Jurnal Agronomi Indonesia
(Indonesian Journal of Agronomy)

ISSN 2085-2916 | e-ISSN 2337-3652
April 2024, *52*(1), 110-121
DOI: https://dx.doi.org/10.24831/jai.v52i1.53751

Research Article

Effect of $CaCO_3$, humic acid, and arbuscular mycorrhizal applications on soybean growth in ultisol of Sijunjung, West Sumatra

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ABSTRACT

Soybean is an important source of cheap protein. However, an adequate supply of soybeans is still challenging in Indonesia. This research aimed to evaluate the morphological, physiological, and yield responses of soybeans from the application of calcium carbonate ($CaCO_3$), humic acid, and arbuscular mycorrhizal fungi (AMF). The research was conducted from December 2022 to April 2023 at the ultisol soil of Sijunjung Regency, West Sumatra. The experiment used a split-split-plot design with three replications. The first factor was AMF and without AMF application. The second factor was humic acid and without humic acid. The third factor was doses of $CaCO_3$ application: 0, ¼, ½, ¾, and 1 time of exchangeable Al. The results showed that $CaCO_3$ application significantly affected root length, shoot dry weight, N-tissue content, soil pH, and exchangeable Al. The application of AMF affected the percentage of AMF colonization. The yield components did not show any significant differences among treatments.
However, the yield was 24.85% higher than that written in the official description. Therefore, the application of $CaCO_3$ or humic acid or AMF could be a potential solution to increase soybean yields in ultisol. It needs further evaluation in the field to support future farming practices.

Keywords: Al-exch; calcium carbonate; N-tissue content; productivity; soil pH

Edited by:
Okti Syah Isyani Permatasari
IPB University

Received:
6 February 2024
Accepted:
1 April 2024
Published online:
27 May 2024

Citation:
Arraudah, R., Purnamawati, H., Ghulamahdi, M. (2024). Effect of $CaCO_3$, humic acid, and arbuscular mycorrhizal applications on soybean growth in ultisol of Sijunjung, West Sumatra. *Jurnal Agronomi Indonesia (Indonesian Journal of Agronomy)*, *52*(1), 110-121

INTRODUCTION

Soybeans (*Glycine max* (L.) Merrill) have become a favorite food for Indonesians and even the world. Soybeans are a high source of vegetable protein, processed in the form of foods such as tempeh and tofu (Astawan & Hazmi, 2016), so it is not surprising that soybeans occupy the third position after rice and corn in Indonesia's national staple food strategic commodities (ICALRRD, 2020). Globally, Indonesia is in fifth position as the country that imports the most soybeans after China, Mexico, Saudi Arabia, and Japan (ASA, 2023). The domestic agricultural crisis is one of the results of the unequal distribution of production land (BPS, 2023). The domestic agricultural crisis is one of the results of the unequal distribution of production land. The average soybean production in several provinces on the island of Sumatra is still low, namely <1 tons ha^{-1}, compared to soybean production on the islands of Java and Sulawesi which has reached 1.5 - 2 tons ha^{-1}, which is above the national average production of 1.54 tons ha^{-1} (BPS, 2023). Efforts to increase production have been carried out in several regions in Indonesia, one of which is West Sumatra Province.

Sijunjung District, West Sumatra Province has ultisol soil (Syofiani et al., 2020), about 316.9 thousand hectares dominated by bushes, plantations, forests, and rice fields (RPRDA, 2022). Ultisol land is still underutilized due to low soil fertility, high exchangeable aluminum (Al-exch) >1.44 cmol kg^{-1} especially Al^{3+} resulting in soil pH <5.5 that is toxic to many plants (Meriño-Gergichevich et al., 2010; Chang et al., 2015; Baquy et al., 2018). Hight Al content in soil altered the elongation of roots (Chang et al., 2015), and exhibited lower root and shoot biomass leaf xylem diameter (Reis et al., 2018).

Increasing ultisol soil fertility can be done by applying soil conditioner or ameliorant which can improve the physical, chemical (Penn & Camberato, 2019; Deressa et al., 2020; Mulyati et al., 2022; Deru et al., 2023) and biological properties of the soil (Guo et al., 2019; Arraudah et al., 2020). The ameliorant that has been widely used is limestone or calcium carbonate ($CaCO_3$) (Mullen et al., 2016; ICATAD, 2021). The application of limestone is dedicated to suppress Al to a level suitable for plant growth. The application depends on the type of plant and soil conditions (Mullen et al., 2016). Applying lime for soybean production can be considered from the critical values of soil pH, Al-exch, and Al saturation (Deressa et al., 2020). The critical value of Al-exch for soybean plants is 2.42 cmol kg^{-1} and Al saturation is 22.75% (Baquy et al., 2018). The addition of $CaCO_3$ can increase the soil pH and affect the soil microbial community (Penn & Camberato, 2019; Guo et al., 2019; Deru et al., 2023). However, the use of agricultural lime in the long term will reduce soil fertility if it is not accompanied by the application of organic material. Therefore, we evaluate humic acid as an alternative to soil conditioners.

Humic acid is a macromolecular organic acid with acidic properties determined by the –COOH carboxylic and –OH phenolic groups, these groups are the most reactive in binding metal cations (Piri et al., 2019). Humic acid can change the absorption of ions in the soil (Piri et al., 2019) increase the pH of the soil, and decrease the Al^{3+} content in acid soil (Deng et al., 2021). The use of 45 mL L^{-1} humic acid in inoculation of 5 g AMF per plant was able to increase soil pH by 31.1%. However, the application of humic acid alone was not able to increase soybean yields in ultisol (Arraudah et al., 2020). Therefore, apart from providing soil ameliorant, it is also necessary to provide biological fertilizer so that the performance of plant roots in absorbing water and nutrients is optimal. One of the potential biofertilizers is arbuscular mycorrhizal fungi (AMF).

Arbuscular mycorrhizal fungi are a symbiotic relationship between fungi and plant growth which plays a role in helping to provide nutrients for plants (Wahab et al., 2023). AMF can make phosphorous (P) available to plants, through organic acids produced by plant roots which will become nutrients for fungi (Oktaviani et al., 2014). AMF will play a role in increasing the rate of photosynthesis and tolerance of photosynthesis to roots (Moelyohadi et al., 2012). Appropriate application of biofertilizers and the use of appropriate soybean varieties can play an important role in promoting optimal plant growth, nodulation, nitrogen fixation, and seed yield (Htwe et al., 2019).

The benefits of $CaCO_3$, humic acid, and AMF were evaluated to increase soybean production in ultisol soil, especially in Sijunjung, West Sumatra. This research aimed to evaluate the morphological, physiological, and yield of soybeans in response to the application of calcium carbonate ($CaCO_3$), humic acid, and arbuscular mycorrhizal fungi (AMF).

MATERIALS AND METHODS

The research was carried out from December 2022 to April 2023 at the community land of Pematang Panjang Village, Sijunjung District, West Sumatra at an altitude of ± 120 meters above sea level. The material used Demas 1 variety, which has been classified as resistant to acid soils (Somantri et al., 2018).

Other materials were liquid humic acid, AMF, $CaCO_3$, insecticide, Urea, SP-36, KCl, kieserite, and cow manure. The research design used a split-split-plot randomized complete block design with three replications of three treatments with 20 experimental units. The first factor was AMF treatment (0 and 2.5 g plant^{-1}), the second factor was humic acid treatment (0 and 40 mL L^{-1}), and the third factor was $CaCO_3$ dosages (0, ¼, ½, ¾, and 1 × exchangeable aluminum).

The land was plowed and 60 plots sized 2 m × 1.5 m each were created. Composite soil was analyzed for Ph_{H2O}, Al-exch, cation exchange capacity (CEC), total nitrogen content (N-total), total phosphorous content (P-total), exchangeable potassium (K-exch), exchangeable calcium (Ca-exch), P-available and C-organic soil (Table 1). The chemical analysis of the soil was carried out before treatment. Based on analysis at the IPB University, Faculty of Agriculture Testing Laboratory, the soil pH was classified as acid with pH 4.47, and CEC was classified as high (26.66 cmol kg^{-1}). The N-total and P-available contents were in moderate status of 0.23% and 7.54 ppm P_2O_5 respectively. The available K-available was 0.55 cmol K kg^{-1} (high), C-organic was low (1.74%), and Al-exch was 9.73 cmol kg^{-1} (Table 1).

Table 1. Soil chemicals before treatment.

Indicators	Value
Soil pH (H_2O)	4.47
Al-exch (cmol kg^{-1})	9.73
CEC (cmol kg^{-1})	26.66
N-total (%)	0.23
K-available (cmol K kg^{-1})	0.55
Ca-exch (cmol Ca kg^{-1})	5.61
P-available (ppm P_2O_5)	7.54
C-organic (%)	1.74

Note: Soil sample was analyzed in the Testing Laboratory of the Agronomy and Horticulture Department of IPB University (2023).

The $CaCO_3$ application was given one month before planting, while AMF and humic acid were given at the planting hole at 2.5 g $plant^{-1}$ and 40 mL L^{-1}, respectively. There were two seeds per hill with a line spacing of 50 cm × 10 cm. Fertilization consisted of cow manure (10 ton ha^{-1}, urea (25 kg ha^{-1}), SP-36 (50 kg ha^{-1}), KCl (50 kg ha^{-1}), and kieserite (50 kg ha^{-1}). Harvesting was at 95 days after planting when 95% of the soybean pods got brownish yellow, the leaves started to fall and the stems and pods started to dry out.

Morphological observations included plant height (3, 4, and 5 WAP), number of leaves (3, 4, and 5 WAP), vertical root length, shoot dry weight, root dry weight, and number of effective root nodules. Physiological observations included root colonization, and uptake of N, P, K, and Al in plant tissue. Observation on yield included seed weight per plant, weight of 100 seeds, seed weight per m^2, total number of pods, number of empty pods, and yield productivity (tons ha^{-1}). Yield productivity was obtained by converting yield per m^2 into tons ha^{-1} with a land use efficiency used in this research of 80% of the total land area used. Environmental data such as light intensity, temperature, and rainfall were obtained from the Meteorology, Climatology, and Geophysics Agency, West Sumatra (Table 2).

Research data was analyzed using variance or F-test at an error level of 5%. Observed variables that were significantly different in the F-test were further tested using the Duncan Multiple Range Test (DMRT) and Orthogonal Polynomials.

Table 2. Environmental climate during planting.

Indicators	Value
Rainfall (mm)	129 – 245.5
Air temperature (°C)	25.3 – 26.5
Intensity of sunlight (cal cm^{-2})	248.8 – 305.7

Source: Meteorology, Climatology and Geophysics Agency, West Sumatra (2023)

RESULTS AND DISCUSSION

The application of AMF, humic acid, and $CaCO_3$ simultaneously or the application of humic acid alone did not have a significant effect on soil chemistry, plant morphology, physiology, and yields. However, a single application of AMF had a significant effect on the percentage of AMF colonization. On the other hand, single $CaCO_3$ application had a significant effect on soil pH and soil Al-exch variables at 4 and 9 weeks after $CaCO_3$ application and N-tissue (Table 3).

Table 3. Summary of analysis of variance in response to soil chemistry, morphology, physiology, and yield of soybean plants from the application of AMF, humic acid, and $CaCO_3$.

Observation variables	Probability							DC
	M	H	C	M x H	C x M	H x C	M x H x C	
Soil pH[1]	-	-	0.27*	-	-	-	-	14.78
Al-exch(1 (cmol kg^{-1})	-	-	<0.01**	-	-	-	-	26.78
Soil pH[2]	0.27	0.36	<0.01**	0.33	0.23	0.94	0.28	10.70
Al-exch(2 (cmol kg^{-1})	0.43	0.11	<0.01**	0.20	0.73	0.59	0.96	51.06
Plant height (cm)	0.94	0.38	0.57	0.38	0.73	0.08	0.58	9.75
Number of leaves	0.41	0.64	0.57	0.58	0.94	0.52	0.34	16.70
Root lenght (cm)	0.29	0.70	<0.01**	0.43	0.22	0.16	0.58	20.24
Shoot dry weight (g)	0.92	0.37	0.02*	0.96	0.54	0.77	0.86	32.78
Root dry weight (g)	0.64	0.30	0.36	0.28	0.30	0.51	0.10	37.69
Number of root nodule	0.54	0.16	0.74	0.10	0.43	0.23	0.46	34.04
Root nodule weight (g)	0.39	0.10	0.72	0.15	0.90	0.43	0.55	17.69
AMF colonization (%)	0.03*	0.59	0.63	0.75	0.14	0.77	0.15	47.67
N tissue (%)	0.66	0.81	<0.01**	0.29	0.48	0.49	0.36	10.06
P tissue (%)	0.32	0.63	0.15	0.92	0.86	0.72	0.64	9.29
K tissue (%)	0.70	0.78	0.65	0.59	0.84	0.48	0.70	14.53
Al tissue (%)	0.60	0.11	0.61	0.94	0.41	0.20	0.23	23.58
Number of total pods	0.51	0.63	0.22	0.51	0.99	0.29	0.24	25.08
Number of filled pods	0.05	0.69	0.16	0.51	0.99	0.26	0.14	24.48
Yield per plant (g)	0.26	0.62	0.70	0.40	0.74	0.09	0.18	25.90
Yield per m^2	0.42	0.37	0.23	0.24	0.70	0.11	0.34	23.64
Weight of 100 seeds (g)	0.27	0.43	0.24	0.27	0.19	0.90	0.37	4.88
Productivity (tons ha^{-1})	0.42	0.37	0.29	0.24	0.70	0.11	0.34	23.64

Note: M = FMA; H = Humic acid; C = $CaCO_3$; DC= Diversity coefficient and *) = indicates the results of analysis of variance are significantly different at α = 5%; **) = indicates the results of analysis of variance are significantly different at α = 1%; [1]) = 4 weeks after application of $CaCO_3$ or at planting time; [2]) = 9 weeks after application of $CaCO_3$ or 5 weeks after planting.

Soil pH and Al-exch

It is known that the soil pH before being given $CaCO_3$ was 4.47 after applying $CaCO_3$ for 4 weeks, the soil pH increased to 6.76 or 51.23% from the application of $CaCO_3$ 1 × Al-exch or equivalent to 9.7 tons ha^{-1} (Figure 1A). Application of $CaCO_3$ after 9 weeks also increased soil pH by 22.45%, namely 5.5 of $CaCO_3$ 1 × Al-exch application or equivalent to 9.7 tons ha^{-1} (Figure 1B). The pH values meet the optimum requirements for soybeans to grow, namely at pH 6.0 – 6.8 (Walangululu et al., 2014).

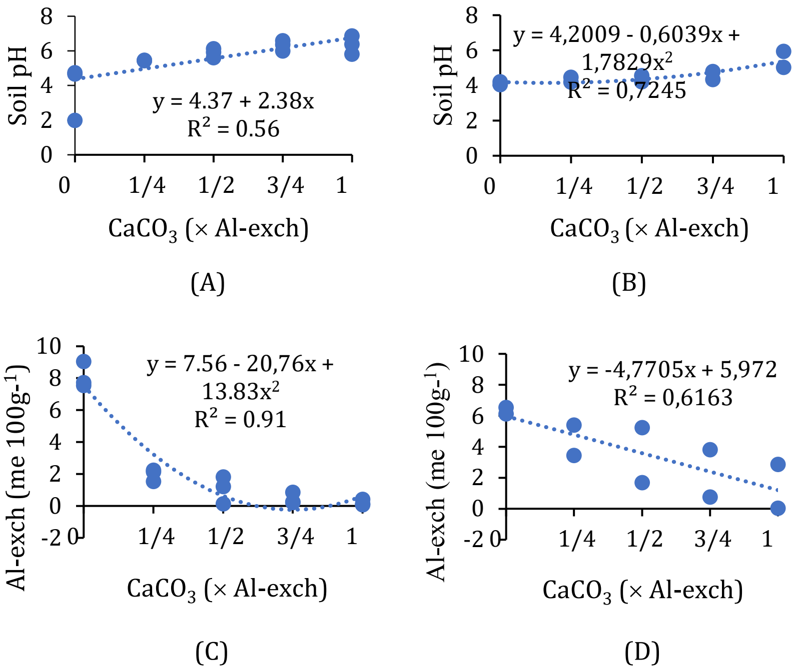

Figure 1. Soil pH at (A) 4 weeks, (B) 9 weeks after application of $CaCO_3$ and Al-exch at (C) 4 weeks, and (D) 9 weeks after application of $CaCO_3$ relationship on $CaCO_3$ dosages.

Al-exch before application of $CaCO_3$ was 9.7 me 100 g^{-1}. The application of $CaCO_3$ at ¾ × Al-exch or equivalent to 7.3 tons ha^{-1} was the best treatment that was able to reduce Al-exch up to 0.2 me 100 g^{-1}. However, the Al-exch can also be reduced up to 0.6 me 100 g^{-1} by applying $CaCO_3$ at ½ × Al-exch or equivalent to 4.8 tons ha^{-1}(Figure 1C). The critical value of Al-exch for soybean plants according to Baquy et al. (2018) is 2.42 cmol kg^{-1}. When the pH is low, the solubility of Al increases in the form of Al^{3+}; the Al^{3+} cation is one of the sources of soil acidity because it combines with the H^+ ion (Fazlina et al., 2021). The principle here is that the position of the Al^{3+} cation can be replaced by Ca^{3+}. When $CaCO_3$ is in the soil solution, it will decompose into Ca^{2+} and CO^-. The CO^- anion will react with the presence of H_2O water which will increase the OH^- ion content. The hydroxyl ion will bind more quickly to the Al^{3+} cation, which makes it easier for Al to precipitate (Syahputra et al., 2015). The decrease in Al concentration results in a smaller amount of Al being hydrolyzed to produce hydrogen ions, indirectly increasing soil pH (Fazlina et al., 2021; Nurzakiah et al., 2021). In other words, the application of $CaCO_3$ is able to reduce Al-exch (Figures 1C and 1D) which is followed by an increase in soil pH (Figures 1A and 1B); in line with Deng et al. (2021). In general, from the chemical analysis carried out on soil pH and Al-exch, the dose of $CaCO_3$ ½ × Al-exch or equivalent to 4.8 tons ha^{-1} has been able to be used as a recommendation to improve the chemical properties of ultisol soil in Sijunjung for soybean plants.

Nevertheless, application of $CaCO_3$ is not always advantageous. In the present research, the correlation between Al-exch and several $CaCO_3$ levels shows a quadratic pattern with the equation $y = 7.56 - 20.76x + 13.83x^2$. It means that adding a particular $CaCO_3$ dose of 7.56 will reduce the soil Al-exch by 20.76 times to the optimum Al-exch value of 0.2, so adding the $CaCO_3$ dose has the potential to increase the soil Al-exch value again by 13.83. The R^2 value 0.91 indicates that 91% of the soil Al-exch can be explained by the equation $y = 7.56 - 20.76x + 13.83x^2$ (Figure 1C). It's important to note that applying $CaCO_3$ higher than the optimum dose may cause excessive soluble Al in the soil. Additionally, environmental conditions such as rainfall can also affect the solubility of Al in the soil. Rainfall can lead to mass flow and dissolve or precipitate compounds in the soil, including Al.

Plant morphology

Plant morphology is a representation of the plant's ability to absorb nutrients from its environment. Morphological observations were carried out precisely at the peak of the vegetative phase or when they were about to enter the reproductive phase. This is based on root activity and nutrients generally decreasing, especially due to reduced carbohydrate supply to the roots because they are diverted to reproductive organs such as flowers, seeds, and fruit (Marschner, 2012). Plant stems have the function of transporting water and minerals from the roots to the leaves and also as a place for the growth of leaves, flowers, and fruit (Sari et al., 2015). In this study, the application of $CaCO_3$, AMF, or humic acid did not significantly impact the height of the soybean, either in single treatments or interactions. The height of the soybean plants at 5 WAP was 34.23 cm; it was higher than in mineral soil as of 24.6 cm according to Gaol et al. (2018) but lower than 43.96 cm reported by Haitami et al. (2020) and 81.3 cm by Somantri et al. (2018). The comparison of soybean plant height between ultisol soil and normal soil with the same variety tends to balance growth in normal soil conditions from the application of $CaCO_3$, AMF, or humic acid. Plant morphology is influenced by genetic and environmental factors. The Demas 1 variety is classified as resistant to acid soil environments (Somantri et al., 2018). Apart from that, it is also supported by the stability of the soil pH and Al-exch when planting, the soil pH has reached 6.76 (Figure 1A).

The application of $CaCO_3$, humic acid, or AMF did not significantly affect the height and number of leaves (Table 3). On the other hand, $CaCO_3$ application independently had a significant effect on root length and shoot dry weight, but it showed a tendency to produce similar results for root dry weight, number of root nodules, and root nodules weight (Table 4).

Table 4. Average root length, root dry weight, shoot dry weight, number of root nodules root nodule weight from $CaCO_3$ dosages.

$CaCO_3$ (× Al-exch)	RL (cm)	RDW (g)	SDW (g)	NRN	RNW (g)
0	13.86b	1.64	11.40c	12.75	0.60
¼	17.96a	1.57	12.89bc	15.03	0.81
½	18.28a	2.10	17.24a	16.94	0.76
¾	18.74a	1.90	16.48ab	13.17	0.64
1	18.71a	1.84	15.59abc	20.00	0.73

Note: RL (root length), RDW (root dry weight), SDW (shoot dry weight), NRN (number of root nodules), RNW (root nodule weight), and values in the same column and treatment followed by the same letter are not significantly different at DMRT α = 5%.

Roots play an important role in supplying nutrients from the soil to plants. Root development is influenced by the environment, like as the presence of microbes in the soil, the chemistry of soil condition or nutrition available, and the type of the root depending on the kind of plant (Tajima, 2021). During our earlier discussion, we noted that $CaCO_3$ has been effective in stabilizing soil pH conditions (Figure 1A and 1B). This has had a positive impact on the growth of roots as well, where the addition of $CaCO_3$ at all doses has resulted in better root development, measuring 17.96 – 18.71 cm as compared to 13.86 cm without $CaCO_3$ (Table 4). Root development was inhibited in the absence of $CaCO_3$ because the soil chemical conditions remained high in Al solubility (Figure 1C and 1D). High Al solubility can inhibit root elongation (Marschner, 2012) because Al will accumulate in the root apoplasm (Kinraide, 1990; Kopittke et al., 2004). Apoplasma will be responsible for cell wall synthesis (Boyer, 2009). This is where growth inhibition occurs because Al^{3+} high affinity easily binds to the cell wall matrix (Marschner, 2012). Therefore, the solubility of Al is one of the first focuses when cultivating soybeans in acid soil, one of which is to avoid inhibiting root development.

Long-term exposure to Al might have adverse effects on the uptake of nutrients and water by plants, ultimately leading to nutritional deficiencies (Marschner, 2012).

According to Nleya et al. (2019), root development will continue until the sixth reproductive phase. The difference in $CaCO_3$ application lies not only in root length but also in shoot dry weight. As per Table 4, applying $CaCO_3$ ½ - 1 × Al-exch produced better shoot dry weight than 0 – ¼ × Al-exch. However, the root dry weight, number of root nodules, and root nodule weight tended to be consistent across all $CaCO_3$ levels (Table 3). This is due to the horizontal root structure in both length and diameter, hence not all longer roots will have larger root diameters than shorter roots. This can affect the number and weight of root nodules because root nodules are formed on root hairs. However, unfortunately, the root diameter was not evaluated in this study, and the conditions in the field may yield mixed results when observing the component's root.

Plant physiology

Arbuscular mycorrhizal fungi (AMF) are known as soil-borne fungi, part of the ancient phylum Glomeromycota (Sun et al., 2018). Arbuscular mycorrhizal fungi play an important role in soil-plant nutrient dynamics (Suri & Chodary, 2013). Some of the benefits of AMF in acid soil are, an increase in seed weight, and increased air content of plants help in the absorption of phosphorus, nitrogen, and potassium (Shi-chu et al., 2019; Parihar et al., 2020), increased biomass, chlorophyll content, plant nutrition (Mathur et al., 2018; Bijalwan et al., 2021), root growth and activation of antioxidant enzymes in the roots (Mathur et al., 2018), and resistance to various abiotic stresses (Sun et al., 2018). AMF colonization is located at the tips of young roots or root hairs. This observation was observed in the fifth week after planting because in this phase the rate of N2 fixation from the air by the roots increased rapidly (Nleya et al., 2019). AMF Colonization is observed under a microscope with the appearance of hyphae and/or spores in the root tissue, indicating that the roots have been infected with mycorrhiza. The percentage of AMF colonization did not show a significant effect from the administration of $CaCO_3$ or humic acid either independently or in interaction. However, there is a significant effect of giving FMA independently (Table 3). The percentage of AMF was better when given AMF 2.5 g plant^{-1} compared to without AMF, namely 44.86% (Table 5). This colonization percentage is considered high because it is greater than 30% (O'Connor et al., 2001).

Table 5. Percentage of colonization in soybean roots from AMF application.

AMF (g plant^{-1})	Colonization of AMF (%)
0	25.17b
2.5	44.86a

Note: Values in the same column and treatment followed by the same letter are not significantly different at DMRT α = 5%.

Other physiological characteristics observed were the content of N (nitrogen), P (phosphorus), K (potassium), and Al (aluminum) in plant tissue. The $CaCO_3$ administration had a significant effect on N-tissue content. The application of 1 × Al-exch $CaCO_3$ gives the highest value for the N-tissue, namely 3.33%, but the application of ½ × Al-exch is enough to give the same results as giving 1 × Al-exch, which is 3.21%. P-tissue content in the range 0.24 – 0.27%, K-tissue content in the range 2.68 – 2.92%, and Al-tissue content in the range 647.92 – 904.43%. Although the P-tissue and K-tissue content is not significantly different, the lowest values are obtained when $CaCO_3$ is not given. Additionally, Al-tissue content is lower when $CaCO_3$ is given (Table 6).

Table 6. Nutrient content of soybean biomass from different $CaCO_3$ dosages.

$CaCO_3$ (× Al-exch)	N (%)	P (%)	K (%)	Al (ppm)
0	2.69c	0.24	2.68	904.43
¼	3.04b	0.26	2.85	727.25
½	3.21ab	0.27	2.75	837.55
¾	3.10ab	0.27	2.92	647.92
1	3.33a	0.27	2.82	723.28

Note: N (N-tissue), P (P-tissue), K (K-tissue), Al (Al-tissue), and values in the same column and treatment followed by the same letter are not significantly different at DMRT α = 5%.

Nitrogen is one of the essential macronutrients for plants, playing a role in the formation of proteins, nucleic acids, chlorophyll, co-enzymes, phytohormones, and secondary metabolism. Its content in plants ranges from 1-5% (Marschner, 2012). The highest N uptake was observed when the plants received 1 × Al-exch, but the uptake was still similar when they received only ½ or ¾ × Al-exch, which ranged from 3.1% to 3.21%. However, in the absence of $CaCO_3$, the N uptake decreased to 2.69% (Table 6). This decrease can be attributed to the unstable pH and Al-exch conditions of the soil, which are still unstable in the absence of $CaCO_3$ (Figures 1A-1D). Low soil pH inhibits nodulation activity and N2 fixation (Marschner, 2012). The N uptake is also related to root and shoot function (Sari et al., 2015; Backtiar et al., 2016). It can be seen that $CaCO_3$ significantly influences root length and thus has an impact on N uptake (Table 3). Differences in N uptake capacity are caused by differences in the level of efficiency of N uptake capacity per unit root length and root weight (Puccio et al., 2021). But, in this study root weight is not significantly different from all treatments (Table 3).

The root development is linked to the activity of nodulation that is associated with AMF. However, when AMF is present along with $CaCO_3$, it only affects the root colonization percentage (Table 3) and not the uptake of plant nutrients. This is because the effectiveness of $CaCO_3$ in providing nutrients to the plants is observed only after four weeks of planting. According to Deru et al. (2023), adding $CaCO_3$ to soil increases N mineralization. Additionally, the availability of air around plant roots greatly affects the ability of plants to absorb nutrients (He et al., 2017). It is known that, during liming or before planting, rainfall is moderate, 129 mm (Table 2), so the application of ½ to 1 × Al-exch $CaCO_3$ works optimally in improving the chemical properties of the soil.

Aluminum is a non-essential element for soybean plants. The maximum Al content in plants is 1 mg per dry weight. When it exceeds, it will inhibit root growth and root hairs from absorbing P (Marschner, 2012). In this study, the uptake of P did not significantly differ across various levels of $CaCO_3$ provided. However, it was observed that the absence of $CaCO_3$ resulted in lower nutrient uptake compared to when $CaCO_3$ was given, particularly with regard to N-tissue and K-tissue content. This trend was inversely proportional to the Al content which tends to be higher when $CaCO_3$ is not given. This is due to the high solubility of Al before the addition of $CaCO_3$ (Figures 1C and 1D).

Yield components

The application of AMF, humic acid, and $CaCO_3$ did not affect the components of soybean yield, such as total number of pods, number of empty pods, weight of 100 seeds, seed weight per plant, seed weight per m^2, and yield productivity (Table 7). The average number of pods per plant from all treatments was 74.29, compared with the total number of pods to empty pods (72.29) only decreasing by 2.7%. In another study, the number of empty pods per plant is around 58.6 (Somantri et al., 2018). In the present study, pod was filled well leading to fewer empty pods. Judging from the weight of 100 seeds, i.e., 10.99 g, the Demas 1 variety was classified as close to medium-seed soybeans; as those in the range of 11-13 g per 100 seeds weight (Somantri et al., 2018). The seed production per plant was 16.15 g and the seed production per m^2 was 391.72 g. Thus, the average yield productivity achieved was 3.13 tons ha^{-1} (Table 7).

The soybean yield from all treatments were not significantly different. In terms of productivity, the yield gained in the present study was 24.85% higher than its potential yield as described in the official description by the Ministry of Agriculture (2.51 tons ha^{-1}). The percentage increase in yield from AMF, humic acid, and $CaCO_3$ independently was 8.06% higher (2.71 tons ha^{-1}), 32.3% (3.32 tons ha^{-1}), and 42.55% (3.58 tons ha^{-1}), respectively.

Environmental stress is a major threat to soybean (Nleya et al., 2019). The ability of soybean roots to absorb nutrients is greatly influenced by the availability of soil humidity (He et al., 2017). Such high productivity is probably due to adequate rainfall and air temperature during the research. According to Mandic et al. (2017), suitable monthly rainfall is 245.5 mm, and the suitable air temperature is 25.6 °C according to Liu et al. (2008). In general, all of the treatments showed that the separate application of $CaCO_3$, 40 mL L^{-1} of humic acid, and 2.5 g $plant^{-1}$ AMF is prospective to increase soybean yields in ultisol soil of Sijunjung. This suggests that the addition of AMF, humic acid, and $CaCO_3$ can increase soybean yield, and should be considered in future farming practices.

Table 7. Average total number of pods, number of empty pods, weight of 100 seeds, seed weight per plant, seed weight per m^2 and productivity from AMF, humic acid and $CaCO_3$ treatments.

Treatments	TNP	NPP	W100 (g)	SWP (g)	SWM (g)	P (tons ha^{-1})
Control	53.47	51.73	10.93	11.70	258.55	2.07
C1	73.27	72.40	10.80	14.02	404.13	3.23
C2	82.40	81.27	11.23	19.45	447.27	3.58
C3	62.47	60.27	10.20	13.77	409.59	3.28
C4	70.13	69.33	11.07	16.94	365.25	2.92
H	66.07	64.47	10.60	13.40	415.08	3.32
H + C1	47.20	45.93	11.47	10.08	296.12	2.37
H + C2	57.40	53.73	11.03	11.58	299.35	2.39
H + C3	91.27	89.27	10.67	20.78	447.54	3.58
H + C4	69.47	67.33	10.83	15.69	370.24	2.96
M	72.80	70.07	11.00	16.85	339.05	2.71
M + K1	68.87	65.53	11.47	14.41	393.47	3.15
M + K2	78.93	77.73	10.97	17.53	340.36	2.72
M + K3	80.07	80.47	11.40	15.41	407.56	3.26
M + K4	74.00	72.00	11.60	15.58	361.60	2.89
M + H	77.13	73.60	10.97	17.16	417.89	3.34
M + H + K1	79.40	76.80	10.93	16.72	436.34	3.49
M + H + K2	90.00	87.93	10.53	19.30	405.68	3.24
M + H + K3	91.27	88.27	10.77	19.49	454.24	3.63
M+ H + K4	100.2	97.73	11.23	23.12	565.17	4.52
Average	74.29	72.29	10.99	16.15	391.72	3.13

Note: TNP (total number of pods), NPP (number of empty pods), W100 (weight of 100 seeds), (seed weight per plant), SWM (seed weight per m^2), and P (productivity), control (whitout treatments), K1 (¼ × Al-exch), K2 (½ × Al-exch), K3 (¾ × Al-exch), K4 (1 × Al-exch), H (40 mL L^{-1}), M (2.5 g $plant^{-1}$).

CONCLUSIONS

Morphological characteristic of soybean was influenced by the dose of $CaCO_3$ on the variables of root length and shoot dry weight. Physiological characteristic was also significantly influenced by the dose of $CaCO_3$ on N-tissue content and the administration of AMF affected the percentage of AMF colonization. Soybean yield did not show any significantly different response from the application of FMA, humic acid, and $CaCO_3$. However, the grain production tended to be 24.85% higher than official description equal to 3.13 tons ha^{-1}. The application of $CaCO_3$, humic acid or AMF is prospective to increase soybean yields in Sijunjung ultisol soil, West Sumatra.

ACNOWLEDGEMENTS

The authors thank the Ministry of Finance of the Republic of Indonesia for funding this research through the LPDP Scholarship.

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