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Effect of Re-Using Vernalization Bulbs on Growth and Development of Lilies

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ABSTRACT

Purpose: *Selecting bulbs based on their quality as planting materials is a critical point in obtaining high qualified growth and development of lilies, however published reports in re-using bulbs on cultivation of lilies were still limited . Objective of the research was to reveal effect of re-using vernalization bulbs on growth and development of lily. The research was conducted in plastic house of Segunung Experimental Gerden, Indonesian Ornamental Crops Research Institute.*

Research Method: *Materials used in the study were lily bulbs harvested from the third cultivation period of Asiatic lily, Lilium 'Delina', L. 'Liana', L. longiflorum and L. 'Renata'; two bulb sizes of them were grade A and B. The experiment was arranged in split plot design with three replications. Main plot was two bulb sizes of the grade A and B; while sub-plot was five lily varieties of Asiatic, Delina, Liana, Longiflorum and Renata.*

Findings: *Results of the study revealed that re-utilizing lily bulbs harvested from third cultivation period still had high potential in obtaining better growth and development of lily both vegetatively and generatively. The such bulbs kept resulting in optimal growth performances in all lilies tested. Higher bulb sizes produced better lily performances grade A > grade B in the most varieables observed, while lily varieties indicated varied growth responses in all variables observed. The best variety of lily growth and preformances vegetatively was showed by Longiflorum, however better aspects was noted on Liana. While in interaction effect, grade A of Delina gave better results in initial influorescent formation after 71.3 days after culture with 102.3 days initial flower buds opened, 100% plants flowered, 3 flower buds per plant, 18.7 cm flower bud diameter and 45 mm bulb diameter. The results of the study gave evidents that re-utilizing lily bulbs derived from third cultivation period still could be applied to produce better quality and quantity of lily flowers.*

Limitations: *The limitations of the research are apparent to the fact that the research was only focused on re-utilization of lily bulbs harvested from third cultivation period.*

Originality/ Value: *Results of the research can be used as important considerations for growers and farmers on producing maximal lily flower on fulfilling market and customer demand under re-utilization of lily bulbs.*

Keywords: *lilium; bulbs, flower production, re-utilization, size, and variety*

INTRODUCTION

Lilies (*Lilium spp*.), as one of the important genera of flower bulbs, belong to family Liliaceae (Asker, 2012). In the floriculture industry, the plants are classified into several main groups based on their morphological, anatomical structures and development patters in one hand, and in the other hand based on their

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growth and environmental conditions (Grassotti and Gimelli, 2011; Roh, 2011; Van Tuyl and Arens, 2011). The main groups are Asiatic hybrids, LA hybrids (*Lilium longiflorum* × Asiayic lily), Oriental hybrids and longiflorum hybrids (*L. longiflorum*), OT hybrids (Oriental × Trumpet lilies) and LO hybrids (*L. longiflorum* × Oriental lilies) (Al-Allaq *et al*., 2014). The lilies are the most important cut flower worlwide, involving in Indonesia. The plants were cultivated commercially in Cianjur, Cisarua, Cihideung-West Java; Batu-East Java and Tabanan-Bali. The flowers are sold between US$ 22.06 to 66.17 per bunch (@ 5 stems). Market and customer demand of the cut flower increase gradually year by year, however availability of qualified-planting materials to develop the plant commercially is constrained.

Conventionally, lilies can be propagated generatively using seeds and vegetatively with bulbils from the stem, bulblets around the stem base and scales (Hartmann *et al*., 1997; Thomson, 2007). In Indonesia, commercial cultivation of lilies was carried out by rich farmers and growers. They generally used imported bulbs ready planted, however production cost for the planting materials was very high. Price of the imported bulbs was US $ 2.57 - 3.53 per bulb and the production cost can not be covered by poor farmers. Therefore the farmers generally utilized un-qualified lili planting materials to reduce the production cost as low as possible. Normally the poor farmers used varied-sizes and non-qualified bulbs or seeds for low production cost. The situation resulted in low quality and productivity of their products leading to less profit obtained in every cultivation period.

In Indonesia, propagation of lilies generally used several methods as described previously. The methods were applied by most of Indonesian farmers, however production of qualified-lilium bulb commercially is never reported. Several *in vitro* propagation researches of lilies were reported using bulb scales as explant source on *Lilium longiflorum* (Priyono and Winarsih, 2000; Priyono, 2001; Haryati, 2015), on Oriental lily (Pramanik and Rachmawati, 2010) and filament of Oriental lily (Kurniati *et al*., 2012). From these study, end products of the researches were micro-bulbs and plantlets. To process the end products till obtaining production bulbs ready for commercial scale cultivation, enlargement process of the micro-bulbs and/or plantlets into production bulbs is significantly addressed and one of the processes in enlargement of them was re-using bulbs.

In the present study, re-using vernalization bulbs on growth and development of lilies was main objective in the research. Furthermore the growth and develpment of lilies derived from re-using vernalization bulbs harvested from the third cultivation period was explored and performed in the study.

MATERIALS AND METHODS

The experiment was conducted at Segunung Experimental Garden of Indonesian Ornamental Crop Research Institute 1100 meter above see level. Planting materials used in the experiment was lilium bulbs harvested from third cultivation period of *Lilium longiflorum*, *Lilium* 'Renata', 'Delina', 'Liana' and 'Asiatic'. After harvesting, the bulbs were sorted and clusterred into two different grades of A and B based on bulb diameters (Table 1) and then vernalized at 4-6°C and 90% relative humidity for 6 weeks. After vernalization, the bulbs were re-planted on cultivation bed prepared previously.

Cultivation beds were prepared by hoing the land areas conventionally using hoe in ± 30 cm in depth. The cultivation areas were then added by burned-rice husk and bamboo moss and fumigated using 98% dazomet for 14 days. After the fumigation, the cultivation areas were made into beds. Size of beds were 1 × 1 m^2 (length and width) with 50 cm distance between each bed and watered sufficiently. Before bulb planting, adding 20 g/m^2 gliocompost was carried out to improve quality of media for bulb growth.

Table 01: Two grade bulbs and five lily varieties tested in the experiment

No.	Variety of lily	Bulb diameter (cm)	
		Grade A	Grade B
1.	Asiatic	5.3 – 7.0	1.8 – 3.1
2.	Delina	3.1 – 5.6	1.2 – 2.0
3.	Liana	4.3 – 6.0	2.6 - 3.9
4.	Longiflorum	5.7 – 7.0	3.8 – 5.3
5.	Renata	4.3 – 5.3	2.0 – 3.7

Vernalized-bulbs in two different grades of A and B successfully prepared were then planted in the cultivation beds with 20 × 20 cm planting distance. The bulbs were planted by making holes in the beds in ± 7 cm in diameter and depth, putting the bulbs in the hole with one bulb for one hole, covering the bulb with media and watering beds with water sufficiently.

After bulb planting, the bulb growth was maintained by watering, weed handing and pest-disease controlling. Watering of lily sufficiently was carried out everyday in the morning manually using plastic pipe. Weed handling was done when growth of weeds inhibited the growth of lily plants. The weed handling was carried out manually by hand. While pest and disease controlling was conducted by spraying pesticide in suggested dosage depending on targets of pests and diseases using Score, Anvil and Preficure. Frequency of pesticide application was depended on pest and disease intensity

Treatments in the experiment were two grades of bulbs and five lily varieties (Table 1). The experiment was arranged in split plot design with two grades of bulbs as main plot and five lily varieties as subplot and three replications. Each treatment consisted of 10 bulbs. Total bulbs used in the study were 300 bulbs.

Variables observed in the study were (1) percentage of bulb growth (%), counted by calculating number of bulbs growth divided by total number of planted-bulbs times by 100%; (2) plant height (cm), measured from base part of stem till tip of shoots using ruler; (3) number of leaves in flower harvesting; (4) stem diameter (mm), measured in widest part of stem using digital caliper; (5) influorescent initiation period (days), counted started from initial planting till early flowering easily observed, (6) initial flower buds opened (days), counted started from initial planting till early flowered buds opened easily observed, (7) number of flower buds per stem, (8) flower diameter (cm) and (9) bulb diameter (cm), measured on widest part of its using digital caliper. Periodical observation was carried out to follow bulb growth response till flower harvesting. All variables were measured in flower harvesting time.

All data collected from the experiments were analyzed by analysis of variance (Anova) using SAS program Release Windows 9.12. Significant differences between means were assessed by Tukey's Studentized Range (HSD) at $p = 0.05$ (Mattjik and Sumertajaya, 2006).

RESULTS AND DISCUSSION

Under periodical observation it was known that vernalized-lilium bulbs produced initial shoots 10 days after culture. The initial shoots grew continually and increased in height of shoots and number of leaves. After 43-145 days of culture, height of shoots and number of leaves reached 43-217 cm and 31-174 respectively. After the height of plants reaching 48-217 cm, influorescent initial formation was noted 27 to 150 days with 50 to 180 days after culture for initial flower buds opened. In the end of experiment, the height of plant was varied from 30-220 cm, 3.5 to 13.0 mm in stem diameter, 30 to 180 leaves per plant, 1 to 4 flower buds per plant, 7 to 19 cm in flower bud diameter and 3 to 7 cm in bulb diameter. Varied-performances of

lilium was recorded on Asiatic and Longiflorum lily under re-utilizing bulbs harvested from third cultivation period.

Statistically two grades of bulbs and five lily varieties gave significant effect on growth and development of lilies. Higher size of bulbs higher effect on growth and development of lilies both vegetatively and generatively. Grade A bulbs showed higher effect on vegetative growth of lilies compared to the grade B bulbs. The grade had height of shoots up to 97.8 cm with 7.9 cm in stem diameter, 99.3 leaves per plant for vegetative growth (Fig. 01A); The grade was also induced high percentage of flowered plants up to 90.6% with 87.6 days influorescent initial formation, 95.2 days initial time for flower buds opened, 2.8 flower buds per plant, 13.8 cm flower bud diameter and 41 mm bulb diameter (Fig. 01B). While based on varieties of lily tested, Longiflorum was the most responsive variety of lily in vegetative growth compared to others. The lily grew with 213 cm in height, 11.8 mm in stem diameter and 268.9 number of leaves per plant (Fig. 02A). The second best response was performed by Liana, while the lowest growth indicated by Delina. Furthermore on generative growth, especially for faster influorescent initial formation and initial time for flower buds opened, Renata was type of lily with the shortest influorescent initial formation of 38 days and 64 days for initial time for flower buds opened; however the variety stimulated lower results in number of flower buds per plant, flower bud diameter and bulb diamater (Fig. 02.B). Though Liana formed influorescent and initiation time for flower buds opened in longer period compared to Renata, Asiatic and Delina, the variety was noted as lily variety with high number of flower buds per plant up to 3.4 buds with smaller flower bud diameter (Fig. 02B). Low results were generally indicated by Asiatic lily

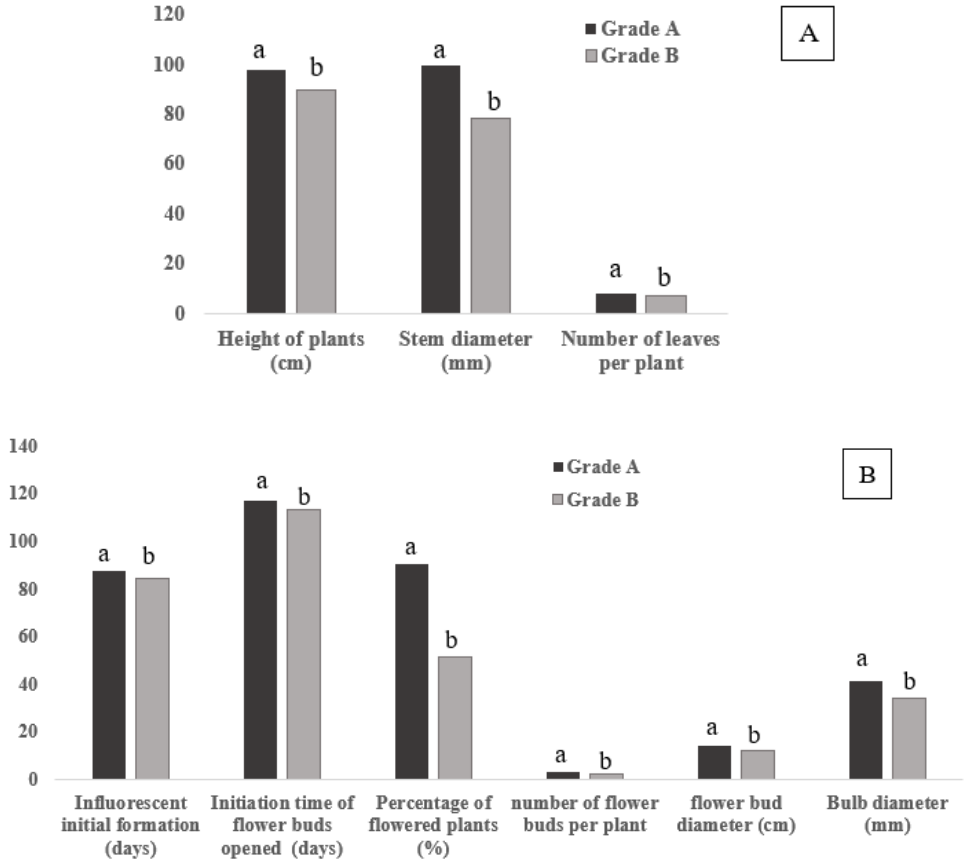

Figure 01: Effect of two grades of bulbs on vegetetive and generative growth of lilies. A. Effect of two grade bulbs on vegetative growth and development of lilies. B. Effect of two grade bulbs on generative growth and development of lilies.

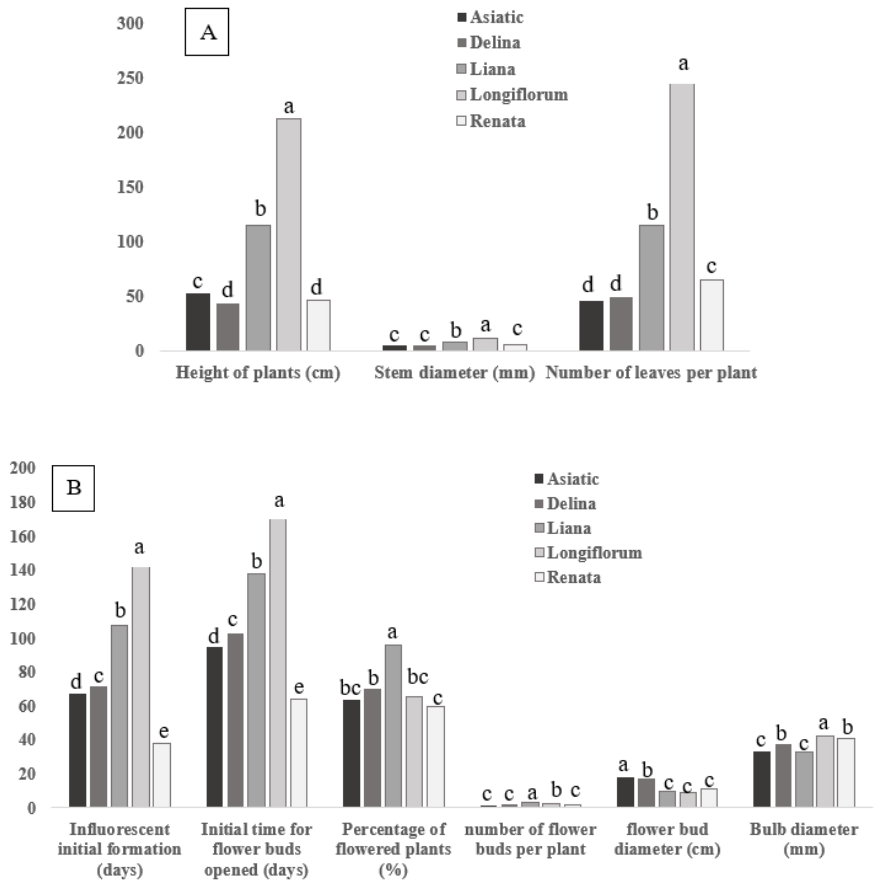

Figure 02: **Effect of five lily varieties on vegetative and generative growth of lilies. A. Effect of five lily varieties on vegetative growth and development of lilies. B. Effect of five varieties of lilies on generative growth and development of lilies.**

Two grades of bulbs and five lily varieties also gave significant interaction effect on growth and development of lilies both vegetatively and generatively. Grade A bulbs of longiflorum was suitable combination treatment in obtaining the best vegetative growth up to 215.8 cm in plant height (Table 02) with the highest stem diameter of 12.3 mm (Table 03) and 174.9 leaves per plant (Table 04), however the combiation resulted in low quality on generative performances. While grade A bulbs of Delina indicated better results on generative growth of lily compared to others. In the generative stage, grade A bulbs of Delina regenerated initial influorescent formation after 71.3 days after culture (Table 05) with 102.3 days initial flower buds clearly opened (Table 06), 100% plants flowered (Table 07), 3 flower buds per plant (Table 08), 18.7 cm flower bud diameter (Table 09) and 45 mm bulb diameter (Table 10). The second best results was indicated by Renata. While Liana gave better results and performances compared to Longiflorum. From the study, it was clearly revealed that better performance lily growth vegetatively and generatively under re-useable vernalization bulbs was indicated by Delina. In the variety, enlargement of bulb size was noted both in grade A and B with 2.9 and 93.2%, respectively (Table 11).

Entirely from the study it was successfully revealed that re-using bulbs derived from the third harvesting period still gave better results on growth and development of lilies. Grade A bulbs produced higher results and performances of lilies on height of plants, stem diameter and number of leaves per plant, percentage of flowered plants, number of flower buds per plant, flower bud diameter and bulb diameter. Addai and Scott (2011) used > 30 g of lily bulbs as planting materials to produce high vegetative biomass, flower production

and bulblet formation. In other studies, 3-4 cm bulb sizes resulted in higher percentage on sprouting up to 96.6% with 55.9 cm plant height, 77.4 number of leaves per plant, 96.3 days for flowering, 3.4 spikes per plant, 1.13 cm thickness of spike, 36.7 cm length of spike, 36.7 florets and 20 days flower longevity of *P. tuberosa* (Ahmad *et al*., 2009). In *Lachenalia*, higher bulb sizes of 5.1-6.0 cm produced higher plant height up to 31 cm, 3 leaves per plant, 31.5 cm leaf leangth, 3.6 cm leaf width, 20.7 cm inforescence length, 21 florets, and 2.7 cm floret length (Kapczyńska, 2014). Furthermore in the study it was also found that different types of lilies indicated different responses on growth and their development with the best performances recorded on Longiflorum in longiflorum type and Delina in Asiatic types. While in another study, Rosabeth gave higher results on vegetative and regenerative growth variables observed compared to Namakwa and Ronina of *Lachenalia* (Kapczyńska, 2014).

Table 02: Intveraction effect of two grade bulbs and five lily varieties of on plant height (cm)

Variety of lily	Two grade bulbs	
	Grade A	Grade A
Asiatic	56.2 c	50.5 c
Delina	57.2 c	30.8 d
Liana	110.7 b	112.3 b
Longiflorum	215.8 a	210.2 a
Renata	49.1 c	43.2 cd
Coefficient of variation (%)	5.65	5.95

Table 03: Interaction effect of two grade bulbs and five varieties of lily on stem diameter (mm)

Variety of lily	Two grade bulbs	
	Grade A	Grade A
Asiatic	5.8 c	4.8 c
Delina	6.2 c	4.3 c
Liana	8.8 b	8.3 b
Longiflorum	12.3 a	11.0 a
Renata	6.2 c	6.3 b
Coefficient of variation (%)	7.83	12.33

Table 04: Interaction effect of two grade bulbs and five varieties of lily on number of leaves per plant

Variety of lily	Two grade bulbs	
	Grade A	Grade A
Asiatic	57.6 d	33.3 cd
Delina	66.8 d	31.3 d
Liana	114.3 b	116.0 b
Longiflorum	174.9 a	162.8 a
Renata	82.6 c	48.4 c
Coefficient of variation (%)	4.77	7.57

Means followed by the same letter in the same column are not significant different based on HSD (p=0.05).

Table 05: Interaction effect of two grade bulbs and five varieties of lily on inflourescent initiatial formation (days)

Variety of lily	Two grade bulbs	
	Grade A	Grade A
Asiatic	69.7 c	64.8 c
Delina	72.7 c	71.3 c
Liana	105.0 b	110.3 b
Longiflorum	145.3 a	145.7 a
Renata	47.3 d	28.7 d
Coefficient of variation (%)	4.10	3.68

Means followed by the same letter in the same column are not significant different based on HSD (p=0.05).

Table 06: Interaction effect of two grade bulbs and five varieties of lily on initial time for flower buds opened

Variety of lily	Two grade bulbs	
	Grade A	Grade A
Asiatic	97.7 c	92.7 c
Delina	102.3 c	103.0 c
Liana	135.0 b	140.7 b
Longiflorum	175.7 a	175.3 a
Renata	73.7 d	54.3 d
Coefficient of variation (%)	4.07	3.61

Means followed by the same letter in the same column are not significant different based on HSD (p=0.05).

Table 07: Interaction effect of two grade bulbs and five varieties of lily on percentage of flowered plants

Variety of lily	Two grade bulbs	
	Grade A	Grade A
Asiatic	95.0 b	33.0 c
Delina	100 a	40.7 c
Liana	100 a	91.7 a
Longiflorum	58.3 c	73.0 b
Renata	100 a	19.3 d
Coefficient of variation (%)	1.41	9.35

Means followed by the same letter in the same column are not significant different based on HSD (p=0.05).

Table 08: Interaction effect of two grade bulbs and five varieties of lily on number of flower buds per plant

Variety of lily	Two grade bulbs	
	Grade A	Grade A
Asiatic	2.3 b	1.3 c
Delina	3.0 ab	1.1 c
Liana	3.3 a	3.5 a
Longiflorum	2.9 ab	2.5 ab
Renata	2.5 ab	1.7 bc
Coefficient of variation (%)	11.89	21.14

Means followed by the same letter in the same column are not significant different based on HSD (p=0.05).

Table 09: Interaction effect of two grade bulbs and five varieties of lily on flower bud diameter (cm)

Variety of lily	Two grade bulbs	
	Grade A	Grade A
Asiatic	17.9 ab	17.7 a
Delina	18.7 a	16.1 a
Liana	9.6 c	9.6 b
Longiflorum	9.5 c	8.3 b
Renata	13.3 bc	9.4 b
Coefficient of variation (%)	14.45	13.50

Means followed by the same letter in the same column are not significant different based on HSD (p=0.05).

Table 10: Interaction effect of two grade bulbs and five varieties of lily on bulb diameter (mm)

Variety of lily	Two grade bulbs	
	Grade A	Grade A
Asiatic	17.9 ab	17.7 a
Delina	18.7 a	16.1 a
Liana	9.6 c	9.6 b
Longiflorum	9.5 c	8.3 b
Renata	13.3 bc	9.4 b
Coefficient of variation (%)	14.45	13.50

Means followed by the same letter in the same column are not significant different based on HSD (p=0.05).

Table 11: Reducing or increasing of bulb diameter in initial culture compared to in harvesting time of two grade bulbs (cm)

Variety of lily	Grade A (cm)		Percentage of reduction / enlargement of bulb size (%)	Grade B (cm)		Percentage of reduction / enlargement of bulb size (%)
	Initial culture	Harvesting time		Initial culture	Harvesting time	
Asiatic	6.15	3.53	- 42.6	2.45	3.30	+ 34.6
Delina	4.37	4.50	+ 2.9	1.63	3.15	+ 93.2
Liana	5.13	3.13	- 38.9	3.35	3.23	- 3.6
Longiflorum	6.37	4.47	- 29.8	4.57	3.73	- 18.4
Renata	4.75	4.53	- 4.6	2.83	3.45	+ 21.9

Under combination treatments it was established that grade A bulbs of Longiflorum indicated high results on vegetative growth and development of lilies. While for generative variables, grade A bulbs of Delina exhibited better responses with high percentage of flowered plants, number of flower buds per plant, number of flower bud diameter and bulb diameter (Table 07, 08, 09 and 10). In different study, 3-4 cm in diameter bulbs of *P. tuberosa* cv. Single resulted in higher percentage of sprouting, plant height, number of leaves per plant, days for flowering, number of spikes per plant, thickness of spike, length of spike, number of florets and flower longevity (Ahmad *et al*., 2009). Bulb sizes of 5.1-6.0 cm of Rosabeth was the best combination to regenerate higher plant height, number of leaves per plant, leaf leangth, leaf width, inforescence length, number of florets and floret length (Kapczyńska, 2014).

CONCLUSION

From the study it can be concluded that re-utilizing bulbs harvested from third cultivation

period kept resulting optimal growth performances of lilies. Higher bulb sizes produced better lily performances in vegetative and generative growth; grade A was better than grade B. Varieties of lily indicated different and varied growth responses in all variables observed. Though best response of lily growth was showed by Longiflorum vegetatively, best generative growth was noted on Liana. While in interaction effect, grade A of Delina was better combination treatment in obtaining better vegetative and generative growth and performances of lily under re-utilization of vernalization bulbs harvesting from the third cultivation period.

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Conflict of Interest

We declare that there is no conflict of interest dealing with authors and Indonesian Agency for Agriculture Research and Development that facilitated and funded the research activities.

Author Contributions

Debora Herlina and Budi Winarto play important roles equally in designing, carrying out and analysing all data regenerating from the research till writing, editing and approving the final manuscript.

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