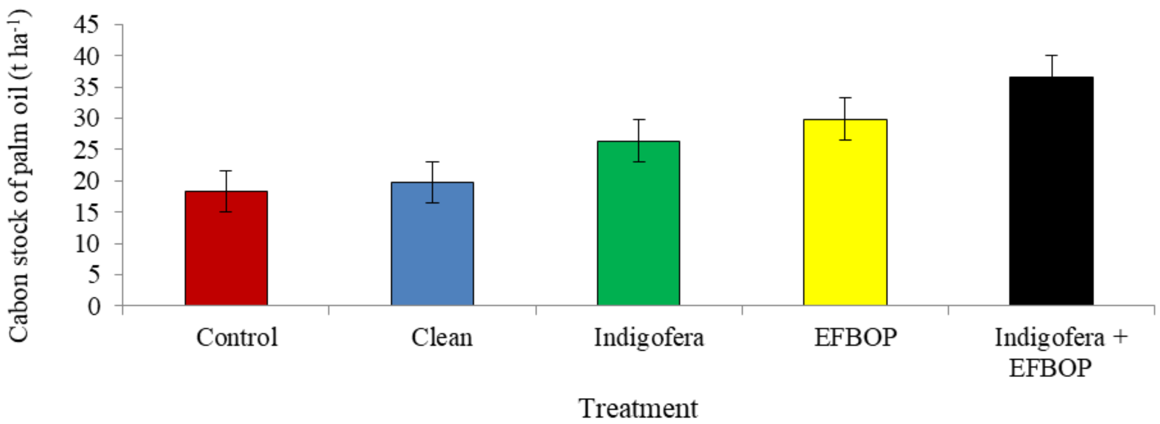

Figure 7. Effects of the treatments on the carbon stocks of palm oil (t ha^{-1})

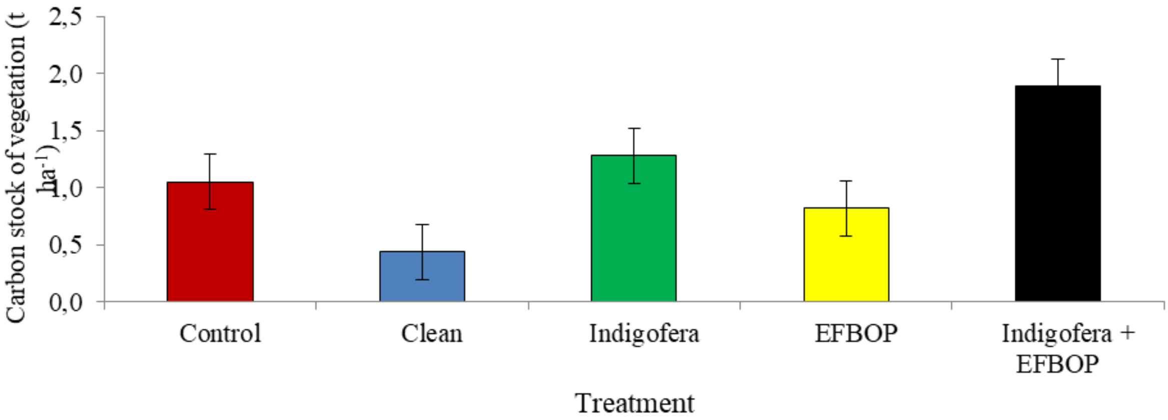

Figure 8. Effects of the treatments on the carbon stocks of vegetation (t ha^{-1})

Table 1. Effects of treatments on respiration and a total population of soil microorganisms

Treatment	Variables	
	Respiration (CO_2-C100^{-1} g soil day^{-1})	Total Microorganism (x 10^5 CFU g^{-1})
Control	61.71	60
Cleaned	65.14	85
Indigofera zollingeriana	66.86	80
EFB	60.00	75
Indigofera zollingeriana + EFB	72.00	105

ing the carbon stocks. The effects of the treatments on respiration and the total population of soil microorganisms can be seen in Table 1.

Soil biological type is directly related to a sustainable farming system because it has an important role in the decomposition process that breaks down complex organic molecules and converts them into available forms to plant (Friedel et al., 2001). Total respiration reflects the activity of soil microorganisms (Pietika et al., 2005). The higher the total soil respiration, the higher the activity of microorganisms in the soil. This study result showed that the treatment of *Indigofera zollingeriana* + EFBOP resulted in higher soil respiration, which was 72.00 CO_2-C100^{-1} g soil on day^{-1} compared to the soil respiration in control, which was only 61.71 CO_2-C100^{-1} g soil of day^{-1}. The increase of microorganisms activity in soil planted with *Indigofera zollingeriana* + EFB is due to its high organic content (Pietika et al., 2005).

The diversity and population of soil microorganisms increased with the treatment of *Indigofera zollingeriana* and EFB. This is in accordance with the research by Broughton & Gross (2000); Malý et al., (2000); Wang et al., (2013), reporting that ground cover plant affects biodiversity and the population of soil microorganism. Gessner et al., (2010) state that soil microorganism population is influenced by litter quality, the amount of nutrients, and plant tissue structure, such as protein and lignin. According to Cesarz et al., (2013), the plant influences the diversity and soil microorganisms population through the carbon supply provided by root exudate.

The treatment of *Indigofera zollingeriana* + EFB resulted in higher carbon stock than other treatments. The highest soil carbon stock was found in the experimental plot planted with *Indigofera zollingeriana* and treated with EFB, which was 81.6 t ha^{-1}, while the lowest soil carbon stock was in the experimental plot cleaned, which was 67.4 t ha^{-1}. Meanwhile, the highest carbon stock of oil palm was also shown in the experimental plot planted with *Indigofera zollingeriana* and treated with EFB, which was 36.60 t ha^{-1}, the lowest one was in the control plot, which was 18.34 t ha^{-1}. The EFB treatment resulted in the highest value of vegetation carbon stock, which was 1.89 t ha^{-1}, while the lowest one was in the cleaned experimental plot, which was 0.44 t ha^{-1} (Hairiah et al., 2001). Carbon stocks of soil, oil palm, and vegetation under oil palm due to the treatments given in the study can be seen in Table 2.

The difference between the treatments given on the carbon stock of soil can be seen in Figure 6. Carbon stock is the amount of carbon stored in an ecosystem at a certain time, both in the soil, plant biomass, and carbon stored in vegetation (Agus, 2011). Ohkura et al., (2003), stated that soil carbon content is affected by the soil's physical properties and the type of vegetation that grows on it. Plants save carbon by absorbing carbon from

Table 2. Effects of the treatments on the carbon stocks of soil, oil palms and vegetation under 5-year-old oil palm

Treatment	Carbon stock (CO_2) (t ha^{-1})		
	Soil	Oil palm	Vegetation
Control	74.4b	18.34b	1.05bc
Cleaned	67.4b	19.75b	0.44d
Indigofera zollingeriana	74.6b	26.36ab	1.28b
EFBOP	72.3b	29.87a	0.82c
Indigofera zollingeriana + EFBOP	81.6a	36.60a	1.89a

Remarks: Values followed by the same letters in the same column are not significantly different based on the DMRT test at α level of 5%.

the air through the process of photosynthesis into constituents of plant tissue. When leaves, twigs, or whole plants die, this material is then returned to the ground and undergoes decomposition (Robert, 2001). Azham (2015) reported that the number of components making up carbon stocks found in vegetation was 5,834 t ha^{-1}, and 22% of the ground cover was shrubs under pioneer vegetation. Thus, some crops must be grown on land to balance the amount of free carbon in the air.

Figures 7 and 8 show that the highest carbon stocks were obtained in the experimental plots planted with *Indigofera zollingeriana* and treated with empty bunches of oil palm.

CONCLUSIONS

The measurement of soil moisture content showed that the experimental plots treated with *Indigofera zollingeriana* + EFB retained water more than the plots with other treatments. The highest soil moisture content in the dry month was shown in July, which was 28.78% at a depth of 20 cm. The highest carbon stocks of oil palm and soil were obtained in the treatment of *Indigofera zollingeriana* + EFB, 81.6 t ha^{-1} and 36.60 t ha^{-1}, respectively. The activity and population of soil microorganisms in the experimental plots treated with *Indigofera zollingeriana* + EFB were higher than in other treatments. The highest respiration was 72.00 CO_2-C 100^{-1} g of soil day^{-1}, and the highest total microorganism was 105 x 105 g^{-1}. The implications of the research results on the current environmental conditions are environmentally friendly, increasing soil fertility and increasing FFB production.

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Utilization of Several Agricultural Wastes Into Briquette as Renewable Energy Source

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ABSTRACT

Tobacco stems contain 56.10% cellulose content, 15.11% lignin, 22.44% hemicellulose, and 44.61% total organic carbon, which can be used as a source of energy or fuel. This study aimed to utilize tobacco stems in a briquette form as alternative energy. The materials used in this study were tobacco stem waste, rice husk, wood charcoal, and coconut shell. The treatments used in this study consisted of T1 (100% of tobacco stems), T2 (80% of tobacco stem + 20% of coconut shell), T3 (80% of tobacco stem + 20% rice husk), and T4 (33.33% of tobacco stems + 33.33% of rice husk + 33.33% coconut shell). The fastest combustion rate was found at T3, 0.12 gram/sec, while T1 and T2 had the same combustion rate. T4, a mixture of various materials, had no significant difference compared to T1, T2, and T3. The highest calorific value of tobacco stem briquettes was in T4 (4127 Kcal/kg), and the lowest was in T1 (2343 Kcal/kg). The combustion rate of these tobacco stem briquettes was longer than that of charcoal briquettes, whose average burning rate is 0.234 grams/second. Overall, this study provides an overview of the best combination to create briquettes from agricultural waste.

Keywords: Briquettes, Tobacco stem, Utilization, Waste

ABSTRAK

Batang tembakau mengandung 56,10% selulosa, 15,11% lignin, 22,44% hemiselulosa, 44,61% total karbon organik yang dapat digunakan sebagai sumber energi atau bahan bakar. Penelitian ini bertujuan untuk memanfaatkan batang tembakau menjadi bentuk briket sebagai energi alternatif. Bahan yang digunakan dalam penelitian ini adalah limbah batang tembakau, sekam padi, arang kayu, dan tempurung kelapa. Perlakuan yang digunakan dalam penelitian ini adalah; T1: 100% batang tembakau, T2: 80% batang tembakau + 20% tempurung kelapa, T3: 80% batang tembakau + 20% sekam padi dan 33,33% batang tembakau + 33,33% sekam padi + 33,33% batok kelapa. Laju pembakaran tercepat terdapat pada T3 yaitu 0,12 gram/detik, sedangkan T1 dan T2 memiliki laju pembakaran yang sama. T4, yang merupakan campuran berbagai bahan, tidak berbeda nyata dengan T1, T2, dan T3. Nilai kalor briket batang tembakau hasil penelitian tertinggi pada T4 sebesar 4127 Kkal/Kg dan terendah pada T1 sebesar 2343 Kkal/Kg. Laju pembakaran briket batang tembakau ini lebih lama dibandingkan briket arang yang rata-rata laju pembakarannya 0,234 gram/detik. Secara keseluruhan, penelitian ini memberikan gambaran kombinasi terbaik untuk menciptakan briket dari limbah pertanian.

Kata kunci: *Briket, Batang Tembakau, Pemanfaatan, Limbah*

INTRODUCTION

The world's energy needs are still dominated by fossil energy, which is increasingly limited. Renewable energy sources are needed to reduce dependence on fossil energy. Biomass energy has a high potential due to its abundant availability worldwide. Biomass waste is found in the agricultural sector. Agricultural waste, which has a carbon content, has the potential to be used as an alternative energy source called briquettes. Agricultural waste, such as husk, straw, and coconut shells, has a carbon content of 1.33%, 2.71%, and 18.80%, respectively (Pancapalaga, 2008). One of the largest agricultural wastes humans often ignore is tobacco stem waste. With a population range of 22,000 trees per hectare and an estimated weight of 0.5 kg of tobacco stems, Indonesia will generate more than 2 million tons of tobacco stem waste (Handayani et al., 2018). Tobacco stems contain

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56.10% cellulose, 15.11% lignin, 22.44% hemicellulose, and 44.61% total organic carbon (Amirudin et al., 2020).

Tobacco is an agricultural plant commodity with high economic value as devised in the cigarette industry sector. East Java is one of Indonesia's largest tobacco producer provinces, with a land area of 108,524 ha in 2015 (Director General of Plantation, 2017). Tobacco leaves are used in the cigarette industry, and the wastes are in the form of leaf stems and tobacco leaf bones. The stems go to waste or are left in the fields to fertilize the soil. However, tobacco stems contain nicotine, which can be a hazardous waste due to soil penetration and cause pollution (Bareschino et al., 2021). The nicotine content of tobacco stems is 0.53 ppm (Obidziński et al., 2017). Sustainable agriculture is agriculture whose management is based on meeting needs without compromising the interests of future generations. Efforts that can be made are post-harvest processing and waste management (Indahsari & Negoro, 2020). The utilization of tobacco waste is still not managed properly, so there needs to be an effort that can be used to treat waste into a material that is beneficial and not harmful to the environment, one of which is processing it into briquettes.

Briquetting is the technology used to convert all agricultural and forestry wastes into solid fuels. Briquettes are formed in cylindrical logs using high mechanical pressure without chemicals or binders (Kanagaraj et al., 2018). Briquettes have a higher thermal value, lower ash content, a more uniform rate of combustion, and are less expensive than coal. Briquettes with low moisture and a high density improve boiler efficiency (Aishwariya & Amsamani, 2018).

Good quality briquettes have standards so they can be used to their needs. Briquette quality is generally determined by physical and chemical properties such as water content, ash content, volatile substances, carbon content, density, compressive resistance, and calorific value (Ren et al., 2019). Because the biomass component of the briquette burns at a lower temperature than the coal, the volatile matter in the coal, which would otherwise be released as smoke at a low combustion temperature, is completely burned (Promdee et al., 2017).

Mixing the raw materials of tobacco stem waste with other raw materials with higher specific gravity than tobacco stems is necessary to produce briquettes with world trade standard quality. This research is expected to determine the composition of briquettes to increase the use value of tobacco plant (*N. tabacum L.*) waste as one of the fuels from renewable energy sources and as an alternative energy substitute for fossil energy. Tobacco waste processed into a briquette can be used as alternative energy by utilizing carbon sources from lignocellulose of tobacco waste, primarily the stems and leaves that are not used. Mixtures of other materials, such as rice husks and coconut shells, are known to improve the quality of tobacco waste briquettes. Thus, in this study, the production of tobacco waste briquettes was given a mixture of these materials.

MATERIALS AND METHOD

The research was conducted from September 2019 – April 2020 by Poultry Research in collaboration with Universal PT Tempu Rejo. All tobacco stem waste materials were provided by Universal PT Tempu Rejo and then processed into briquettes.

Briquette samples were made by adding the same amount of adhesive to each treatment. The adhesive used was 10% tapioca flour with 3000 psi press pressure. The treatments used in this study were T1 (100% of tobacco stems), T2 (80% of tobacco stem + 20% of coconut shell), T3 (80% of tobacco stem + 20% rice husk) and T4 (33.33%

Figure 1. The production process of agricultural waste briquettes: a. Fresh tobacco stem waste; b. Drying tobacco stem; c. Charcoal process; d. Charcoal of several agricultural wastes; e. Charcoal sifting process; f. Briquette process; g. Press machine; h. Briquette

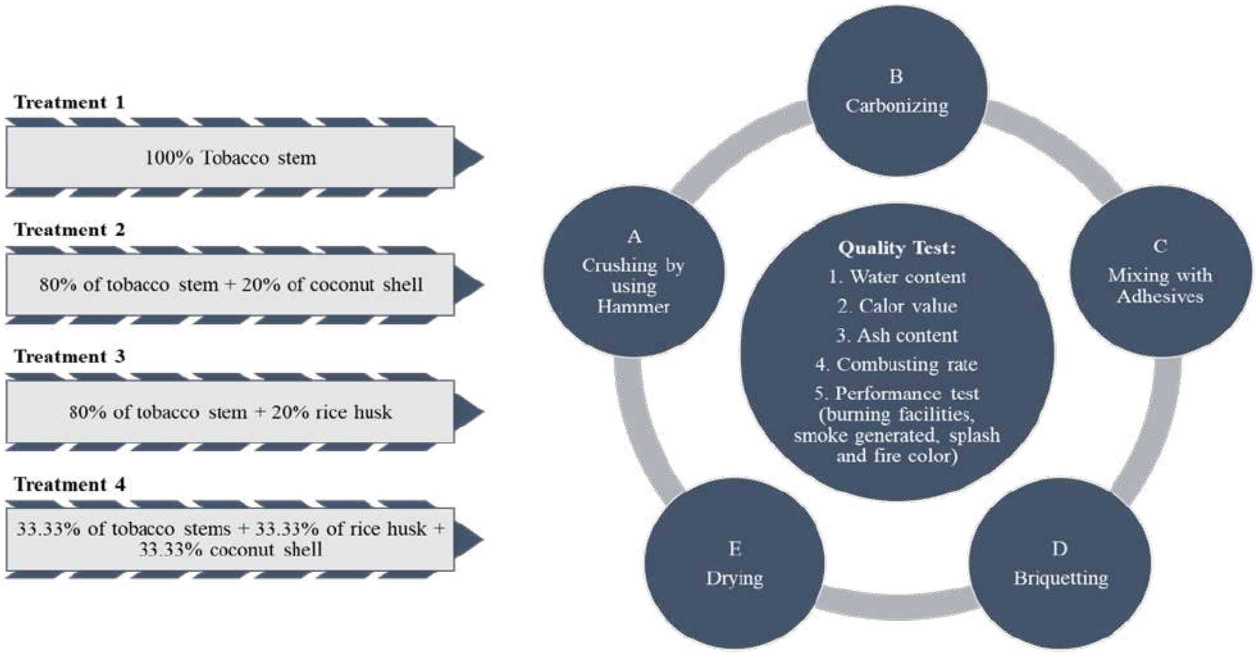

Figure 2. Road map research.

of tobacco stems + 33.33% of rice husk + 33.33% coconut shell) (Figure 1).

The raw materials prepared include dry tobacco stems, rice husks, and coconut shells (Figure 3). Using dry ingredients can accelerate the drying process compared to wet ingredients because they have low water content. The carbonizing process was done using a 250L can. The carbonizing was performed by burning crushed material in a closed 250L can with a slit for gas exchange, then it burnt on combusting stove. The sifted charcoal of tobacco stem, rice husks, and coconut shells was mixed with tapioca flour adhesive by 10% of raw material per unit of briquette according to each treatment.

Figure 3. Raw material of briquette waste: a. Tobacco stem and leaf charcoal; b. Rice husk charcoal; c. Coconut shell charcoal

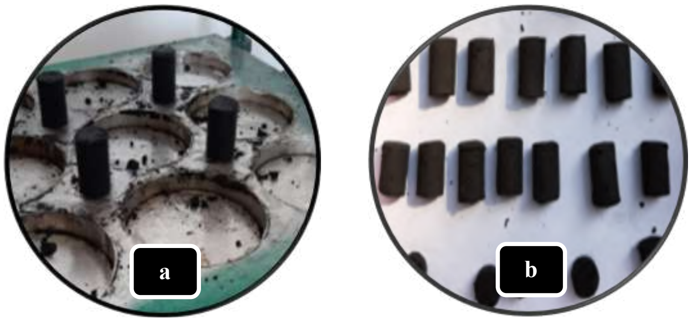

Figure 4. a. Pressing briquette; b. Briquette size, 2.25 cm x 3.25 cm

Each treatment mixed with the adhesive material (tapioca flour) was stirred evenly until the entire briquette dough turned black. After homogenization, the mixed materials were pressed using a pressing machine. The diameter size of briquettes was 2.5 cm with a length of 3.5 cm. The drying temperature used was 60 °C for 24 hours (Figure 2). The purpose of drying is to reduce the water content in briquettes by the provisions of the applicable briquette water content according to SNI 01-6235-2000 of 8%.

The sample's water content was determined using the oven method by weighing the material with an analytic scale of 5 grams in an aluminum dish. The material was then dried in the oven at a temperature of 105 °C until constant weight was achieved (4 hours), cooled in a desiccator, and weighed again (Ekpete & Horsfall, 2011). Density is influenced by the amount of pressure applied, affecting the efficiency of burning briquettes as fuel. According to Falemara et al., (2018), briquette mass density is calculated by weighing the briquette sample and then dividing the weight by the volume of the briquette sample. The density of briquettes can be expressed by the formula density (g/cm^3).

The sample was weighed (5 grams), put into a porcelain dish, and then heated until no smoke was generated. It was then blown in the furnace at 600 °C to become ash, cooled in the desiccator, then immediately weighed after reaching room temperature. The burning rate of the samples was determined at a certain mass of charcoal briquette combusted in the air. The stopwatch was set, and the total time required for the samples to burn completely to ashes was recorded (Kongprasert et al., 2019). The calorific value of biomass fuel is the amount of heat energy that can be released in each unit of mass of the fuel when it burns completely (in units of Kcal/Kg). The principle of determining calorific value is measuring the energy generated in the combustion of one gram of charcoal by measuring changes in fluid temperature at a fixed volume performed in a closed vessel (Simiyu et al., 2017).

RESULTS AND DISCUSSION

The quality of the briquettes from tobacco waste is presented in Table 2. Figure 4a shows the tobacco stem briquette processing using a pressing machine. Cylindrical briquettes are produced because it has higher density and produces higher energy. Figure 4b shows the briquette drying process after pressing with a machine.

Based on the result, the water content of tobacco stem waste with the addition of various materials showed significantly different results. The T4 showed the lowest water content, followed by T2, T1, and T3. The water content of T2 and T4 briquettes was more moderate than 8%, as required by the minimum wood charcoal briquettes. The standard minimum percentage of water content, according to SNI 01-6235-2000, explained that the water content of briquette is 8% (Radam et al., 2018). The highest water content (8.62%) was obtained in the T3, a mixture of 80% tobacco stem waste and 20% rice husk. Similar to the research conducted by Saeed et al., (2021), the water content in rice husks ranged from 6% - 10%.

According to Nurek et al., (2019), the material with low moisture shows a weaker interaction between particles. The increase in humidity (to a certain value) strengthens this effect, but the mechanical properties deteriorate, adversely affecting the agglomeration process. The disturbance of the compaction process causes this by the increased amount of generated steam. Previous research showed that the water content of tobacco waste briquettes in Indonesia was lower than the tobacco stem briquettes made in Henan, China, which was 10.84% (Xinfeng et al., 2015).

Ash content of all treatments showed a significant difference. Ash content of T1, T2, T3, and T4 45.93%, 31.69%, 36.83%, and 37.73%, respectively. Ash content of T2 is the lowest than others, but this result is different from the study conducted by Bot et al., (2021), who reported that the ash content in coconut shells was 10.02%, lower than the results of this study. Ash generated from this study do not meet the standards set by SNI 01-6235-2000 (<8%), Japanese (3-6%), and ISO 17225 (3.3-11.7%) for bio-briquettes standards (Ifa et al., 2020). High ash content is caused by high silica content in the material, which is nondegradable. This silica causes low heating and carbon values (Putri & Andasuryani, 2017).

The mass density of briquettes made from tobacco waste is presented in Table 1. All treatments showed very low densities, with the highest value of 0.73 gram/cm^3 (T4), and the lowest value of 0.63 (T3). These results were supported by Lingulеasa et al., (2017), reporting that the tobacco stem briquette density was 0.89 gram/cm^3. This study's results differed from Tanko et al., (2020), mentioning that the density of rice husks and coconut shells mixture ranged from 1.5 - 3 grams/cm^3, two

Table 1. Physical properties of agricultural waste briquettes

Treatments	Water Content (%)	Ash Content (%)	Mass Density (gram/cm^3)	Combusting Rate (gram/minute)	Calor Value (Kcal/kg)
T1	8.34^c	45.93^d	0.71^b	0.16^b	2343^a
T2	7.76^b	31.69^a	0.72^b	0.16^b	3782^c
T3	8.62^d	36.83^b	0.61^a	0.12^a	2997^b
T4	6.34^a	37.73^c	0.73^b	0.13^{ab}	4127^d

Note: 100% tobacco stem (T1), 80% of tobacco stem + 20% of coconut shell (T2), 80% of tobacco stem + 20% rice husk (T3), 33.33% of tobacco stems + 33.33% of rice husk + 33.33% coconut shell (T4). Means followed by different letters in the same column are significantly different based on Duncan's 5% test.

Table 2. Performance test of the briquettes

Treatments	Smoke Generated	Splash and Fire Color
T1	Negative	No fire
T2	Negative	No fire
T3	Negative	No fire
T4	Negative	No fire

Note: 100% tobacco stem (T1), 80% of tobacco stem + 20% of coconut shell (T2), 80% of tobacco stem + 20% rice husk (T3), 33.33% of tobacco stems + 33.33% of rice husk + 33.33% coconut shell (T4).

Figure 5. Briquette fire burn process

to four times denser than the results of this study. This density is influenced by the structure and size of the material. Smaller particle sizes can expand the surface area to the bond between particles, so it is related to briquette hardness. The higher the density, the higher the briquette hardness.

The combusting rate of the briquette shows how fast the briquette is burning. The fastest combustion rate in T3 was 0.12 gram/sec, while T1 and T2 had the same combustion rate. T4, a mixture of various materials, had no significant difference compared to T1, T2, and T3. The combustion rate of these tobacco stem briquettes was longer than charcoal briquettes whose average burning rate is 0.234 grams/second (Putri & Andasuryani, 2017). According to Aljarwi et al., (2020), the greater the pressure (solid), the higher the calorific value and the rate of combustion of the briquettes.

The highest calorific value of tobacco stem briquettes was obtained in T4, which was 4127 Kcal/kg, while the lowest was in T1 (2343 Kcal/kg). The results of the calorific value of T4 were similar to Suryaningsih & Nurhilal (2018). The calorific value in a mixture of rice husks and coconut shells ranged between 4107 - 4886 Cal/Gram. The calorific value of all treatments was lower than the SNI 01-6235-2000 standard for wood charcoal briquettes, which is a minimum of 5000 Kcal/kg (Radam et al., 2018). Purwono et al., (2010) explained that the heating value of charcoal briquettes from tobacco stems pyrolyzed for ninety minutes with a 4-ton pressure was 5438.9 Kcal/kg. A shorter pyrolysis time and greater pressure can reduce the caloric value of briquettes (Purwono et al., 2010).

In using briquettes, odor and visible smoke are not wanted. Briquettes should go through a burning test since these effects can be created using specific binders (Borowski et al., 2017). The briquette performance is presented in Table 2. The smoke generated by this briquette was negative, meaning no smoke was generated (Figure 5). Moreover, this briquette didn't have a splash when it was burned, and the flame did not appear.

CONCLUSIONS

The highest calorific value of tobacco stem briquettes was 4127 Kcal/kg, resulting in T4 (33.33% of tobacco stems + 33.33% of rice husk + 33.33% coconut shell), and the lowest water content also found in T4, which was 6.34. T3 resulted in the highest water content value compared to other treatments, which was 8.62%. All treatments do not generate smoke and sparks, so they can be used as briquettes for renewable energy. The implication of this research is to provide an overview of the best combination to create briquettes from agricultural waste.

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Effects of Foliar Application of Oil Palm Empty Fruit Bunch Ash Nanoparticles on Stomatal Anatomy of Potato Leaf Plants (*Solanum tuberosum* L.)

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ABSTRACT

The productivity of potatoes (*Solanum tuberosum* L.) in Indonesia is still low. Fertilization needs to be done to increase potato productivity. This study aimed to examine the effects of applying oil palm empty fruit bunch (OPEFB) ash nanoparticles on the anatomy of potato stomatal and leaf cells (*Solanum tuberosum* L.). The research was carried out from March to June 2021 in the Sumberejo Village, Ngablak District, Magelang Regency, Central Java, and at the Agrobiotechnology Laboratory, Faculty of Agriculture, University of Muhammadiyah Yogyakarta. The study used a single-factor treatment design with a Randomized Completely Block Design (RCBD). The treatments tested included the foliar application of OPEFB ash nanoparticles at several concentration, consisting of 0% (control), 0.1%, 0.2%, 0.3%, and 0.4%. The results showed that foliar application of nanoparticles OPEFB ash affected stomatal anatomy, namely guard cell width, stomatal aperture, and density. The application of OPEFB ash nanoparticles with a concentration of 0.3% was most effective in increasing the opening of stomata because it affects the activity of the photosynthetic process.

Keywords: Nano fertilizer, Oil palm empty fruit bunch ash, Potassium, Stomata

ABSTRAK

Tingkat produktivitas tanaman kentang (Solanum tuberosum L.) di Indonesia masih rendah. Pemupukan perlu dilakukan untuk meningkatkan produktivitas kentang. Penelitian ini bertujuan untuk menguji pengaruh penyemprotan nano partikel abu tandan kosong kelapa sawit (TKKS) terhadap anatomi stomatal dan sel daun tanaman kentang (Solanum Tuberosum L.). Penelitian dilaksanakan pada bulan Maret hingga Juni 2021 di lahan desa Sumberejo, kecamatan Ngablak, kabupaten Magelang, Jawa Tengah dan di Laboratorium Agrobioteknologi Fakultas Pertanian, Universitas Muhammadiyah Yogyakarta. Penelitian menggunakan Rancangan Acak Kelompok Lengkap (RAKL) faktor tungga terdiri dari 5 perlakuanl. Perlakuan yang diuji meliputi penyemprotan foliar partikel nano abu TKKS dengan konsentrasi 0% (kontrol); nano TKKS konsentrasi 0,1%; konsentrasi 0,2%; konsentrasi 0,3%; konsentrasi 0,4%. Hasil penelitian menunjukkan bahwa aplikasi foliar partikel abu TKKS berpengaruh terhadap anatomi stomata yaitu lebar sel penjaga, bukaan stomata dan kerapatan stomata. Aplikasi partikel nano abu TKKS dengan konsentrasi 0,3% paling efektif dalam pembukaan stomata yang mempengaruhi proses fotosintesis.

Kata kunci: *Pupuk nano, Abu tandan kosong kelapa sawit, Kalium, Stomata*

INTRODUCTION

Potato (*Solanum tuberosum* L.) is one of the important food ingredients for humans and the main vegetable crop, after rice, wheat, and corn. In Indonesia, potato plants have become one of the priority foods to be developed as a source of carbohydrates to support food diversification. The demand for potatoes is increasing yearly, along with changes in lifestyle and the development of the potato processing industry (Isra, 2020). However, potato production in Indonesia has not been able to meet the demand for potatoes due to the increasing demand for potatoes. In contrast, potato production in Indonesia fluctuates from year to year. National potato production in 2019 (1.31 million tons/ha) has increased compared to 2018 (1.28 million tons/ha). However, potato production in 2020 decreased by 1.28 million tons/ha (Badan Pusat Statistik, 2020). The decrease in production will impact potato productivity, which is also low. One factor that leads to low potato production is

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the lack of nutrients that potato plants need.

Potato production is greatly affected by nitrogen, phosphorus, and potassium nutrients. The major functions of K in plants are controlling enzyme activity, cation-anion homeostasis, and membrane polarization. These are based on its osmotic nature, which is why it is needed for cell extension, turgor regulation, and stomatal movement (Warnita, 2019). One important role of K in the stomatal functions is stimulating enzyme to starch synthase for starch synthesis. Potassium also plays a role in the the stomatal aperture to meet the needs of CO_2 and water vapor for photosynthesis and in the stomatal closing to prevent excessive water loss from plant tissues. Suppose the function of the stomatal guard cells is not optimal. In that case, drought stress can occur in plants because K-deficient plants can significantly reduce the net CO_2 assimilation rate (Naumann et al., 2020).

The type of stomata of potato leaves belongs to the Amphistomatic type, which is located on both leaf surfaces, but stomata are mostly found on the underside of the leaves (abaxial). The anatomical structure of stomata is closely related to the ontogeny of the epidermis or the type of epidermis because stomata are from the modification of some of the epidermal cells. There are several factors that affect the opening and closing of stomata, including sunlight, potassium, availability of CO_2, water and temperature (Driesen et al., 2020). If these components are met, it will affect plant physiological processes such as transpiration, photosynthesis, and respiration that occur in leaf stomata.

The need for potassium (K) in potato plants can be fulfilled by the application of inorganic (synthetic) fertilizers or organic fertilizers (fertilizers derived from organic waste). One of the organic wastes that can be used as a potassium (K) fertilizer source is oil palm empty fruit bunch ash. According to the results of research by Efendi et al., (2020), OPEFB ash contains nutrients such as total N of 0.05%, P_2O of 54.79%, K_2O of 36.48%, MgO of 2.63%, CaO of 5.46%, Mn of 1,230 ppm, Fe of 3450 ppm, Cu of 183 ppm, and Zn of 28 ppm, with pH ranging from 11.9 to 12.0. Based on the research by Azizah (2019), the application of OPEFB ash nanoparticles with a concentration of 0.4% increased the shallots' productivity.

The fertilization for potato plants can be done through the soil or the leaves (foliar application). Leaf fertilization will be effective if the particle size of the fertilizer material is smaller than the leaf stomatal pore size. The effective absorption of nutrients in OPEFB ash through the leaves requires technological innovation by reducing particle size through nano-fication.

Nanotechnology is a technique for creating materials, functional structures, and devices at the nanometer scale. Fertilizers with -size has properties and abilities far superior to the starting material, such as being easily absorbed by plants with slow-release fertilizers (Ratih et al., 2021). For this reason, nanoparticles of OPEFB ash are needed as a source of potassium fertilizer that can be used to meet the K needs in the stomata of potato plants. In addition, research on the application of OPEFB ash nanoparticles on potato plants has never been carried out. Thus, this study aimed to determine the effects of foliar application of OPEFB ash nanoparticles on the stomatal anatomy of potato plants.

MATERIALS AND METHODS

Study area

Field research was carried out in Sumberejo Village, Ngablak Sub-district, Magelang District, Central Java, with coordinates of -7.4018090 LS 110.3908880 east longitude starting from March to June 2021. Observations were made in the field and in the Agrobiotechnology Laboratory, Faculty of Agriculture, University of Muhammadiyah Yogyakarta.

Experimental design

Experimental research was conducted with a single factor treatment arranged in a Randomized Completely Block Design (RCBD), consisting of five concentrations of OPEFB nanoparticles (0% (control), 0.1%, 0.2%, 0.3%, and 0.4%. Each treatment consisted of five replications, in which there were 25 plants in each unit. Thus, there were 125 potato plants. Each experimental unit contained a physiological plot consisting of three physiological plants. OPEFB ash nanoparticles were applied 20 days after planting, and the next application was carried out once every 10 days.

The data collected include stomatal length, stomatal guard cell width, stomatal aperture, stomatal density, cell wall thickness, and leaf cell area. Observations were made at each phase of potato plant growth at 40, 65, and 75 days after planting. Sampling was carried out directly in the sun without picking the leaves to keep the stomatal cells open using the replica method. As for the cross-section of the leaf, thin slices were made with a transverse direction in the thickness of the leaf, and the incisions were observed through an Olympus CX-22LEDRFS1 computer microscope with a magnification of 400x.

Statistical Analysis

The data obtained from this study were analyzed using Statistic Analysis System (SAS) 9.0 applications. Analytical method with Analysis of Variance (ANOVA) significance level of 5%. Means comparison between treatments was tested using Duncan Multiple Range Test (DMRT) at 5%.

RESULTS AND DISCUSSIONS

The results showed that foliar application concentration of OPEFB nanoparticles did not significantly affect the stomatal length during the vegetative phase and tuber ripening phase, but there was a significant difference observed in the tuber initiation phase (Table 1). Further test results showed that foliar application of OPEFB ash nanoparticles at a concentration of 0% produced the longest stomata of 50.78 µm. It was significantly different compared to that in the 0.4% concentration treatment.

Based on these results (table 1), the stomatal length is included in the very long category >25 µm (Makin et al., 2022). Foliar application of OPEFB nanoparticles at a concentration of 0% resulted in the longest stomatal length compared to that at a concentration of 0.4%. OPEFB ash nanoparticles contain potassium, which can maintain cell turgidity and cause guard cells to expand, resulting in the elongation of cellulose microfibrils outward. In the process of stomatal opening and closing, stomatal elongation occurs only in the cellulose microfibrils or cellulose fine fibers contained in the guard cell walls in a radial shape; this arrangement pattern is referred to as radial mycelation. The shape of the pattern allows only the long stretching of the cellulose microfibrils, but the two ends of the guard cells stick together so that when the cellulose microfibrils elongate, the thick abdominal wall limits the stretching. As a result, guard cells will bend and open, which affects the stomatal width instead of the stomatal length (Pautov et al., 2018). The stomatal length is in a fixed state when the stomatal aperture is based on the hardening of the stomatal poles, and polar clamping occurs (Carter et al., 2017). Thus, the stomatal length is related to the stomatal width, where the process becomes a single entity that affects the size of the stomatal porous. The longer cellulose microfibril

Table 1. Stomatal length (μm) as affected by various concentrations of OPEFB ash nanoparticles under different phases

Foliar application concentration	Stomatal Length (μm)		
	Vegetative Phase (40 day)	Bulb Initiation Phase (65 day)	Tubers Ripening Phase (75 day)
0 %	44.66 ± 5.55 a	50.78 ± 8.80 a	47.03 ± 5.66 a
0.1 %	40.40 ± 4.89 a	40.83 ± 3.97 b	42.85 ± 4.09 a
0.2 %	40.34 ± 6.34 a	39.69 ± 3.56 b	42.44 ± 8.70 a
0.3 %	38.76 ± 4.23 a	47.00 ± 7.15 ab	51.63 ± 7.73 a
0.4 %	41.40 ± 6.12 a	40.83 ± 4.20 b	50.08 ± 5.35 a
CV	12.80	11.95	13.93

Remarks: Means followed by the same letters are not significantly different based on the DMRT test at the level of 5%

Table 2. Stomatal aperture (μm) as affected by various concentrations of OPEFB ash nanoparticles under different phases

Foliar application concentration	Aperture Stomatal (μm)		
	Vegetative Phase (40 day)	Bulb Initiation Phase (65 day)	Tubers Ripening Phase (75 day)
0 %	2.86 ± 0.61 a	2.76 ± 0.33 a	2.54 ± 0.41 ab
0.1 %	2.76 ± 0.43 a	2.77 ± 0.59 a	2.23 ± 0.46 b
0.2 %	2.50 ± 0.42 a	2.24 ± 0.59 a	2.37 ± 0.45 b
0.3 %	3.12 ± 0.10 a	2.76 ± 0.48 a	3.03 ± 0.75 a
0.4 %	2.71 ± 0.50 a	2.45 ± 0.65 a	2.82 ± 0.32 ab
CV	15.70	21.58	16.60

Remarks: Means followed by the same letters are not significantly different based on the DMRT test at the level of 5%

will experience withdrawal due to the widening of the guard cells outwards.

There was no significant effect of OPEFB ash nanoparticle concentration on the stomatal length because the higher the concentration of OPEFB nanoparticle ash, the smaller the stomatal length. Lu et al., (2017) reported that K deficiency decreased stomatal length.

Based on Table 2, the concentration of OPEFB ash nanoparticles did not significantly affect the stomatal aperture in the vegetative and tuber initiation phase, but there was significant effect in the tuber ripening phase. Further test results showed that foliar application of OPEFB ash nanoparticles at a concentration of 0.3% had the largest stomatal aperture of 3.03 μm, but it was not significantly different from the stomatal aperture at a concentration of 0%.

Based on the results of the analysis, it was shown that the foliar treatment of OPEFB ash nanoparticles had an effect on the stomatal aperture. This was because the stomatal aperture occurred as a result of activity in guard cells that require potassium to maintain turgor pressure so that the stomatal are open. According to Barita et al., (2018), potassium plays a role in stimulating water absorption, thereby affecting the increase in cell turgor pressure; if the high cell turgor pressure is maintained, the stomata can be maximally open wider and longer. Potassium has a role in the process of opening and closing of stomata, which is influenced by several factors, namely the mechanism of turgor, the presence of osmotic pressure, accumulation of potassium ions, accumulation of abscisic acid, and environmental factors, such as sunlight, temperature, humidity and CO_2 concentration (Ratnasari et al., 2020). The opening of stomata results from activity in the stomatal guard cells, where there are cell organelles

Table 3. Guard cell width (μm) as affected by concentrations of OPEFB ash nanoparticles under different phases

Foliar application concentration	Guard Cell Width (μm)		
	Vegetative Phase (40 day)	Bulb Initiation Phase (65 day)	Tubers Ripening Phase (75 day)
0 %	11.30 ± 1.54 a	10.75 ± 1.86 a	8.03 ± 1.74 b
0.1 %	11.46 ± 1.40 a	9.51 ± 2.04 a	8.79 ± 1.80 b
0.2 %	10.75 ± 2.33 a	9.65 ± 1.27 a	8.97 ± 2.12 b
0.3 %	10.81 ± 1.80 a	10.66 ± 0.57 a	8.26 ± 1.24 b
0.4 %	12.66 ± 1.88 a	9.47 ± 1.74 a	11.26 ± 1.51 a
CV	15.81	15.57	14.65

Remarks: Means followed by the same letters are not significantly different based on the DMRT test at the level of 5%

in these guard cells.

Cell organelles that play an active role in stomatal opening are vacuoles, which play a role in cell turgidity and shape. Stomata will open if the two guard cells experience increased cell turgor pressure. Turgor pressure is a condition where the cell expands because water from the surrounding cells enters it. Water movement is influenced by water potential, in which high water potential will go to cells with lower water potential. For stimulating water to enter the guard cell, the solute in the cell must be increased. According to Abidin (2022), the main solutes that mediate cell osmoregulation are K^+ and sucrose because of their high mobility of K^+ and solubility. The guard cells accumulate large amounts of K+ in the vacuole. Accumulating K+ in the vacuole against the electrochemical gradient (Lu et al., 2017) produces sufficient turgor for stomatal opening.

Foliar application of OPEFB ash nanoparticles at a concentration of 0.3% significantly affected the stomatal pore opening of potato plants. Likewise, according to Lu et al., (2017, an increase in potassium concentration to 0.12% showed a significant effect on the stomatal opening of *Brassica napus* leaves.

Foliar application of OPEFB ash nanoparticles did not significantly affect the width of guard cells in the vegetative and tuber initiation phase, but a significant effect was observed in the tuber ripening phase (Table 3). Further test results showed that foliar application of OPEFB ash nanoparticles at a concentration of 0.4% had the highest guard cell width of 11.2660 μm compared to other OPEFB ash nanoparticle concentrations.

The results showed that the foliar application of OPEFB ash nanoparticles affected the width of the guard cells. This is because the content of OPEFB ash nanoparticles in the form of potassium can maintain the turgidity of the vacuole cells in the guard cells, where the guard cells can change shape and size, which is reversible. The mechanism of guard cell dilation occurs due to changes or regulation of turgor, which is influenced by the theory of K ion movement or pump. The leaves absorb potassium by diffusion through ion exchange. According to Jasmi (2018), the main function of K is to activate enzymes and maintain cell water. K^+ ions support the activity of phosphorylase enzymes, which play a role in converting starch into glucose. Glucose plays a role in the osmotic potential of cells, which will move water to guard cells. As a result, the turgor pressure of the guard cells increases, and the stomata open by dilating the guard cells. Thus, when K^+ ions increase in guard cells, the activity of converting starch to glucose also increases (Advinda, 2018). Guard cells will also increase the osmotic potential of their cells, thereby increasing

Table 4. Stomatal density (mm^{-2}) as affected by various concentrations of OPEFB ash nanoparticles under different phases

Foliar application concentration	Stomatal density (mm^{-2})		
	Vegetative Phase (40 day)	Bulb Initiation Phase (65 day)	Tubers Ripening Phase (75 day)
0 %	486.8 ± 97.31a	308.89 ± 44.02 b	411.11 ± 82.03 a
0.1 %	442.2 ± 89.72a	331.11 ± 69.12 ab	468.89 ± 70.89 a
0.2 %	431.2 ± 187.81a	428.89 ± 63.64 a	382.22 ± 94.80 a
0.3 %	419.8 ± 136.40a	362.22 ± 64.12 ab	364.44 ± 101.95 a
0.4 %	406.8 ± 150.43a	426.66 ± 90.13 a	326.66 ± 51.88 a
CV	5.61	18.61	21.95

Remarks: Means followed by the same letters are not significantly different based on the DMRT test at the level of 5%

Table 5. Cell wall thickness (μm) as affected by various concentrations of OPEFB ash nanoparticles under different phases

Foliar application concentration	Leaf Cell Wall Thickness (μm)		
	Vegetative Phase (40 day)	Bulb Initiation Phase (65 day)	Tubers Ripening Phase (75 day)
0 %	5.23 ± 0.38 a	5.54 ± 0.06 a	4.98 ± 0.51 a
0.1 %	5.17 ± 0.21 a	5.33 ± 0.32 a	5.04 ± 0.32 a
0.2 %	5.19 ± 0.31 a	5.47 ± 0.10 a	5.32 ± 0.15 a
0.3 %	5.11 ± 0.12 a	5.37 ± 0.28 a	5.41 ± 0.16 a
0.4 %	5.53 ± 0.62 a	5.63 ± 0.32 a	5.50 ± 0.13 a
CV	7.54	4.82	5.77

Remarks: Means followed by the same letters are not significantly different based on the DMRT test at the level of 5%

turgor pressure and forming guard cell dilation.

This change in the shape of the guard cells occurs because the back cell wall is thin and elastic, protruding away from the opening, while the front cell wall will be straight or concave, the entire cell will appear bent, and openings with an increased size are formed (Roux & Leonhardt, 2018). When the guard cells widen, the metabolic activity in the cell will also be easy with the accumulation of ions or materials needed in the process. Thus, this mechanism proves that it can affect cell activities such as photosynthesis, respiration, transpiration, and another cell metabolism.

Foliar application of OPEFB ash nanoparticles at a concentration of 0.4% significantly affected the width of the stomatal guard cells in potato leaves. Research by Lu et al., (2017) showed that the application of potassium fertilizer significantly affected the width of the stomatal guard cells on *Brassica napus* leaves.

In table 4, the foliar application of OPEFB ash nanoparticles has not a significant effect on the stomatal density in vegetative and ripening phases of tubers, but a significant effect was found in tuber initiation phase. Further test results showed that the foliar application of OPEFB ash nanoparticles at a concentration of 0.2% had the highest stomatal density of 428.89 mm^{-2}, but was not significantly different compared to that at the concentrations of 0.4%, 0.3% and 0.1 %. Meanwhile, the 0.4% concentration differed significantly from the 0% concentration treatment.

Based on the results, concentration of 0% and 0.4% had significantly different results, and the

Table 6. Leaf cell area (μm²) as affected by various concentrations of OPEFB ash nanoparticles under different phases

Foliar application concentration	Leaf Cell Area (μm²)		
	Vegetative Phase (40 day)	Bulb Initiation Phase (65 day)	Tubers Ripening Phase (75 day)
0 %	3.05 ± 0.15 a	3.25 ± 0.08 a	3.22 ± 0.41 a
0.1 %	3.32 ± 0.16 a	3.30 ± 0.18 a	3.18 ± 0.21 a
0.2 %	3.32 ± 0.09 a	3.68 ± 0.82 a	3.27 ± 0.26 a
0.3 %	3.36 ± 0.26 a	3.44 ± 0.14 a	3.05 ± 0.32 a
0.4 %	3.15 ± 0.09 a	3.11 ± 0.23 a	3.29 ± 0.23 a
CV	5.33	12.27	7.91

Remarks: Means followed by the same letters are not significantly different based on the DMRT test at the level of 5%

average stomatal density was classified as moderate, which was in the range of 300 – 500/mm² (Claudia et al., 2020). This is because in general, stomatal density is related to stomatal size. High potassium concentrations can widen stomata and cause stomatal density to be quite high. According to Sihotang (2017), if the size of the stomatal is larger, the distance between the stomatal gets further by 20 times its diameter so that the evaporation process can take place optimally. This is evidenced by the width of the stomatal guard cells resulting from the foliar application of OPEFB ash nanoparticles at a concentration of 0.4%, showing the highest value compared to other concentrations (Table 4). Stomatal density affects two important processes in plants, namely transpiration and photosynthesis. Plants with high stomatal density have a higher transpiration rate than plants with low density. Because more stomata per unit area mean more CO_2 can be taken in and more water can be released. (Mercyana et al., 2021). Foliar application of OPEFB ash nanoparticles at various concentrations of potassium significantly affected the stomatal density of potato plants. Pratama et al., (2020) reported that the application of potassium fertilizer at a concentration of 0.3% significantly affected the stomatal density of oil palm plants experiencing drought stress.

Foliar application of OPEFB ash nanoparticles had no significant effect on cell wall thickness (Table 5). Based on the analysis results, the responses of leaf cells to the foliar application of OPEFB ash nanoparticles were not significantly different. This result is because potassium only affects the activity of the phosphorylase enzyme in stomatal guard cells that are not related to leaf cells. However, Widiyawati (2019) mentioned that thickening of the epidermal tissue was a structural defense response of plants against pathogen attacks. Epidermal cells are the outermost cell network as a place of penetration of pathogens. Structural defenses when attacked by pathogens include thickened epidermal cell structures that affect the stomatal surface and thickened cell walls to inhibit pathogen penetration so that pathogens do not damage deeper cell layers. However, in this case, the foliar application of OPEFB ash nanoparticles did not affect the cell walls' thickness, so pathogens would easily attack potato plants due to lack of protection. The results also showed that the foliar application of OPEFB ash nanoparticles on potato plants showed no significant effect on the thickness of the leaf epidermal cells. Likewise, research of Lu et al., (2016) reported that cell wall thickness was not affected by K nutrition.

Based on Table 6, foliar application of OPEFB ash nanoparticles had no significant effect on potato leaf cell area. This is because the leaf cells had enough water when taking sample in the morning. According to Saragih dan Ardian (2017), the content of OPEFB in the form of potassium can affect the optimal leaf cell area if the condition of the plant lacks water. Cell enlargement will also be hampered due to a decrease in the rate of photosynthesis because in these conditions, there is a decrease in the availability of nutrients, inhibition of protein synthesis so that the leaf area also decreases. Potassium will play a role in regulating the availability of sufficient water for cell enlargement. Enlargement of leaf cells becomes inhibited if the water content is low due to the need for turgor pressure for cell enlargement. The results of photosynthesis support the work of plant tissue cells in differentiation so that it will accelerate the growth and development of the plant, forming parts such as leaves. But in this case, the high potassium treatment did not affect the leaf area of potato plants. Foliar application of OPEFB ash nanoparticles on potato plants did not significantly affect the leaf cell area. However, Lu et al., (2016) reported that the leaf area was significantly down-regulated under K deficiency conditions.

CONCLUSIONS

Based on this research, it can be concluded that foliar application of oil palm empty fruit bunch ash nanoparticles on potato plants can affect stomatal anatomy, including the width of stomatal guard cells, stomatal opening (aperture), and stomatal density. Meanwhile, the application did not affect the anatomy of potato plant leaf cells. Also, foliar application of oil palm empty fruit bunch ash nanoparticles at a concentration of 0.3% is the most effective in increasing the stomatal aperture because it affects the activity of the photosynthetic process.

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Effects of Mycorrhiza Doses and Manure Types on Growth and Yield of Cassava in Gunungkidul

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ABSTRACT

Gunungkidul is a production center of cassava (***Manihot esculenta*** Crantz), a carbohydrate source and raw material for food industry. AMF inoculation in cassava plants is known to increase biomass production. However, little studies have been conducted on the response of cassava to mycorrhizal inoculation and organic fertilizer. Therefore, this study was aimed at examining the effects of AMF inoculation and types of manure on the AMF colonization and yield of cassava in Gunungkidul. The research was carried out by planting cassava in Alfisol Gunungkidul arranged in a randomized complete block design with two factors, AMF doses of 25g; 50; and 75g/plant; and types of manure i.e. cow, goat, and poultry manure, for five months period. Rhizosphere soil and root samples were analyzed for AMF colonization and the spores number. The results showed that AMF-infected cassava roots combined with cow or goat manure application produced more spores than poultry manure. AMF infection and manure, thus, significantly resulted in better root proliferation, root forehead weight, tuber diameter, and cassava products, than the absence of both treatments. Cow manure combined with AMF at a dose of 25 g/plant significantly affected the dry weight of cassava roots. This study implies that applying AMF and manure provide a substantial contribution on the growth and production of cassava.

Keywords: AMF, Cassava, Gunungkidul, Manure

ABSTRAK

Gunungkidul merupakan sentra singkong (Manihot esculenta Crantz), sebagai salah satu sumber kabohidrat dan bahan baku industri di Indonesia. Penelitian ini bertujuan untuk mengkaji pengaruh dosis inokulasi Mikoriza (Arbuscular Mycorrhizal Fungi-AMF) dan jenis pupuk kandang terhadap kolonisasi pada akar, pertumbuhan dan hasil singkong di Gunungkidul. Metode penelitian yaitu singkong ditanam di lahan Alfisol Gunungkidul dengan rancangan acak kelompok lengkap dan diberi perlakuan faktorial dosis AMF (25g, 50, 75g/tanaman) dengan jenis pupuk kandang (sapi, kambing, ayam). Tanah rhizosfer tanaman singkong dan sampel akar dianalisis kolonisasi Mikoriza dan jumlah sporanya. Parameter pertumbuhan tanaman dan hasil singkong selama 5 bulan dilakukan dianalisis. Hasil menunjukkan bahwa AMF menginfeksi akar singkong 100% dan aplikasi pupuk kandang sapi atau kambing menghasilkan spora lebih banyak dari pupuk kandang ayam dan nyata lebih baik terhadap proliferasi akar, berat kering akar, diameter ubi dan hasil ubi singkong. Pupuk kandang sapi dengan dosis AMF 25g/tanaman nyata saling berpengaruh terhadap berat kering akar tanaman singkong, sehingga disarankan penggunaan pupuk kandang sapi dengan mikoriza ini pada budidaya singkong karena dapat meningkatkan pertumbuhan dan hasil.

Kata kunci: *Mikoriza, Singkong, Gunungkidul, Pupuk Kandang*

INTRODUCTION

Symbiotic associations between Arbuscular Mycorrhizal Fungi (AMF) with plant roots often occur in almost 80% of terrestrial plants (Brundrett & Tedersoo, 2018; Zhang et al., 2019). AMF symbiosis with plants plays an essential role in the absorption of minerals, especially phosphorus ions exposed to soil and micronutrients, and increases the plant's resistance to pathogens, drought stress, and heavy metals so that it is potentially used as an environmentally friendly biological fertilizer (Jiang et al., 2017; Ryan & Graham, 2018). AMF inoculation in cassava plants can increase biomass production (De Bauw et al., 2021). Still, the variety strongly influences the association, species, and number of AMFs and their cultivation techniques (Ryan & Graham, 2018). The research of (Saputro

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et al., 2016) showed that the provision of a 75-gram crude AMF/plant was the most effective dose for the growth of *Albizia chinensis* plant. Some AMF genera associated with cassava are the genus *Glomus* sp., *Gigaspora*, sp., and *Acaulospora* sp. According to (Lone et al., 2017), the crude inoculum dose is AMF 20 grams/plant for agriculture.

Gunungkidul is a production center of cassava, a food source of carbohydrates in Indonesia used as raw material for the food processing industry, animal feed ingredients, and bioethanol (Hidayat et al., 2016; Ogundare, 2017). Cassava plants are easy to grow. However, fertilization is needed in its cultivation to get the optimal yield, and synthetic fertilizers are usually widely used. According to (Biratu et al., 2018b), the effect of synthetic fertilization on cassava depends on previous cropping patterns, soil type, and season. The application of 2.8 tons/ha of manure and *NPK* fertilizer (100:22:83) at the beginning of the wet season increased a higher yield compared to the application at the end of the dry season. The soil in Gunungkidul is weathering limestone with low organic matter content. The soil is infertile, dry, and fragile during the dry season. For this reason, it is necessary to study the proper organic matter to be applied for the sustainability of cassava cultivation in Gunungkidul by AMF inoculation.

Environmental factors, such as temperature, humidity, pH, and organic matter, affect the development of AMF (Ryan & Graham, 2018; Valverde Barrantes et al., 2017). Cow manure improves soil fertility and cassava production (Ognalaga et al., 2017). In intensive agriculture, the AMF population is lower than in low-input systems. In contrast, according to (Chandhana & Kerketta, 2021), goat manure can increase cassava weight and protein content. The advantages of chicken manure applications are improving soil physical properties, water binding capacity, organic matter, and soil nutrient content (Biratu et al., 2018a; Biratu et al., 2018b). For this reason, this study aimed to examine the effects of mycorrhizal fungi inoculation dose and type of manure on the colonization of roots, growth, and yield of cassava in Gunungkidul.

MATERIALS AND METHOD

The research was conducted in Alfisol soil in Gunungkidul, arranged in a randomized complete block design consisting of two factors. The first factor was AMF dose (25g, 50, and 75g/plant), and the second was the type of manure (cow, goat, and poultry). Each treatment combination was replicated three times, each consisting of eight plants.

Gunungkidul indigenous AMF inoculum was obtained by multiplication of the trapping method for three months, then applied in the planting hole before planting cassava seeds with a spacing of 1x1 m. The type of manure treatment was given a week before planting (Selvakumar et al., 2016)

The number of infections was observed using microscopic analysis, according to the method of Kormanik &McGraw, and calculated based on the AMF colonization in the roots of cassava plants. The amount of AMF spores was calculated by extracting 100 g of rhizosphere soil using the wet sieving and decanting technique (Selvakumar et al., 2016). Dry root weight, the number of primary and secondary roots, plant height, and the number of leaves were determined when the plants aged 1, 2, and 3 months. Meanwhile, the length, diameter, number, and weight of the tubers were determined by harvesting 5-month-old plants.

Statistical analysis

The data of AMF colonization and the number of spores, root dry weight, number of primary and secondary roots, plant height, number of leaves, tuber's length, diameter, number, and weight were analyzed using analysis of variance. If there was a significant difference between treatments, the data were subjected to Duncan Multiple Range Test at a significance level of 5%.

RESULTS AND DISCUSSION

AMF colonization of cassava roots

Mycorrhizae colonize cassava plants by infecting roots (Straker et al., 2010). The percentage of internal hyphae formation, external hyphae, arbuscular, or vesicles on the roots indicate mycorrhizal colonization at the roots of cassava plants (Ryan & Graham, 2018). Based on microscopic analysis of roots colonized by AMF, this study reported compatibility between mycorrhizae and cassava plant roots, as indicated by mycorrhizal colonization of cassava roots by 100%. However, there was no interaction between AMF doses and types of manure. The AMF dose or types of manure did not affect anything. The development of AMF colonization is presented in Figure 1.

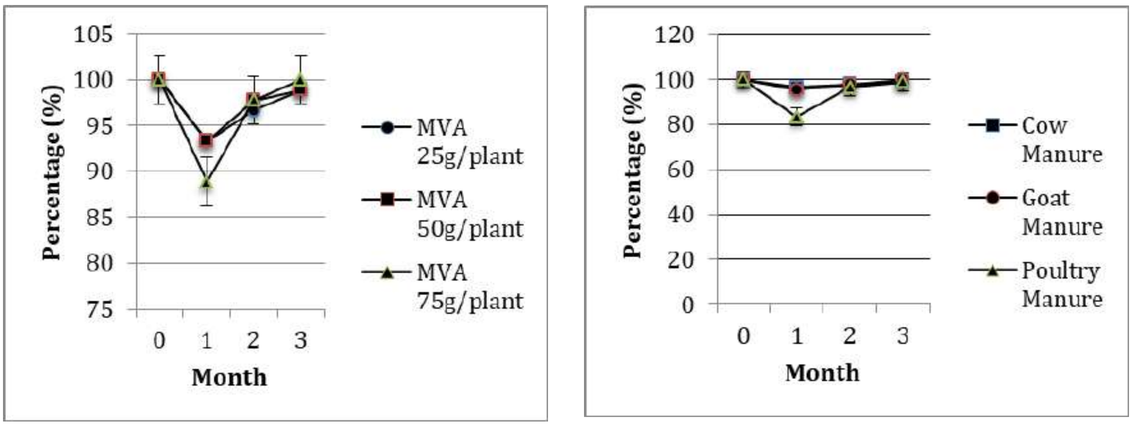
This result showed that the compatibility of Gunungkidul indigenous mycorrhizae with cassava roots was excellent, as indicated by the percentage of AMF colonization at a dose of 25 g/plant, which was not significantly different from that at doses of 50 g/plant or 75 g/plant (Table 1). The colonization percentage was slower with the higher AMF dose due to the competition between spores.

From trapping results, AMF first infected 100% of corn plants. However, after the inoculation of cassava plants, the percentage of AMF colonization decreased in the first month. It is because the

Table 1. The percentage of mycorrhizal colonization and spores at the roots of cassava plants in the 3rd month

Treatments	Mycorrhizal Colonization (%)	Number of spores (spores / 100g of soil)
Mycorrhizal Dosage:		
25 g/plant	100 a	62.3 a
50 g/plant	100 a	67.2 a
75 g/plant	100 a	62.2 a
Manure types:		
Cow manure	100 p	66.6 p
Goat manure	100 p	69.6 p
Poultry manure	100 p	52.8 q
Interaction	(-)	(-)

Remarks: Means followed by different letters are significantly different based on the F test at a significance level of 5%; (-) indicates no interaction between treatments



(a) (b)

Figure 1. Development of AMF colonization as affected by (a) AMF dose and (b) Type of manure

AMF infection process was taking place at the root of the plant, and it turned out that the 75 g dose resulted in the lowest colonization (88%). Likewise, AMF colonization in plants treated with poultry manure was the lowest (83%) compared to those treated with cow manure (96%) and goat manure (95%). Later in the next 2nd and 3rd months, the overall colonization reached 100%, and there was no mutual influence and a significant difference between treatments (Table 1).

The number of AMF spores and their diversity

AMF symbiosis with plant roots gets energy from the host and develops to produce spores. In the third month, AMF dose and type of manure did not influence the number of spores (Table 1). Still, the highest number was in cow manure (66.6 spores/100 g of soil) and goat manure (69.6 spores/100 g of soil), which was significantly different from that in poultry manure (52.8 spores/100 g of soil). While the development of the number of spores in the rhizosphere of cassava plants over three months showed an increasing number of spores, there was no significant interaction effect between AMF doses and types of manure (Figure 2).

AMF spore production increased rapidly after the 2nd month, reaching 66.78 spores/100g of rhizosphere soil, but it was not affected by the AMF dose. In contrast, cow manure (67 spores/100g of rhizosphere soil) and goat manure (68 spores/100g of rhizosphere soil) were the best organic matter to increase the number of AMF spores compared to poultry manure (53 spores/100g of rhizosphere soil). Based on the identification of spore types, it was dominated by *Glomus* sp., although some *Gigaspora* sp. and *Acaulospora* sp. also existed.

The AMF in symbiosis with plant roots obtains energy from the host and develops to produce spores (Zhang et al., 2019). The three-month observations showed that the number of spores in the cassava rhizosphere increased, along with the increase in the percentage of AMF colonization, which was not affected by various AMF doses (Figure 2). However, the number of spores in plants fertilized with cow or goat manure was significantly higher ($p < 0.05$) than those of poultry manure. Cow manure and goat manure were the best organic materials to increase the number of mycorrhizal spores and could replace one another. According to (Begoude et al., 2016), the type of fertilization in cassava cultivation affects the indigenous AMF population. (Biratu et al., 2018b) support the statement by stating that chicken manure weakens the appearance and composition of cassava nutrients.

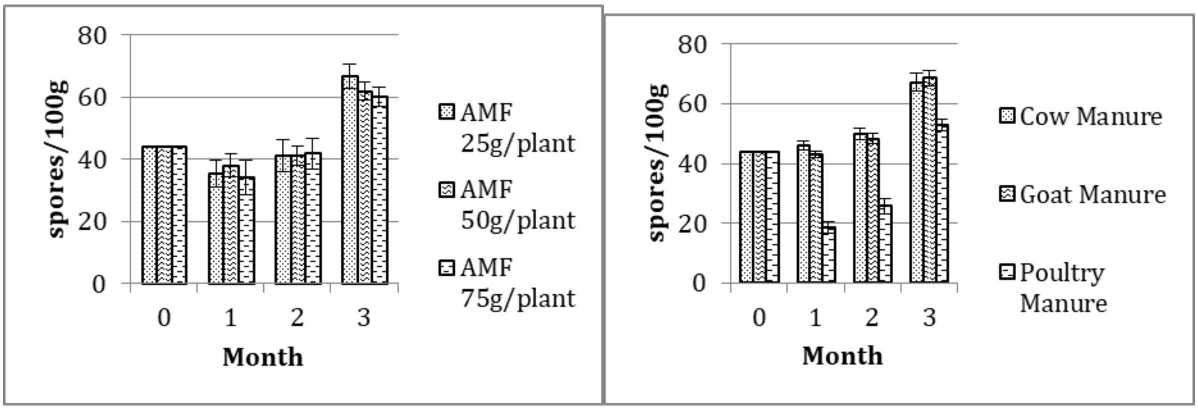
Identification of AMF

The type of spores identified was dominated by *Glomus* sp., although several Gigaspora sp. and Acaulospora sp. were observed. According to (Begoude et al., 2016), *Glomus* sp. dominates the tropics and is usually present in soils. The previous study by (Astuti et al., 2020) showed that the genus *Glomus* sp., *Gigaspora* sp., and *Acaulospora* sp. identified the indigenous AMF spores of Gunungkidul. (Lopes et al., 2019) supported the result by showing that AMF colonization in cassava plants could reach 93%, usually from the genus *Glomus*, *Gigaspora*, and *Acaulospora*.

AMF association in cassava roots

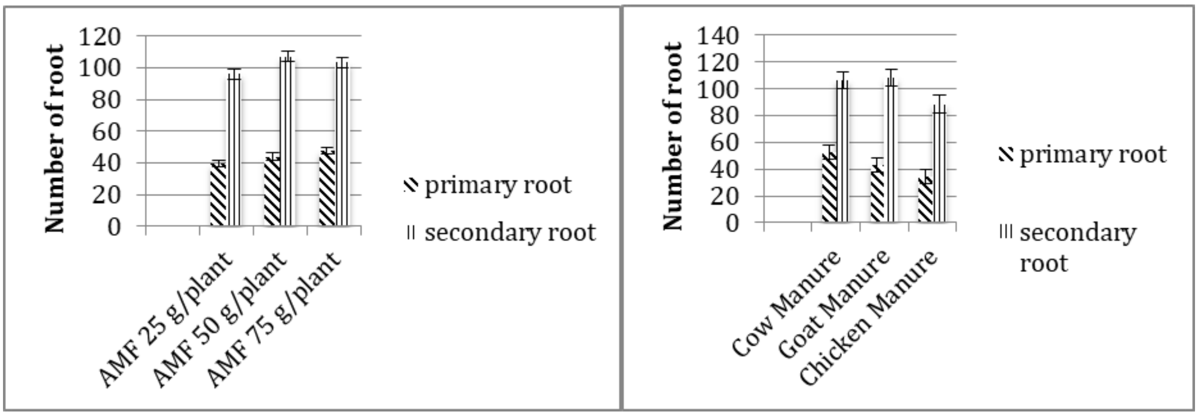
Mycorrhizal spore infection into the roots of cassava plants will stimulate root branching (Zhang et al., 2019). AMF infection affects the root in terms of length, dry weight, and proliferation, as indicated by the number of primary and secondary roots (Figure 3).

Mycorrhizal spore infection into the roots of cassava plants will stimulate root branching. Various AMF doses showed the same effect on the number of spores, so the number of primary and secondary



(a) (b)

Figure 2. Number of spores as affected by (a) AMF dose and (b) Type of manure



(a) (b)

Figure 3. The average number of primary and secondary roots at the 12th week as affected by (a) AMF dose and (b) Type of Manure

roots was also not affected by AMF doses. However, in the treatment of manure types, the number of spores was high, so the number of primary and secondary roots was also high. The primary and secondary roots of plants fertilized with cow and goat manure were significantly higher than those fertilized with poultry (Figure 3).

The results showed a correlation between the number of spores with the number of primary and secondary roots. However, the treatment of AMF doses did not affect the number of spores and the number of primary and secondary roots. Meanwhile, the type of manure significantly affected the number of spores and the number of primary and secondary roots. The high number of spores in the treatment of cow and goat manure stimulated the number of primary roots (52.71 and 43.28, respectively) and secondary roots (108.43 and 108.57, respectively). This result was significantly higher than in poultry manure, which produced primary and secondary roots of 34.33 and 88.50, respectively. Another effect observed was on the root dry weight of cassava plants, which was the highest in the treatment of cow manure with an AMF dose of 25 g (2.96 g) (Table 2).

AMF infection affected root proliferation, so the root dry weight increased. There was an interaction effect of AMF dose and type of manure on the root dry weight of cassava plants, which was the highest (2.96g) at AMF dose of 25 g/plant combined with

Table 2. Average root dry weight at week 12 (gram)

AMF Dose	Types of Manure			Average AMF Dose
	Cow	Goat	Poultry	
25 g/plant	2.96 a	2.14 bc	1.30 bc	2.13
50 g/plant	1.28 bc	1.27 bc	1.99 ab	1.51
75 g/plant	0.76 c	0.85 c	1.43 bc	1.01
Average Types of Manure	1.66	1.42	1.57	(+)

Remarks: Means followed by different letters are significantly different based on the DMRT test at a significance level of 5%; (+) indicates an interaction between treatments

Table 3. Average growth and products of cassava

Treatments	Height (cm)	Number of leaves (strands)	Number of tubers /plant	Diameter of tuber (cm)	The length of tuber (cm)	Weight of tuber/ plant (kg)	Cassava yield (ton / Ha)
AMF Dose:							
25 g/plant	237.44 a	224.67 a	11.22 a	**32.20 a**	22.52 a	**3.87 a**	**38.76 a**
50 g/plant	234.67 a	224.89 a	11.78 a	31.18 ab	22.50 a	3.79 ab	37.92 ab
75 g/plant	236.45 a	216.45 a	11.56 a	29.71 b	21.54 a	3.65 b	36.53 b
Types of Manure:							
Cow	242.45 p	218.00 p	13.78 p	31.15 p	22.44 p	3.86 p	38.63 p
Goat	225.22 p	224.33 p	10.33 q	30.47 p	21.34 p	3.34 q	33.40 q
Poultry	240.44 p	219.67 p	10.44 q	29.46 p	22.79 p	3.41 q	34.13 q
Interaction	(-)	(-)	(-)	(-)	(-)	(-)	(-)

Remarks: Means followed by different letters are significantly different based on the DMRT test at a significance level of 5%; (-) indicates no interaction between treatments

cow manure, while the lowest was 0.76-0.85 g at AMF dose of 75g / plants combined with cow or goat manure (Table 2).

Cassava growth and yield

The analysis of variance showed no interaction effect of AMF dose and the type of manure (cow, goat, and poultry) on all growth variables and cassava yields. Still, each factor affected the yield of cassava independently (Table 3).

The AMF dose of 25-50 g per plant resulted in the highest value of tuber diameter and tuber weight per plant, reaching 37.92-38.76 tons per hectare, compared to 75g/plant (36.53 tons). Meanwhile, the application of cow manure significantly increased the number of tubers and the weight of tubers per plant, resulting in the highest values per hectare (38.63 tons) compared to goat or poultry manure (34.13 tons).

The results of this study indicated that the treatment of Gunungkidul indigenous AMF doses and types of manure on cassava plants could increase root proliferation and dry weight so that it had a significant effect on the tuber. However, the effects on the plant growth, height, and number of leaves were not significant (Table 3). The application of AMF at a dose of 25 g/plant had the most significant effect on the diameter of the tuber (32.20 cm) and the most substantial cassava yield (3.87 kg/ plant). At the same time, cow manure affected the highest number of tuber (13.78 tuber/plant) and the highest cassava yield (3.86 kg per plant). This result is in line with the opinion of (Lehmann et al., 2017) that AMF symbiosis in plants can increase nutrient absorption and resist drought stress, thereby increasing plant growth and yield.

CONCLUSIONS

The results showed that AMF-infected cassava roots and cow or goat manure application produced more spores than poultry manure. AMF infection and manure, thus, significantly resulted in better root proliferation, root forehead weight, tuber diameter, and cassava products, than the absence of both treatments. Cow manure combined with AMF at a dose of 25 g/plant significantly affected the dry weight of cassava roots. This study implies that applying AMF and manure provide a substantial contribution on the growth and production of cassava.

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Fertilizers for Improving the Growth Characteristics and N Uptake of Wild *Rorippa indica* L. Hiern in Different Soil

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ABSTRACT

Rorippa indica L. Hiern is a local vegetable that is widely consumed by Dayak's tribe in Central Kalimantan, Indonesia. It is mostly traditionally cultivated without fertilizers, resulting in low productivity. The research aimed to investigate the effect of fertilizers and soil type on the growth characteristics and N uptake of *R. indica*. The experiment was arranged in a factorial completely randomized design consisting of two factors with four replications. The first factor was fertilizer application (control, 20 t ha^{-1} of chicken manure, and 600 kg ha^{-1} of NPK), and the second was soil type (peat and Ultisol). The results revealed that the interaction of fertilizers and soil type gave a non-significant effect on all variables observed, except N uptake. The application of NPK increased the plant height and number of leaves significantly. Compared to control, the increment was 112.50% and 130.32%, respectively, and chicken manure application increased the dry weight (327.87%), N total (310.16%), and N uptake of plants by 478% in peat soil and 228% in Ultisol. This finding concludes that 20 t ha^{-1} of chicken manure can be applied to increase the productivity of *R. indica*.

Keywords: Inorganic fertilizers, Organic fertilizers, Peat, *Rorippa indica*, Ultisol

ABSTRAK

Rorippa indica L. Hiern merupakan sayuran lokal yang dikonsumsi oleh masyarakat Dayak di Kalimantan Tengah. Sayuran ini tumbuh liar tanpa asupan pupuk dalam budidayanya berakibat pada rendahnya hasil tanaman. Tujuan penelitian ini adalah mempelajari pengaruh pemberian pupuk dan perbedaan tanah sebagai media tumbuh terhadap pertumbuhan dan serapan N dari R. indica. Rancangan penelitian menggunakan rancangan acak lengkap faktorial dan diulang sebanyak 4 kali. Faktor pertama adalah pemupukan (kontrol, 20 t ha^{-1} kotoran ayam, dan 600 kg ha^{-1} NPK), sementara itu faktor kedua adalah tipe tanah yang digunakan (gambut dan Ultisol). Hasil penelitian menunjukkan bahwa tidak terjadi interaksi antara pemupukan dan tipe tanah pada semua parameter pengamatan, kecuali serapan N. Pemberian NPK mampu meningkatkan tinggi tanaman (112.50%) dan jumlah daun (130.32%) dibandingkan kontrol. Aplikasi pupuk kotoran ayam meningkatkan berat kering tanaman (327.87%), N total (310.16%), dan serapan N sebanyak 478% pada tanah gambut dan 228% pada Ultisol. Pemupukan dengan kotoran ayam sebanyak 20 t ha^{-1} dapat diaplikasikan untuk meningkatkan produktivitas R. indica.

Kata kunci: *Pupuk anorganik, Pupuk organik, Gambut, Rorippa indica, Ultisol*

INTRODUCTION

Brassicaceae is a family that includes many important crop plants, ornamentals, and weeds (Liu et al., 2011). The Rorippa Scop. is one of the Brassicaceae, comprising approximately 80 species, including *Rorippa indica* (L) Hiern, *R. palustris* (L) Besser, *R. integrifolia* Boulos (Marzouk et al., 2016), *R. cantoniensis* (Lour.) Ohwi (Liu et al., 2012), *R. fluviatilis* (E.Mey.ex Sond), *R. nudiuscula* (Welcome & Van Wyk, 2019; Moteetee et al., 2019), *R. islandica*, *R. subumbellata* and *R. nasturtium* (Baskin & Baskin, 2014). *R. indica* L. Hiern belongs to the family of Brassicaceae or Cruciferae. The vascular plant (Hwang et al., 2013; Lee et al., 2013; Jang et al., 2013; Yoon et al., 2013) has the common names of watercress and field cress. Many are found in Asia, South and North America (Xu & Deng, 2017), and India, with a dense population (Ananthi & Kumari, 2013). In the various report, *R. indica* was regarded as a wild plant (Bandopadhyay et al., 2013; Nag & Hasan, 2016; Takabayashi & Shiojiri,

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2019;), weed (Hamdani & Nuryanti, 2011; Nasu & Momohara, 2016; Nazir et al., 2016; Sarkar et al., 2016; Hwang et al., 2017), animal forage (Marzouk et al., 2016), phytoremediator (Cui et al., 2013), medicinal plant (Ananthi & Kumari, 2013; Long-Ze et al., 2014; Siew et al., 2014; Zhang et al., 2014; Dutt et al., 2015; Marzouk et al., 2016; Sengupta et al., 2018; Yang et al., 2020; Lin et al., 2021), companion species in the rice field (Kim et al., 2019), and wild edible plant (Moteetee et al., 2019; Iyda et al., 2019).

R. indica is a local vegetable consumed by Dayak's tribe in Central Kalimantan. Leaves of *R. indica* have been used by local people to fulfill their need for sources of vitamins and minerals. They consume the vegetable in clear soup with corn, milk soup, and salad. The vernacular name is segau (Chotimah et al., 2013). *R. indica* is not specifically cultivated, and it tends to grow wild. Their natural habitat is on the roadsides, burned land, valley, riverbanks, wetland, gardens and, rice fields. The *R. indica* also dominates the perennially and seasonally flooded areas as well as flood plain areas (Liu et al., 2020). It grows best on sandy soils, and full sunlight favors better flowering. It has a simple leaf, bright yellow petals, and round seeds, with a flowering age of 46 – 51 days. *R. indica* has jagged leaves, and its taste is slightly bitter. The plant height was around 1.9 and 0.6 m when planted in lowland next to the water and upland, respectively. The seed is brownish-red, with an ovoid shape and a size of 0.8–1 x 0.7–0.8 mm.

The government recognizes the requirement for systematic wild plant species conservation, which pays more attention because wild plant species will be of value for the future for securing this vast reservoir of diversity for agriculture and food security. The wild plants are experiencing widespread genetic erosion and even extinction (Vincent et al., 2013). The germplasm rescue and the land conversion for oil palm plantation and the forest fire every year in Central Kalimantan become a basic consideration for domesticating *R. indica* to improve food security significantly. Amelioration is our target to underpin their genetic adaptation to a diverse range of habitats.

Based on personal communication with farmers, the decrease in *R. indica* yields could reach 0.25 tons per ha due to constraints in the use of growing media. The abundant soil in Central Kalimantan is peat and Ultisol. These soils are sometimes grouped as marginal soils due to their acidity and low natural fertility (Prasetyo et al., 2016; Maftu'ah & Nursyamsi, 2019). Fertility can be improved by amelioration applications, including inorganic and organic fertilizers. Alakhyar et al., (2019) investigated the effects of six organic fertilizers concentration of 0%, 20%, 40%, 60%, 80%, and 100% on the *Brassica juncea* L production, and the optimum concentration obtained was 70.85% to produce a weight of 73 g per plant. The attempts to develop a prominent yield were conducted by Mir et al., (2010) using the combination of phosphorous and potassium on the mustard yield, and 60 kg P_2O_5 ha^{-1} and 60 kg P_2O_5 + 60 K_2O ha^{-1} were proven to improve the seed yield. Therefore, fertilizer use is the key factor in maintaining soil quality, enhancing soil nutrients, and increasing crop production. There is only preliminary information on the fertility requirement of wild plant *R.indica*. Optimum nutrients amount has a major impact not only on crop productivity but also on nutritional value. Hence, the study was conducted to determine the effects of fertilizers (organic and inorganic NPK) and the soil type on the growth characteristics and nitrogen uptake of *R.indica*.

MATERIALS AND METHODS

The research was conducted from January to June 2018 at the Greenhouse of the Department of

Agronomy, University of Palangka Raya (S 2°12'42" E 113°54'15"). Peat and Ultisol were obtained from Kalampangan Palangka Raya and Pundu Katingan District, taken at a depth of 20 cm. The experiment was arranged in a factorial completely randomized design, consisting of two factors with four replications. The first factor was fertilizers application (control, 20 t ha^{-1} chicken manure, 600 kg ha^{-1} NPK (16-16-16), and the second factor was soil type (peat and Ultisol). The peat soil has soil pH (H_2O) of 3.35, N-total (Kjeldahl) of 0.64%, organic C of 57.01% (Walkey and Black), available P (Bray I) of 165.67 ppm, exchangeable K (NH_4OAc pH 4.8) of 0.63 cmol/kg, exchangeable Ca (NH_4OAc pH 4.8) of 2.11 cmol/kg, and base saturation of 13.09%, respectively. Meanwhile, Ultisol has soil pH (H_2O) of 4.25, total N of 0.17%, organic C of 2.65%, available P (Bray I) of 53.53 ppm, K of exchangeable 0.22 cmol/kg, exchangeable Ca (NH_4OAc pH 4.8) of 0.93 cmol/kg, and base saturation of 9.60%. The chemical properties of chicken manure are 1.39% total N, 872.40 ppm total P, 15752.42 ppm total K, 8549.53 ppm total Ca, and 5366.33 ppm total Mg.

The seeds used were obtained from the farmer in Seruyan District. Before planting, the seeds were planted at the seedbed for 21 days with husk charcoal media, and then planting was done by placing one seedling per polybag. Fertilizers were applied at planting and repeated four times in seven days. Chicken manure was added to as much as 20 t ha^{-1} (227g/5 kg peat/polybag; 74g/12 kg Ultisol/polybag), and NPK was applied to as much as 600 kg ha^{-1} (6.82 g/5 kg peat/polybag; 2.22 g/12 kg Ultisol/polybag) by placing it around 5 cm from root. Watering was carried out twice a day using 250 ml glass. Weeding was manually performed by pulling out the weeds. *R.indica* was harvested 60 days after planting (DAP) by pulling out the whole plant. The observed growth characteristics include plant height, number of leaves, leaf area (measured by leaf area meter), plant dry weight, and total N of tissues measured at 35 DAP and determined by HNO_3-$HClO_4$ wet extraction. N uptake is the total N of tissues multiplied by plant dry weight. The samples for total N of tissues were analyzed at the Soil Laboratory of the University of Lambung Mangkurat. In addition, the chemical properties of soil were analyzed at the Analytical Laboratory of the University of Palangka Raya. The collected data were subjected to ANOVA, followed by LSD test with 5% significance levels using SPSS statistical package.

RESULTS AND DISCUSSION

Chemical properties of soils

The application of organic and inorganic fertilizers increased the chemical properties of both soils (Table 1). Compared to control, the pH, N, P, K, Ca and base saturation of peat increased by 36.72%, 78.13%, 458.22%, 379.37%, 248.82% and 160.35%, respectively, while the increment of Ultisol were 57.65%, 70.59%, 154.08%, 1272.73%, 312.90% and 135.31%, respectively. Generally, soil fertility increased with the application of fertilizers compared to the initial media properties before treatments. The increasing pH due to 20 t ha^{-1} chicken manure and 600 kg ha^{-1} NPK indicated that fertilizer application on *R.indica* could provide plant nutrient content. Increased soil pH affects the increase in negative soil charge. The soil charge of both peat and Ultisol is pH-dependent (Lesbani & Badaruddin, 2012). The functions of negative soil charge are to bind the cations present in the soil, resulting in reduced leaching and enlarged storage capacity of nutrients in the soil. The foregoing is shown by increasing exchangeable K, exchangeable Ca, and base saturation in both types of soil. The base saturation in both types of soil was from 13.09% to 34.08% in peat and 9.60% to 22.59%

Table 1. The chemical properties of soils as affected by fertilizer application

Properties	Peat	Ultisol
pH H20 (1:2,5)	4.58	6.70
Total N (%)	1.14	0.29
Organic C (%)	54.22	0.88
P-Bray I (ppm)	924.8	136.01
Exc. K (cmol/kg)	3.02	3.02
Exc. Ca (cmol/kg)	7.36	3.84
Base Saturation (%)	34.08	22.59

Table 2. The plant height, number of leaves, leaf area of *R. indica* at 35 (DAP) and dry weight as well as N content of tissue as affected by the application of fertilizers

Fertilizers	Plant height (cm)	Leaf number	Leaf area (cm)	Dry weight (g)	N-total tissue (%)
Control	16.79 a	15.63 a	13.70 a	0.61 a	3.11 a
20 t ha-1 chicken manure	31.53 b	32.00 b	21.34 b	2.61 c	3.34 a
600 kg ha-1 NPK	35.68 b	36.00 b	19.85 b	1.49 b	3.38 a
LSD 0.05	5.43	5.74	3.30	0.85	2.72
Soil					
Peat	26.64	26.67	19.34	1.50	3.34
Ultisol	29.35	29.08	17.26	1.64	3.21
LSD 0.05	3.65	3.86	2.22	0.57	1.83

Remarks: Means followed by the same letter sat the same column are not significantly different based on LSD test at a level of α 0.05

in Ultisol, respectively. The rate of increase in pH is also closely related to plant biomass. This can be seen from the significant differences in the dry weight of *R.indica*. In the Ultisol, chicken manure given at a dose of 20 t ha^{-1} produced the maximum plant weight of 2.61g, followed by NPK at 1.49g, while the plants without fertilizer produced the minimum plant dry weight of 0.61 g (Table 2).

The increase in negative soil charge also increased the availability of soil P. The high positive charge in both soils has a strong binding to soil P, causing its availability to be very low. Decreasing the positive charge will release phosphate compounds into the soil. In the Ultisol, the presence of both Al and Fe compounds induces P unavailable to plants due to P compounds being fixed and difficult to release (Khan et al., 2014). The rising pH value results in the declining mobility of metal Al and Fe (Ch'Ng et al., 2014) and the release of fixated P to the soil. In the peat soil, the rising pH value exhibits the decline of the toxic organic acids' activity for plants. An increase in soil pH stimulates soil microorganisms to actively aid in the breakdown of organic matter (Ichriani et al., 2021) into an organic, which is more available to plant growth, such as the availability of P. The soil microorganisms' activity produces phytohormones, vitamins, and amino acids that can release soil P (Chakkaravarthy et al., 2010). Chicken manure contains high total nutrients.

In contrast to other organic fertilizers, chicken manure decomposes relatively quickly. The activity can be diminished by the provision of binding agents, such as ash (Haryoko, 2012). The improvement of soil chemical conditions will provide the best atmosphere for growing media that support plant growth.

Growth characteristics

The interaction effect of fertilizer and soil types was not significant on the growth characteristics. Compared to control, applying organic and inorganic fertilizers increased the growth characteristics of *R. indica* (plant height, number of leaves, leaf area, dry weight) (Table 2). Moreover, Table 2 shows that the increment of plant height, number of leaves, and leaf area are 87.79%, 104.73%, and 55.77%, respectively, in organically fertilized pots. Meanwhile, those in NPK fertilized pots are 112.51%, 130.33%, and 44.89%, respectively. The highest biomass or dry weight was obtained in the treatment of 20 ton ha^{-1} chicken manure, followed by 600 kg ha^{-1} NPK and control.

The case of low soil fertility is considered as one of the most important constraints on improving crop production, including *R. indica*. Fertilizers can provide sufficient nutrients for good plant growth. The data showed that these treatments had significant effects (Table 2). Therefore, it is concluded that using fertilizers in the *R. indica* cultivation should be encouraged. The use of fertilizers for vegetable crops that belong to the Brassicaceae family was confirmed by Olaniyi & Ojetayo (2011). The slightest growth response of the unfertilized cabbage might be due to the low nutrient availability during the growth period. The vegetable crop performance could be linked to genetic and environmental influences, including climatic conditions, nutrient source, and soil fertility. The use of fertilizers is attributed to the availability of nutrients, thus increasing plant growth. The field experiment by Jankowski et al., (2019) reported that *Camelina sativa* (L.) Crantz, Brassicaceae fertilized with N produced taller, thicker, and more branched shoots. Fertilizer nitrogen higher than 120 kg ha^{-1} is recommended in the *C. sativa* seed production.

The chemical properties of both soil types also showed improvements due to fertilization (Table 1). This is inconsistent with Uka et al., (2013), reporting that inorganic fertilizers, such as NPK, worsen soil degradation, thereby generating higher acidity, nutrient imbalance and low crop yield. Whilst, organic fertilizer promotes the gradual release of nutrients over time. Jankowski et al., (2019) conveyed Brassicaceae's fertilizers and soil fertility needs, previously Cruciferae or Crucifers. Throughout their life cycle, Brassica crops require certain nutrients in varying amounts to support optimal growth and reproduction. The optimal growth and reproduction can be achieved if the soil is healthy. Healthy soil will have a greater capacity to uptake fertilizers, and nutrient uptake will be more balanced. To maintain healthy soil is cultivation practices, such as applying manure and compost, using soil cover, and crop rotation.

The plant height was higher compared to the results obtained by Syahid et al., (2013), who reported that *R.indica* grown in peat soil treated by adding chicken manure combined with charcoal husk resulted in the plant height of 23.4 cm 5 weeks after planting. Both results of the experiment are still shorter evidently if compared to those in their natural habitat. The plant reached 0.6 m in upland and 1.9 m in the lowland next to the water. Table 2 shows that NPK fertilizer plays a role in the first stage of plant growth, especially in the elongation of stems and leaf formation. The role of organic fertilizer is in leaf area development and the formation of plant biomass. This is in line with Uka et al., (2013) research on okra (*Abelmoschus esculentus*). The fastest growth rate occurred in the first three weeks due to NPK fertilizer, while the plants treated with organic fertilizer grew taller from the sixth week up to the end of the experiment in the tenth week. The highest values of plant growth and yield were found in *Raphanus sativus*, an edible root vegetable that belongs to the Cruciferae family, treated with NPK. (Kiran et al., 2016). Syahid et al., (2013) reported that the administration of chicken manure combined with husk charcoal to *R.indica* resulted in a large number of leaves of 9.3. Yuseda (2012) reported that *R.indica* planting in mineral soil produced the highest leaf number of 10.70.

Concerning leaf area expansion, the organic fertilizer improved the leaf area of *R. indica* at 35 DAP (Table 2). The organic nutrient source has been reported by Lim & Vimala (2012) to improve both vegetable quality and soil chemical, physical, and biological properties. This has an important effect on the high charge of organic matter for retaining nutrients and preparing them available to the plants (Diacono & Montemurro, 2010). The availability of nutrients on plant roots increases plant growth (Uka et al., 2013).

Amelioration with chicken manure at a dose of 20 t ha^{-1} produced maximum dry weight of 2.61 g, followed by those treated with NPK of 1.49 g compared to control (Table 2). Organic manures, like chicken manures, promote microbial degradation and the gradual release of nutrients over time, while NPK results in soil degradation due to loss of inorganic matter, which leads to higher acidity, nutrient imbalance, and low crop yield (Adewole & Ilesanmi, 2012). In the rhizosphere, organic fertilizer can help shape the microbial composition and recruit beneficial bacteria into the rhizosphere (Lin et al., 2019).

N uptake

Both fertilizer applications gave no significant increase in the total N of *R. indica* (Table 2). Moe et al., (2019) declared that nutrient uptake characteristics generally varied with the cultivar, soil type, environment, and fertilizers used. The nutrient content in soil, particularly N, P, and Zn, could also be improved by applying cattle manure on leafy vegetables due to reducing soil acidity and increasing soil electrical conductivity without affecting the growth and yield of the leafy vegetables (Mantovani et al., 2017). Total N in the tissues of *R. indica* impacted by both fertilizers (Table 2) was higher than N contents of *Diplazium esculentum*, the wild edible fern collected from Bangladesh, at 13.97 mg/g (Zihad et al., 2019). Ntuli (2019) also recorded the nutrient content of nine rare wild leafy vegetables consumed by rural communities in northern KwaZulu-Natal from 3.89-6.29% N. The sufficient nitrogen leaf content varies from a low of 2.00 to a high of 5.00% of the dry weight.

The interaction effect of fertilizers and soil type was significant. On average, both fertilizers increased the N uptake of *R. indica* in both soil types. The highest N uptake occurred on *R. indica* treated by organic fertilizer in Ultisol (Table 3). The nutrient uptake by plants is strongly influenced by the level and availability of nutrients in the soil (Nugraha, 2010). Soil organic matter is a predominant source of N crops. The suitability of organic matter to finetune the nutrient supply to the crop requirement is characterized by the fast N availability provided (Tei et al., 2020). Biofertilizer was also recommended for providing N nutrient in lettuce grown in a Ultisol (Stamford et al., 2019).

Table 3. The N uptake (g/plant) of plants as affected by soil types and fertilizers

Fertilizer	Growing media (soil types)	
	Peat	Ultisol
Control	1.24 a	2.49 a
20 t ha-1 chicken manure	7.17 c	8.17 c
600 kg ha-1 NPK	5.44 b	4.65 b

Remarks: Means followed by the same letter sat the same column are not significantly different based on LSD test at a level of α 0.05

CONCLUSION

Application of inorganic and organic fertilizer improved soil properties, growth characteristics, and N uptake of *R. indica* in both soil types. All observed variables were not affected significantly by the type of soil. Compared to control, the application of 20 t ha^{-1} chicken manure increased the dry weight and N uptake, while the 600 kg ha^{-1} NPK increased plant height and number of leaves. The maximum dry weight (2.61g/pot) and N uptake (8.17 g/plant) were obtained from the application of 20 t ha^{-1} chicken manure (75.17% and 75.70% higher than NPK fertilizer). These results confirm that chicken manure, as an environmentally friendly ameliorant, can be applied to improve the productivity of *R. indica*.

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Inoculation of Merapi Indigenous Rhizobacteria as A Substitute Compost for Application in Rice Cultivation on Coastal Sandy Under Drought Stress

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ABSTRACT

This study aimed to determine the role of the indigenous rhizobacteria from Merapi as a substitute for compost in rice cultivation on coastal sandy land under drought stress. The study was a single-factor experiment, with types and doses of compost as treatments, arranged in a completely randomized design (CRD) consisting of seven treatments and three replications. The seven treatments tested were cow manure compost at doses of 30 and 40 tons/ha, chicken manure compost at doses of 30 and 40 tons/ha, Azolla compost at doses of 20 and 30 tons/ha, and without compost as a control treatment. Each experimental unit consisted of three plants for destructive sampling, three sample plants, and a substitute plant. The application of cow manure compost at a dose of 30 tons/ha to the rice plants inoculated with MB and MD isolates of Merapi indigenous rhizobacteria resulted in the best growth at five weeks after planting, which was not significantly different from that without compost application. This result indicated that the rice plants cv. Segreng Handayani inoculated with Merapi indigenous rhizobacteria, cultivated on coastal sandy soil under drought stress, even without the application of compost, could give the same responses as the plants treated with various types and doses of compost.

Keywords: Coastal sandy, Compost, Indigenous rhizobacteria, Merapi

ABSTRAK

Penelitian ini bertujuan untuk mengetahui peran Rhizobacteri indigenous *Merapi sebagai substitusi kompos pada budidaya padi di lahan pasir pantai yang mengalami cekaman kekeringan. Penelitian ini merupakan eksperimen faktor tunggal yaitu jenis dan takaran kompos yang disusun dalam Rancangan Acak Lengkap (RAL) yang terdiri dari tujuh perlakuan dan tiga ulangan. Ketujuh perlakuan yang diuji adalah kompos kotoran sapi dengan dosis 30 dan 40 ton/ha, kompos kotoran ayam dengan dosis 30 dan 40 ton/ha, kompos Azolla dengan dosis 20 dan 30 ton/ha, dan tanpa kompos sebagai perlakuan kontrol. Setiap satuan percobaan terdiri dari tiga tanaman korban, tiga tanaman sampel, dan satu tanaman pengganti. Aplikasi kompos kotoran sapi dosis 30 ton/ha pada tanaman padi yang diinokulasi MB dan MD isolat Rhizobakteri* indigenous *Merapi menghasilkan pertumbuhan terbaik pada umur lima minggu setelah tanam, yang tidak berbeda nyata dengan tanpa aplikasi kompos. Hasil ini menunjukkan bahwa tanaman padi varietas Segreng Handayani yang diinokulasi dengan Rhizobakteri* indigenous *Merapi yang dibudidayakan di tanah pasir pantai di bawah cekaman kekeringan, bahkan tanpa aplikasi kompos, dapat memberikan respons yang sama dengan tanaman yang diperlakukan dengan berbagai jenis dan dosis kompos.*

***Kata kunci:** Pasir pantai, Kompos, Rhizobakteri indigeneus, Merapi*

INTRODUCTION

The rice production in Indonesia in 2017 was 70.61 million tons of dry unhusked rice ready for milling (GKG), which increased by 0.67 million tons (0.94%) compared to rice production in 2016 (BPS, 2017). The improvement of rice production has not been able to keep up with the increasing need for national rice, so efforts are still needed to increase rice production.

Efforts to increase rice production can be achieved through intensification and extensification. Increasing rice production by extensification can be done using marginal lands, including coastal sandy land. The utilization of coastal sandy land to increase rice production is faced with several limiting factors, including low water storage ability, high infiltration and evaporation, very low fertility

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and organic matter content, and low water use efficiency. Wibisana et al., (2020) found that using filter cake compost at a dose of 5 tons ha^{-1} was more efficient, and it could provide efficiency of 0.097 tons per kg of cane at a dose of 76.76% inorganic fertilizer. Intensification efforts to increase soil fertility and rice yields in coastal sandy land include adding organic fertilizers, using microbial biotechnology in the form of biological fertilizers, and using superior cultivars tolerant to environmental stress (upland rice cv. Segreng Handayani). Kristamtini & Prajitno (2009) stated that the Segreng Handayani cultivar is one of the superior upland rice cultivars tolerant to water stress, but its production has only reached 3-4 tons/ha.

Organic fertilizers added to the coastal sandy land can improve the soil's physical, chemical, and biological properties. The role of organic matter in soil physical properties includes stimulating granulation, improving soil aeration, and increasing water retention capacity. The potential organic fertilizers in coastal sandy land include cow and chicken manure and Azolla compost. Mertikawati et al., (1999) and Hasibuan (2015) found that providing compost of cow manure, chicken manure, gliricidia leaves, and angsana leaves at a dose of 30 ton/ha could improve the physical properties (soil moisture, soil porosity, and bulk density) and chemical properties (soil pH, organic C, and soil organic matter) of soil. Ratnasari et al., (2020) found that organic matter significantly increased water availability in the soil, thereby supporting the growth and yield of upland rice. Yovita (2012) reported that chicken manure compost at a dose of 20 tons/hectare showed the best results on the growth and yield of sweet corn plants on peat soil. Meanwhile, the research by Kustiono et al., (2012) showed the application of Azolla at a dose of 6 tons/hectare compost on rice cv. Ciherang on Inceptisol soils produced 8.69 tons/hectare of grain.

Merapi indigenous rhizobacteria isolates have the potential to be used as biological fertilizers, especially for rice plants in fields with limited water. It is supported by the research of Agung Astuti et al., (2014a), reporting that rice plants inoculated with the Merapi indigenous rhizobacteria with a watering frequency of six days gave the same results as rice plants without inoculation with a daily watering frequency. Rhizobacteria isolates can produce growth hormones and osmoprotectants, increasing plant resistance to drought stress and fixing N from the air. Therefore, it is necessary to study the types and doses of compost to be added to the coastal sandy soil to improve soil fertility and increase the yield of rice inoculated with Merapi indigenous rhizobacteria isolates. Applying the inoculum of Merapi indigenous rhizobacteria is expected to reduce the use of organic fertilizer.

MATERIALS AND METHODS

Experimental design

This study used upland rice plants cv. Segreng Handayani (a local variety of Gunung Kidul), MB and MD isolates of Merapi indigenous rhizobacteria (collection of Ir. Agung Astuti, M.Si), LBA (Luria Bertani Agar) platting media, LBC (Luria Bertani Cair) isolate propagation media, cow manure compost, chicken manure compost, Azolla compost, coastal sandy soil for planting media. The study was a single-factor experiment, with types and doses of compost as treatments, arranged in a completely randomized design (CRD). The treatments applied were various types of compost with different doses on Segreng Handayani rice plants that were inoculated with Rhizobacteri indigenous Merapi in drought stress for two days, consisting of seven treatments and three replications. The seven treatments tested were cow manure compost at doses of 30 and 40 tons/ha, chicken manure compost at doses of 30 and 40 tons/ha, Azolla compost at doses of 20 and 30 tons/ha, and without compost (control). Each unit consisted of three plants for destructive sampling, three sample plants, and a substitute plant.

Rhizobacteria were identified and characterized according to Bergey's Manual of Determinative Bacteriology. Colony morphology observations include colony color, diameter, edge shape, internal structure, and elevation. Cell morphology observations include cell shape, gram properties, catalase, and aerobics.

Statistical Analysis

The data were analyzed using analysis of variance (ANOVA) at P≤0.05. The data were then subjected to Duncan's Multiple Range Test (DMRT) at P≤0.05 to determine the difference between treatments.

RESULTS AND DISCUSSION

Identification and characterization

Identification is carried out to ensure that the bacteria used are the same as the bacteria that have been determined. Identification of the indigenous rhizobacteria of Merapi includes colony and cell characterization. Colony characterization was carried out by culturing MB and MD isolates on the LBA medium using the surface platting method.

The identification and characterization of the MB and MD isolates of Merapi indigenous rhizobacteria follow the results of research by Agung Astuti (2016), reporting that the color of the isolates is white (MB) and creamy white (MD). The diameter is 0.4 cm (MB) and 1.4 cm (MD). Meanwhile, the colony form is circular (MB) and amuse (MD) with an edge shape of entire (MB) and filamentous (MD). The elevation is law convex (MB) and convex rugose (MD), and the inner structure is coarsely Granular (MB) and arborescent (MD). The cell's shape is bacillus (MB) ad coccus (MD). Both isolates are negative-Gram bacteria.

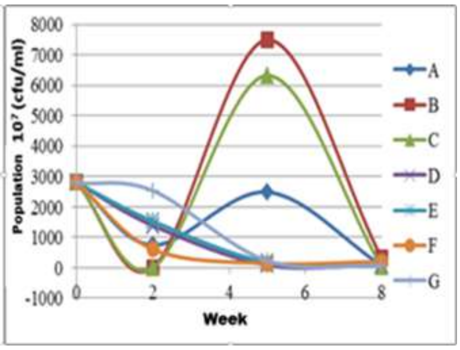
The MB and MD isolates of Merapi indigenous rhizobacteria were Gram-negative. It means that the indigenous rhizobacteria of Merapi can accumulate glycine betaine. Glycine betaine is a compound accumulated by Gram-negative bacteria under drought stress. According to Brock (1997), the characterization of rhizobacteria cells is Gram-negative with a diameter of 0.5 to 0.9 µm long 1.2 - 3.0 µm, without spores. It follows this study's gram characteristics of the Merapi indigenous rhizobacteria isolates.

The population dynamics of Merapi indigenous rhizobacteria

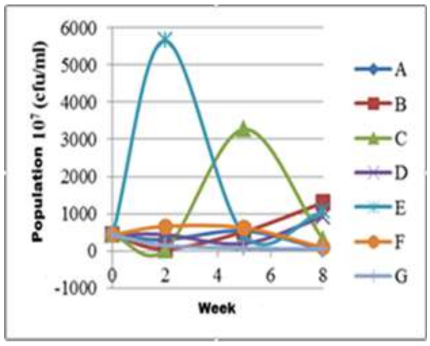
The population dynamics of Merapi indigenous rhizobacteria were observed in the starter mixture and nursery as well as at 2, 5, and 8 weeks after planting. The number of colonies was counted using the Total Plate Count (TPC) method. The population of Merapi indigenous rhizobacteria, both MB and MD isolates, increased from the time of the starter mixture to the nursery phase in the Greenhouse (Figure 1). It is assumed that the population of MB and MD isolates experienced a growth phase, and they were able to pass the environmental adaptation phase in the nursery.

The Merapi indigenous rhizobacteria in the starter mixture reached 48.33x107 CFU/ml (MB and MD). In the nursery planting media (in the Greenhouse), the MB isolate population increased to 2796x107 CFU/ml, and the MD isolates increased to 426.67x107 CFU / ml. Meanwhile, the population of other bacteria in the soil was 196x107 CFU/ml, so the total bacteria in the nursery was 3418.67x107 CFU/ml (Figure 1).

MD isolates had a faster adaptation ability compared to MB isolates. This result is consistent with the research of Agung_Astuti et al., (2014a), reporting that the development of MD isolates in the first



(a)



(b)

Figure 1. (a) MB isolates and (b) MD isolates of Merapi indigenous *Rhizobacteria* inoculated in rice plants cv. Segreng Handayani. A: Cow manure compost 30 ton/ha, B: Cow manure compost 40 ton/ha, C: Chicken manure compost 30 ton/ha, D: Chicken manure compost 40 ton/ha, E: Azolla compost 20 ton/ha, F: Azolla compost 30 ton/ha, G: Control (without compost/only inoculated with indigenous rhizobacteria).

week was faster than that of MB isolates. It proves that MD isolates can adapt to week 2, and the population decreases until week 8. The population growth of MD isolates in treating Azolla compost at 20 tons/ha was very good initially. Meanwhile, the population growth in the treatment of chicken manure compost at 30 tons/ha was very good in the end, but the growth of the isolates was slow. In the treatment of cow manure 30 and 40 tons/ha, the growth of MD isolates was low until the 5th week, then increased, while in Azolla compost, 30 tons/ha, the growth of isolates was prolonged and did not even develop.

From week 2 to week 5, the population of Merapi indigenous rhizobacteria experienced a log phase. The increase in the population of MB isolates is supported by the research of Agung Astuti et al., (2014b), mentioning that the MB and MD isolate experienced development from weeks 4 to 6.

The population of Merapi indigenous rhizobacteria in the treatment of Azolla 20 ton/ha experienced exponential growth from week 0 to week two so that it could pass the adaptation phase. This increase was dominated by the MD isolate colony of Merapi indigenous rhizobacteria, which was 5656.67x107 CFU/ml (Figure 1b).

In the span of week 2 to week 5, the MB isolates experienced a log phase in the treatment of cow manure compost at 30 and 40 tons/ha, while in the treatment of 30 tons/ha of chicken manure compost, both MB and MD isolate experienced a log phase (Figure 1a and 1b). Thus, MD isolates had a faster adaptation ability compared to MB isolates.

The root growth of rice plants cv. Segreng Handayani

Roots are important in supporting plants to grow upright and absorb nutrients and water for plant metabolic processes. Root growth is influenced by environmental conditions, such as soil texture and type, air, and soil cultivation (Gardner et al., 1991).

There was a significant effect of rhizobacteria inoculation with the addition of various types and doses of compost on the proliferation, length, fresh weight, and dry weight of the roots of rice plants cv. Segreng Handayani was cultivated on coastal sandy soil under drought stress at week 5 with each P-value of 0.0032, 0.0203, 0.0003, and 0.0040, respectively (Table 1), but not at week 8 (Table 2) with a P-value higher than 0.05. The application of cow manure compost at 30 ton/ha and 40 ton/ha, Azola compost at 20 ton/ha, and without compost (only inoculation of MB and MD isolates) under drought stress resulted in the higher proliferation and longer roots compared to other treatments at

Table 1. Effect of types and doses of compost on the root proliferation, root length, root fresh weight, and root dry weight at week 5

Treatments	Root proliferation (+)	Root length (cm)	Root fresh weight (g)	Root dry weight (g)
A	3.00 a	46.00 a	42.48 a	7.16 a
B	2.67 a	45.00 ab	25.80 a	4.46 ab
C	1.00 c	22.63 bcd	0.75 b	0.30 c
D	1.33 bc	20.83 cd	2.19 b	0.60 c
E	2.33 ab	33.33 abcd	9.15 b	2.13 bc
F	1.00 c	11.67 d	0.25 b	0.11 c
G	3.00 a	39.97 abc	26.24 a	5.69 ab

Remarks: Means followed by the same letters in the same column are not significantly different according to DMRT at $P \leq 0.05$. A: Cow manure compost 30 ton/ha, B: Cow manure compost 40 ton/ha, C: Chicken manure compost 30 ton/ha, D: Chicken manure compost 40 ton/ha, E: Azolla compost 20 ton/ha, F: Azolla compost 30 ton/ha, G: Control (without compost/only inoculated with indigenous rhizobacteria).

Table 2. Effect of types and doses of compost on the root proliferation, root length, root fresh weight, and root dry weight at week 8

Treatments	Root proliferation (+)	Root length (cm)	Root fresh weight (g)	Root dry weight (g)
A	4.00 a	55.33 a	55.41 a	8.70 a
B	3.67 a	44.00 a	49.37 a	7.35 a
C	3.33 a	37.17 a	52.24 a	7.12 a
D	3.67 a	47.33 a	50.54 a	6.91 a
E	3.00 a	35.67 a	31.75 a	4.17 a
F	3.00 a	34.00 a	34.74 a	5.50 a
G	4.00 a	39.50 a	52.55 a	7.02 a

Remarks: Means followed by the same letters in the same column are not significantly different according to ANOVA at $P \leq 0.05$. A: Cow manure compost 30 ton/ha, B: Cow manure compost 40 ton/ha, C: Chicken manure compost 30 ton/ha, D: Chicken manure compost 40 ton/ha, E: Azolla compost 20 ton/ha, F: Azolla compost 30 ton/ha, G: Control (without compost/only inoculated with indigenous rhizobacteria).

week five. Meanwhile, the application of cow manure compost at both doses and without compost resulted in higher fresh and dry weight of the roots.

The application of cow manure compost at 30 and 40 tons/ha, Azolla compost at 20 tons/ha, and without compost significantly increased the proliferation, length, and fresh and dry weight of roots. It is suspected that mixed MB + MD isolates can stimulate root development with these treatments, thereby increasing soil fertility and improving plant growth. In addition, the development of root proliferation is also influenced by IAA produced by rhizobacteria in the roots. According to Agung Astuti (2014 a), MB isolates have a strong ability to break down NH_4^+ to Nitrite (NO_2^-) or Nitrate (NO_3^-) and to break down organic or inorganic N into ammonia, besides also having resistance to very high osmotic pressure (NaCl> 2.75M). Meanwhile, MD isolate is very strong in dissolving phosphate and resistant to osmotic pressure (NaCl> 2.75M). However, at week 8, the roots' proliferation, length, and fresh and dry weight were the same in all treatments. This result is because, from weeks 5 to 8, there was an increase in the population of rhizobacteria, especially MD isolates. Chavez et al., (2019) 's research results stated that soil fertility strongly influenced plant microbiota.

The shoot growth of rice plants cv. Segreng Handayani

Plants experience biomass growth in forming their body parts. Plant biomass includes all plant materials from photosynthesis (Sitompul and Guritno, 1995 in Apriyanti, 2007). The effect of

Table 3. Effect of types and doses of compost on the plant height, shoot fresh weight, shoot dry weight at week 5 and number of tillers

Treatments	Plant height (cm)	Shoot fresh weight (g)	Shoot dry weight (g)	Number of tillers
A	51.81 a	44.32 a	9.86 a	23.78 a
B	50.22 a	31.68 ab	7.10 a	18.78 a
C	45.33 ab	4.73 c	1.37 c	14.33 a
D	39.39 bc	6.23 c	1.42 c	16.17 a
E	50.53 a	18.38 bc	4.21 bc	18.55 a
F	29.95 c	1.28 c	0.46 c	6.56 a
G	52.48 a	40.61 a	10.32 a	18.56 a

Remarks: Means followed by the same letters in the same column are not significantly different according to DMRT at P≤0.05. A: Cow manure compost 30 ton/ha, B: Cow manure compost 40 ton/ha, C: Chicken manure compost 30 ton/ha, D: Chicken manure compost 40 ton/ha, E: Azolla compost 20 ton/ha, F: Azolla compost 30 ton/ha, G: Control (without compost/only inoculated with indigenous rhizobacteria).

Table 4. Effect of types and doses of compost on the plant height, shoot fresh weight, shoot dry weight at week 8, number of tillers, and anthesis period

Treatments	Plant height (cm)	Shoot fresh weight (g)	Shoot dry weight (g)	Number of tillers	Anthesis period (days)
A	65.49 a	100.69 a	22.77 a	29.67 a	63.67 cd
B	55.72 ab	84.16 a	18.42 a	24.22 a	69.33 bc
C	53.59 ab	84.40 a	18.28 a	26.11 a	71.00 b
D	48.17 bc	90.08 a	19.75 a	23.11 a	70.50 b
E	63.16 ab	53.18 a	11.66 a	31.11 a	68.33 bcd
F	37.03 c	54.71 a	11.35 a	9.00 a	77.00 a
G	62.56 ab	85.56 a	18.56 a	22.77 a	63.00 d

Remarks: Means followed by the same letters in the same column are not significantly different according to DMRT at P≤0.05. A: Cow manure compost 30 ton/ha, B: Cow manure compost 40 ton/ha, C: Chicken manure compost 30 ton/ha, D: Chicken manure compost 40 ton/ha, E: Azolla compost 20 ton/ha, F: Azolla compost 30 ton/ha, G: Control (without compost/only inoculated with indigenous rhizobacteria).

rhizobacteria inoculation with the addition of various types and doses of compost on plant height was in line with the effect on the root development (proliferation, root length, fresh root weight, and root dry weight) because the better the root development, the more water and nutrient absorbed, thereby resulting in optimum plant growth. The nutrients in cow manure compost at 30 and 40 tons/ha can increase the biomass weight of the rice plants. Besides, the treatment can provide energy and nitrogen for the indigenous rhizobacteria to fertilize the plants. According to Rao (1994), the roots of rice plants can provide exudates in the form of organic compounds needed for soil microorganisms.

There was a significant effect of rhizobacteria inoculation with the addition of various types and doses of compost on the plant height and the shoot fresh and dry weight at week 5, but no significant effect on the number of tillers (Table 3). The application of cow manure compost at 30 and 40 tons/ha, chicken manure compost at 30 tons/ha, Azolla compost at 20 tons/ha, and control (without compost) produced higher plants compared to the application of chicken manure compost at 40 ton/ha and Azolla compost at 30 ton/ha. The shoot fresh and dry weight of plants treated with cow manure compost at 30 and 40 tons/ha and of control (without compost) plants were significantly higher than those treated with other treatments. Meanwhile, at week 8, the application of cow manure compost at 30 and 40 tons/ha, chicken manure compost at 30 tons/ha, Azolla compost at 20 tons/ha, and control produced significantly higher plants compared to the application of chicken manure and Azolla compost at 40 and 30 ton/ha, respectively (Table 4).

Conversely, the treatments had no significant effect on the fresh and dry weight of the shoots at week 8. This result is in line with the development of the roots that were not significantly affected by the treatments because shoot growth is influenced by the root's ability to absorb water and nutrients for growth. The results of the research by Khalimi et al., (2014) also showed that the application of PGPR could stimulate plant growth and increase plant resistance to pathogenic fungi.

There was no significant effect of the inoculation of rhizobacteria with the addition of various types and doses of compost on the number of tillers in week 5 (Table 3) as well as at week 8 (Table 4). However, all treatments resulted in a higher number of tillers compared to the description of the cultivar (9 - 11) due to the role of the indigenous rhizobacteria that can produce phytohormones such as IAA. Organic materials from cow manure, chicken manure, and Azolla compost can be used as nutrients that help the growth of indigenous rhizobacteria in the roots so that they can fertilize plants. Research by Agung Astuti et al., (2014b) reported that inoculation of Merapi indigenous rhizobacteria (MB + MD isolates) produced a higher number of tillers, reaching 12-16 tillers, compared to those without inoculation, producing only 9.27 tillers (Utami et al., 2009).

There was no significant effect of the inoculation of rhizobacteria with the addition of various types and doses of compost on the anthesis period (Table 4). The control treatment resulted in an earlier anthesis period. However, it was not different from the application of cow manure compost at 30 tons/ha (63.67 days) and Azolla compost at 20 tons/ha (68.33 days). This result is because the different types of compost have different levels of ability to bind water, especially in coastal sandy soil, which has a high porosity. Hence, the soil is prone to drought. Drought can affect morphology, physiology, and activities at the molecular level of rice plants, such as delayed anthesis, reduced distribution and allocation of dry matter, reduced photosynthetic capacity due to stomata closure, restriction in metabolism, and damage to chloroplasts (Farooq et al., 2009).

The yield of rice plants cv. Segreng Handayani

The productivity of rice plants is affected by the interaction between genetic factors and the environment (Yoshida, 1981). According to the Analysis of Variance, there was no significant effect of the inoculation of rhizobacteria with the addition of various types and doses of compost on the harvesting age, the number of panicles and weight of seeds per hill, the weight of 1000 seeds, and the grain yield of rice plants cv. Segreng Handayani in coastal sandy soil under drought stress (Table 5) with a P value> 0.05. However, the application of cow manure compost at 30 tons/ha, Azolla compost at 20 tons/ha, and control tended to result in earlier harvesting age, a higher number of panicles and weight of seeds per hill, and higher grain yield compared to other treatments. The application of cow manure compost at 30 tons/ha resulted in a higher number of panicles per hill (29.11), although it was not different compared to other treatments (Table 5). This result is in line with the anthesis period in the application of cow manure compost at 30 tons/ha and control (without compost), which was earlier than other treatments. Besides, the root growth (root proliferation, root length, and fresh and dry weight of roots) and shoot growth (plant height and fresh and dry weight) at week 5 were higher. According to Purwaningsih and Kristamtini (2009), the harvesting age of rice plants cv. Segreng Handayani is 109 days after planting. It is suspected that the application of cow manure compost at 30 tons/ha can improve growth because the nu-

Table 5. Effect of types and doses of compost on the harvest age, number of panicles per hill, the weight of seeds per hill, the weight of 1000 seeds, and grain yield (ton/ha)

Treatments	Yield				
	Harvest age (days)	Number of panicles per hill	Weight of seeds per hill (g)	Weight of 1000 seeds (g)	Grain yield (ton/ha)
A	104.67 a	29.11 a	17.46 a	20.65 a	4.25 a
B	107.33 a	24.33 a	14.90 a	17.03 a	3.63 a
C	109.00 a	23.72 a	15.59 a	18.01 a	3.80 a
D	107.00 a	25.34 a	15.59 a	20.50 a	3.90 a
E	107.33 a	27.83 a	17.21 a	18.64 a	4.19 a
F	111.00 a	22.50 a	8.38 a	17.64 a	2.04 a
G	104.67 a	27.89 a	16.93 a	20.50 a	4.13 a

Remarks: Means followed by the same letters in the same column are not significantly different according to ANOVA at $P \leq 0.05$. A: Cow manure compost 30 ton/ha, B: Cow manure compost 40 ton/ha, C: Chicken manure compost 30 ton/ha, D: Chicken manure compost 40 ton/ha, E: Azolla compost 20 ton/ha, F: Azolla compost 30 ton/ha, G: Control (without compost/only inoculated with indigenous rhizobacteria).

trients are absorbed optimally, thereby providing sufficient energy and nitrogen for the indigenous rhizobacteria of Merapi so that the plants can be harvested earlier.

Meanwhile, the inoculation of Merapi indigenous rhizobacteria with the addition of cow manure and Azolla compost at 30 and 20 tons/ha, respectively, and without the addition of compost resulted in a higher seed weight per hill, the weight of 1000 seeds and grain yield compared to other treatments. Merapi indigenous rhizobacteria can produce NO_3^- and NH_4^+ ions through a mineralization process to form complex materials, such as amino acids and nucleic acids, that plants can directly absorb and use. In addition, Merapi indigenous rhizobacteria can fertilize plants because they can produce IAA to become biological fertilizers for plant growth (Agung Astuti, 2014a). This result is also supported by the research results of Chaves et al., (2019), stating that of the 41 strains of rhizobacteria studied, 86% can produce IAA, and only 14% are high phosphorus solubilizing bacteria. This is in line with research results by Tuhuteru et al., (2019), mentioning that PGPR isolates BrSG.5 (*Burkholderia seminali)s* tested could produce IAA 41.41 mg kg-1, isolate BP25.2 (*Bacillus methylotrophicus*) was effective at producing N (0.05%), while isolate BP25.7 (*Bacillus subtilis*) was effective at producing residue P (0.22 ppm).

Overall, the effects of the inoculation of Merapi indigenous rhizobacteria added with various types and doses of compost on rice plants cv. Segreng Handayani cultivated on coastal sand soil under drought stress were observed on the root proliferation, root length, root fresh and dry weight, plant height, shoot fresh and dry weight, and anthesis period. The application of cow manure compost at 30 tons/ha and control (without compost) resulted in better responses than other treatments. Meanwhile, the treatments had no significant effect on the grain yield. Nevertheless, the application of cow manure and Azolla at 30 and 20 tons/ha, respectively, and control (without compost) tended to produce higher grain yield than other treatments. It is suspected that the application of cow manure compost at 30 tons/ha can improve vegetative and generative growth more quickly because nutrients are absorbed optimally, thereby providing energy and nitrogen for the indigenous rhizobacteria of Merapi so that the photosynthesis process runs optimally. In addition, the indigenous rhizobacteria of Merapi can fertilize plants because they can produce IAA. According to Agung Astuti (2014a), rhizobacteria isolates have the potential to be used as biological fertilizers. It can be seen from their ability to produce growth hormones and osmoprotectants that can increase plant resistance to drought stress and fix N from the air. The application of Azolla compost at higher doses tends to enhance the denitrification process. Denitrification is the process of reducing nitrogenous oxides, especially nitrite and nitrate, to nitrogen, N_2O, and N_2 (Tiedje, 1988 in Picone et al., 2014). Besides, the research site's environmental temperature (volatilization) is quite high, thereby releasing N and ammonium in the soil into the air as gases. Volatilization is the change of ammonium to ammonia gas, and this process occurs mostly in soils with a pH greater than 7.5 with a sand texture (Budiyanto, 2009). Control treatment (without compost) was found to give the same effect as the application of cow manure compost at 30 ton/ha. The coastal sandy soil used in this research may already contain sufficient organic matter for developing rhizobacteria to develop and play a role in increasing soil fertility, growth, and yield of rice plants. This is in line with the research results of Ningrum et al., (2017), reporting that the combination of treatments without composting rabbit manure and 30 ml of PGPR were able to replace the combination treatment of 10 tons ha-1 of rabbit manure compost and 20 ml on the yield of cobs per hectare.

CONCLUSION

Applying 30 tons/ha of cow manure compost to rice plants inoculated with Merapi indigenous rhizobacteria isolates MB+MD in coastal sandy soil gave the best growth at week five and was not significantly different from that without compost. The provision of various types and doses of compost did not significantly affect the rice yield inoculated by Merapi indigenous rhizobacteria in coastal sandy soil. The inoculation of the indigenous rhizobacteria Merapi can substitute the application of compost in rice cultivation on coastal sandy soil under drought stress.

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