PERFORMANCE EVALUATION AND VARIABILITY ANALYSIS FOR MAJOR GROWTH AND FLOWERING TRAITS OF *Lilium longiflorum* THUNB. GENOTYPES

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ABSTRACT

The performance evaluation and variability estimation are some of the most important basic steps in any breeding scheme and are usually taken as a pre-breeding task. This experiment was carried out to evaluate the performance and estimate the variability for major growth and flowering traits of Sixteen *Lilium longiflorum* Thunb genotypes in a randomized complete block design (RCBD) with three replications. The estimated analysis of variance (ANOVA) demonstrated prevailing significant variability for studied ten growth and flowering traits of all 16 genotypes. The experiment was conducted to quantify the genetic variability, heritability, and genetic advance as a percentage of the mean (GAM), for understanding the breeding potentiality of *L. longiflorum* genotypes based on their performance for growth and flowering traits, so that pre-breeding scheme could be executed properly. The estimated genotypic coefficient of variation (GCV) and phenotypic coefficient of variation (PCV) value, along with heritability estimate and GAM, revealed that almost all studied traits besides flower diameter were controlled by additive gene action. For these traits, improvement would be possible through the selection after cross-breeding among these genotypes. The choice of genotypes for the hybridization scheme could be made based on the mean performance of those genotypes for the particular traits of the breeding scheme.

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1 Introduction

Lilium longiflorum Thunb. is one of the species of the genus *Lilium* which consists of more than 110 species (Mc Rae, 1998; Dhiman et al., 2018). It belongs to the family Liliaceae and is found endemic to the southernmost Japanese Ryukyu Islands and the eastern seacoast and satellite islands of the mainland of Taiwan (Miller, 1993; Okazaki, 1996; Mc Rae, 1998; Hiramatsu et al., 2001). Though it was first reported in Europe in 1974, and it became famous as Easter lily worldwide; in the USA, it is used as a potted flower and used to force for the special day 'Easter' usually falls in April. It is taxonomically grouped into the subsection of the section Leucolirion or trumpets, representing 11 species (Dhiman et al., 2018). Nowadays, these are widely regarded as species of great importance for world Horticulture. *L. longiflorum* cultivars are famous for their large and trumpet-shaped flowers, besides strong fragrant white colored-flowers, and due to the early flowering habit year-round forcing cultivation capability are some of the distinguishing characteristics (Mc Rae, 1990). It is one of the primary sources of cut flowers during religious holidays in countries like the Netherland, Israel, Japan, and Korea (Grassotti & Gimelli, 2011).

According to Matthews (2007) until 2005, more than 14000 cultivars have been developed and cultivated worldwide. Since then, hundreds of new cultivars have been grown and registered every year. *Lilium* breeding doesn't have so long a history. It has started in the first decennia of the 20th century by producing the intersectional hybrids of the Sinomartagon section firstly in Japan and the USA (VanTuyl & Arens 2011). After the development of appropriate pollination, embryo rescue and polyploidization technology interspecific hybridization has been started (Ki-Byung & Van Tuyl, 2006; Van Tuyl et al., 2002). There are seven primary lily hybrids viz. O, A, L, T, LA, LO, and OT. Among these hybrid groups, Orientals (O) are supposed to be the first hybrid groups which later on replaced by OT hybrids. Based on this experience, it was predicted that Asiatic (A) hybrids largely would be replaced by the LA hybrids, and the longiflorums (L) hybrids replaced by LO hybrids (Van Tuyl & Arens, 2011; Dhiman et al., 2018). In this context, many commercial cultivars have been produced by interspecific hybridization to get the diverse cut flower traits and mainly propagated by vegetative methods (Mc Rae, 1998). In the context of recent breeding trends of interspecific hybridization, longiflorum hybrids and their commercial cultivars play a vital role in developing new cultivars in the competitive flower market of the world. There are so many cultivars available in longiflorum hybrids with their distinct growth and flowering traits. With these aspects, it deserves crucial to evaluate the performance of those cultivars in terms of growth and flowering habits, which would be beneficial for breeders to decide on the further development of an interspecific hybridization scheme. This study evaluated the growth and flowering traits of 16 cultivars/breeding lines to monitor their performance for these traits and for screening out them for the further breeding program (Table 1).

Table 1 List of used *Lilium longiflorum* Thunb. genotypes and their special remarks

S.N.	Genotype	Denotation	Flower color	Special remarks
1	Bright tower (BT)	CV	White	Upward directional, Mid flowering habit
2	Faith	CV	Pink	Upward, early flowering habit
3	Global beauty	CV	White	Upward, Mid flowering habit
4	Global village	CV	White	Upward, early flowering habit
5	Georgia	CV	White	Side facing, very early flowering habit
6	Gelria	CV	White	Side facing, very early flowering habit
7	Lankon	CV	White, Brown mixed	Very early and pendant
8	Hinomoto upward(HU)	Breeding line	White	Upward facing, early flowering habit
9	Ishaghaki (IS)	Breeding line	White	Upward directional, Mid flowering habit
10	White American (WA)	CV	White	Upward facing, early flowering habit
11	White heaven	CV	White	Upward directional, early flowering habit
12	White tower	CV	White	Upward directional, early flowering habit
13	White trumph	CV	White	Side facing, Mid flowering habit
14	Watch up	CV	White	Upward directional, early flowering habit
15	Woori tower	CV	White	Upward facing, early flowering habit
16	12-1	Breeding line	White	Upward facing, late-flowering habit

2 Materials and Methods

2.1 Plant material

The plant materials employed for this study represented 13 different cultivars (CVs) and three breeding lines of *L. longiflorum* (Table 1). These cultivars and breeding lines were obtained from the floricultural breeding laboratory under the Department of Horticulture, Kangwon National University (KNU), South Korea.

2.2 Experimental setup and raising the plant materials

The commercial-sized bulbs (10-15mm) of all genotypes selected in this experiment were grown for the evaluation inside the plastic house laid out in the randomized complete block design (RCBD) with three replications (Table 1). The genotypes were grown in multiple beds measuring 30m in length and 1 m in width containing well-crushed coco-peat as growth media. The bulbs of all genotypes were planted using the planting net maintaining plant to plant and row to row equidistance of 12.5cm. Each bed contained four drip irrigation hoses fitted along the lengthwise of the bed for the provision of irrigation. All the plant saplings of all genotypes were grown well, adopting standard seedlings growing cultural practices followed by local lily grower farmers to produce healthy plant saplings to achieve the proper results.

2.3 Morphological observation and sampling

The morphological observation of all studied growth and flowering traits has been carried out during the main flowering season viz. June (4th week onward), July, and August (up to the third week) for selecting the four reliable plants from each replication. The measurement of all ten traits was carried out based on the guidelines of lily register checklists as provided by Mathews (2007) and following the general method of Rai et al. (2018), sampling four plants from each replication and in total 12 plants of each genotype.

2.4 Statistical analysis

The data measured for all studied growth and flowering traits of all genotypes were first prepared using MS-Excel-2013 and then employed for analysis using TNAUSTAT statistical packages. The estimation of Variance (ANOVA), F test, and other variance components were carried out following the plant breeding general tools of the TNAUSTAT statistical package for the randomized complete block design (RCBD). Likewise, Duncan's multiple range test was estimated at the 5% level of significance for the separation of estimated means of the treatments adopting the available designs, DMRT comparison tools of the TNAUSTAT statistical package (Manivannan, 2014). Furthermore, variance components viz. phenotypic, genotypic, and environmental variance and their variance coefficients were estimated according to the formula and detail methods provided by Burton & Devane (1953). Likewise, broad-sense heritability (H^2), genetic advance (GA), and genetic advance as percent of the mean (GAM) were estimated based on the formula given by Allard (1960).

3 Results and Discussion

3.1 Analysis of variance (ANOVA) and components of variance

The analysis for variance (F test; Table 2) demonstrated that all the studied traits viz. height of the plant, diameter of the stem, leaf numbers, length of leaves, width of the leaves, flower number, duration of flowering, diameter of flower, length of bud, and the axis of flower revealed genotypes are significantly varied with each other (P<0.01). The results indicated that 16 studied genotypes were different in their potentiality to perform variable growth and flowering traits. The genotypic variance, phenotypic variance, environmental variance, and its components, genetic advance, and genetic advance as percent of the mean are presented in table 2. The estimated PCV for all studied growth and flowering traits was found slightly higher than GCV, indicating the impact of environmental effect on the expression of these traits. The magnitude of the estimated PCV and GCV of the studied growth and flowering traits in this study are ranged between 11.15 and 6.73 (lowest for flower diameter), and 45.08 and 45.01 (the highest for the angle of flower) respectively (Table 2). It is proof of the existence of substantial broadly based genetic variability. Based on classification as provided by Deshmukh et al. (1986), PCV and GCV values can be categorized into three categories i.e. high (>20%), medium (10-20%), and low (<10%). According to the estimated PCV and GCV values in this experiment, out of the ten studied growth and flowering traits, four traits viz. number of leaves, leaf width, number of flowers, and the angle of flower had demonstrated higher PCV and GCV which ranged from 23.46 to 45.08. According to Khan et al., (2009), high PCV demonstrated the existence of huge scope for selection of the traits under study. On the other hand estimated higher GCV indicates the presence of exploitable genetic variability for these traits which may helpful for selection (Yadav, 2009). The results indicated that the tested genotypes had the more outstanding and practical possibility for improvement if these traits are considered during selection and hybridization. On the other hand, growth and flowering traits viz. plant height, stem diameter, leaf length, days to flowering, and bud length had demonstrated moderate PCV and GCV value which had ranged from 12.32 to 19.98, thereby indicating the acceptable improvement could be obtained through selection and of these traits. While only one remaining trait viz. flower diameter demonstrated a moderate PCV value (11.15) and lower GCV value (6.73). Though GCA provides information on the genetic variability existing in quantitative traits, this is only not the criteria to determine the amount of variation that was heritable, but GCA, along with the heritability estimates, would give an exact picture of heritable variation.

Table 2 ANOVA and estimation of a genetic component for growth and flowering traits in *L. longiflorum* genotypes

Traits /Parameter	G. Mean ± SEM	LSD (5%)	CV (%)	F test	PCV (%)	GCV (%)	ECV (%)	H^2 (%)	GA	GAM (%)
Plant height(cm)	78.77±1.20	11.53	1.98	**	19.98	19.88	1.98	99.02	25.43	40.75
Stem diameter	7.72±0.10	0.98	1.92	**	16.26	16.14	1.92	98.61	2.41	33.02
No. of leaves	56.87±1.20	11.40	4.99	**	28.27	27.82	4.99	96.88	45.31	56.41
Leaf length	11.98±0.20	1.70	4.65	**	19.22	18.65	4.65	94.15	8.65	37.28
Leaf width	1.95±0.0	0.30	4.14	**	23.82	23.46	4.14	96.98	1.29	47.59
No. of flowers	3.33±0.1	0.70	6.95	**	27.95	27.07	6.95	93.82	3.57	54.02
Days to flowering	78.43±0.8	8.30	0.53	**	15.04	15.03	0.53	99.88	6.83	30.93
Flower diameter	49.4±0.24	2.39	8.89	**	11.15	6.73	8.89	36.42	26.36	8.36
Bud length	14.8±0.1	1.30	2.32	**	12.53	12.32	2.32	96.58	5.46	24.93
Angle of flower	58.95±1.93	19.05	2.65	**	45.08	45.01	2.65	99.65	25.65	92.55

Abbreviation ANOVA=Analysis of variance, G.Mean=General Mean, SEM=standard error of mean, SD=Least significance difference, CV=Coefficient of variation, PCV=Phenotypic coefficient of variation, GCV=Genotypic coefficient of variation, ECV=Environmental coefficient of variation, H^2=Heritability, GA=Genetic advance, GAM=Genetic advance as percentage of mean

3.2 Heritability

It is one of the most important insights to decide for the breeders on selecting and improving any breeding scheme. According to Ndukauba et al. (2015), heritability provides an idea of the extent of genetic control for the expression of a particular trait and the reliability of phenotype in predicting its breeding value. Based on estimated heritability, it is easier to understand whether their presence between parents and offspring for a particular trait has strong (having high heritability) or low level of (low heritability) resemblances (Ranjan & Gautam, 2018).

Based on the classification provided by Singh (2001), the obtained heritability can be categorized into very high, moderately high, medium, and low categories. In the current study, the estimation of broad-sense heritability for all studied growth and flowering traits had ranged from 36.42% (for flower diameter) to 99.88% (for days to flowering). Based on heritability classification, all the studied traits besides flower diameter had possessed very high heritability. There should have less environmental impact on the observed variation of these traits (Eid, 2009). The heritability estimation indicates the presence of a positive response of improvement through the selection of these traits.

3.3 Genetic advance and Genetic advance as percent of the mean (GAM)

The knowledge of heritability and genetic advance is more useful since heritability alone does not indicate the amount of genetic improvement resulting from the selection of individual genotypes (Gebregergs & Mekbib, 2020). According to Ullah et al. (2012), higher heritability coupled with higher genetic advance is more useful than heritability alone. Further, according to the classification of Johnson et al. (1955), genetic advance as percentage of mean (GAM) can be categorized into three groups i.e. high (>20%), moderate (10-20%), and low (0-10%). In this experiment, the estimated GAM had ranged from 8.36 (for flower diameter) to 92.55 (Table 2). Furthermore, almost all growth and flowering traits under study viz. plant height, stem diameter, number of leaves, leaf length, leaf width, number of flowers, days to flowering, bud length, and angle of flower had possessed high GAM while remaining one trait flower diameter had recorded low GAM as we have achieved high heritability along with high GAM for nine growth and flowering traits that these traits were controlled by additive gene action and thereby indicated the most effective condition for the selection process for the further breeding schemes (Tazeen et al., 2009; Ndukauba et al., 2015).

3.4 Mean performances

The estimated mean performance for ten growth and flowering traits demonstrated huge variability in these traits among the studied 16 *L. longiflorum* genotypes (Table 3). The plant height is one of the essential traits from the cut flower point of view; in this study, global beauty and white tower were found tallest than other remaining genotypes. Likewise, among the 16 genotypes, *L. longiflorum* cultivar 'White American (WA)' and breeding line IS (Ishaghaki) were found shortest for their mean performance in plant height. In the second estimated trait, i.e., stem diameter, we have discovered the thickest stem for white triumph while the thinnest stem was jointly recorded for Gelria and Georgia. Likewise, *L. longiflorum* cultivar' Global village' possessed the highest number of leaves while IS had the lowest number of leaves in their stems. The longest leaves were recorded for white heaven, and the shortest leaves were measured for the global village and Lankan.

Table 3 The mean performance for major growth and flowering traits of *L. longiflorum* genotypes

Traits /Genotypes	Plant height	Stem diameter	No. of leaves	Leaf length	Leaf width	No. of flowers	Days to flowering	Flower diameter	Bud length	Angle of flower
Bright tower	87.5[c]	6.65[i]	48.6[f]	11.8[gh]	2.3[b]	2.5[fg]	81.7[f]	49.40[bc]	14.7[ef]	63.75[g]
Faith	62.8[g]	9.69[b]	57.3[e]	11.1[hi]	1.7[e]	2.6[fg]	74.3[i]	45.15[c]	14.8[e]	85.00[a]
Global beauty	100.4[a]	9.16[c]	83.4[b]	12.1[fg]	2.3[b]	3.4[d]	88.5[e]	59.80[a]	16.3[bc]	67.50[f]
Global village	71.1[e]	7.28[g]	93.5[a]	8.0[k]	1.1[g]	3.6[d]	68.5[k]	50.46[bc]	10.9[j]	82.50[ab]
Georgia	61.6[g]	6.29[j]	44.2[fg]	12.1[fg]	2.1[c]	2.4[gh]	63.6[l]	44.70[c]	12.8[h]	18.13[i]
Gelria	65.5[f]	6.20[j]	43.3[gh]	14.1[bc]	1.7[e]	2.8[f]	68.8[k]	45.13[c]	16.4[bc]	26.88[h]
Lankon	92.3[b]	7.23[g]	71.1[c]	7.7[k]	1.5[f]	3.6[d]	60.4[m]	44.78[c]	11.7[i]	11.67[j]
Hinomoto upward	68.8[e]	6.72[hi]	41.3[hi]	11.7[gh]	1.6[ef]	2.5[fg]	73.5[j]	48.78[bc]	13.5[g]	78.75[cd]
IS	53.8[h]	6.94[h]	38.5[i]	14.6[b]	2.0[cd]	2.2[h]	75.4[h]	46.73[bc]	15.9[cd]	68.33[f]
White American	56.1[h]	6.66[i]	47.4[fg]	13.2[cd]	1.9[d]	3.1[e]	73.1[j]	47.16[bc]	16.7[b]	71.25[e]
White Heaven	92.6[b]	8.33[d]	45.6[fg]	15.6[a]	2.4[b]	2.5[fg]	89.4[d]	55.27[ab]	17.3[a]	28.12[h]
White tower	98.1[a]	8.18d[e]	69.9[cd]	13.0[de]	1.7[e]	4.3[c]	74.3[i]	49.99[bc]	15.6[d]	72.50[e]
White trumph	84.9[c]	10.57[a]	58.9[e]	13.9[bc]	2.0[cd]	4.7[b]	76.3[g]	54.26[ab]	15.6[d]	26.27[h]
Watch up	90.2[b]	7.70[f]	54.3[e]	10.7[i]	2.1[c]	3.5[d]	90.1[c]	49.30[bc]	14.1[f]	81.27[bc]
Woori tower	92.8[b]	8.03[e]	65.4[d]	12.9[ef]	1.7[e]	4.6[b]	92.0[b]	50.72[bc]	16.3[bc]	76.25[d]
12-1	81.8[d]	7.91[ef]	47.2[fg]	9.1[j]	3.1[a]	5.0[a]	104.8[a]	48.77[bc]	14.2[ef]	85.00[a]
G Mean	78.8	7.72	56.9	12.0	1.9	3.3	78.4	49.40	14.8	58.95
SEM	1.2	0.10	1.2	0.2	0.0	0.1	0.8	0.24	0.1	1.93

In the case of another growth trait, leaf width; the broadest leaves were measured for 12-1, while narrowest leaves were found in the stem of 'global village.' Additionally, for more numbers of flowers that deserved vital importance from the cut flowers point of view, the highest numbers of flowers were found for 12-1, while the fewest number of the flower were recorded in the stem of IS. Lankan has very early flowering habit with average days to flowering 60.4 days while very late flowering cultivar was recorded for 12-1 with 104.8 days to the flowering period from transplanting bulbs. On average, most enormous flowers were found for global beauty, while smaller flowers were recorded for Faith, Georgia, Gelria, and Lankan. The longest bud was measured for white heaven, while the shortest buds were calculated for the global village. The last and most important trait, ' flower direction,' is synonymously used as the attitude of the floral axis; most upward-facing flowers were found for Faith while pendulous flower habit 'Lankan' flowers consisted lowest flower angle. The mean performance evaluation is one of the insights which emphasize the selection of the progeny based on the superior performance of a particular trait of interest. Rai et al. (2018) revealed varied mean performances are due to the genetic variability of the studied genotypes.

Conclusions

Results of the study can be concluded that all the sixteen genotypes were statistically significant for all the studied growth and flowering traits, thereby revealing the presence of an enormous amount of genetic variability. The characteristics viz. number of leaves, leaf width, numbers of flowers, and the angle of flower had possessed high GCV and PCV along with high heritability and GAM while traits viz. plant height, stem diameter, leaf length, days to flowering, and bud length had demonstrated moderate GCV and PCV value along with high heritability and GAM thereby indicating the potentiality for the selection and improvement of these traits. On the other hand, as flower diameter had possessed lower GCV and PCV coupled with low heritability and GAM, there is a limitation for this trait for selection and improvement through cross-breeding.

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Conflict of Interest

There is no conflict of interest between the authors about the content of the article

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