

Oil Palm (*Elaeis guineensis* Jacq.) Shaded Model at Immature Stage II and Application of Arbuscular Mycorrhizal Fungi (AMF) on Growth and Yields of Soybean (*Glycine max* (L.) Merrill

(Model Berlorek Kelapa Sawit (*Elaeis guineensis* Jacq.) pada Peringkat Tidak Matang II dan Penggunaan Kulat Mikoriza Arbuskul (AMF) pada Pertumbuhan dan Hasil Kacang Soya (*Glycine max* (L.) Merrill)

CUCU SUHERMAN^{1*}, F INDRI², SANTI ROSNIAWATY¹ & MOCHAMMAD ARIEF¹

¹Department of Agronomy, Faculty of Agriculture, Universitas Padjadjaran, 45363 Jatinangor, Sumedang, West Java, Indonesia

²Study Program of Agrotechnology, Faculty of Agriculture, Universitas Padjadjaran 45363 Jatinangor, Sumedang, West Java, Indonesia

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ABSTRACT

The practice of intercropping soybeans among oil palm trees is an approach used to optimize plantation land. Oil palm (OP) at the second immature stage (IS-2) or the age of 2 years provides shade of around 40%. This research aimed to observe soybean growth and yields under shade conditions close to those of oil palm shading conditions at IS-2 and to determine the optimal dosage of Arbuscular Mycorrhizal Fungi (AMF) to apply to the soybeans. The shade intensity treatment used artificial shading nets close to actual shade intensity in OP plantation, i.e., 40% shading. The experiment was conducted at the Experimental Station, Faculty of Agriculture, Universitas Padjadjaran, from October 2019 to January 2020. The experimental design used was a split-plot design with shade intensity as the main plots (0 and 40%) and AMF dosage as subplots, consisting of six treatments of 0, 2, 4, 6, 8, and 10 g/plant, with each treatment repeated four times. The results showed that at 10 weeks after planting, the interaction effect of 40% shade intensity with a dosage of 10 g AMF per plant resulted in the highest chlorophyll index. Independently, the AMF dosage of 10 g per plant produced the maximum plant height, number of productive branches, dry weight of plants, and number of seeds per pod.

Keywords: Arbuscular Mycorrhizal Fungi; immature plant; oil palm; shade intensity; soybean

ABSTRAK

Amalan penanaman kacang soya di antara pokok kelapa sawit merupakan pendekatan yang digunakan untuk mengoptimalkan tanah ladang. Kelapa sawit (OP) pada peringkat belum matang ke-2 (IS-2) atau berumur 2 tahun memberikan teduhan sekitar 40%. Penyelidikan ini bertujuan untuk melihat pertumbuhan dan hasil kacang soya di bawah keadaan teduhan yang hampir dengan keadaan teduhan kelapa sawit di IS-2 dan untuk menentukan dos optimum Kulat Mikoriza Arbuskul (AMF) untuk digunakan pada kacang soya. Rawatan keamatan teduhan menggunakan jaring teduhan tiruan hampir dengan keamatan teduhan sebenar di ladang OP, iaitu 40%. Uji kaji telah dijalankan di Stesen Eksperimen, Fakulti Pertanian, Universitas Padjadjaran, dari Oktober 2019 hingga Januari 2020. Reka bentuk uji kaji yang digunakan adalah reka bentuk plot terpisah dengan keamatan teduhan sebagai plot utama (0 dan 40%) dan dos AMF sebagai subplot, yang terdiri daripada enam rawatan 0, 2, 4, 6, 8 dan 10 g/tanaman, dengan setiap rawatan diulang empat kali. Keputusan menunjukkan bahawa pada 10 minggu selepas penanaman, kesan interaksi 40% keamatan teduh dengan dos 10 g AMF setiap tumbuhan menghasilkan indeks klorofil yang paling tinggi. Secara tidak langsung, dos AMF sebanyak 10 g setiap tumbuhan menghasilkan ketinggian tumbuhan, bilangan cawangan produktif, berat kering tumbuhan dan bilangan biji setiap buah yang maksimum.

Kata kunci: Kacang soya; kelapa sawit; keamatan teduhan; Kulat Mikoriza Arbuskul; tumbuhan tidak matang

INTRODUCTION

Oil palm plantations in Indonesia have reached a total area of more than 14.6 million hectares, representing both the largest plantation in Indonesia and the world in 2019 (Direktorat Jenderal Perkebunan 2019). The planting distance between oil palm trees is always large. Therefore, uncultivated open space remains between trees, especially those aged between one and two years (first and second immature stages, IS-1 and IS-2). During IS-1, there is around 75% open space, whereas, in IS-2, approximately 60% of the land area is open space (Mawarni 2011; Nuraisah et al. 2019). These open land areas have significant potential for the cultivation of annual crops, such as soybean, using an intercropping system.

Soybeans require full sunlight. However, there are shade-tolerant varieties that can be grown under shade intensities of 33-50% (Chairudin & Sabaruddin 2015; Soverda 2012). Mostly, oil palm plantations are located on marginal sites, such as where plants are grown in inceptisols. Therefore, there are typically some disadvantages in terms of soil fertility, e.g., a lack of phosphorus (P). P is essential for plant growth, and the combination of P and compost is needed to improve plant growth and yield (Mubarok et al. 2020a). To improve growth under such conditions, the addition of Arbuscular Mycorrhizal Fungi (AMF) in the intercropping system can help soybean roots to absorb nutrients and minerals, despite being grown in acidic soils (Kanno et al. 2006; Shukla et al. 2009).

One of the key advantages of AMF, as stated by Bolan (1991), is its ability to convert phosphorus (P) from unavailable forms (i.e., Al – P, Fe – P, Ca – P, Mg – P) to available forms (i.e., $H_2PO_4^-$, HPO_4^{2-}). Furthermore, Davies et al. (1996) proved that the AMF could replace approximately 50% of phosphate, 40% of nitrogen and 25% of potassium. Application of AMF to soybeans at a dose of 10 g/plant was reported to increase root expansion, root nodules and seed weight (Sukmasari et al. 2018). Another study showed that AMF application to soybean plants at a dose of 8 g/plant gave the best effect on the plant height, leaves number, chlorophyll index, seed weight, and 1,000-soybean seeds weight. Meanwhile, a dose of 4 g/plant had the maximum impact on net assimilation (LAN) and relative growth rate (RGR) (Suherman et al. 2012). The advantage of shading conditions under an intercropping system is that AMF can develop under low air temperatures and shading. Schenck and Schroder (1994) reported that appropriate AMF development and colonization temperatures in between

28-34 °C, with the optimal expansion of vesicles at 35 °C. Soybean cultivation among oil palm trees has potential advantages and disadvantages, depending on palm leaf expansion and soybean adaptability. The ‘Arjasari’ Soybean cultivar can be categorized as high temperature and low light intensity-tolerant plants, which have the potential to adapt easily to growth under shade treatments. In this study, we used an artificial shading net, which simulates conditions close to actual shading in oil palm plants at IS-2 (two years old), i.e., 40% light shading or 60% light transmission, so that the physiological and morphological responses of soybeans can be observed in detail.

Overall, this research aims to observe the growth and yield of soybeans grown under shading conditions close to the shade conditions of oil palm plantations at IS-2 and to determine the optimal dosage application of AMF.

MATERIALS AND METHODS

This study was conducted at the Ciparanje Experimental Station, Faculty of Agriculture, Universitas Padjadjaran, Indonesia, from October 2019 to January 2020. This site is located at ~752 m above sea level. The plant material used was the Arjasari cultivar, grown in inceptisols with shade treatments of 0% (i.e., without shade) and 40% intensity. The experiment used a split-plot design with shading intensity as the main plot and AMF as a sub-plot. The shading intensity of 40% was set using an artificial vinyl shade; this value was designed to be as close as the actual shade value of oil palm shoots during IS-2. The shading net intensity was adjusted to 40% or 20,300 lux, while light intensity in open fields was 30,560-35,011 lux. Therefore, the net provided shade ranges between 33.6-42%. The shading intensity value was chosen based on the actual shading conditions observed in palm trees in a previous study. The subplots consisted of six treatment levels, namely, no AMF treatment and AMF treatment at doses of 2, 4, 6, 8, and 10 g/plant, for a total of 12 treatments, consisting of six plants per treatment. All treatments were repeated four times.

Measurements of plant height and the number of trifoliolate leaves were conducted at 2, 3, 4, 5, and 6 weeks after planting (WAP), with the number of productive branches and dry matter weight measured at 12 WAP. Stomatal conductance was measured by using a porometer (Decagon, Inc. US). The leaves chosen were the second leaves from the top of plants at 4, 8, and 12 WAP. The leaf chlorophyll index was measured using a chlorophyll meter (Opti Sci., US) at the second leaf from the top of the

plant at 4, 6, 10, and 12 WAP. Chlorophyll fluorescence measurements were made using a fluorescence meter (Hansatech Instrument). The leaf observed was the second leaf from the base of the plant. Measurements were made at 4, 6, 10, and 12 WAP. Both the stomatal and chlorophyll fluorescence responses were measured at the beginning, middle and last growing stages, represented by measurements at minimum values of 4, 8, and 12 WAP. The P uptake of the plants was analyzed at 6 WAP; at this point, the plant is beginning to move to the reproductive stage, where more P uptake is possible than in the vegetative stage. Data collected for this parameter were not analyzed statistically due to a lack of sample. However, these results can nonetheless provide a general description of this parameter.

P uptake analysis was carried out using the Wet Digestion Method (Pequerul et al. 1993). Samples were taken from the shoot of the plant and dried at 75 °C for 48 h. Each sample (0.5 g) was removed and ground in a blender before it was placed in 25 mL Kjedahl ash, to which 5 mL of HNO₃ was added. Once mixed, 0.5 mL of HClO₄ was added to the flask, which was then shaken gently so that all the sample was wetted; the mixture was then left overnight. The flask was then heated in a smoke room over low heat, with the heat slowly increased to a temperature of 350 °C. The digestion was complete when white steam emerged, and a clear extract was obtained (about 4 h); 50 mL of distilled water was added, and the mixture was shaken for a while before being transferred to a distillation tube. A comparison was also made using a blank sample.

For P absorption measurements, 1 mL of extract and standard series were pipetted into each test tube, to which 9 mL of 0.25% La solution was then added and the mixture was shaken until homogeneous. P absorption was measured using a standard series flame photometer as a comparison in the Atomic Absorption Spectrophotometry (AAS) room. P absorption was measured using the following equation (%):

$$\begin{aligned} &= \text{ppm curve} \times \text{mL extract} / 1000 \text{ mL} \times 100\text{g} / \text{example} \\ &\times F_p \times f_k F_k \\ &= \text{ppm curve} \times 50/1000 \times 100/250 \times 10 \times F_k \\ &= \text{ppm curve} \times 0.2 \times F_k \end{aligned}$$

where F_p is the dilution factor (if any); F_k is the moisture content correction factor = $100 / (100 - \text{moisture content percentage})$. At the end of the experiments (24 WAP), the pod number, seed number per pod, seed weight and 100-seed weight were measured.

The collected data were analyzed using the Analysis of Variance (ANOVA) approach with the F test at a 95% confidence interval. If the difference in the average treatment effect was found to be significant, then further tests were conducted with Duncan's Multiple Range Test (DMRT) at a 95% confidence interval.

RESULTS AND DISCUSSION

PLANT HEIGHT

Statistical analysis showed no interaction effect between shade and AMF treatment. However, both shade intensity and AMF dosage independently significantly influence plant height at all measurements (Table 1). Shade treatment of 40% increased plant height significantly more than the 0% shade treatment. Treatments using different AMF dosages at 2 WAP showed that f_6 treatment (10 g AMF/plant) produced higher plants than f_1 , f_2 , and f_3 treatments. Likewise, plant height under f_6 treatment at 6 WAP was higher than the other dosages.

NUMBER OF TRIFOLIATE LEAVES

There was a significant interaction effect between shade intensity and AMF dosage on the number of trifoliolate leaves observed only at 2 WAP. However, the shaded and AMF treatments alone, i.e., 3, 4, 5, and 6 WAP did not show any significant difference in the number of trifoliolate leaves even there was no significant interaction effect (Table 2).

NUMBER OF PRODUCTIVE BRANCHES

There was no interaction effect observed between shade intensity and AMF dosage on the number of productive branches. However, there was a significant difference observed in the case of treatment solely with AMF, where the f_6 treatment (10 g AMF/plant) showed a significantly higher number of productive branches than the other treatments (Table 3).

LEAVES CHLOROPHYLL INDEX

The value of the chlorophyll index at 10 WAP was significantly affected by the interaction of shade and AMF dosage in shaded conditions. Increasing AMF dosage increased the leaves' chlorophyll index at 10 WAP (Table 4). In the shade-only tests (i.e., no AMF dosage, Table 5), shade significantly affected the leaf chlorophyll index. Chlorophyll index values were higher with shade treatment than without shade in all measurements. Meanwhile, no significant differences were observed under different AMF dosage treatments (Table 5).

TABLE 1. Independent effect of shade intensity and AMF dosage on the height of soybean plants at 2 WAP, 3 WAP, 4 WAP, 5 WAP and 6 WAP

Treatments	Plant height (cm)				
	2 WAP	3 WAP	4 WAP	5 WAP	6 WAP
Shade intensity					
n_1 (40% shade)	19.54 a	29.07 a	37.64 a	67.47 a	88.03 a
n_2 (0% shade)	14.85 b	24.97 b	34.90 b	47.80 b	58.22 b
AMF dosage					
f_1 (control)	14.67 c	22.35 c	29.77 b	49.23 c	62.50 c
f_2 (2 g AMF/plant)	15.10 bc	23.03 bc	30.04 b	49.38 c	63.58 bc
f_3 (4 g AMF/plant)	15.51 bc	23.83 abc	32.29 ab	51.04 ab	66.77 b
f_4 (6 g AMF/plant)	16.01 ab	23.96 ab	31.10 ab	52.65 ab	65.75 b
f_5 (8 g AMF/plant)	16.05 ab	24.79 b	32.67 ab	52.80 ab	64.38 bc
f_6 (10 g AMF/plant)	16.85 a	26.15 a	34.71 a	54.71 a	69.13 a

Note: An average value followed by the same letter in the same column shows no significant difference according to Duncan's multiple range test at a 95% confidence interval

TABLE 2. Independent effect of shade intensity and AMF dosage on the number of soybean trifoliolate leaves at 3, 4, 5, and 6 WAP

Treatments	Number of trifoliolate leaves			
	3 WAP	4 WAP	5 WAP	6 WAP
Shade intensity				
n_1 (40% shade)	2.03 a	3.21 a	8.46 a	11.61 a
n_2 (0% shade)	1.99 a	3.35 a	8.22 a	11.60 a
AMF dosage				
f_1 (control)	2.00 a	3.13 a	7.83 a	11.62 a
f_2 (2 g AMF/plant)	1.96 a	3.13 a	8.75 a	11.63 a
f_3 (4 g AMF/plant)	2.04 a	3.21 a	8.00 a	11.79 a
f_4 (6 g AMF/plant)	2.00 a	3.30 a	8.58 a	11.58 a
f_5 (8 g AMF/plant)	2.00 a	3.33 a	8.17 a	10.92 a
f_6 (10 g AMF/plant)	2.04 a	3.60 a	8.71 a	12.08 a

Note: The average value followed by the same letter in the same column shows no significant difference according to Duncan's multiple range test at a 95% confidence interval

TABLE 3. Independent effect of shade intensity and AMF dosage on number of productive branches at 12 WAP

Treatments	Number of productive branches
Shade intensity	
n ₁ (40% shade)	4.70 a
n ₂ (0% shade)	4.82 a
AMF dosage	
f ₁ (control)	3.54 d
f ₂ (2 g AMF/plant)	4.21 c
f ₃ (4 g AMF/plant)	4.50 bc
f ₄ (6 g AMF/plant)	4.92 bc
f ₅ (8 g AMF/plant)	5.09 b
f ₆ (10 g AMF/plant)	5.78 a

Note: The average value followed by the same letter in the same column shows no significant difference according to Duncan's multiple range test at a 95% confidence interval

TABLE 4. Interaction between shade intensity and AMF dosage on soybean leaves chlorophyll index at 10 WAP

Shade intensity	Leaves Chlorophyll Index (CCI)					
	AMF dosage					
	f ₁ (control)	f ₂ (2 g AMF/ plant)	f ₃ (4 g AMF/ plant)	f ₄ (6 g AMF/ plant)	f ₅ (8 g AMF/ plant)	f ₆ (10 g AMF/ plant)
n ₁ (40% Shade)	26.80 a C	25.44 a C	25.99 a C	34.81 a B	38.70 a AB	40.50 a A
n ₂ (0% Shade)	32.90 a A	37.05 a A	37.33 a A	34.64 a A	34.14 a A	37.33 a A

Note: The average value followed by the same letter in the same column shows no significant difference according to Duncan's multiple range test at 95% confidence interval; capital letters are read horizontally while lowercase letters are read vertically

TABLE 5. Independent effect of shade intensity and AMF dosage on soybean leaves chlorophyll index at 4 WAP, 6 WAP, and 12 WAP

Treatment	Leaves Chlorophyll Index (CCI)		
	4 WAP	6 WAP	12 WAP
Shade intensity			
n ₁ (40% shade)	19.94 a	28.11 a	36.81 a
n ₂ (0% shade)	18.36 b	25.20 b	26.64 b
AMF dosage			
f ₁ (control)	17.88 a	24.63 a	28.68 a
f ₂ (2 g AMF/plant)	18.20 a	25.74 a	29.74 a
f ₃ (4 g AMF/plant)	18.46 a	25.98 a	31.99 a
f ₄ (6 g AMF/plant)	19.48 a	27.14 a	32.83 a
f ₅ (8 g AMF/plant)	20.01 a	27.51 a	32.85 a
f ₆ (10 g AMF/plant)	20.89 a	28.95 a	34.29 a

Note: The average value followed by the same letter in the same column shows no significant difference according to Duncan's multiple range test at a 95% confidence interval

STOMATAL CONDUCTANCE

Based on Table 6, the shaded treatment significantly increased stomatal conductance at 4 and 12 WAP.

However, AMF treatment showed no significant differences in stomatal conductance under all treatments.

TABLE 6. Independent effect of shade intensity and AMF dosage on soybean leaves stomata conductance at 4 WAP, 8 WAP, and 12 WAP

Treatment	Stomata conductance (mmol H ₂ O/m ² s)		
	4 WAP	8 WAP	12 WAP
Shade intensity			
n ₁ (40% shade)	452.94 a	263.25 a	192.55 a
n ₂ (0% shade)	363.94 b	212.30 a	125.63 b
AMF dosage			
f ₁ (control)	391.66 a	197.20 a	176.82 a
f ₂ (2 g AMF/plant)	431.74 a	227.07 a	151.27 a
f ₃ (4 g AMF/plant)	460.91 a	235.86 a	158.50 a
f ₄ (6 g AMF/plant)	388.84 a	240.64 a	162.01 a
f ₅ (8 g AMF/plant)	429.66 a	244.64 a	133.62 a
f ₆ (10 g AMF/plant)	347.84 a	281.21 a	172.33 a

Note: The average value followed by the same letter in the same column shows no significant difference according to Duncan's multiple range test at a 95% confidence interval

CHLOROPHYLL FLUORESCENCE

The shade treatment yielded significantly lower values of

chlorophyll fluorescence. However, the AMF treatments did not significantly affect this value (Table 7).

TABLE 7. Independent effect of shade intensity and AMF dosage on chlorophyll fluorescence at 4 WAP, 6 WAP, 8 WAP, and 12 WAP

Treatment	Chlorophyll fluorescence (index fv/fm)			
	4 WAP	6 WAP	10 WAP	12 WAP
Shade intensity				
n ₁ (40% shade)	0.74 b	0.73 b	0.75 b	0.74 b
n ₂ (0% shade)	0.79 a	0.79 a	0.79 a	0.76 a
AMF dosage				
f ₁ (control)	0.72 a	0.77 a	0.76 a	0.75 a
f ₂ (2 g AMF/plant)	0.78 a	0.75 a	0.76 a	0.76 a
f ₃ (4 g AMF/plant)	0.77 a	0.76 a	0.76 a	0.73 a
f ₄ (6 g AMF/plant)	0.77 a	0.76 a	0.77 a	0.76 a
f ₅ (8 g AMF/plant)	0.77 a	0.76 a	0.76 a	0.75 a
f ₆ (10 g AMF/plant)	0.76 a	0.77 a	0.78 a	0.76 a

Note: The average value followed by the same letter in the same column shows no significant difference according to Duncan's multiple range test at a 95% confidence interval

ABSORPTION OF PHOSPHATE IN PLANTS

Based on Table 8, under n_1 conditions (40% shade), the P_2O_5 value in plants (0.59%) tends to be lower than under n_2 conditions (0% shade) (0.75%). However, P absorption was higher in the n_1 treatment at 0.07 g/plant than n_2 with

0.06 g/plant. In terms of varying AMF dosages, f_2 (2 g AMF/plant) treatments yielded the highest percentage of P_2O_5 uptake based on its dried matter, followed, in order by f_6 (10 g AMF/plant), f_3 (4 g AMF/plant), f_4 (6 g AMF/plant), f_5 (10 g AMF/plant), and f_1 (control).

TABLE 8. Independent effect of shade intensity and AMF dosage on phosphate uptake at 6 WAP

Treatment	P_2O_5 (%)	Absorption of phosphate (P) (g)
Shade intensity		
n_1 (40% shade)	0.59	0.07
n_2 (0% shade)	0.75	0.06
AMF dosage		
f_1 (control)	0.45	0.04
f_2 (2 g AMF/plant)	0.83	0.10
f_3 (4 g AMF/plant)	0.71	0.06
f_4 (6 g AMF/plant)	0.64	0.07
f_5 (8 g AMF/plant)	0.63	0.05
f_6 (10 g AMF/plant)	0.76	0.07

DRY WEIGHTS

There was no interaction effect observed between shade intensity and AMF dosage in the plant dry weight variables. However, in terms of independent effects, AMF treatment had a significant effect on the plant dry weight, whereas no significant change was observed with varying

shade intensity (Table 9). Based on Table 9, at both 6 and 12 WAP, the f_6 treatment (10 g AMF/plant) gave significantly higher dry root weight results than other treatments. However, the 12 WAP result was not found to be significantly different from treatments f_4 and f_5 . The dry shoot weight at 12 WAP gave the best results with the f_6 (10 g AMF/plant) treatment than other treatments.

TABLE 9. Independent effect of shade intensity and AMF dosage on soybean dry weights of roots and shoot at 6 WAP and 12 WAP

Treatment	Dry weight (g)			
	Root		Shoot	
	6 WAP	12 WAP	6 WAP	12 WAP
Shade intensity				
n_1 (40% shade)	8.17 a	18.30 a	14.68 a	38.99 a
n_2 (0% shade)	9.53 a	18.95 a	14.86 a	40.41 a
AMF dosage				
f_1 (control)	5.49 c	16.91 b	11.34 c	36.13 b
f_2 (2 g AMF/plant)	5.97 c	17.70 b	13.09 bc	37.98 b
f_3 (4 g AMF/plant)	8.13 c	17.03 b	14.20 abc	41.58 b
f_4 (6 g AMF/plant)	8.09 c	18.64 ab	15.14 ab	39.50 b
f_5 (8 g AMF/plant)	11.56 b	19.68 ab	17.13 ab	38.07 b
f_6 (10 g AMF/plant)	14.01 a	21.94 a	17.73 a	44.94 a

Note: The average value followed by the same letter in the same column shows no significant difference according to Duncan's multiple range test at a 95% confidence interval

PLANT YIELD COMPONENTS

Based on Table 10, statistical analysis showed that there was no interaction effect between shade intensity and AMF dosage on the number of pods and the number of seeds per pod. In terms of independent effects, the shade intensity and AMF dosage treatments individually had the same effect on the number of pods per plant, i.e., no significant differences. However, for the effects of independent treatments on the number of seeds per pod, shade intensity did not show any changes. AMF dosage f_6 (10 g AMF/plant) treatment yielded the highest number of seeds per pod than other treatments.

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differences. For the effects of independent treatments on the number of seeds per pod, shade intensity did not show any changes. However, AMF dosage f_6 (10 g AMF/plant) treatment yielded the highest number of seeds per pod than other treatments.

The statistical analysis results showed that there were no interaction effects between shade intensity and AMF dose on pod weight and the weight of 100 grains. In terms of independent effects, there were no statistically significant differences with varying shade intensity. However, for varying AMF dosages, the f_6 (10 g AMF/plant) and f_5 (8 g AMF/plant) treatments gave significantly higher pod weights than the f_1 treatment (control), but with no statistical differences from f_2 , f_3 and f_4 treatments. For the weight of 100 grains variable, the f_6 (10 g AMF/plant) treatment showed a weight of 100 pods significantly higher than for the f_1 (control), f_2 and f_3 (6 g AMF/plant) treatments, but the same as the f_4 and f_5 treatments.

TABLE 10. Effect of shade intensity and AMF dosage on the plant yield components at 12 WAP

Treatment	Number of pods per plant	Number of seeds per pod	Pod weight (g)	Weight of 100 grains (g)
Shade intensity				
n_1 (40% shade)	32.38 a	2.04 a	32.20 a	16.36 a
n_2 (0% shade)	38.25 a	2.10 a	37.08 a	16.55 a
AMF dosage				
f_1 (control)	31.13 a	1.99 b	25.50 b	14.77 c
f_2 (2 g AMF/plant)	31.50 a	2.01 b	32.59 ab	16.07 bc
f_3 (4 g AMF/plant)	33.63 a	2.02 b	33.27 ab	16.21 bc
f_4 (6 g AMF/plant)	33.63 a	2.05 b	34.78 ab	16.84 ab
f_5 (8 g AMF/plant)	39.88 a	2.08 b	37.72 a	17.26 ab
f_6 (10 g AMF/plant)	42.13 a	2.24 a	43.96 a	17.57 a

Note: The average value followed by the same letter in the same column shows no significant difference according to Duncan's multiple range test at a 95% confidence interval

DISCUSSION

Soybean plants grown in shaded conditions were taller than plants grown in an unshaded environment. This is because the plants under shade display the presence of symptoms of etiolation. Etiolation occurs when a lack of sunlight causes the production and distribution of

plant hormone such as the high levels of auxin and lower level of cytokinin (Mubarak et al. 2020b; Nuraini et al. 2021; Rosniawaty et al. 2020), thereby stimulating cell extension and increasing the crop height by more than 25% (Widiastuti et al. 2004).

In terms of AMF dosage treatment, in general, the f_6 (10 g AMF/plant) treatment showed the greatest results compared to other treatments. This is interpreted to be because of the increased number of spores, resulting in more hypha, with a corresponding increase in the area for absorption of water and nutrients. This finding is consistent with Niswati et al. (2011), who found that the application of 10 g AMF/plant can increase plant growth compared to other treatments with lower doses. According to Sinwin et al. (2006), mycorrhiza infections increased the height of the plant due to the increased nutrient intake through increased mass flow.

At 3 to 6 WAP, shade intensity and AMF dosage did not significantly affect the number of trifoliolate leaves. This may be because shaded conditions do not have any specific effect on trifoliolate leaf frequency compared to unshaded conditions. Thus, it is suggested that it is possible to use an intercropping system to grow soybeans under shaded conditions. Taiz and Zeiger (2002) reported that plants increase their efficiency of light capture by increasing leaf area without increasing the number of trifoliolate leaves. This change is suggested to occur to maintain the balance of photosynthesis usage.

The observed increases in plant height, chlorophyll index and stomatal conductance under shading and AMF application indicate that, morphologically and physiologically, the plants were not significantly affected by the shading treatment. This result is in accordance with a study by Chairudin and Sabaruddin (2015), who reported that shading-tolerant traits tend to lead to improvement in morphological traits such as plant height and leaf thickness. According to Evans and Poorter (2001), the leaves of soybeans become thinner and the surface area of the leaf becomes wider in shade-tolerant varieties, with the aim of increasing the area of light absorption and light harvester tissues to optimize the process of photosynthesis. Fan et al. (2019) stated that compared with leaves under normal light (CK) treatment, leaves under shading treatment exhibited decreased palisade and spongy tissue thicknesses but significantly increased cell gap.

The f_6 (10 g AMF/plant) treatment gave the best results in terms of the number of productive branches of the plant, possibly due to the larger amount of spores than under other treatments, indicating that the AMF has a substantial opportunity to colonize the plant roots. Inoculation with AMF or PSF increased nutrient uptake and improved the gas exchange and Chl fluorescence parameters (Zai et al. 2021).

The chlorophyll index values of soybean leaves at 10 WAP showed that AMF application gave better results under 40% shade compared to plants grown without shade. It is suspected that the development of the AMF improved water availability for the plants. The micro-climate in the shaded condition means water availability can be maintained due to lower evaporation rates than unshaded conditions. According to Nopphakat et al. (2022), the AMF spores can grow and develop well in soil with sufficient water availability. The percentage of root infections is lower in limited water conditions because mycorrhiza will remain dormant in the form of spores (Suherman 2008). Thus, higher water availability provides better development opportunities for AMF.

The soybean plants grown under 40% shade had higher chlorophyll index and stomatal conductance values than those grown under 0% shade. This result is thought to be a crop response to low light intensity or low light tolerance. Wang et al. (2021) stated that, in lettuce, the lighting direction had a profound effect on the morphological characteristics, especially in leaf morphology and stomatal characteristics where the plants adapted to the changing lighting environments.

Absorption of P in 40% shade conditions tends to be greater than in unshaded conditions. A different micro-climate causes more water availability in planting conditions under shade, where more groundwater availability leads to higher absorption (Rosi et al. 2016). The increased abundance of water serves to dissolve nutrients and mineral substances so that they are easily absorbed by plants. The treatments with various AMF doses showed higher percentages of P_2O_5 than observed in the control experiment. The application of AMF increased P uptake in plants compared to those without AMF application. This finding is in agreement with the research results of Hasanudin (2002), who identified that with increasing application of mycorrhiza to soybeans, the absorption of N, P, and K on the leaves increased, compared to plants without AMF application.

The dry weight results did not differ in soybeans planted under and without shade. This was presumed to relate to the soybean plants' low light intensity tolerance so that the process of photosynthesis and plant growth could continue optimally in the shade. In soybeans that are sensitive to low light intensity, the dry weight of the plant would be significantly reduced. In terms of AMF dosage, the f_6 treatment (10 g AMF/plant) generally showed the greatest result compared to the other treatments, likely due to the increased likelihood of

AMF symbiosis causing the plant roots to become larger. The symbiosis between the AMF and the plant roots increased the root length and the rooting system. The introduction of mycorrhiza increased the ability of plants to absorb water and nutrients, as indicated by an increase in the number and dry weight of the roots (Jannah 2011).

Regarding the number of pods, the shade intensity treatment and AMF dosage produced similar results, potentially because the number of pods is controlled more strongly by genetic factors than environmental ones. Barmawi et al. (2013) state that the characteristics of flowering age, harvest age, and the number of pods per plant have a high heritability value. This high heritability value indicates that genetic factors are more instrumental in controlling a trait compared to environmental factors. Therefore, in this case, the treatment types do not exhibit a significant effect.

The 40% shade treatment had the same effect as 0% shade on the variation in the number of seeds per pod, the weight of the pod, and the 100-grain weight. It is suspected that this is because the plants are low-light-tolerant. Plant tolerance is characterized by an increase in chlorophyll index (Table 4), stomatal conductance (Table 6) and chlorophyll fluorescence (Table 7). However, the yields of shaded plants show no significant differences in comparison to those grown under full light intensity conditions (Table 10). The change in the morphological character and physiology of the leaves is a form of crop mechanism that allows photosynthesis to be maintained in shaded conditions with optimal production (Evans & Poorter 2001). Soverda (2012) also states that a shade level of 50% in shade-tolerant soy plants does not significantly decline the number of pods, seed size, or seed crops. A plant's tolerance to shade is determined by its ability to carry out the process of photosynthesis, usually in shaded conditions.

The AMF dosage that yielded the best results was the f_6 (10 g AMF/plant) treatment, both in the number of seeds per plant and 100-grain weight. Similarly, the best results for pod weight of pods were achieved using the f_6 (10 g AMF/plant) and f_5 (8 g AMF/plant) treatments. This is because, at higher dosages, the AMF has increased opportunities to form hypha, and symbiotic relationships with the plant will be increasingly greater. Plant-microbe interactions improve the ability of biological inocula involving AM fungi and bacteria to enhance the sustainability of agricultural crops in P-limited conditions (Jiang et al. 2021). The increased absorption of N, P, and K can increase the rate of photosynthesis, which will then increase production. AMF

also has a phosphatase enzyme that converts P from forms that are unavailable to plants (Oktaviani et al. 2014). Damanik et al. (2011) state that P plays an essential role in the body of plants, including cell division, the formation of flowers, fruits and seeds, and improving the quality of crop yields. The research results of Suherman et al. (2012) stated that the AMF administration increased the number of pods compared to treatments without AMF. Sukmasari et al. (2018) also stated that a dose of 10 grams of mycorrhizal per polybag increased the number of seeds per plant and the 100-grain soybean weight.

CONCLUSION

Based on the results of this study, we suggest that an intercropping system between soybeans and oil palm plants is possible with 2-year old oil palm plants without any significant decrease in yields. Moreover, this approach can improve the plants' physiological traits, which is useful for improving yields in future adaptations. The addition of AMF can improve soybean growth and yields due to increases in soil nutrients, especially P.

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REFERENCES

- Barmawi, M., Yushardi, A. & Sa'diyah, N. 2013. Daya waris dan harapan kemajuan seleksi karakter agronomi kedelai generasi f_2 hasil persilangan antara Yellow Bean dan Taichung. *Jurnal Agrotek Tropika* 1(1): 20-24.
- Bolan, N.S. 1991. A critical review on the role of mycorrhizal fungi in the uptake of phosphorus by plants. *Journal Plant Soil* 134(2): 189-207.
- Chairudin, E. & Sabaruddin, D. 2015. Dampak naungan terhadap perubahan karakter agronomi dan morfofisiologi daun pada tanaman kedelai (*Glycine max* (L.) Merrill). *Jurnal Floratek* 10(1): 26-35.
- Damanik, M.M.B., Hanum, H. & Syarifuddin. 2011. *Kesuburan Tanah dan Pemupukan*. Medan: USU Press.
- Davies, F.T., Svenson, S.E., Henderson, J.C., Phavaphutanon, C., Duray, S.A., Olalde, P.C., Meier, E. & Bo, S.H. 1996. Non-nutritional stress acclimation of mycorrhizal woody plants exposed to drought. *Tree Physiology* 16(2): 985-993.
- Direktorat Jendral Perkebunan. 2019. *Statistik Perkebunan Kelapa Sawit Indonesia Tahun 2017 - 2019*. Departemen Pertanian. Jakarta.

- Evans, J.R. & Poorter, H. 2001. Photosynthetic acclimation of plants to growth irradiance: The relative importance of specific leaf area and nitrogen partitioning in maximizing carbon gain. *Plant, Cell & Environment* 24(3): 755-767.
- Fan, Y., Chen, J., Wang, Z., Tan, T., Li, S., Li, J., Wang, B., Zhang, J., Cheng, Y., Wu, X., Yang, W. & Yang, F. 2019. Soybean (*Glycine max* L. Merr.) seedlings response to shading: leaf structure, photosynthesis and proteomic analysis. *BMC Plant Biology* 19(1): 34. <https://doi.org/10.1186/s12870-019-1633-1>
- Hasanudin. 2002. Peningkatan kesuburan tanah dan hasil kedelai akibat inokulasi mikrobia pelarut fosfat dan azotobacter pada ultisol. *Jurnal Ilmu-Ilmu Pertanian Indonesia* 4(2): 97-103.
- Jannah, H. 2011. Respons tanaman kedelai terhadap asosiasi fungi mikoriza arbuskula di lahan kering. *Jurnal Ganec Swara* 5(2): 28-31.
- Jiang, F., Zhang, L., Zhou, J., George, T.S. & Feng, G. 2021. Arbuscular mycorrhizal fungi enhance mineralisation of organic phosphorus by carrying bacteria along their extraradical hyphae. *New Phytologist* 230(1): 304-315.
- Kanno, T., Saito, M., Ando, Y., Macedo, M.C.M., Nakamura, T. & Miranda, C.H.B. 2006. Importance of indigenous arbuscular mycorrhiza for growth and phosphorus uptake in tropical forage grasses growing on an acid, infertile soil from the Brazilian savannas. *Journal Tropical Grasslands* 40(5): 94-101.
- Mawarni, L. 2011. Kajian awal varietas kedelai tahan naungan untuk tanaman sela pada perkebunan kelapa sawit. *Jurnal Ilmu Pertanian* 5(2): 54-59.
- Mubarok, S., Nuraini, A., Wicaksono, F.Y. & Qonit, M.A.H. 2020a. Effects of compost and phosphorus on growth and yield of sweet corn (*Zea mays saccharata*). *Research on Crops* 21(4): 671-675.
- Mubarok, S., Kusumiyati, Fauzi, A.A., Nuraini, A., Rufaidah, F. & Qonit, M.A.H. 2020b. Effect of benzyl amino purine and 1-methylcyclopropene in maintaining rooting quality of chrysanthemum (*Chrysanthemum morifolium* Ramat cv. 'White Fiji') cuttings. *Research on Crops* 21(1): 141-150.
- Niswati, A., Nugroho, S.G., Utomo, M. & Suryadi. 2011. Pemanfaatan mikoriza vasikular arbuskular untuk mengatasi pertumbuhan akibat cekaman kekeringan. Ilmu Tanah Fakultas Pertanian Universitas Lampung. Lampung.
- Nopphakat, K., Runsaeng, P. & Klinnawee, L. 2022. *Acaulospora* as the dominant arbuscular mycorrhizal fungi in organic lowland rice paddies improves phosphorus availability in soils. *Sustainability* 14: 31. <https://doi.org/10.3390/su14010031>
- Nuraini, A., Nugroho, P.S., Sutari, W., Mubarok, S. & Hamdani, J.S. 2021. Effects of cytokinin and paclobutrazol application time on growth and yield of G2 potato (*Solanum tuberosum* L.) medians cultivar at medium altitude in Indonesia. *Agriculture and Natural Resources* 55: 171-176.
- Nuraisah, A., Suherman, C., Ariyanti, M., Nuraini, A. & Soleh, M.A. 2019. Pertumbuhan, hasil, dan karakter fisiologis padi yang diberi pupuk hayati pada pertanaman kelapa sawit belum menghasilkan. *Jurnal Kultivasi* 18(3): 1004-1009.
- Oktaviani, D., Hasanah, Y. & Barus, A. 2014. Pertumbuhan kedelai (*Glycine max*. L. Merrill) dengan aplikasi fungi mikoriza arbuskular (FMA) dan konsorsium mikroba. *Jurnal Online Agroekoteknologi* 2(2): 905-918.
- Pequerul, A., Pérez, C., Madero, P., Val, J. & Monge, E. 1993. A rapid wet digestion method for plant analysis. In *Optimization of Plant Nutrition*, edited by Frago, M.A.C., Van Beusichem, M.L. & Houwers, A. *Developments in Plant and Soil Sciences* 53: 3-6.
- Rosi, A., Niswati, A., Yusnaini, S. & Salam, A.K. 2016. Penentuan dosis dan ukuran butir pupuk fosfat super terbaik untuk mendukung pertumbuhan dan serapan p tanaman kedelai (*Glycine max*[L.] Merrill). *Jurnal Agrotek Tropika* 4(1): 70-74.
- Rosniawaty, S., Anjarsari, I.R.D., Sudirja, R., Harjanti, S.P. & Mubarak, S. 2020. Application of coconut water and benzyl amino purine on the plant growth at second cantering of tea (*Camellia sinensis*) in lowlands area of Indonesia. *Research on Crops* 21(4): 817-822.
- Schenck, N.C. & Schroder, V.N. 1994. Temperature response of *Endogene* mycorrhiza on soybean roots. *Mycologia* 66: 600-605.
- Shukla, A., Kumar, A., Jha, A., Chaturvedi, O.P., Prasad, R. & Gupta, A. 2009. Effects of shade on arbuscular mycorrhizal colonization and growth of crops and tree seedlings. *Agroforest Systems* 76(1): 95-109.
- Sinwin, R.M., Mulyati, & Lolita, E.S. 2006. Peranan kascing dan inokulasi jamur mikoriza terhadap serapan hara tanaman jagung. *Jurnal Jurusan Ilmu Tanah Fakultas Pertanian Universitas Mataram* 3(2): 1-8.
- Soverda, N. 2012. Pengaruh naungan terhadap kandungan nitrogen dan protein daun serta pertumbuhan dan hasil tanaman kedelai. *Jurnal Agroekoteknologi* 5(1): 1-9.
- Suherman, C. 2008. Pertumbuhan benih cengkeh (*Eugenia aromatica*) kultivar Zanzibar yang diberi fungi mikoriza arbuskula dan pupuk majemuk NPK. *Jurnal Agrivigor* 8(1): 50-58.
- Suherman, Iradhatullah, R. & Akhsan, M.A. 2012. Aplikasi mikoriza vesikular arbuskular terhadap pertumbuhan dan produksi tanaman kedelai (*Glycine max* (L.) Merrill). *Jurnal Galung Tropika* 1(1): 1-6.
- Sukmasari, Miftah, D., Wijaya, A.A. & Herdiana, I. 2018. Pertumbuhan tanaman kedelai (*Glycine max* (L.) Merrill) dengan pemberian pupuk hayati konsorsium dan fungi mikoriza arbuskular. *Prosiding Konser Karya Ilmiah Tingkat Nasional Tahun 2018*.
- Taiz, L. & Zeiger, E. 2002. *Plant Physiology*. Tokyo: The Benjamin Cumming Publishing Company Inc.
- Wang, M., Wei, H. & Jeong, B.R. 2021. Lighting direction affects leaf morphology, stomatal characteristics, and physiology of head lettuce (*Lactuca sativa* L.). *International Journal of Molecular Sciences* 22(6): 3157. <https://doi.org/10.3390/ijms22063157>

Widiastuti, L., Tohari & Sulistyarningsih, E. 2004. Pengaruh intensitas cahaya dan kadar daminosida terhadap iklim mikro dan pertumbuhan tanaman krisan dalam pot. *Jurnal Ilmu Pertanian* 11(1): 35-42.

Zai, X.M., Fan, J.J., Hao, Z.P., Liu, X.M. & Zhang, W.X. 2021. Effect of co-inoculation with arbuscular mycorrhizal fungi and phosphate solubilizing fungi on nutrient uptake and photosynthesis of beach palm under salt stress environment. *Scientific Report* 11: 5761. <https://doi.org/10.1038/s41598-021-84284-9>.

*Corresponding author; email: cucu.suherman@unpad.ac.id