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STUDIES OF SHADING LEVELS AND NUTRITION SOURCES ON GROWTH, YIELD AND ANDROGRAPHOLIDE CONTENT OF SAMBILOTO (*Andrographis paniculata* Ness)

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ABSTRACT

Growth and biochemical content of medicinal crops are influenced by agroecosystems characteristics. The objective of this research was to determine the optimum shading level and type of fertilizer as sources of nutrition on the growth, yield, and andrographolide content of sambiloto. The experiment used Split Plot Design with basic design of Randomized Complete Block Design arranged with two treatment factors and three replications. The first factor as the main plot was shading levels, namely without shading, 25% shading, 50% shading, and 75% shading. The second factor as the sub plot was sources of nutrition represented by type of fertilizer, namely NPK fertilizer, cow stable fertilizer, and compost fertilizer. The result of research indicated that shading level and the kind of nutrition influenced some growth and yield variables such as number of leaves, number of branches, plant height, plant dry weight and simplisia weight, and andrographolide content. Interaction of shading level at 25% and straw compost fertilizer performed best in growth characteristics, while the highest andrographolide content resulted from the treatment combination of 50% shading level and straw compost fertilizer.

Keywords: fertilizer,nutrition, shading, andrographolide, *Andrographis paniculata*

INTRODUCTION

Indonesia is a tropical country that has diverse plant and is the second largest mega center of biodiversity after Brazil. It is estimated there are about 30,000-40,000 species of flowering plants found in Indonesia and about 1,000 plants of which have been used as medicinal ingredients (Kosela, 1998).

Nowadays people tend to consume drugs derived from plants. This is because medicinal plants have mild side effects and more affordable prices. In addition, there is a global issue "back to nature" for drug consumption, and supporting a strong social culture in the use of traditional medicine. Medicinal plant development prospects quite bright views of the potential aspects of flora, fauna, climate, and soil as well as aspects of industrial development of traditional medicines and cosmetics.

Sambiloto (*Andrographis paniculata* Ness) is a nutritious crops as antifever, antibiotics, antiperitrik, anti-inflammatory, antiswollen, anti-diarrheal, antitumor and hepatoprotective (Heyne, 1987; Winarto, 2003). Sambiloto that is continuously harvested from agro-forestry trees can become extinct (Hanan, 1996; Anonymous, 2001; Muliawati, 2002; Winarto, 2003; Januwati, 2004). To overcome the destruction proper cultivation is required. The needs of sambiloto plant as raw materials of traditional pharmaceutical and phytopharmaca continue to increase, but the quantity of sambiloto harvest farmers is uncertain.

The virtue of sambiloto plant lies on its secondary metabolites (andrographolide). Because of the importance andrographolide as active ingredient drug in sambiloto plant, it is necessary to study fertilization, the level of shade, and water stress so that the growth, yield and andrographolide content can be increased. The problem raised in this research was how to cultivate Sambiloto in suitable agro-climate for the growth and development to obtain the biochemistry content of sambiloto plant or maximum secondary metabolites.

This study was aimed to examine the effect of fertilizer types and levels of shading on growth, yield and andrographolide content of sambiloto.

MATERIALS AND METHODS

This research is a field experiment followed by determination of andrographolide in the laboratory. The research was conducted during wet season of 2008 in Sukoharjo Central Java region with elevation of 86 m above sea level, and with climate type C. Materials used were cow stable fertilizer, straw compost fertilizer (straw compost was sprayed 10 ml of EM4 in one liter of water), vertisol type of soil, polybag, sambiloto seed, paranet 25%, 50% and 75%, NPK compound fertilizer with 16-16-16 compositions. The equipment used were leaf area meter, flux meter, electric scales, ovens, sprayer, plastic hose, gauge rolls, saws, scythes and stationery.

The research design applied was Split Plot Design with basic design of Randomized Complete Block Design with three replications. The main plot was the level of shade comprising four levels, i.e. not shaded , shaded by 25% (A2), shaded by 50% (A3), and shaded by 75% (A4). The sub plot was the kind of organic compost which comprised three levels, namely NPK compound fertilizer (P1), cow stable fertilizer (P2), and straw compost (P3). So there were 12 combinations of treatments with three replications, and each treatment consisted of nine plants. The data obtained were analyzed by analysis of variance (ANOVA) using F test at 5% level. Duncan multiple range test (DMRT) at 5% level would be applied if there was significant influence among the treatments .

RESULTS AND DISCUSSION

Growth in Different Shading Levels and Nutrition Sources

Plant growth is influenced by environmental factors such as radiation, temperature, soil fertility, and so forth. The result of this research indicated that the optimum growth of sambiloto required shading treatment which was shown by the lower magnitude of some growth variables in without shading condition (full radiation). In the other words, this plant needed shading for better growth. However, it would reduce plant growth when the level of shading was more than 50%, which may be due to dramatic reduction of photosynthesis affected by the decrease of solar radiation. The average of some growth variables at the treatments of shading level is presented in Table 1.

Analysis results in table 1, shows that all shaded plant had higher growth than the non shaded one. In the other word, sambiloto plant needed shading during its growth with shading level of 25% to 75%.

Plants that grow in the shade tend to have elongated growth due to long segment of stem composed of thin-walled cells, larger intercellular spaces and fewer transport tissue and binding tissue. This can be caused by the activity of auxin (Boardman, 1977).

Table 1. Plant height, branches number, leaves number, and leaf area on shading treatments

Shade treatments	Plant height (cm)	Branches number	Leaves number	Leaf area (cm2)
0%	29.37 a	11.92 b	96.79 b	629 a
25%	38.20 b	13.26 b	121.33 c	752 a
50%	36.38 b	12.80 b	106.03 bc	646 a
75%	39.98 b	9.29 a	80.50 a	562 a

Ramarks = The value followed by same letter at the same column is non significant different at 5% of DMRT.

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The response of leaves number indicated that shading increased number of leaves to the level of 50%; however, the trend decreases when the shading level was more than 50%. Differences in the number of leaves on the plant in the shade were influenced by the different light intensities obtained. Plants received the appropriate intensity of light would lead to the achievement of balance in the body between the transpiration in the leaves of plants with water and mineral absorption by plant roots, so plant growth and development will be perfect (Sulandjari *et al*., 2005).

However, the average leaf area among the levels of shading treatments was not significantly different, but it was still consistently the highest average leaf area obtained in 25% shading treatment (A2). Maintenance of large leaf area is important to ensure a high carbon input and solar interception for photosynthesis. Kaspar and Bland (1992) stated that plants that receive the appropriate light intensity improve nutrient absorption of plant through soil temperature and root extension.

The effect of nutrition sources which is represented by the type of fertilizer used on some growth variables are presented on Table 2. The effect of fertilizer treatment on plant height showed the treatment of cow stable fertilizer treatment (P2) and straw compost (P3) were higher than that of NPK compound fertilizer (P1). The result indicated that the organic nutrition were more suitable for optimum growth of the medicinal plant, especially for good practices.

Figure 1 illustrates the interaction effect of shading treatment and nutrition sources on number of branches. The number of leaves were based on the interaction of fertilizer types and shading level as presented in Figure 2. As we can see in Figure 2, the average of leaves number on the A2P3 treatment majority (25% shade and compost fertilizer) was 139.7, and the smallest number of leaves on the A4P2 treatment (75% shade + compost) was74.83.

Average of fresh and dry weight of plants treated on the shading levels is presented in Table 3.

The environmental condition such as shade is affected for sambiloto plant growth. A good crop development will add to the amount of plant biomass. The availability of macro nutrients, especially N, P and K in sufficient quantity and balance will improve the vegetative growth. Solar radiation with proper light intensity, light quality and duration of exposure is an important element for plants. When the received light intensity is low, then the amount of light received by each leaf surface area within a specified time is low as well. The lack of light results in metabolism disorder, causing decrease in the rate of photosynthesis and carbohydrate synthesis. Conversely, if the light intensity is too high then the plant can suffer from high temperature stress or drought stress.

Analysis results for dry weight of plant showed that the shading level of 75% was the lowest (Table 3).Plants receive the appropriate intensity of light will lead to the achievement of balance in the body between the transpiration in the leaves of plants with water and mineral absorption by the roots perfectly. Plants experiencing water stress would result in increased body temperature, increased CO_2 compensation point, and the carboxylase enzyme was more responsive to the oxygen because the enzyme was amphoteric and functioned as oxygenase. Oxygenase activities ultimately led to the increase of photorespiration which caused the decreased results of net photosynthesis (Salisbury and Ross, 1992). Shading reduces the primary radiation which is active in photosynthesis, which resulted in the decrease of net assimilation (Lambers *et al*., 1992), so that the photosynthates are stored in the storage organs like roots decreased (Schaffer, 1996), resulting in decreased plant dry weight.

Table 2. The average of plant height, leaf area, and root length on type of fertilizer treatments

Treatments	**Plant height (cm)**	**Leaf area (cm2)**	**Root length (cm)**
P1	30.52 a	624 a	16.25 a
P2	38.74 b	627 a	17.29 a
P3	38.68 b	592 a	17.71 a

Ramarks= The value followed by same letter at the same column is non significant different at 5% DMRT

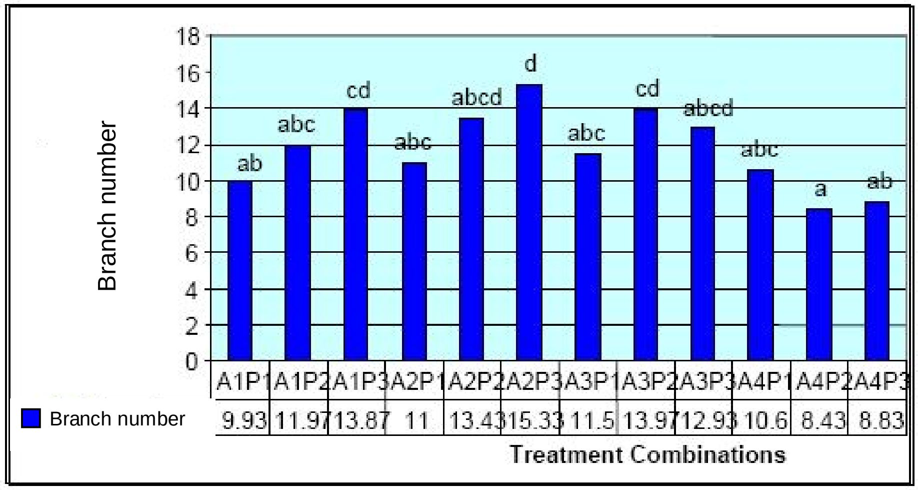

Figure 1. Histogram of branch number on several treatment combinations.

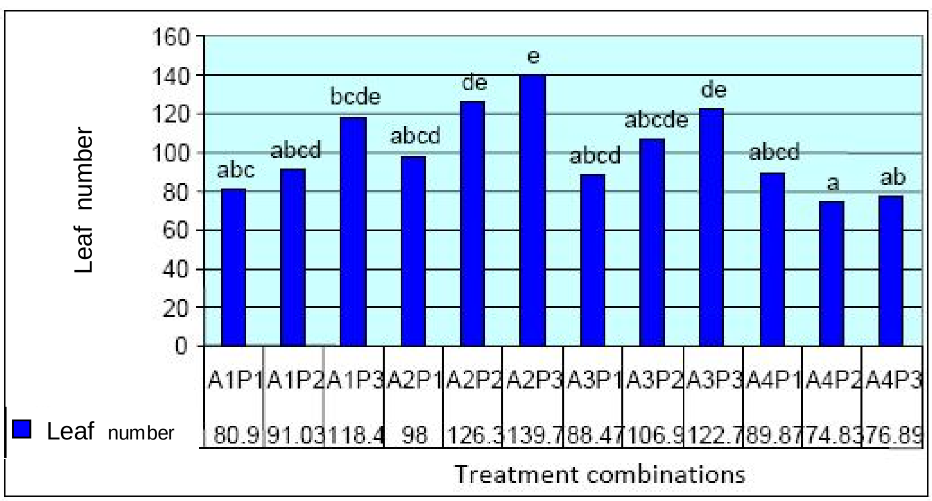

Figure 2. Histogram of leaf number on several treatment combinations.

Pujiasmanto *et al*. (2007) states that the sambiloto herbs that grow in medium land have relatively higher morphology (60-125 cm) than those in lowlands (40-90 cm) and medium land (20-60 cm). The leaves are also wider in the secondary plateau. Andrographolide content in middle plateau is also higher (2.27%), than that in lowlands (1.37%), high plateaus and plains (0.89%).

Table 3 also shows that the mean relative growth rate in the non-shading treatment was significantly different from 25% shading treatment, shaded by 50%, and shaded by 75%. The highest relative growth rate was 25% treatment.

Relative growth rate illustrates the increase in plant dry weight in the interval of time, in conjunction with its initial weight. Decline in relative growth rate occurs due to differences in the rate of photosynthesis. The process of photosynthesis will be disturbed if the water is not available for plants. Decrease in the photosynthetic process occurs due to an increase of stomata diffusion and non-stomata barriers. Decline in relative growth rate is also due to the slow growth of vegetative organs.

Andrographolide Content in Different Level of Shading and Nutrition Sources

Secondary metabolit is very important biochemical compound that is produced by medicinal plants, and the most important secondary metabolite produced from sambiloto plant is andrographolide levels.

Analysis result for andrographolide content as affected by the combination of shading level and type of fertilizer is presented in figure 3, where we can see that the highest average of andrographolide concentrations obtained in 50% shaded and straw compost was 1.07%, and the lowest in not-shaded and NPK fertilizers was 0.26%.

This result indicates that this plant is suitable in term of growth, and is able to produce secondary metabolites under shading condition and organic nutrition application.

As a secondary metabolite compound, alkaloids are compounds from plants that occur naturally, have a basic character and contain at least one nitrogen atom forming part of a cycle system (Aerts and Verpoorte, 1992). This compound is a natural compound that is alkaline and contains one or more nitrogen atoms. Usually, the nitrogen atom in the alkaloid is in the cyclic system, but there is also a nitrogen atom bound beyond cyclical system (Hashimoto and Yamada, 1994).

Fertilization can increase the growth and alkaloid content, which increases N supply, resulting in higher accumulation of alkaloids. According to Herms and Mattson (1992), those environmental conditions that suppress the growth will increase the synthesis of secondary metabolites in the roots as a defensive reaction. Secondary metabolite biosynthesis is controlled by the amount and kinds of enzymes, so that its activity is strongly influenced by environmental factors, particularly temperature and humidity, whereas carbohydrate as a result of assimilate is a precursor.

CONCLUSIONS AND SUGGESTION

The result of research indicated that shading level and the kind of nutrition influenced some growth and yield variables such as number of leaves, number of branches, plant height, plant dry weight and simplisia weight, and andrographolide content .Interaction of shading level at 25% and straw compost fertilizer performed best in growth characteristics, while the highest andrographolide content resulted from the treatment combination of 50% shading level and straw compost fertilizer. The other important point that can be concluded is that sambiloto plant needed shading at the range of 25% to 50% for optimum growth and andragrapholide production. Moreover, it was indicated that organic fertilizer exhibited a potential sources of nutrition for medicinal plant.

Table 3. The average of fresh weight of plant, dry weight of plant, simplisia weight, relative growth rate, and andrographolide content on shading level treatments

Treatments	**Fresh weight of plant (g)**	**Dry weight of plant(g)**	**Simplisia weight (g)**	**Relative growth rate (**g/g/week)	**Andrographolide content (%)**
0%	17.80 a	5.92 b	5.66 a	0.71 b	0.47 b
25%	25.40 a	7.24 b	6.58 a	0.81 c	0.76 c
50%	19.80 a	6.30 b	5.85 a	0.58 a	0.86 c
75%	19.20 a	5.49 a	5.03 a	0.55 a	0.42 a

Remarks = The value followed by the same letter in the same column is non significant different at 5% of DMRT.

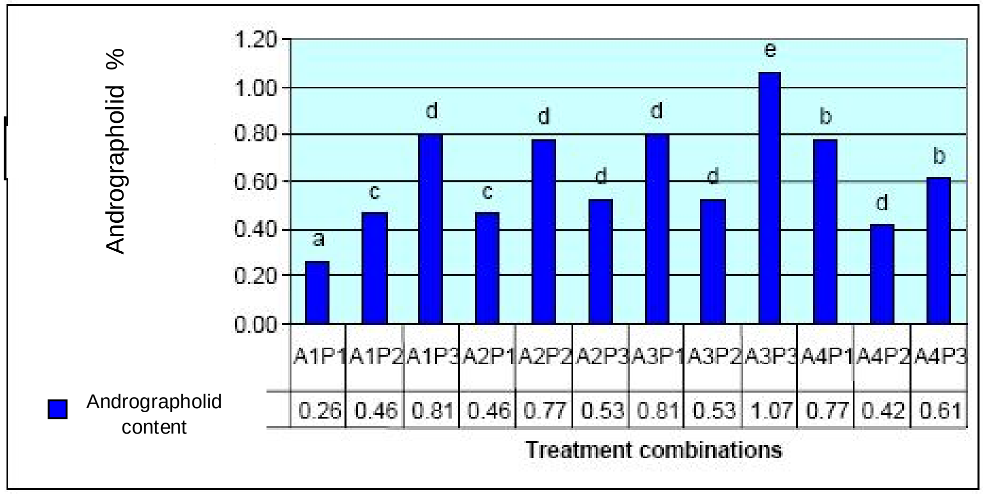

Figure 3. Histogram of andrographolide content on several treatment combinations.

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