Genetic Parameters and Physiology of *Phalaenopsis* Flowering in Different Directions of Sunlight in Tropical Lowlands

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Abstract

Flowering in *Phalaenopsis* is influenced by temperature, with a requirement for temperatures below 26°C, leading to the usual practice of initiating it in tropical highlands. Light also influences the initiation process by playing a significant role in activating the flowering Locus T (FT) gene. Therefore, this study aimed to determine the flowering response of *Phalaenopsis* in lowlands in two directions of sunlight exposure. The experiment was conducted in the lowlands of Magelang, Central Java, Indonesia, using a Randomized Complete Block Design (RCBD). The first factor was *Phalaenopsis* hybrids: KHM 2283, KHM 2508, GL 13540, DF 1622, and Big Chili, while the second factor was sunlight direction (east and west). Across all measured characteristics, it was observed that there was a low Coefficient of Genetic Variability (CVG). The Coefficient of Phenotypic Variability (CVP) for flowering was higher than the CVG value. Heritability analysis showed that vegetative growth exhibited high herit ability. However, flowering characteristics were categorized as low heritability, suggesting that the phenotype was strongly influenced by the environment, specifically lighting. These results contribute to alternative methods for increasing *Phalaenopsis* flowering production in tropical lowlands, indicating that eastward sunlight shows potential as a prerequisite for flower production.

Keywords: *Phalaenopsis*, flowering LOCUS T, genetic variability, phenotypic variability, heritability

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Introduction

Phalaenopsis orchid is a popular flower plant, with high economic value, playing a crucial role in the floriculture industry worldwide as the most widely grown orchid (Khatun et al., 2020). In Indonesia, it is known as the moon orchid and is declared a national flower, according to the Presidential Regulation of The Republic of Indonesia Number 4 of 1993 on National Animals and Flowers (Setiawan, 2013).

The growing temperature range of *Phalaenopsis* is 28-35 °C during the day and 20-24 °C at night, following a specific developmental path from the juvenile to the adult phase, indicating its readiness to flower (Cho et al., 2020). Commercially, *Phalaenopsis* in the juvenile to adult phase is usually cultivated in lowlands, but flowering initiation is carried out in the highlands. Previous studies on flowering induction caused by cold

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temperature treatment have been carried out extensively (van Tongerlo et al., 2021). Paradiso and Pascale (2014)stated that flower induction of *Phalaenopsis* can be successfully achieved at 17°C to 19°C. Consequently, in the tropics, highlands are preferred for flowering initiation (De, 2020), as lowland temperatures, which can be more than 29°C, are not considered suitable. According to Kim et al. (2015)and Wang et al. (2017), exposure to temperatures above 29°C in *Doritis x Phalaenopsis* can inhibit flowering. This cultivation method causes the grower to have two greenhouses for nursery and flowering (Dewi et al., 2015).

Light intensity related to the direction of the sun is another factor influencing the initiation and development of *Phalaenopsis* inflorescence. Generally, light serves as an energy source for photosynthesis and plays a crucial role in regulating various aspects of plant growth and development (Yang et al., 2017), including flowering induction (Magar et al., 2018). The regulation of flowering time is an essential target in orchid flower production (Sidhu et al., 2021). Light is related to the quantity and quality obtained, including intensity, day length, and wavelength transformed by plants. The intensity of light received affects photosynthesis and other metabolic processes (Shamshiri et al., 2018). When all physiological and environmental aspects meet, the initiation of flower buds occurs in mature plants due to cell differentiation, which develops into the peduncle. In tropical areas, the intensity and direction of sunlight are determined by the east and west direction, with these variations significantly affecting plant growth and physiology (Magar et al., 2018).

Genetic factors also influence flowering in *Phalaenopsis*, where parameters such as variability and heritability determine the uniqueness of genetic resources and predict the ability of flowering gene expression to correlate with the environment. This genetic diversity can be identified by observing phenotypic variations in each hybrid studied, while the influence of environmental factors in regulating flowering is determined by estimating heritability. Moreover, heritability is a measure of the variability of a character appearance in a population caused by the role of genetic factors (Priyanka and Jaiswal, 2017). The heritability value of a character needs to be known whether it is primarily influenced by genetic factors or the environment. The estimation also provides information on the proportion of phenotypic variations that can be inherited (Al-Naggar et al., 2019). A high heritability estimation indicates success in selecting parents with optimal flowering ability. Therefore, this study aimed to determine the flowering response of *Phalaenopsis* in lowlands in two directions of the sunlight and genetic parameters in controlling *Phalaenopsis* flowering.

Materials and Methods

Study Site and Design

This study was conducted in Magelang, Central Java Province, Indonesia, from June to October 2022. The experiment was carried out using the CI-340 Handheld Photosynthesis System, measuring cups, lux meters, flowerpots, rulers, and stationery. The *Phalaenopsis* hybrids selected were KHM 2283, KHM 2508, GL 13540, DF 1622, and Big Chili, all of which were three years old after acclimatization.

A Nested Design with three replicates, with the main plot focusing on sunlight direction, consisting of east and west. Meanwhile, the subplot included various hybrids, namely KHM 2283, KHM 2508, GL 13540, DF 1622, and Big Chili. The two-way sun lighting treatment was carried out by placing the orchid treated with T1 to the east to receive optimum sunlight from morning to noon. This was followed by the placement of T2 in the west to receive optimum sunlight from noon to evening. The climatic factor observed was light intensity at the *Phalaenopsis* placed on the east and west sides of the building. For each trial unit, a single plant was used for sampling, and the observed variables were Photosynthetic Active Radiation, stomatal conductance, internal CO_2 content, leaf temperature, transpiration using the CI-340 Handheld Photosynthesis System, plant height, as well as the number of pseudo bulbs, leaves, internodes, and flowers, including flower diameter. Plant care was carried out by applying AB mix orchid fertilizer at a 50 mL/day dose, while bactericidal and fungicide applications were performed once a week.

Table 1
Mean square of expectations based on analysis of variance

Variation sources	Mean square	Mean squares of expextations
Repetition	M1	-
Genotype (G)	M2	$\sigma_e^2 + r\sigma_{dg}^2 + rd\sigma_g^2$
Light direction (E)	M3	$\sigma_e^2 + r\sigma_{dg}^2 + rg\sigma_d^2$
Interaction of G x E	M4	$\sigma_e^2 + r\sigma_{dg}^2$
Error	M5	σ_e^2

Table 2
The internal CO_2, leaves temperature, external leaves temperature, transpiration, stomatal conductance, and Photosynthetic Active Radiation on five *Phalaenopsis* hybrids in different sun directions

Sun direction	Internal leaves temperature (°C)	External leaves temperature (°C)	Transpiration (mmol m^{-2} s^{-1})	Stomatal conductances (mmol m^{-2} s^{-1})	CO_2 internal (µmol mol^{-1})	PAR (µmol m^{-2} s^{-1})
East	30.73 ± 0.43	29.76 ± 0.57	0.047 ± 0.02	2.98 ± 1.15	584.69 ± 10.46	78.03 ± 2.43 a
West	30.42 ± 0.52	29.50 ± 0.54	0.035 ± 0.01	2.22 ± 1.05	601.84 ± 26.00	29.03 ± 1.88 b
Sig.	0.135	0.087	0.723	0.794	0.984	0.000
CV	2.17	3.05	11.12	18.21	5.87	8.36
Genotype						
KHM 2283	30.55 ± 0.52	29.68 ± 0.56	0.075 ± 0.03	4.68 ± 1.54	595.65 ± 35.87	47.15 ± 1.54
KHM 2508	30.72 ± 0.41	29.62 ± 0.62	0.045 ± 0.02	2.75 ± 1.21	593.13 ± 11.75	69.02 ± 1.21
GL 13540	30.13 ± 0.74	29.23 ± 0.74	0.033 ± 0.01	2.09 ± 1.09	594.53 ± 19.32	37.58 ± 1.09
DF 1622	30.95 ± 0.29	29.97 ± 0.34	0.030 ± 0.01	1.82 ± 0.78	588.60 ± 14.10	63.88 ± 0.78
Big Chili	30.55 ± 0.42	29.65 ± 0.53	0.025 ± 0.01	1.67 ± 0.89	594.42 ± 10.10	50.03 ± 0.89
Sig.	0.225	0.442	0.549	0.687	0.196	0.720
CV	1.67	1.38	4.90	7.59	3.81	14.89

Notes: The numbers followed by the same letter in the same variable and factor were not significantly different according to Duncan's multiple range test at level 5%.

Table 3
Vegetative and generative characters on five *Phalaenopsis* hybrids in different sun directions

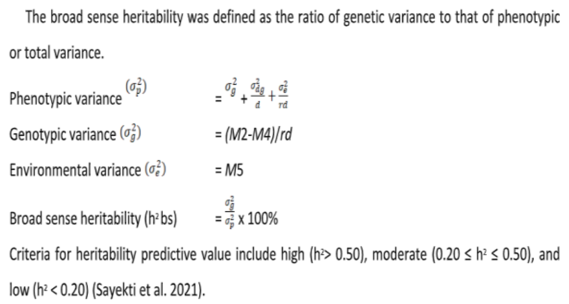
Sun direction	Length of leaves (cm)	Number of leaves	Flowering percentage (%)	Number of flowers	Length of flower stem (cm)	Flower width (cm)
East	24.03 ± 0.76	6.73 ± 0.55	100.0 ± 0.00 a	8.53 ± 1.39 a	65.31 ± 6.95 a	11.09 ± 0.14 a
West	24.76 ± 1.43	6.80 ± 0.69	73.33 ± 46.18 b	4.20 ± 2.92 b	38.27 ± 6.95 b	10.73 ± 0.07 b
Sig.	0.805	0.333	0.000	0.000	0.000	0.000
CV	6.78	10.79	11.77	16.13	4.10	2.43
Genotypes						
KHM 2283	23.48 ± 0.63 b	8.00 ± 0.57 a	83.33 ± 28.87	6.50 ± 2.67	61.15 ± 2.50	11.80 ± 0.06
KHM 2508	23.65 ± 1.41 b	5.67 ± 0.57 c	100.00 ± 0.00	6.16 ± 1.44	53.88 ± 7.40	9.93 ± 0.30
GL 13540	27.21 ± 0.68 a	6.33 ± 0.28 bc	83.33 ± 28.87	5.83 ± 2.29	50.22 ± 3.45	10.50 ± 0.06
DF 1622	21.58 ± 1.00 b	7.00 ± 0.57 b	100.00 ± 28.87	8.33 ± 1.76	49.75 ± 14.67	11.25 ± 0.06
Big Chili	23.57 ± 1.75 c	6.83 ± 1.07 b	66.67 ± 28.87	5.00 ± 2.60	43.95 ± 2.47	11.08 ± 0.06
Sig.	0.000	0.000	0.890	0.253	0.744	0.439
CV	4.59	9.91	14.01	12.64	6.06	1.38

Notes: The numbers followed by the same letter in the same variable and factor were not significantly different according to Duncan's multiple range test at level of 5%.

Data analysis

The quantitative performance data were analyzed using Analysis of Variance (ANOVA), and when there were significant differences, a further test was carried out using Duncan's Multiple Range Test at a level of 5%.

Phenotypic and genotypic variances were estimated by extracting the components from the analysis of variance (Table 1). The Coefficient of Genetic Variance (CGV) and Coefficient of Phenotypic Variance (CPV) were calculated using the Mishra et al. (2019) method. Variability criteria were determined based on the value of the coefficient of variation with a range of 0 - 100% quartiles, namely low (0% ≤ 25%), rather low (25% ≤ 50%), quite high (50% ≤ 75%), and high (75% ≤ 100%). The broad-sense heritability was defined as the ratio of genetic variance to that of phenotypic or total variance.



The broad sense heritability was defined as the ratio of genetic variance to that of phenotypic or total variance.

Phenotypic variance (σ_p^2) $= \sigma_g^2 + \frac{\sigma_{dg}^2}{d} + \frac{\sigma_e^2}{rd}$

Genotypic variance (σ_g^2) = (M2-M4)/rd

Environmental variance (σ_e^2) = M5

Broad sense heritability (h^2bs) $= \frac{\sigma_g^2}{\sigma_p^2} \times 100\%$

Criteria for heritability predictive value include high ($h^2 > 0.50$), moderate ($0.20 \leq h^2 \leq 0.50$), and low ($h^2 < 0.20$) (Sayekti et al. 2021).

Results

Microclimate and Physiological Activity Affected by Different Sun Direction

In this study, the analysis of variance showed a significant influence of sunlight direction on PAR values, while no discernible impact was observed on other microclimatic and physiological variables. Stomatal conductance values indicated a trend towards being higher in plants exposed to the east-sun direction compared to the west-sun, but this disparity did not reach statistical significance. However, internal and external temperatures, as well as leaf transpiration rates, showed similar tendencies in plants exposed to both east and west sunlight direction, as presented in Table 2. Despite similar CO_2 levels in the leaves, ranging from 584.69 to 601.84 µmol mol^{-1}, the eastern direction generated a PAR of 78.03 µmol m^{-2} s^{-1}. Meanwhile, the western direction only reached 29.03 µmol m^{-2} s^{-1}, indicating that leaves exposed to the eastern sunlight direction showed high photosynthetic activity.

Flower Development of *Phalaenopsis* under Different Sun Direction

The variations in leaf characteristics are determined by the genotype of each plant, where factors such as leaf size and quantity are correlated with genetic traits of the individual type. This vegetative parameter remains unaffected by the direction of sunlight due to the maturity of the plants used, having reached their maximum vegetative growth (Table 3).

In this study, the flowering percentage of KHM 2508 and DF 1622 showed that all the orchids in this study produced flowers, both in the east and west direction of sunlight. However, KHM 2283 and GL 13540 showed that the percentage of flowers grown, on average, in the direction of east and west sunlight was 83.33%. This average indicated that there was a decrease in the number of flowering plants exposed to western sunlight. Meanwhile, at *Big Chili*, there was a decrease in the most significant percentage, namely 66.67%, indicating fewer flowering plants in the western sunlight. This showed that the sensitivity of each genotype to the quality of light produced from the east and west was different (Table 3).

The light energy received by *Phalaenopsis* from the east also accelerated inflorescence development and the time of anthesis. The flower development of four genotypes, including KHM 2508, GL 13540, DF 1622, and *Big Chili*, has a faster anthesis time, with an average of 80.27 days when exposed to eastern light, while under western light, the time was slower, averaging 86.73 days (Fig. I.).

The quality of the produced flowers was related to the quantity and size of the flowers, impacting the sales quality of *Phalaenopsis*. In this study, *Phalaenopsis* exposed to eastern light yielded an average of 8.53 flowers, while those under

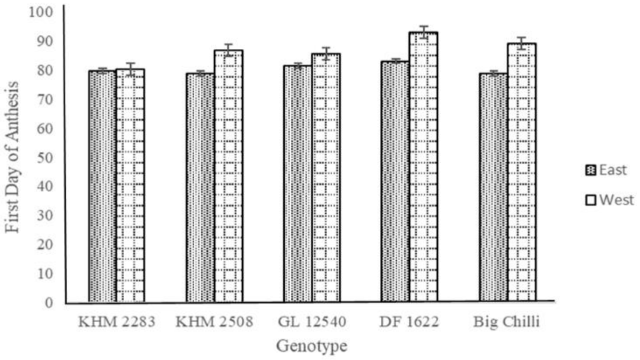


Fig. I. First Day of Anthesis on five *Phalaenopsis* hybrids in different sun directions

Fig. II. Difference responses of flowering performance in *Phalaenopsis* genotypes a). East direction exposure b). west direction exposure.

western light produced an average of 4.20 (Table 3). Generally, flowers numbering fewer than six are considered substandard and are often sold at a lower price (Fig. II).

Genetic Variability and Heritability of Flowering *Phalaenopsis*

The results of genetic and phenotypic variability analysis are presented in Table 4. All the observed characteristics showed a low CGV, indicating limited genetic variation within the tested *Phalaenopsis*. Furthermore, there was low phenotypic diversity of leaf characters among the tested genotypes, with a mean value of the leaf length ranging from 21.58 (DF 1622) to 27.21 (GL 13540). The average leaf number ranged from 5.67 (KHM 2508) to 8.00 (KHM 2283), while the average leaf width was between 6.58 (GL 13540) and 9.59 (Big Chili).

The result of heritability is shown in Table 5. In flowering variables, the CPV value showed an increase compared to the CGV; thereby, the heritability of flowering characters was relatively low due to the influence of the environment.

Discussion

Performance of Flowering Phalaenopsis at Different Sunlight Directions

Solar radiation is the energy source that initiates the photosynthesis process. Several results have indicated a positive correlation between Photosynthetically Active Radiation (PAR) and net photosynthesis. A linear trend in the relationship between increasing PAR and the augmentation of net photosynthesis has also been observed (Ngatui et al., 2017). This phenomenon is attributed to the enhanced absorption of light energy, leading to an increase in the photosynthetic process in plants (Yang et al., 2017).

The internal CO_2 content did not show any significant differences, but the sunlight direction treatment influenced PAR, potentially enhancing photosynthesis in orchid plants exposed to the eastern sun direction. Although observations indicated no discernible effects of light direction on CO_2 content, leaf temperature, and humidity, the PAR incorporates light within the 400–700 nm wavelength range in the light spectrum, which is essential for plant photosynthesis, as chlorophyll absorbs light within this wavelength range.

These results indicate the impact of sunlight direction on both the quality and quantity of energy that plants receive, emphasizing its crucial role in plant metabolism. The eastern light direction yielded a higher PAR value compared to the western light direction. This light orientation facilitated increased photosynthetic activity in plants exposed to the east, resulting in a more substantial transfer of photosynthates within the plants. These photosynthates played an important role in triggering flowering; thus, plants receiving eastern light showed faster flowering initiation compared to those exposed to western light.

An increase in the number of flowers was accompanied by a rise in PAR. Based on observations, at a PAR value of 29.03 $\mu mol/m^2/s$, the average flower number was 4.20, which increased to 8.53 at 78.03 $\mu mol/m^2/s$. Wang et al. (2020)stated that net primary production usually has a linear relationship with PAR. The role of light in flower induction is indicated by its impact on the production of florigen, a signal molecule that induces flowering (Goretti et al., 2020). Florigen, which is flowering LOCUS T (FT) protein expressed in the phloem of the leaves and transferred to the apical shoot, initiates flower development. Furthermore, it transforms meristematic vegetative organs into reproductive organs (Tsuji, 2017). According to Wang et al. (Wang et al., 2021), florigen is related to the photoperiodic phenomenon, which is controlled by the quantity and duration of light. In long photoperiodicity plants, an appropriate photoperiod triggers the differentiation of plant shoots into flower buds.

Light is one of the primary factors that regulate the initiation of flowering in ornamental plants (Kamelia et al., 2018). The eastern light direction results in higher PAR, affecting both the quantity and quality of flower production (Horváth et al., 2020). Additionally, sunflower inflorescences absorb maximum light energy when the plant is oriented towards the east, receiving approximately 10–50% more than the western sun direction. Takács et al. (2022)also stated that energy received from the eastern direction of sunlight led to maximum energy absorption, potentially accelerating the sunflower seed maturation process. Increasing the light energy, the plant receives in *strawberry* plants can also shorten the harvest time (Yoshida et al., 2016).

The activation of florigen by light occurs initially in the leaves and is transported to the shoot (Shim and Jang, 2020). Mascheretti et al. (2015)also explained the presence of florigen in *corn*, which was expressed in young leaves and transferred to plant shoots. When the quantity and quality of the provided light signal reached an adequate threshold, the ID1 gene would be translocated to the apex, becoming a potential candidate gene responsible for regulating florigen synthesis (Goretti et al., 2020). Subsequently, the photoreceptors responsible for regulating gene signals within the vascular tissue of leaves transmit signals to flowering LOCUS T (FT) protein at the cellular level along with the shoot apical meristem (SAM). Within the SAM, signals activate two genes at specific cellular regions, controlled by the secondary cellular protein TERMINAL FLOWER 1 (TFL1), which ensures the translation of the flowering signal into the SAM. Wang et al. (2017) also stated that the *Phalaenopsis aphrodite*

LEAFY (PhapLFY) gene accumulated in floral meristem primordia to stimulate floral initiation.

The transmission of a spectrum consisting of various wavelengths towards photoreceptors generates specific signals capable of activating the expression of genes associated with physiological and metabolic activities (Paradiso and Proietti, 2022). In *Arabidopsis*, classified as a long-day plant, exposure to an appropriate duration of light can enhance the initiation of flowering. Molecular genetic methods have also been used in *Arabidopsis* to identify the genes necessary for receiving light quantity signals, and some proteins are specifically included in regulating flowering. Other components also simultaneously participate in the light signal transduction process (Shim and Jang, 2020). Flowering time in *rice* is affected by environmental factors such as photoperiod, temperature, and light intensity, which influence the activation of the flowering LOCUS T (FT) gene, a critical factor in the transition from the vegetative phase to the generative phase (Kutsher et al., 2021). Additionally, it has been reported in *Oncidium* orchids that FT mRNA is expressed in axillary buds, leaves, pseudobulbs, and flowers (Dewi et al., 2015).

The variation in sensitivity of each genotype to eastern and western sunlight direction treatment showed that the genetic potential of each plant in responding to light signals was different. This indicates that the eastern sunlight direction was the threshold of light needed and appropriate for initiation and flowering development. Besides that, a variation occurred in the first day of anthesis that was attributed to the higher solar energy received by plants with eastern sun exposure, which **is** related to light quantity such as intensity and photoperiod. However, it was also influenced by light quality, including the composition of received wavelengths, by controlling primary and secondary metabolism (Cho et al., 2017). This photochemical reaction process occurred through the conversion of light energy into chemical energy by synthesizing ATP and NADPH, which are used in the production of carbon atoms within the Calvin cycle (Shafiq et al., 2021).

The impact of photoperiod on each stage of the flower has been documented by Sidhu et al. (2021), including tissue differentiation and the differentiation of flower organs. The maturation of floral organs towards anthesis requires energy from photosynthesis. Plants require an increasing photosynthate supply, derived from the conversion of light energy into chemical energy, during the organ differentiation process. In *strawberry* plants, Yoshida et al. (2016)found that increased exposure to high-energy light reduced harvesting time. This study also indicated that the eastward sun exposure played a significant role in accelerating *Phalaenopsis* flowering development.

Genetic Variability and Heritability of Flowering *Phalaenopsis*

Flower quality is one of the selection parameters in *Phalaenopsis* plant breeding. The number of flowers on one stalk determines the quality of the plant. A higher number of blooms per spike is often seen as a positive quality. The *Phalaenopsis* used in this study was included in the Grandiflora type, with the criteria of large leaves, wide flowers, and no branches. Therefore, the quantitative characteristics of the plants used include low diversity.

The presence of genetic and phenotypic diversity plays a significant role in determining the effectiveness of plant breeding efforts. When genetic and phenotypic diversity exists within a population, it indicates that there are variances in genotype and physical trait values among individuals within that population. An increase was observed in the value of the phenotypic coefficient of variation on flowering characters. This was identified in the average number of flowers, which ranged from 2.33 (*Big Chili*) in the western sunlight direction to 8.33 (*DF 1622*), flower width from 9.93 cm (*KHM 2508*) to 11.80 cm (*KHM 2283*), the number of flower stalks from 0.67 (*Big Chili*) to 1.17 (*KHM 2508*), average number of flowers blooming from 2.00 (*DF 1622*) to 4.33 (*KHM 2508*), and flower stalk length from 43.95 (*Big Chili*) to 61.15 (*KHM 2283*). The contrast between CPV and CGV illustrated that the observed traits were affected by environmental factors such as light (Sayekti et al., 2021), particularly in *Phalaenopsis* flowering development. The extensive range of traits offers valuable opportunities during the selection process, aligning well with the anticipated improvements in plant characteristics.

Heritability is a genetic parameter utilized to gauge the degree of inheritance of a trait within a plant population or an estimate that quantifies the extent to which genetic factors contribute to the variability in the expression of a trait within the population. When a trait exhibits a high heritability value, it means that genetic factors have a substantial influence on the trait's expression, making it easily inheritable. In such cases, genetic factors play a more significant role in governing the trait than environmental factors. The heritability value represents the influence of genetic factors that impact the expression of a character (phenotype). In this study, heritability analysis showed that leaf length, width, and the number of leaves were categorized in the high category (>0.20). A high heritability value indicated that genetic capability influenced the performance of a character and showed the impact of genetic diversity on the observed variation in phenotype among the tested genotypes. The relatively low heritability category included the number of flowers per stem, width, stems, and number of blooms, but flower stem length belonged to the medium category (0.279). Low heritability values suggested that the diversity of plant characteristics was a combination of genetic and environmental factors, but the proportion of environment was more dominant than genetic factors (Terfa and Gurmu, 2020).

In this study, it was discovered that sunlight direction significantly contributed to *Phalaenopsis* flowering. The light energy produced in the east and west direction of the sunlight influenced metabolic processes and the activation of the flowering LOCUS T gene, inducing and impacting flower development (Legris et al., 2019)). The activity of the FT gene also resulted in the generation of a protein known as FT protein, which played an important role in stimulating vegetative growth to flowering.

Conclusions

Eastward light was more important in increasing the number of flowers, the time of anthesis, and the length of the inflorescence in *Phalaenopsis*. Furthermore, these results contributed to an alternative method for enhancing *Phalaenopsis* flowering in tropical lowland regions, indicating that the eastern light direction was optimal for achieving successful flowering production.

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