

## Photosynthetic response and chlorophyll content of *Spathoglottis plicata* Blume on different altitudes

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**Abstract.** This study investigated the effect of altitudes on photosynthetic parameters and chlorophyll content of terrestrial orchids (*Spathoglottis plicata* Blume). The population and sample of this study were *Spathoglottis plicata* Blume (*S. plicata* Blume) purple and white flowers with 3 different altitudes 95m (1<sup>st</sup>), 600 (2<sup>nd</sup>), 980 (3<sup>rd</sup>) m above sea level. The response of photosynthesis can be increased through the photosynthetic rate of two types *S. plicata* Blume at different altitudes. The result of photosynthetic rate of the *S. plicata* Blume purple and white flowers tend to go up with the height of the place. However, this is not followed by higher temperatures and light intensity. Because the intensity of light and temperature at the time of sampling rises with increasing altitude The chlorophyll content of *S. plicata* Blume purple flower depends on fluctuation, in the contrast to the chlorophyll content of *S. plicata* Blume white flower tend to go up with rising altitude. The photosynthesis response of both terrestrial orchids was highest at the highest altitude (980 asl). Statistical tests show that the rate of photosynthesis of *S. plicata* Blume at the highest place is significantly different from the other two locations. And the chlorophyll content at the three altitudes is significantly different.

### 1. Introduction

*Spathoglottis plicata* Blume is known as terrestrial orchids that has beautiful and attractive flower shapes and colors. This species is an exotic ground orchid including the family Orchidaceae. *Spathoglottis plicata* (*S.plicata*) is able to adapt to a less favorable environment so that it is easy to maintain. In Nature, this species is found from 125 to 1650 m above sea level [1]. *Spathoglottis plicata* is able to live at a low to moderate altitudes from grassland and open forest [2]. The plant produces flower all the year round and the flower stalks carry 10-50 flowers which bloom continually for several months. According to [3] in Java, there are more than 731 orchid species, 231 orchids are endemic species and distributed evenly. Endemicity is caused by a variety of topographic patterns on this island. Yogyakarta has a diverse topography from the lowlands to the highlands with the altitude varies between 0-100m above sea level to 100-500m above sea level (asl). Large distribution area results in a high diversity of *Spathoglottis*. Environmental factors will affect physiological processes in plants [4].

There are a number of *Spathoglottis* orchid species found in Indonesia include *S. plicata*, *S.aurea*, *S. unguiculata*, and *S. angustorum*. The pink-purple *S. plicata* orchid is a standard type of orchid that has spread widely. And the white *S. plicata* orchids are still very rare [5]. This plant is easy to grow in open places and on marginal lands, where competition with other plants is not too strong. The beauty

of the colorful flowers and the ease in cultivating this orchid can be used as a model plant for the study of orchids [6]. Altitude variations and climate differences in a region can cause a quite high species diversity. The difference in altitude will affect the environment of the plant, which causes the difference in distribution of orchid plants [7]. Environmental factors will affect physiological processes in plants. All physiological processes will be affected by temperature and some processes depend on light [4]. Photosynthesis rates and chlorophyll content of plants can vary in every location, because they are influenced by internal factors such as metabolism, physiology and external factors such as fertility levels and others environmental factors. Physiological of the plant is influenced by environmental conditions and genetic factors. Both of these factors will interact during the life cycle of the plant, thus giving rise to different external forms in one species [8]. This research was determined to study the photosynthetic responses and the chlorophyll content of purple and white *S. plicata* Blume at 3 different altitudes.

## 2. Materials and Methods

### 2.1. Study Area

This research was conducted in 3 locations based on different altitudes, during March to June 2018. The first location was in Dongkelan village, Mantrijeron District, Yogyakarta City (95 asl). The second location was in Boyong Hamlet, Hargobinangun Village (600 asl), and the third location was in Turgo Hamlet, Purwobinangun Village (980 asl).

### 2.2. Materials

Li-cord (LI-6400 Portable Photosynthesis System version 5) were used to adjust the photosynthetic process. Some other laboratories equipment such as mortar, grinder, digital scales, 50 ml volumetric flask, 50 ml erlenmeyer, funnel, measuring cup, centrifuge, cuvette, UV spectrophotometer, test tube, thermometer, hygrometer, soil tester, lux meter, GPS to measure the chlorophyll content and stomatal morphology.

### 2.3. Photosynthesis rate analysis

Photosynthesis data of *S.plicata* Blume was measured using a Li-cord device (LI-6400 Portable Photosynthesis System Version 5 with unit is  $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ ). The first method was measuring the intensity of light, then data was inserted into the Li-cord device. The next step, the leaf bleed was inserted into special chamber then the measurement was made. The sampling was done in the morning until noon (starting from 09.00 to 13.00 pm).

### 2.4. Chlorophyll content analysis

Half a gram of leaf sample was finely sliced and gently mixed with a clean pestle and mortar. A 4-5 mL of 96% ethanol was added to the mixture. The materials were further grind gently. Thereafter, the sample was centrifuged at 4000 rpm for 3 minutes. The supernatant was transferred to 50 mL volumetric flask. The final volume was made up to 50 mL with addition of 96% ethanol. The color absorbance of the solution was estimated by a spectrophotometer using 649 and 665 nm wavelength against the solvent. Ethanol 96% was used as a blank (Wintermans and de Mots Methode).

For the calculations we use a formula:

$$\text{Chl a} = 13.7 \text{ OD (665)} - 5.76 \text{ OD (649)}$$

$$\text{Chl b} = 25.8 \text{ OD (665)} - 7.6 \text{ OD (649)}$$

$$\text{Total Chl} = 20.0 \text{ (649)} + 6.10 \text{ OD (665)}$$

### 2.5. Data Analysis

Quantitative data were analyzed with one-way variants (One way Anova). The results of significant analysis were followed by the DMRT (Duncan Multiple Range Test) test with a confidence level of

95%. Data from the measurement of chlorophyll, photosynthesis and microclimate were carried out by correlation analysis.

### 3. Results and Discussion

The physiology of plant is influenced by environmental conditions and genetic factors. Geographical differences such as differences in altitude will cause differences in climate and weather such as temperature, humidity, and rainfall [9]. Temperature differences in each height range cause differences in metabolic processes. Ground orchid growth is influenced by microclimate and edaphic conditions due to the influence of altitude. Table 1 presents the microclimate conditions and edaphic factors at the three observations sites. Table 1 shows that the higher altitude, the lower the air temperature and light intensity. And the higher the altitude, the higher the soil moisture and pH.

**Table 1.** Microclimate conditions and edaphic factors of *S. plicata* Blume in 3 different locations

Environment variables	Location based on height		
	Loc I (low)	Loc II (temperate)	Loc III (high)
Temperature (°C)	30±1,41	27,5±0,71	25±0,71
Light intensity (Lux)	1993±544,47	1974,5±320,32	1862,5±1169,7
Humidity (%)	56,5±1,41	52,5±1,41	68±1,41
Soil moisture (%)	70±4,24	85±2,83	89±4,95
Soil pH	6,5±0,14	6,9±0,71	7±0,14

Temperature is one of the most important factors affecting photosynthesis. This is because there are enzyme activities that are affected by temperature. Photosynthesis response shows a clear relationship. Photosynthesis response of *S. plicata* Blume purple flowers can be seen on table 2 and *S. plicata* Blume white flowers on table 3.

**Table 2.** Photosynthesis Response of *S.plicata* Blume purple flowers

Location based on height	Rate of photosynthesis ( $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ )
L I (low)	149,17±11,13 <sup>b</sup>
L II (temperate)	137±23,39 <sup>b</sup>
L III (high)	171,50±5,24 <sup>c</sup>

**Table 3.** Photosynthesis Response of *S.plicata* Blume white flowers

Location based on height	Rate of photosynthesis ( $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ )
L I (low)	139,17±3,74 <sup>a</sup>
L II (temperate)	149,50±21,57 <sup>a</sup>
L III (high)	178,50±20,87 <sup>b</sup>

Note : The mean value  $\pm$  SD followed by different letters in the same column indicate significant difference according to Duncan's Multiple Range Test (DMRT) at a significant  $\alpha = 0.05$

Photosynthetic response can be seen from the rate of photosynthesis of *S. plicata* purple flowers on table. 2. Based on altitude, there is a tendency for an increase in photosynthesis rate for *S.plicata* white flowers, even though location 2 shows decrease for *S.plicata* purple flowers. While the chlorophyll content can be seen in table 4 and 5.

**Table 4.** Chlorophyll content of *S.plicata* Blume purple flowers

Chlorophyll content (mg/l)	Location based on height		
	L I	L II	L III
<b>Chlorophyll a</b>	15,59±0,79 <sup>a</sup>	33,07±0,89 <sup>c</sup>	21,98±0,85 <sup>b</sup>
<b>Chlorophyll b</b>	7,63±0,42 <sup>a</sup>	16,50±0,42 <sup>c</sup>	10,76±0,40 <sup>b</sup>
<b>Total Chlorophyll</b>	12,31±0,42 <sup>a</sup>	21,98±0,94 <sup>c</sup>	17,40±0,96 <sup>b</sup>

**Table 5.** Chlorophyll content of *S.plicata* Blume white flowers

Chlorophyll content (mg/l)	Location based on height		
	L I	L II	L III
<b>Chlorophyll a</b>	10,71±1,42 <sup>a</sup>	20,14±0,38 <sup>b</sup>	21,98±0,85 <sup>b</sup>
<b>Chlorophyll b</b>	5,32±0,78 <sup>a</sup>	10,06±0,17 <sup>b</sup>	12,94±1,88 <sup>c</sup>
<b>Total Chlorophyll</b>	7,41±0,41 <sup>a</sup>	13,33±0,63 <sup>b</sup>	19,27±2,72 <sup>c</sup>

Note : The mean value ± SD followed by different letters in the same column indicate significant difference according to Duncan's Multiple Range Test (DMRT) at a significant  $\alpha = 0.05$

Spathoglottis orchids are plants that have high adaptability to a less favorable environments. In this research, focused on the effect of altitude on the rate of photosynthesis and leaf chlorophyll content. Areas with different altitudes will show differences in environmental factors both microclimate and edafic factors. The spathoglottis orchid used in this study is a purple and white flowering Spathoglottis that exists at 3 different altitudes (95, 600, 980 m asl).

Photosynthesis is an important process for plants and only takes place in cells that have photosynthetic pigments. The photosynthetic pigments are in the chloroplast through chlorophyll as a device. And the process of photosynthesis is greatly influenced by light intensity and temperature. Table 1 shows that the higher the place the lower the air temperature and light intensity. Light intensity is a very important growth factor and closely related to the activity of assimilation and stomatal opening. According to [10], physiologically light has an influence on orchids both directly and indirectly. The direct effect is the process of photosynthesis and the indirect effect in on growth, germination, and flowering. According to [11], the percentage of sunlight requirements for each type of orchid is different types of orchids. Epiphytic orchids generally require low sunlight intensity, which is around 25-50%. And terrestrial orchids require a higher amount which is around 60-75%. According to [1], average air temperature will decrease with increasing altitude. Temperature is one of the most important environmental factors, which can affect photosynthesis because photosynthetic activity is sensitive to pressure caused by temperature. However, at high temperature, CO<sub>2</sub> is less soluble in chloroplast water, thereby reducing the rate of photosynthesis, drought stress and stomatal closure occur so that CO<sub>2</sub> cannot enter the leaves [12].

In this study, the response of photosynthesis can be increased through the photosynthetic rate of two types *S. plicata* Blume at different altitudes. The result of the photosynthetic rate of the *S. plicata* Blume purple and white flowers tend to go up with the height of the place (Table.2 and 3). However, this is not followed by higher temperatures and light intensity. Because the intensity of light and temperature at the time of sampling rises with increasing altitude.

In higher plants such as *S. plicata* Blume, chlorophyll a and chlorophyll b are the main photosynthetic pigments that plays a role in absorbing sunlight on the photosynthesis process. According to [6], the chlorophyll content of each plant is different and it can be seen in the color of the leaf. If chlorophyll deficient in the leaves, it will reduce the rate of photosynthesis. The data of chlorophyll content of *S. plicata* Blume purple flower depend on fluctuation, otherwise to the chlorophyll content of *S. plicata* Blume white flower tend to go up with rising altitude. For *S. plicata* Blume purple flowers it is not supported by high content of chlorophyll. However, in this case that chlorophyll content did not affect the value of photosynthesis rate, this can be caused by there are

more factors that influence the rate of photosynthesis such as air temperature, humidity, soil pH which increases with increasing photosynthesis rate. And for *S. plicata* Blume white flower, chlorophyll content can affect the rate of photosynthesis. According to [13], photosynthesis is a complex process that occurs in plants because many factors affect both internal and external factors. The factors such as oxygen, chlorophyll content, temperature, water content, light intensity, CO<sub>2</sub> concentration etc. The high chlorophyll content will certainly result in a maximum photosynthesis reaction. The data of photosynthetic rate of *S. plicata* Blume white flower in this study tends to increase with increasing chlorophyll content at a higher place.

The photosynthesis response of both *S. plicata* Blume was highest at the highest altitude (980 asl). Statistical tests show that the photosynthetic rate and chlorophyll content of the *S. plicata* Blume purple and white flowers at the three altitudes are significantly different. This can be seen from different letters in the same column at table 2, 3, 4, and 5. In the tables show that the rate of photosynthesis at the highest place is significantly different from the other two locations. Although the highest photosynthesis rate is not followed by high light intensity and temperature, this may be due to the orchid plants requiring a maximum temperature of 28°C and a minimum temperature of 15. Because an increase in temperature that exceeds can cause dehydration in orchids and inhibit its growth. And the chlorophyll content at the three altitudes is significantly different. However the trends shown in the two types of orchids are not the same. For *S. plicata* Blume white flowers, the higher place the higher the chlorophyll content, and different from the purple flower. This shows that the response of each orchid variety is different. The results show that altitude can affect the photosynthetic response and chlorophyll content of *S. plicata* Blume purple and white flowers. Altitude variations and climate differences in a region can cause a quite high species diversity. The difference in altitude will affect the environment of the plant, then will affect of plant physiological processes. Altitude including physiological factors that greatly affect the climate. Altitude affects the temperature and light intensity. The temperature and intensity of the light will get smaller with the higher place to grow. Plant physiology is influenced by environmental conditions and genetic factors. Both of these factors will interact during the life cycle of the plant, thus giving rise to different external forms in one species.

#### 4. Conclusion

The photosynthesis response of both terrestrial orchids was highest at the highest altitude (980 asl). For *S. plicata* Blume purple flowers it is not supported by high amounts of chlorophyll. The chlorophyll content of *S. plicata* Blume purple flowers tends to fluctuate, whereas in *S. plicata* Blume white flowers shows that the higher the chlorophyll content the higher the rate of photosynthesis. The photosynthesis response of both terrestrial orchids was highest at the highest altitude (980 asl). Statistical tests show that the photosynthetic rate at the highest place is significantly different from the other two locations. And the chlorophyll content at the three altitudes is significantly different. This shows that altitude can affect the photosynthetic response and chlorophyll content of *S. plicata* Blume purple and white flowers.

#### 5. References

- [1] Chikmawati T 2013 Anatomical and Cytological Features of *Spathoglottis plicata* from Java Island *The journal of Tropical Life Science* **3**(2) 87-90
- [2] Sangadji S 2001 *Pengaruh Iklim Tropis di Dua Ketinggian Tempat Berbeda* (Yogyakarta: UGM Press)
- [3] Comber J B 2001 *Orchids of Java* (Thailand: Bentham-Moxon Trust The Royal Botanical Gardens Kew Charoen Slip Press)
- [4] Hossain M M 2013 Multiple regeneration pathways in *Spathoglottis plicata* Blume-A Study in vitro *South African Journal of Botany* **85** 56-62.
- [5] Thompson S A and Wright F W 1995 *Spathoglottis plicata* (Orchidaceae): New to Dominica, Another Record from the Lesser Antiles *Caribbean J. Sci.* **31**(1-2) 148-149.

- [6] Effendi K and Widyastoeti S 2002 *Profile and Development Strategy Commodities Orchid* (Jakarta: Ornamental Plants Research Center)
- [7] Wiharto M 2009 *Klasifikasi Vegetasi Zona Sub Pegunungan Gunung Salak, Bogor, Jawa Barat. Disertasi* (Bogor: Sekolah Pascasarjana IPB)
- [8] Kiswara W 2011 *Vegetasi Lamun (seagrass) di Rataan Terumbu Pulau Pari, Pulau-Pulau Seribu Buletin Ilmiah Oseana 25*
- [9] Muhdi 2004 *Pengaruh Elevasi terhadap Pertumbuhan dan Kualitas Kayu* (Medan: Universitas Sumatra Utara)
- [10] Fitter A H and Hay R K M 1998 *Fisiologi Lingkungan Tanaman* (Yogyakarta: Gadjah Mada University Press)
- [11] Iswanto H 2002 *Petunjuk Perawatan Anggrek* (Jakarta: Agro Media Putri)
- [12] Nazaruddin 1999 *Dasar-dasar Fisiologi Tanaman* (Jakarta: Fakultas Pertanian Universitas Hasanuddin dan Yayasan Forest Indonesia)
- [13] Loveless A R 1999 *Prinsip-prinsip Biologi Tumbuhan untuk Daerah Tropik I* (Jakarta: Gramedia)