

Article

Alternatives for Circular Bioeconomy in Organic Farming under Excessive Nutrients (Goat manure and Arbuscular Mycorrhizal Fungi): A Case Study in Indonesia

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Abstract: A case study in Indonesia of circular bioeconomy implementation was investigated by managing livestock wastes, especially goat manure (GM), which an excess of its availability may be adverse to the environment. The efficacy of this scenario to control pollution or to increase productivity still needs to be proven. Hence, this research aimed to study the possibility of circular bioeconomy implementation using biotic and abiotic resources in Indonesia under excessive nutrients (GM and mycorrhizal) on *P. angulata* production. Outdoor factorial container experiment was carried out using a randomized complete block design in Central Java, Indonesia. Treatments included four levels of GM (0, 10, 20, 30 g plant⁻¹) and four levels of mycorrhizal (0, 10, 20, 30 g plant⁻¹) applied in the soil with six replications. This case study revealed that the use of mycorrhizal inoculant and GM indicated no significant difference to most of *P. angulata*'s growth and yield parameters. The implementation of circular bioeconomy through integrated farming of *P. angulata* was not an instant solution for economic and environmental optimization, but can be considered as a way to tackle environmental problem due to the excessive livestock wastes. The environmental sustainability can be achieved step by step, without hindering farmers' income.

Keywords: sustainable agriculture; *Physalis angulata*; integrated farming; phosphorus content



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1. Introduction

Indonesia has potential in developing agricultural sector since it has huge areas of arable farming land. As a tropical country, Indonesia is located at the edge of the Pacific Rim, which makes it a site of numerous volcanoes. With its vast and abundant fertile soils, Indonesia is a major producer of a wide range of tropical agricultural products around the world [1]. However, the agriculture systems implemented in Indonesia mostly are traditional or conventional system based on local wisdom which caused many problems, including environmental issues. For instance, farmers usually have livestock animals [2], especially rearing goats, but the waste is poorly managed. Nevertheless, in reality, the management of livestock waste is necessary to avoid harmful effects on the environment [3]. Livestock rearing should be implemented as an integrated farming system which is efficient and eco-friendly. Recent data given by Indonesian Statistical Center Bureau shows that there is increasing population of livestock, especially goat from 13.6 million in 2013 to 15.3 million in 2020. If each goat could produce 2–5 kg feces per a day, it is estimated that approximately 30.6–76.5 million tons of goat manure production per year is produced. The amount of livestock waste will rise drastically if it was added by the other livestock,

including cattle, cows, sheep, poultry, etc. These animal wastes emit bad smells to the environment, which together with the environment of the micro planes provides a suitable environment to cause various diseases [4].

The excessive nutrients from goat livestock waste can be used as a fertilizer to support plant growth and yield, especially for *Physalis angulata*. This species is often characterized as an invasive weed of crops, especially a host of the causal agent of tomato bacterial spot disease *Xanthomonas campestris* pv. *Vesicatoria*, but its fruit has attracted renewed interest and used as a medicinal plant due to its bioactive compounds. Rengifo and Vargas [5] revealed that the plant contained flavonoids, alkaloids, and many different types of plant steroids which can be used for antitumor, anti-melanoma, antimalarial, anti-inflammatory, anticancer, and antibacterial.

Goat manure has great potential as a source of organic fertilizer because it contains beneficial nutrients to improve soil fertility. Organic fertilizer improved the soil nutrient status and shaped distinct bacterial communities [6]. It encourages agro-ecosystem health which is used to reduce the environmental hazard of chemical fertilizers used continuously. Solid goat manure contains nutrients as shown in Table 1 [7].

Table 1. Characteristics of goat manure (GM).

Parameters	Concentration
pH	7.9 ± 0.1
N (%-TS)	2.8 ± 0.1
C (%-TS)	43.9 ± 0.3
NO ₃ ⁻ -N (mg/L)	12.7 ± 1.1
NH ₄ ⁺ -N (mg/L)	398.0 ± 9.9
PO ₄ ³⁻ (mg/L)	1230 ± 0.4

All values except, nitrogen and carbon are percentage of total wet sample weight (mean value, $n = 3$). Triplicates of 1.0 g of each feedstock (wet weight w/w) were diluted and multiplied by 10 and reported as mean ± SD.

The utilization of goat livestock waste also can be combined by the use of arbuscular mycorrhizal fungi (AMF) to provide extra nutrients to the plants. Mycorrhizal fungi is one of natural bio-fertilizer which can associate with majority of plants under natural conditions. Ninety percent of land plant species are colonized by one or more of the mycorrhizal fungi species ranging from flowering to non-flowering plants, while only a few plant families do not form this association [8]. Bagy et al. [9] reported that AMF has potential to promote plant growth and decrease the disease severity which could be used as a biocontrol agent to provide competitive economic outcomes for sustainable crop protection systems.

The developing organic farming system using goat manure and AMF in *P. angulata* production can be considered as an alternative way to handle environmental pollution caused by excessive livestock wastes. This can be considered as an implementation of circular bioeconomy in farming activities, especially in a case study of Indonesia. Rearing animals and doing farming will be more beneficial under this circular paradigm to increase farmer's income and preserve the environment (Figure 1).

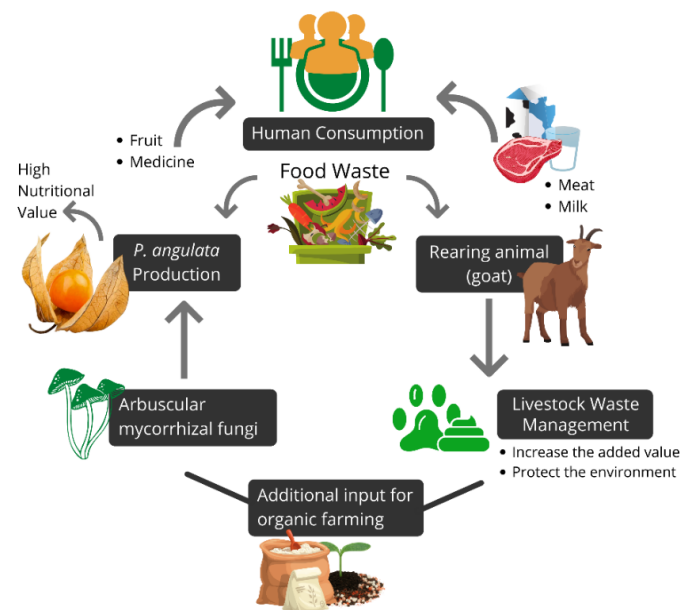


Figure 1. The concept of circular bioeconomy in organic farming. Production of *P. angulata* under excessive nutrients of goat manures and AMF.

Furthermore, research showed that organic farming has another advantage to the plants which can improve phytochemical and antiradical properties compared to conventional farming system. Based on Khalid et al. [10], the phenolic acids and flavonoids contents are considerably high in organic fertilizer and fungi followed by organic fertilizer, biochar, and fungi treated plants.

In terms of production aspect, usage of organic fertilizer for cultivation of *P. angulata* had shown different results. Mycorrhization of bamboo plantlets and the application of organic manure in small doses have cumulative positive effects on plant growth [11]. In contrast, there was no interaction between application of chicken, goat, and cow manure with the level of arbuscular mycorrhizal fungi dosage on the growth and yield of the young ginger [12]. However, in terms of environmental aspect, the use of organic fertilizer produced by livestock wastes, especially goat manure, for farming activity needs to be proven the effectiveness to control the pollution which indirectly may increase the productivity of the plant. Therefore, this research aimed to study the possibility of implementing circular bioeconomy in Indonesia using excessive nutrients (goat manure and mycorrhizal) on *P. angulata* production.

2. Materials and Methods

2.1. Experimental Design and Treatments

An outdoor container experiment was conducted in the lowlands (around 100 m above sea level) at the experimental field of Nahdlatul Ulama University, Central Java, Indonesia from July to October 2020. *P. angulata* seeds used were obtained from the rice field around the experimental area, while AMF was derived from Laboratory for Biotechnology, Agency for the Assessment and Application of Technology (BPPT) containing *Gigaspora* sp., *Glomus* sp. dan *Acaulospora* sp. (spore density was 400 spores per 10 gr carrier from zeolite, while *Sorghum bicolor* and *Pueraria javanica* roots were the hosts). Goat manure used was fresh manures that were left for one month and matured naturally. The study was laid out in a 4×4 factorial arrangement in randomized complete block design to minimize the effect of land contour and sunlight intensity. The treatments were replicated six times, so there were 96 plants (containers) in total based on the varying levels of mycorrhizal and goat manure served as the first and the second factor ($0, 10, 20, 30 \text{ g plant}^{-1}$).

2.2. Cultural Management and Practices

The size of the polybag used was 30 × 40 cm or equal to 6 kg of air-dry soil. The soil used was inceptisols derived from the experimental area which was sterilized by solarization for 3 days. The polybag was kept on the field with 0.5 m distance between plants within a row and 1 m between blocks. Seed dressing was done together with treatment application. Thinning was performed at germination completion (21 days after sowing, the plants had approximately 4–5 cm high and bearing 2–3 pairs of leaves) to maintain one plant per polybag throughout the study.

The cultural practices consisted of manual watering in the absence of rain to keep the soil at its field capacity and weed control, mainly at early stages of development. Biopesticide derived from neem oil was applied throughout the study to control pests. The fruits were collected when they were greenish-yellow and ripe for harvest started from 10 to 12 weeks after sowing.

2.3. Data Gathering and Analysis

2.3.1. Collection and Characterization of Soil Samples

The experimental area was subjected to soil analysis, which was done before and after the study. Soil analysis was performed at Analyses Laboratory, Department of Agronomy and Horticulture, Faculty of Agriculture, IPB University. Soil analysis included soil pH H₂O using pH meter, water content (%) using gravimetric method, percentage (%) of organic carbon using Walkley-Black method and spectrophotometer UV-VIS, total nitrogen using Kjeldahl method and titrimetry, available phosphorus (ppm P₂O₅) using the Olsen method and spectrophotometer UV-VIS, and exchangeable potassium (cmol K/kg) using NH₄OAc pH 7.0 for extraction method and AAS.

2.3.2. Growth and Yield Components

Growth was assessed at four weeks after sowing. The following characteristics were assessed: plant height, leaf number, branch number, and stem diameter. The yield parameters observed were: total fruit weight per plant, fruit number per plant, fruit diameter, fruit weight without calyx, and fruit weight with calyx. Then, the plant materials were packed in paper bags, separating the shoot and root, then dried in the oven at a temperature of 60 °C until the weight was constant to determine shoot dry mass. The percentage of AMF infection on plant roots was determined using clearing and staining method; roots cut were washed and heated in 10% KOH solution at 90 °C for 10 min, then rinsed with 1N HCl 1 and distilled water. A modified procedure for staining of AMF was applied using blue ink as a substitute of Trypan blue.

2.3.3. Nutrient Content in Goat Manure and P Content in Plants

Goat manure sample was analyzed to determine the water content (%), pH, percentage (%) of organic carbon (gravimetric analysis), percentage of N total using Kjeldahl method, P₂O₅ using Spectrophotometer UV-VIS, and K₂O using AAS. Then, total phosphorus content in plants (%) was measured using wet ashing method with HNO₃ 65% + HClO₄ 60% for digestion process and determined by spectrophotometer UV-VIS.

2.3.4. Data Analysis

The data were analyzed using analysis of variance (ANOVA) in a factorial experiment on randomized complete block design (RCBD). Significant differences among treatment means were determined through Duncan Multiple Range Test ($p < 0.05$).

3. Results

3.1. Soil and Goat Manure Characteristics

The results of the analysis revealed that the initial soil pH was 6.31, with 2.28% organic carbon, 0.24% total nitrogen, 124.25 ppm available P, and 0.28 cmol K/kg exchangeable K. These indicated that the soil pH was moderately acidic, moderate in organic carbon and

total nitrogen, very high in P, and low in K. The analysis of soil samples after harvesting resulted in 2.10% organic carbon, 0.21% total nitrogen, 112.60 ppm available P, and 0.20 cmol K/kg exchangeable K. The pH also changed to 6.56.

After the addition of AMF and GM, the condition of the soil remained stable which showed nutrient balance in the soil. The slight decrease was due to the use of nutrients by plants. The plants might use extra nutrients from goat manure, i.e., 31.57% the organic carbon, 2.71% N total, 1.72% P₂O₅, and 3.40% K₂O. The pH of goat manure was 8.12.

3.2. Growth Parameters

To define the effects of organic fertilizer utilization on the growth of *P. angulata*, the observation was done six weeks after sowing. Mycorrhiza treatment showed no effect on growth parameters of *P. angulata*. The growth of *P. angulata* were similar with presence or absence of mycorrhiza. However, the application of 30 g mycorrhiza tended to increase all growth parameters (Table 2). Basically, the AMF inoculation was not related to plant growth, but it also depends on the plant and soil condition because AMF play a role as facilitators in the absorption of nutrients. In the study of Liu et al. [13], strong growth response to AMF inoculation might be related to the larger net photosynthesis and leaf area, apparently resulting from a higher plant P and N uptake. However, goat manure application has increased some of the growth parameters. Plant height and stem diameter were significantly higher at P2 and P3 level, when compared to the control (P0).

Table 2. The growth of *P. angulata* using AMF and GM.

Mycorrhizal (M)	Plant Height (cm)	Leaf Number	Branch Number	Stem Diameter (cm)
M0 (no mva)	33.54 ± 5.655 a	91.88 ± 29.320 a	21.08 ± 4.422 a	0.70 ± 0.114 a
M1 (10 g)	33.83 ± 6.093 a	87.13 ± 24.004 a	19.96 ± 3.850 a	0.69 ± 0.111 a
M2 (20 g)	34.52 ± 4.652 a	90.42 ± 27.335 a	21.13 ± 2.968 a	0.70 ± 0.083 a
M3 (30 g)	36.84 ± 5.714 a	101.08 ± 24.484 a	21.96 ± 2.331 a	0.75 ± 0.108 a
Goat manure (P)				
P0 (no manure)	32.04 ± 4.783 b	86.92 ± 17.257 b	20.13 ± 2.659 a	0.66 ± 0.081 b
P1 (10 g)	34.33 ± 4.838 ab	89.67 ± 27.466 b	21.21 ± 3.934 a	0.69 ± 0.108 ab
P2 (20 g)	35.90 ± 6.687 a	91.13 ± 29.746 b	20.88 ± 4.246 a	0.73 ± 0.112 a
P3 (30 g)	36.44 ± 5.216 a	102.79 ± 28.346 a	21.92 ± 2.903 a	0.75 ± 0.105 a
M × P	ns	ns	ns	ns

Data are presented as the mean ± standard deviation (STDEV, $n = 6$). Different letters within column indicate significance at $p < 0.05$ according to Duncan's test.

P. angulata treated with mycorrhizal and goat manure grew significantly week by week (Figure 2). However, there was no interaction between the two factors. Statistically, the application of mycorrhiza and goat manure also revealed no effect on plant height of *P. angulata*. Kapoulas et al. [14] stated that AMF inoculation should have great potential in enhancing the growth and yield even in high soil P, but Zhang et al. [15] said that AM colonization and plant growth significantly increased at low organic fertilizer rate and decreased or had no significant differences at the highest fertilizer rate compared to the uninoculated plants. The complexity and interaction of plant genotypes and AMF, the method and time of inoculation, the production system, and environmental conditions may show different results [14].

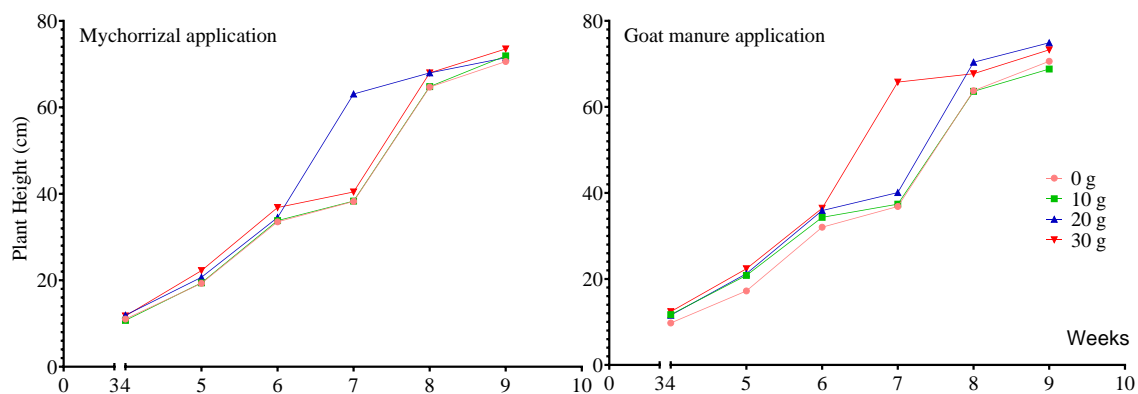


Figure 2. Height of *P. angulata* (cm) during the experiment.

3.3. Yield Parameters

The yield parameters showed that *P. angulata* was affected neither by the varying rates of mycorrhizal nor goat manure, except for total fruit weight per plant (Table 3). The highest rate of GM (P3) resulted in the highest total fruit weight showing similar tendency with figures of fruit number per plant. Statistical analysis also revealed no significant difference in the interaction of the two factors. Nutrient adequacy in the soil has supported *P. angulata* growth and yield.

Table 3. The yield of *P. angulata* using AMF and GM.

Mychorriza (M)	Fruit Number per Plant	Fruit Diameter (mm)	Fruit Weight without Calyx (g)	Fruit Weight with Calyx (g)	Total Fruit Weight per Plant (g)
M0 (no mva)	20.21 ± 5.985 a	11.29 ± 0.761 a	0.93 ± 0.164 a	1.03 ± 0.180 a	18.19 ± 5.988 a
M1 (10 g)	22.08 ± 5.867 a	11.24 ± 0.924 a	0.94 ± 0.190 a	1.06 ± 0.224 a	20.63 ± 6.006 a
M2 (20 g)	19.79 ± 6.627 a	11.31 ± 0.565 a	0.93 ± 0.103 a	1.05 ± 0.209 a	18.32 ± 6.281 a
M3 (30 g)	22.96 ± 8.800 a	11.30 ± 0.545 a	0.93 ± 0.133 a	1.04 ± 0.139 a	21.43 ± 9.208 a
Goat manure (P)					
P0 (no manure)	21.21 ± 5.816 a	11.11 ± 0.668 a	0.90 ± 0.146 a	1.03 ± 0.240 a	19.15 ± 5.760 ab
P1 (10 g)	19.54 ± 7.366 a	11.21 ± 0.589 a	0.89 ± 0.123 a	0.99 ± 0.137 a	17.48 ± 6.940 b
P2 (20 g)	20.58 ± 5.030 a	11.38 ± 0.795 a	0.95 ± 0.152 a	1.08 ± 0.192 a	19.42 ± 5.094 ab
P3 (30 g)	23.71 ± 8.690 a	11.44 ± 0.742 a	0.99 ± 0.159 a	1.08 ± 0.168 a	23.23 ± 8.772 a
M × P	ns	ns	ns	ns	ns

Data are presented as the mean ± standard deviation (STDEV, $n = 6$). Different letters within column indicate significance at $p < 0.05$ according to Duncan's test.

Similar to growth parameters, the application of 30 g goat manure also showed the best result on the total fruit weight per plant, although there was no significant difference between the application of 20 g goat manure and without manure (Table 3).

3.4. Biomass Production

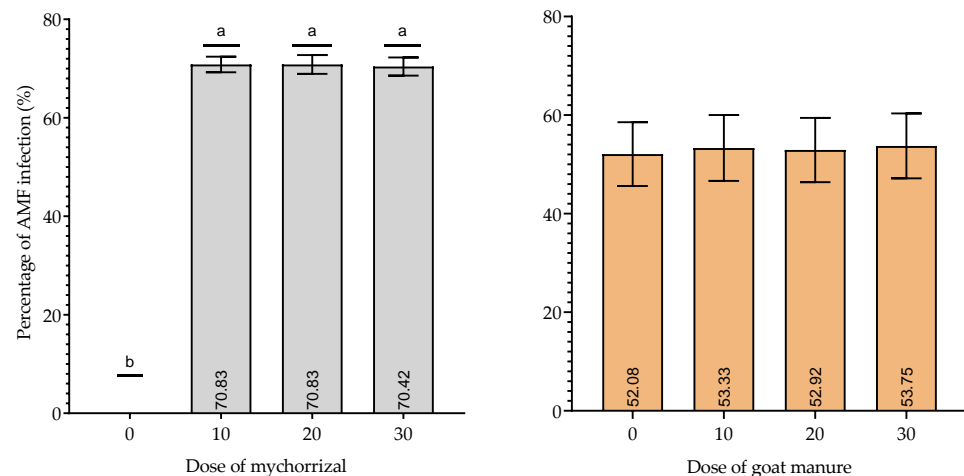
The application of goat manure did not affect all parameters of fresh and dried weight per plant, but there was a significant difference on shoot dried weight of plant using mycorrhiza (Table 4).

Table 4. Biomass production of *P. angulata* using AMF and GM.

Mychorriza (M)	Shoot Fresh Weight per Plant (g)	Shoot Dried Weight per Plant (g)	Root Fresh Weight per Plant (g)	Root Dried Weight per Plant (g)
M0	19.66 ± 4.694 a	2.73 ± 0.522 b	5.68 ± 2.706 a	0.77 ± 0.248 a
M1	19.65 ± 6.301 a	2.83 ± 0.581 ab	6.20 ± 2.744 a	0.84 ± 0.302 a
M2	16.40 ± 5.965 a	2.6 ± 0.482 b	5.53 ± 2.587 a	0.79 ± 0.296 a
M3	19.85 ± 6.831 a	3.06 ± 0.732 a	6.10 ± 2.688 a	0.90 ± 0.397 a
Goat manure (P)				
P0	17.76 ± 4.543 a	2.66 ± 0.555 a	5.34 ± 2.452 a	0.76 ± 0.267 a
P1	17.66 ± 5.247 a	2.65 ± 0.493 a	5.33 ± 2.438 a	0.78 ± 0.333 a
P2	20.10 ± 7.929 a	2.97 ± 0.686 a	6.30 ± 2.969 a	0.86 ± 0.321 a
P3	20.03 ± 5.991 a	2.95 ± 0.616 a	6.55 ± 2.660 a	0.88 ± 0.334 a
M × P	ns	ns	ns	ns

Data are presented as the mean ± standard deviation (STDEV, $n = 6$). Different letters within column indicate significance at $p < 0.05$ according to Duncan's test.

There was no interaction between AMF and goat manure treatment to the infection of mycorrhiza on roots. The application of various doses of goat manure also did not affect root infection of *P. angulata*. However, the application of mycorrhiza showed better result on root infection of AMF than the application of goat manure alone, although statistically it has no significant difference (Figure 3). Control treatment (no AMF application) revealed that the infection of AMF on plant roots will not happen if there was no AMF inoculation. In addition, no mycorrhiza colonization was observed by Gao et al. [16] in the non-mycorrhiza plant.

**Figure 3.** Percentage of AMF Infection on roots of *P. angulata*.

The condition of AMF infection on plant roots did not ensure the P content in plants after harvesting. The application of 0, 10, 20, and 30 g mycorrhiza had 0.5%, 0.56%, 0.49%, and 0.50% of P content, respectively. Comparing to tomato as a same member of *P. angulata* family (Solanaceae), the P content of *P. angulata* has fulfilled the nutrient sufficiency which is 0.25–0.75% of phosphorus in sufficiency range [17]. On the other hand, there were significant contributions of AMF colonization in the roots to the amount of P taken up by the plants, both maize and sorghum [18]. Principally, as an invasive plant, *P. angulata* plants responded positively to NPK levels, because it withstands low-resource availability and even at a higher level of nutrients [19].

4. Discussion

The soil condition used in this study was already fertile which combated the effects of mycorrhiza application. High available soil phosphorus (P) status, often limits AM fungal development and results in the C-costs to the host plant outweighing any benefits from AM colonization [20]. Moreover, the intense use of manure as fertilizer also may lead to high soil P concentration [14]. It is a dilemma because unmanaged livestock waste can enter to the surface water through erosion and runoff, while the application of goat manure on fertile soil with AMF accumulates P legacy in the soil.

The application of goat manure and AMF increased the soil pH to neutral (6.56) because the pH of goat manure itself was 8.12. This status created favor soil conditions for plants to absorb nutrients. Applying organic residues altered soil pH because they release hydroxyl ions (OH^-) during decomposition and increased soil pH to near neutral [21].

The increased dosage of goat manure had a positive correlation to growth parameters especially for plant height, leaf number, and stem diameter (Table 2). The application of organic manure can improve soil nutrient status due to a large stock of carbon and nutrients, a diverse soil microbial community, and a high cation exchange capacity [22]. The soil condition used has provided proper environment for soil microorganisms, such as soil organic carbon, NO_3^- , and available phosphorus which has also been proven to be a key factor affecting the change in the soil bacterial community [23].

The highest dosage of goat manure (30 g) also showed the highest total fruit weight per plant (Table 3). There is no dose at which manure becomes toxic to plants [24]. This result also was shown in growth parameters where plants get more manure, the growth increased. Linear with Cavagnaro [25], the addition of compost had a substantial impact on plant growth, increasing significantly, and linearly, with increasing rates of compost application.

Neither growth nor yield of *P. angulata* were affected by the application of mycorrhiza. Chu et al. [26] stated that the contribution of mycorrhiza P uptake pathway to plant P uptake was significantly affected by soil P availability; it was greater at medium than low and high Olsen P due to inhibition of the expression of phosphate transporter gene induced by mycorrhiza colonization. In line with Xu et al. [27], AMF acquires nutrients from organic matter for host, influence decomposition and alter the bacterial and fungal communities, and that these effects were modulated by the soil P availability.

Samanhudi et al. [12] obtained a similar result in which the treatment of various AMF doses did not significantly affect the stover dry weight of the young ginger plant due to high P content in the soil. Strike influence of mycorrhiza is common in phosphorus-deficient soil. This is supported by the observation of Andrea et al. [28] on the plant growth that the effects of mycorrhiza depended on the soil P status resulting in growth depression and promotion in high and low P soil, respectively.

The effect of mycorrhiza can be shown on shoot dried weight parameter which had the highest number of 3.06 g per plant in the 30 g application of mycorrhiza (Table 4). Since AMF inoculation enhanced both nutrient uptake and photosynthesis, it is highly likely that AMF-induced increased synthesis of photosynthates contributed to both below-ground and above-ground biomass accumulation [29]. This result also was in line with Muktiyanta et al. [30] study that AMF dose given had significant effect on dry weight of biomass because mycorrhiza symbiotic with plant roots can increase water and phosphorus uptake to be exploited by leaves and stems.

Higher above-ground biomass yield stimulated by inoculant application seems to be related to higher fruit number and total fruit weight per plant although the difference was not significant statistically. In line with Stoffel et al. [31], the efficiency of mycorrhiza inoculant for increasing grain yield of soybean had a positive correlation with higher biomass yield. However, the observed AMF-induced plant yield responses in shoot dried weight were not reflected by the vegetative plant growth phase where no AMF effects were observed.

The level of AMF infection was high around 50–70%. It has correlation to soil physical and chemical properties because the soil provides growth environment to the spores, includ-

ing water content, temperature, soil texture, pH, organic carbon, available P, exchangeable K, and nitrogen [32]. Moreover, the addition of goat manure and zeolite as a carrier of AMF also pointed effect of AMF infection through the changes in soil conditions. Zeolite amendment can improve water productivity, prevent soil nutrient losses, and change the soil texture in the long term [33,34].

5. Conclusions

The implementation of circular bioeconomy regarding nutrient cycle with inclusion of biotic and abiotic resources in the soil based on goat livestock system was studied. While waste management and AMF application as additional inputs of nutrients to *P. angulata* production is not the most profitable solution, however, it can be considered as a way to tackle environmental problems sourcing from excessive livestock waste. Based on the case study of *P. angulata* production, the application of AMF, either individually or in combination with GM showed slightly significant difference in growth and yield parameters because of unbalanced soil fertility settings. The observed vegetative plant growth phase responses in several parameters were not reflected in the plant yield and vice versa. Unbalancing nutrients in fecund soils due to the application of organic fertilizer together with AMF might show negative effect on nutrient uptake in plants where both organic fertilizers have their characteristic in terms of providing extra nutrients for plants. The effects of AMF and GM on the production of *P. angulata* vary depending on environmental factors, including the soil nutrient status. Therefore, the concept of circular bioeconomy in this case might not be an instant solution for the sole aspect of production, but environmental sustainability can be achieved gradually. To this end, there is a particular need, such as soil management practices and innovation of livestock-based fertilizer products for further information to overcome the excessive nutrient availability in agricultural areas of Indonesia together with the appraisal of additional land resources along with their economic and biophysical utilization that shall be included to increase the effectiveness of nutrient management within the circular bioeconomy of the livestock sector.

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