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Effect of watering interval and planting media composition on growth, chlorophyll, and flavonoid content of sambiloto (*Andrographis paniculata Nees*.)

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ABSTRACT

Sambiloto (*Andrographis paniculata* Ness.) is a plant rich in natural antioxidants (flavonoid and andrographolides) that had great importance to medicine industry. Drought stress affects the content of secondary metabolites played vital roles as antioxidants. Improving the quality of sambiloto by elevating the flavonoid, can be achieved through drought stress. Moreover, drought stress also affects growth. By utilising marginal lands such as ultisols to cultivate sambiloto, soil amendments is needed to improve fertility. This study aims to investigate the effect of drought stress by various of watering intervals and the composition of planting media on the growth, chlorophyll content, and flavonoids in sambiloto plants. The experimental design was a factorial randomized block design with two factors and three replications. The first factor was the watering time interval, namely watering once every 2 days, watering once every 3 days, watering once every 4 days, and watering once every 5 days. The second factor was the composition of the planting media, namely ultisol soil + chicken manure (1:1/v:v), ultisol soil + rice husk charcoal + chicken manure (2:1:1/v:v:v), ultisol soil + rice husk charcoal + chicken manure (1:1:1/v:v:v), and ultisol soil + rice husk charcoal + chicken manure (1:2:2/v:v:v). The results showed that the combination of watering intervals once every 2 days with the media composition of ultisol soil + rice husk charcoal + chicken manure (1:2:2/v:v:v) or the media composition of ultisol soil + rice husk charcoal + chicken manure (1:1:1/v:v:v) produced a higher shoot root ratio and fresh weight of the inflorescence compared to other treatments. This study suggest that watering interval once every 5 days with the composition of ultisol soil media + rice husk charcoal + chicken manure (1:2:2/v:v:v) have the potential to optimize the flavonoid contents of sambiloto.

Keywords: irrigation, medicinal plant, organic material, secondary metabolites, ultisol.

INTRODUCTION

Sambiloto (*Andrographis paniculata* Ness.) is a medicinal plant used as a raw material for making herbal medicine and medicine (Pribadi, 2009). Sambiloto contains secondary metabolites played major roles as antioxidants such as flavonoids, tannins, saponins, alkaloids, and andrographolides. Natural antioxidants derived from medicinal plants are bioactive compounds that can ward off the harmful effects of free radicals and oxidative stress (Kuntorini et al., 2024), which cause several diseases due to cell damage and diseases such as diabetes, cancer, and hypertension (Astuti et al., 2023). The benefits of sambiloto include relieving flu, strengthening the body's immune system, relieving inflammation and fever, and lowering blood sugar (Rais, 2015).

The demand of sambiloto is a comparatively large as a results of its increasing usage as medicinal plants. Kemala et al. (2003) reported that the traditional medicine industry in Indonesia requires Sambiloto as a raw material of 33.47 tons of dried simplicia or 709.60 tons of wet biomass. In addition, Pribadi (2009) estimated the need for Sambiloto in 2008 for the herbal medicine industry reaches 134 tons simplicia or 939 tons wet biomass. Meanwhile, according to the Badan Pusat Statistik (2023) the production of Sambiloto in Indonesia has decreased from year to year. The production of Sambiloto in 2021 to 2023 respectively was 1930.37 tons, 1751.81 tons, and 708.35 ton. This is due to the production of Sambiloto comes from wild plants with diverse environmental conditions, leading to variations in the quality of the resulting simplicia (Pujiasmanto et al., 2007).

Secondary metabolites play a vital role to enhancing the quality of medicinal plants (Sholikhah, 2016). By serving as antioxidants, these metabolites (including alkoloids, phenolics, and flavonoids) have substansial roles to response drought stress (Muthusamy & Lee, 2024). The state of drought stress occurs when the plant environment does not receive enough water, while water is an essential component in growth and nitrogen mineralization materials for plant absorption (Rusmayadi and Budianto, 2009). Several authors have been reported that drought stress may cause the increase of secondary metabolites in various plants. Tran and Le (2022) found an increment the flavonoid contents in fish mint (*Houttuynia cordata* Thumb.) after 7 days of moderate drought stress treatment (65% field capacity/FC). Additionally, *Amaranthus tricolor* cultivated under severe drought stress (30% FC) and moderate drought stress (60% FC) showed an increase in flavonoid and phenolic concentrations compared with the control (Sarker and Oba, 2018). Thus, improving the quality of Sambiloto can be done by providing drought stress. However, drought stress also provides a physiological response followed by plant morphological changes (Sujinah and Jamil, 2016). The longer the drought stress decreases the growth and dry weight of the plant (Anggraini et al., 2015).

Planting media in organic fertilizer and applying organic silica can reduce the impact of drought stress (Shi et al., 2016). Chicken manure is an organic fertilizer containing the elements N (1.72%), P (1.82%), K (2.18%), Ca (9.23%) and Mg (0.86%) (Tufaila et al., 2014), while organic silica is found in rice husk charcoal containing SiO_2 (52%), C (31%), K (0.3%), N (0.18%), F (0.08%) and Ca (0.14%) (Dharmasika et al., 2019). These organic materials can be ameliorants on marginal land, including Ultisol. Ultisol is a type of soil that has low nutrients, namely low nitrogen content (0.06%), very high phosphorus (325 ppm), deficient potassium (0.16%), and magnesium (0.24 Meq 100 g)$^{-1}$) (Sukarman et al., 2022), and this has an impact on decreasing plant production (Arimbawa et al., 2024). According to research by Murtilaksono et al. (2022), 120 grams of chicken manure per plant gave the highest results in plant height, number of leaves, fresh weight, and dry weight of sambiloto inflorescence. Similarly, it has also been found by Pertiwi et al. (2023), where the addition of 7.5 tons ha^{-1} of rice husk ash can increase 3.3% of flavonoid content in dayak onion plants. Therefore, research needs to be done to investigate the composition of the proper planting media and more efficient water use in increasing physiological indicators and sambiloto flavonoid content in ultisol soil. This study aims to examine the differences in watering time intervals compared to the composition of the planting media to determine the best physiological and flavonoid indicators in sambiloto.

MATERIALS AND METHODS

This research was conducted from January to December 2024 located in Banjarbaru, South Kalimantan Province, Indonesia. The seeds of Sambiloto used in this study were Sambina variety. The seeds were germinated in a mixture of rice husk charcoal and chicken manure soil for 4 weeks. The plants were then grown in varying planting media composition. After 2 weeks, the plants subjected to four levels of watering interval for 4 weeks.

This study used a randomized block factorial design with two factors and three replications. The first factor was the watering time interval consisting for 4 levels, namely watering once every 2 days, watering once every 3 days, watering once every 4 days, and watering once every 5 days. The second factor was the composition of planting media consisting 4 levels, namely, ultisol soil + chicken manure (1:1/v:v), ultisol soil + rice husk charcoal + chicken manure (2:1:1/v:v:v), ultisol soil + rice husk charcoal + chicken manure (1:1:1/v:v:v), and ultisol soil + rice husk charcoal + chicken manure (1:2:2/v:v:v).

The observation variables are leaf area (mm^2), fresh weight of the inflorescence (g), shoot root ratio, leaf chlorophyll (mg/L), and flavonoid (%). For leaf chlorophyll quantification, the optical density of the leaf extract was read at 645 nm and 663 nm with a spectrophotometer. The concentrations of chlorophyll were calculated according to the formula by Arnon (1949). The flavonoid quantification according to $AlCl_3$ method by Chang et al. (2002). The extract absorbance was measured at 415 nm with a spectrophotometer.

The observation results were represented as mean of the three replications. The data were analyzed by analysis of variance using the F test at the 5% and 1% significance levels. If the Analysis of Variance results showed the significant effects of treatment, it is continued with Duncan's Multiple Range Test (DMRT) at the 5% significance level.

RESULTS AND DISCUSSIONS

Leaf area (mm^2)

Analysis of variance in plant leaf area shows that the interaction between watering and planting media composition had no significant effect. Meanwhile, the single factors of watering interval and planting media composition show a significant effect. The effect of single factors of watering interval and planting media composition on leaf area can be seen in Table 1 and Table 2.

Based on Table 1, watering once every 2 days had the highest leaf area compared to other watering interval treatment. It is considered that watering once every 2 days provides the plants with sufficient water in the planting medium. This results also indicated that increasing the watering interval decreased leaf area of Sambiloto. This is supported by Tran and Le (2022), which showed that fish mint (*H. cordata* Thumb.) with well-watered condition (85% FC) had the highest leaf area compared to mild drought stress (75% of FC), moderate drought stress (65% FC), and severe drought stress (55% FC). Similar results also observed in *Abelmoschus esculentus*, where watering once in 3 days showed the highest leaf area compared to other treatments (5 and 7 days). Meanwhile, watering once in 7 days had the lowest leaf area (Fernando et al., 2021).

Table 1. The effect of watering intervals on the leaf area of sambiloto (mm^2)

Watering interval	Leaf area (mm^2)
2 days	2012.28^{d}
3 days	1520.07^{c}
4 days	1229.75^{b}
5 days	878.10^{a}

Note: the averages with the same superscript sign show no difference based on DMRT at the α = 0.05 level.

According to Acharya and Assmann (2009), drought stress promotes ABA (abscisic acid) accumulation, which inducees the closure of stomata to reduces water loss. Peters et al. (2018) stated that the closure of stomata causes reduction of CO_2 intake by leaves, thereby reduceing the transpiration rate. Due to less CO_2 availability in the leaves, the rate of photosynthesis decreases significantly (Zheng et al., 2019). Moreover, drought stress lead reductions in leaf pressure turgor, which results in decreased cells expansion and elongation (Coussement et al., 2021). In addition, Harwati (2007) mentioned that leaf area development depends on water. If the plant's water requirements are supplied, leaf area development will be more significant. Hence, water shortage in sambiloto causes a reduction in leaf area.

Based on Table 2, the planting media treatment of ultisol soil composition + rice husk charcoal + chicken manure (1:2:2/v:v:v) were not significantly different from the ultisol soil composition + rice husk charcoal + chicken manure (1:1:1/v:v:v). The treatment were higher than the ultisol soil composition + rice husk charcoal + chicken manure (2:1:1/v:v:v), and ultisol soil + chicken manure (1:1/v:v) in the observation of leaf area. This is thought to be due to the content of nutrients, especially nitrogen in the composition of ultisol soil + rice husk charcoal + chicken manure (1:2:2/v:v:v) which was 0.79% and the composition of ultisol soil + rice husk charcoal + chicken manure (1:1:1/v:v:v) which is 0.58%

Table 2. The effect of media composition on the leaf area of sambiloto (mm^2)

Planting media composition (m)	Leaf area (mm^2)
Ultisol soil + chicken manure (1:1/v:v)	1259.63^{a}
Ultisol soil + rice husk charcoal + chicken manure (2:1:1/v:v:v)	1351.19^{ab}
Ultisol soil + rice husk charcoal + chicken manure (1:1:1/v:v:v)	1459.54^{bc}
Ultisol soil + rice husk charcoal + chicken manure (1:2:2/v:v:v)	1569.82^{c}

Note: the averages with the same superscript sign show no difference based on DMRT at the α = 0.05 level.

which was higher than the composition of ultisol soil + rice husk charcoal + chicken manure (2:1:1/v:v:v) which was 0.52%, and ultisol soil + chicken manure (1:1/v:v) which was 0.45% supported the development of sambiloto leaf area. Acoording to Rusmayadi et al. (2023), the nutrient N can play a role in the composition of chlorophyll and support photosynthesis activity, so that it can increase the development of leaf meristematic tissue, one of which is in the development of leaf area. Asman et al. (2023) stated that using rice husk charcoal and chicken manure can increase the leaf area of plants.

Fresh weight of inflorescence (g)

The interaction between watering and the media, the single factor of watering, and the composition of the planting media each have a very significant effect on the fresh weight of the Sambiloto inflorescence. The effect of the interaction of watering with the planting media composition can be seen in Table 3.

Based on Table 3, the treatment of watering once every 2 days in the planting media composition of Ultisol soil + rice husk charcoal + chicken manure (1:1:1/v:v:v) was no different with the planting media composition of ultisol soil + rice husk charcoal + chicken manure (1:2:2/v:v:v). The treatment produced a higher fresh weight of inflorescence compared to other combination treatments. It indicated that the treatment of watering once every 2 days provides sufficient water content for the plants, thereby ensuring optimal cell turgor and contributing to plant biomass growth. This is supported by the treatment of ultisol media + rice husk charcoal + chicken manure (1:2:2/v:v:v) and the composition of ultisol soil + rice husk charcoal + chicken manure (1:1:1/v:v:v) which is suspected that the addition of rice husk charcoal and chicken manure can improve the condition of ultisol soil which tends to be acidic and poor in nutrients so that it can increase the fresh weight of sambiloto. This is also supported by Larasati et al. (2020), watering once every 2 days can increase plant height, number of leaves, fresh weight, and dry weight of sambiloto. According Jali et al. (2022), using rice husk biochar and chicken manure can increase plants' diameter and fresh weight. Watering once every 5 days resulted a lower fresh weight of plants than other watering treatments. This is related to plants experiencing drought stress, which causes in disrupted growth and a decrease in fresh weight. Similarly, Han et al. (2024) stated that drought stress causes a decrease in plant morphology, such as the fresh weight of plants.

Shoot root ratio

Analysis of variance on shoot root ratio (SRR) shows the interaction of watering with the planting media composition, and the single factor of watering each has a very significant effect. On the other hand, the effect of the planting media composition is not significantly different. The effect of the interaction of watering with the planting media composition can be seen in Table 4.

Based on Table 4, the treatment of watering every 2 days in the planting media composition of ultisol soil + rice husk charcoal + chicken manure (1:1:1/v:v:v) was no different with the planting media composition of ultisol soil + rice husk charcoal + chicken manure (1:2:2/v:v:v). The shoot root ratio (SRR) of sambiloto was higher than other combination treatments. It showed that watering every 2 days enables the growth of the upper part (leaves and stems) and the lower part (roots) of the plant. Additionally, the presence of organic materials such as chicken manure and rice husk charcoal which have the nutrients needed by plants can support the growth of sambiloto.

Table 3. The effect of interaction between watering and the planting media composition on the fresh weight of sambiloto inflorescence (g)

Watering interval	Planting media composition			
	Ultisol soil + chicken manure (1:1/v:v)	Ultisol soil + rice husk charcoal + chicken manure (2:1:1/v:v:v)	Ultisol soil + rice husk charcoal + chicken manure (1:1:1/v:v:v)	Ultisol soil + rice husk charcoal + chicken manure (1:2:2/v:v:v)
2 days	83.70^{d}	105.18^{e}	123.68^{f}	118.38^{ef}
3 days	67.83^{d}	58.48^{d}	79.78^{d}	72.65^{d}
4 days	40.60^{bc}	42.57^{bc}	51.07^{c}	48.56^{c}
5 days	23.46^{a}	27.94^{ab}	36.49^{abc}	36.18^{abc}

Note: The averages with the same superscript sign show no difference based on DMRT at the α = 0.05 level.

Table 4. The effect of interaction between watering and planting media composition on the shoot root ratio of sambiloto

Watering interval	Planting media composition			
	Ultisol soil + chicken manure (1:1/v:v)	Ultisol soil + rice husk charcoal + chicken manure (2:1:1/v:v:v)	Ultisol soil + rice husk charcoal + chicken manure (1:1:1/v:v:v)	Ultisol soil + rice husk charcoal + chicken manure (1:2:2/v:v:v)
2 days	5.01^{cde}	5.16^{de}	5.44^{ef}	6.39^{f}
3 days	4.37^{bcde}	4.64^{bcde}	4.75^{bcde}	4.6^{bcde}
4 days	3.98^{abcd}	3.89^{abc}	4.05^{abcd}	4.14^{abcd}
5 days	3.82^{abc}	3.63^{ab}	3.59^{ab}	3.08^{a}

Note: The averages with the same superscript sign show no difference based on DMRT at the α = 0.05 level.

According Wafa et al. (2023), the provision of chicken manure has a significant effect on the shoot ratio on marginal dry soil. Moreover, Wasis & Prihanto (2023) stated that plants that are deficient in certain nutrients and minerals will increase root growth to obtain water in the inner part of the planting medium, resulting in a low root shoot ratio value. In addition to the availability of optimal nutrients in the composition of the media, such as manure and rice husk charcoal, it can support plant growth through photosynthesis and transpiration. Thus, the biomass accumulation in the shoot part causes an enormous root shoot ratio value.

Chlorophyll (mg/L)

Chlorophyll ratio A/B

The interaction of watering with planting media and the single factor of planting media shows no significant effect, and the watering time has a very significant effect. The effect of the interaction of watering with the composition of planting media can be seen in Table 5.

Table 5 showed that watering once every 2 days had the highest chlorophyll A/B ratio compared to other watering treatmets. It showed that the water content at the watering interval of once every 2 days was sufficient for the plants, so the chlorophyll A content was higher than chlorophyll b. Meanwhile, watering once every 5 days had the lowest chlorophyll A/B ratio. It showed that the chlorophyll A content was lower than chlorophyll B. These results indicated that the water stress causes a decrease in the ratio of chlorophyll A/B, as a result of the proportion of chlorophyll B increases due to water stress. Trukhachev et al. (2022) stated that the increase in the proportion of chlorophyll B in drought stess is directed to help chlorophyll A to increase photosynthesic activity. Chlorophyll A is the primary pigment in photosynthesis (Viljevac et al., 2013). In contrast, chlorophyll B is an accessory pigment that collects light energy and transfer it to chlorophyll A (Nobel, 2009).

Table 5. The effect of watering on chlorophyll ratio A/B of sambiloto

Watering interval	Chlorophyll ratio A/B
2 days	1.179^{d}
3 days	1.139^{c}
4 days	0.961^{b}
5 days	0.868^{a}

Note: The averages with the same superscript sign show no difference based on DMRT at the α = 0.05 level.

According to research by Maisura et al. (2014), drought condition treatments to 8 rice varieties with moisture content (50%) cause an increase in chlorophyll B, causing the chlorophyll A/B ratio content in the plants to decrease. Similarly, Gadoum et al. (2019) reported that the lowest chlorophyll A/B ratio in *Ceratonia siliqua* L. was recorded in severe drought stress (40% FC) in *Ain Sefra's* ecotype compared to moderate stress (60% FC) and control (100% FC). Somtrakoon et al. (2022) reported that peanut plants under low water condition causes the chlorophyll A content to be greater than the chlorophyll A.

Total chlorophyll (mg/L)

The effect of interaction between watering and media composition is not significantly different from the total chlorophyll content of sambiloto. At the same time, the single factors of watering interval and planting media composition have a very significant effect. The effect of watering interval and media composition on total chlorophyll content can be seen in Table 6 and Table 7.

Based on Table 6, watering once every 2 days showed the highest number of total chlorophyll compared to other treatments. It indicated that the water content at the watering interval of once every 2 days was sufficient for the plants. It affects the amount of plant chlorophyll because more frequent watering enables plants to maintain an efficient photosynthesis process, thereby increasing chlorophyll production. Conversely, insufficient watering can cause stress in plants, resulting in a decrease of total chlorophyll. According to research by Mafakheri et al. (2010), drought stress applied in three chickpea cultivars significantly decreased chlorophyll A, chlorophyll B, and total chlorophyll. The decrease in chlorophyll levels occurs due to plant adaptation to reduce or prevent high ROS content to suppress cell death due to ROS attack when stressed by drought (Putri et al., 2022).

Based on Table 7, the treatment of ultisol + rice husk charcoal + chicken manure (1:1:1/v:v:v) planting media composition was no different with the ultisol + rice husk charcoal + chicken manure (1:2:2/v:v:v) planting media composition. The treatment were higher than other combination treatments. The result indicated that the combination of chicken manure with rice husk charcoal provides sufficient plant nutrition, thus supporting an increase in total plant chlorophyll. According to Monteoliva et al. (2021), organic materials with higher nitrogen nutrients, such as chicken manure, can increase the total chlorophyll content in leaves. Similar results also observed on melons, where combining chicken manure and rice husk charcoal media can increase total leaf chlorophyll in sandy soil (Susanti et al., 2024).

Table 6. The effect of watering interval on total chlorophyll content of sambiloto

Watering interval	Total chlorophyll (mg/L)
2 days	2.319[b]
3 days	2.272[a]
4 days	2.262[a]
5 days	2.248[a]

Note: The averages with the same superscript sign show no difference based on DMRT at the α = 0.05 level.

Flavonoid (%)

Analysis of variance shows the interaction of watering with media composition, and each single factor significantly affects the flavonoid content of sambiloto. The effect of the interaction of watering with the planting media composition can be seen in Table 8.

Based on Table 8, watering treatment once every 5 days in the planting medium of ultisol soil composition + rice husk charcoal + chicken manure (1:2:2/v:v:v) have higher flavonoid content compared to other combination treatments. Watering interval every 5 dayscan cause plants to experience plant stress, which may trigger an increase in flavonoid production as an adaptive response. Flavonoids often function as defense compounds against environmental stress. This is supported by Larasati et al. (2020), which states that the less often Sambiloto are watered, the flavonoid content in Sambiloto increases. Meanwhile, the planting media composition containing more rice husk charcoal than other treatments is thought to contain more organic silica, thus supporting the formation of flavonoids in Sambiloto. According to Utami et al. (2020), silica in the planting medium can increase the total flavonoids of Binahongs, namely 0.59%. Silica can manipulate a series of physiological and biochemical processes related to the activity of plant antioxidant formation. Applying silica can stimulate antioxidant activity and synthesis, thereby stimulating the formation of higher antioxidant compounds. Pinedo-Guerrero et al. (2020) stated that silica application can increase flavonoid content in salinity stress. Silica can regulate the expression of many genes, including enzyme activity such as PAL

Table 7. The effect of planting media composition on total chlorophyll content of sambiloto

Planting media composition	Total chlorophyll (mg/L)
Ultisol soil + chicken manure (1:1/v:v)	2.165[a]
Ultisol soil + rice husk charcoal + chicken manure (2:1:1/v:v:v)	2.250[b]
Ultisol soil + rice husk charcoal + chicken manure (1:1:1/v:v:v)	2.329[c]
Ultisol soil + rice husk charcoal + chicken manure (1:2:2/v:v:v)	2.335[c]

Note: The averages with the same superscript sign show no difference based on DMRT at the α = 0.05 level.

Table 8. The effect of interaction between watering and planting media composition on the flavonoid content of sambiloto (%)

Watering interval	Planting media composition			
	Ultisol soil + chicken manure (1:1/v:v)	Ultisol soil + rice husk charcoal + chicken manure (2:1:1/v:v:v)	Ultisol soil + rice husk charcoal + chicken manure (1:1:1/v:v:v)	Ultisol soil + rice husk charcoal + chicken manure (1:2:2/v:v:v)
2 days	0.09[a]	0.12[ab]	0.13[bc]	0.13[bc]
3 days	0.15[c]	0.23[d]	0.24[d]	0.26[d]
4 days	0.32[e]	0.32[e]	0.32[e]	0.33[e]
5 days	0.39[f]	0.42[fg]	0.43[g]	0.46[h]

Note: The averages with the same superscript sign show no difference based on DMRT at the α = 0.05 level.

(phenylalanine ammonia lyase), by increasing the ascorbic acid-glutathione cycle and enzyme activity. According to Kim et al. (2017), plants that have experienced abiotic stress and silica application can induce stress tolerance by regulating ROS formation. When ROS is formed in plants due to water stress, the plants will signal the formation of flavonoid compounds (antioxidants) as a defense, where flavonoid compounds can help protect plants from oxidative damage and cell stability formed in secondary metabolism.

The planting media composition ultisol soil + rice husk charcoal + chicken manure (1:2:2/v:v:v) has a higher N concentration (0.79%) than other planting media compositions. Kováčik and Klejdus (2014) reported that nitrogen significantly affects the regulation of the secondary metabolic pathway involved in the synthesis of flavonoids as antioxidants. Thus, plants that get enough nitrogen can increase the flavonoid content. Kurniawan et al. (2014) stated that higher N absorption will encourage the enzyme that forms flavone compounds to be more optimal in increasing phenolic compounds and flavonoid content in plants. The provision of chicken manure given at 10.08 tons ha^{-1} gives the best results in higher total phenolic content in white mugwort plants (Thepsilvisut et al., 2022).

CONCLUSIONS

Increasing the watering intervals from 2 days to 5 days significantly increased flavonoid contents but significantly decreased leaf area, fresh weight of inflorescence, shoot root ratio, chlorophyll A/B ratio, and total chlorophyll. The treatment of the watering interval once every 2 days with the composition of ultisol soil media + rice husk charcoal + chicken manure (1:2:2/v:v:v) the composition of ultisol soil media + rice husk charcoal + chicken manure (1:1:1/v:v:v) was greater than other treatments in the observation of the fresh weight of inflorescence and shoot root ratio. The combination of watering intervals once every 5 days with the media composition of ultisol soil + rice husk charcoal + chicken manure (1:2:2/v:v:v) produced the highest flavonoid content, namely 0.46%.

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