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Vegetative Propagation of Amaryllis (*Hippeastrum × johnsonii*) by Different Cutting Methods

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Abstract

Amaryllis (*Hippeastrum×johnsonii*) is commonly propagated by three methods: seeds, offset bulblets, and twin scaling. Since the number and size of bulblets produced by these methods are low, we conducted an experiment to evaluate different bulb cutting methods for propagation. For this purpose, bulbs (circumference of 30 - 35 cm) were sectioned or notched into 8, 12, or 16 segments or twin-scaled into 48, 72, or 96 segments for bulblet formation. Our results show that the largest number of bulblets was produced by the twin scaling method, but they take longer to grow to a final, commercial size. Sectioning and notching resulted in larger, but fewer bulblets than twin-scaling. Compared to notching, sectioning provided more space for the bulblets to grow, and is therefore the recommended method. While increasing the number of sections cut from a single bulb resulted in a larger number of bulblets, the diameter of the bulblets decreased. Therefore, sectioning the bulb into 8 segments was the best method for producing an acceptable number of vigorous bulblets.

Additional key words: Hippeastrum, cutting methods, bulblet, propagation.

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Introduction

Amaryllis (*Hippeastrum × johnsonii*), a perennial herbaceous plant and member of the Amaryllidaceae family, has become an internationally popular bulbous plant in recent years due to its large beautiful flowers and elegant green foliage (Ye and Shi, 2008). It is propagated in four different ways: seeds, offset bulblets, twin scaling, and tissue culture (Sultana et al., 2010; Siddique et al., 2006; Ilczuk et al., 2005). Seeds are usually used to develop new varieties due to the large variation in the characteristics of flowers, plant shape, and flowering time (Zhang et al., 2013). Offset division is applied to cultivars that produce at least three bulblets per year. In most cases, propagation is performed by a method known as twin scaling (Vijverberg, 1981). However, the small daughter bulbs developed from twin-scales need approximately three years to produce mature bulbs (Okubo, 1993). *In vitro* propagation through tissue culture has been successfully used for bulbous plants, including Amaryllis (Amani et al., 2015). However, compared to other methods, in vitro propagation is costly (Zhang et al., 2013). In the commercial propagation of bulbous plants, the long growth period required for producing a salable flowering bulb is a major concern. Additionally, scale rottenness is a major factor leading to the loss of a large number of scales during the propagation process. Therefore, choosing the propagation method that maximizes the proliferation rate while reducing losses to scale rotting is extremely important for achieving optimum economic efficiency. In previous investigations, different methods were assayed for the propagation of Amaryllis, including chipping, notching, and twin scaling, with different results reported. Some researchers showed that notching and chipping the bulb are the best methods for bulb propagation (Zhu et al., 2005; Ephrath et al., 2001) while some recommended the twin scaling method (Andrade-Rodriguez et al., 2015). Furthermore, none of the studies examined the scooping method. Thus, the best method for Amaryllis propagation remains to be determined. In this study, common cutting methods such as sectioning, notching, and twin scaling were examined along with the scooping method to determine the most suitable method for the commercial propagation of Amaryllis.

Materials and Methods

Mature Amaryllis bulbs (*Hippeastrum ×johnsonii*) of a similar size (circumference of 30 - 35 cm) were selected for this experiment. Bulb preparation involved the complete removal of the first two outermost scales as well as all the roots, and the top third of the bulb was cut off. Bulbs were then cut according to the specific treatment: For treatments 1 to 3 (sectioning), the mother bulbs were longitudinally cut into eights, twelfths, or sixteenths, respectively. For treatments 4 to 6 (notching), the mother

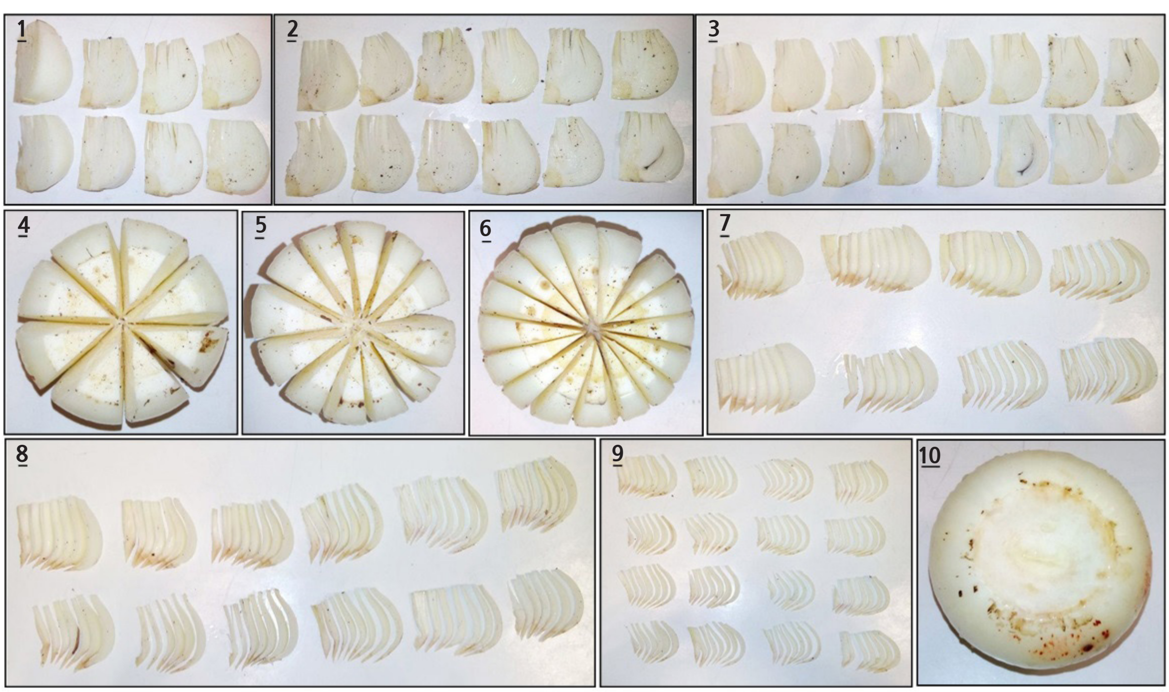

Fig. 1. Different cutting methods for the propagation of Amaryllis (1, 2 and 3: sectioning the bulb into 8, 12, and 16 sections, respectively. 4, 5,and 6: notching the bulb into 8, 12, and 16 sections, respectively. 7, 8,and 9: cutting the bulb into 48, 72, and 96 twin scales, respectively. 10: scooping.)

bulbs were longitudinally cut into eights, twelfths, or sixteenths, respectively. In this method, the segments remained attached together at the top of the bulb. For treatments 7 to 9 (twin scaling), the mother bulbs were longitudinally cut into eights, twelfths, or sixteenths followed by separating each part to 48, 72, or 96 twin scales, respectively. As the mother bulbs were large and mature, numerous twin scale layers were obtained from one bulb. In treatments 1 to 9, the sectioning of the bulbs was performed so that each section contained a portion of the mother bulb's basal plate. For treatment 10 (scooping), the entire basal plate of the mother bulb was removed (Fig. 1).

All segments of the mother bulb were dipped into 0.1% Carbendazol solution for 25 min. Surface water was removed by dabbing with sterile tissue paper. After four days of curing, the segments of the mother bulb were planted in boxes filled with vermiculite in an incubator at a temperature of 23℃ for a period of five months. At the end of this period, the propagated material was removed and examined for the following properties: Number, weight, and diameter of bulblets produced from each mother bulb; percentage of rotten scales; length and number of leaves produced by each plant (for leaf length, the longest leaf of each plant was measured); volume, length, and number of roots produced by each plant (for root length, the average root length was measured). Treatments were statistically compared by ANOVA with the JMP 8 program and the means were compared using an LSD test with a 5% significance threshold.

Results

The different cutting methods had a significant influence on bulblet diameter. Sectioning the bulb into eight sections produced the largest diameter bulblets compared to other methods. Bulblets with the smallest diameter were produced when bulbs were cut into 96 twin scales. Increasing the number of sections in the sectioning, notching, and twin scaling methods led to a reduction in bulblet diameter. Cutting the mother bulbs into smaller segments led to an increase in the total number of bulblets produced. The highest number of bulblets was produced by the twin scaling method. Here, cutting the bulbs into 96 twin scales led to the maximum number of bulblets (Fig. 2).

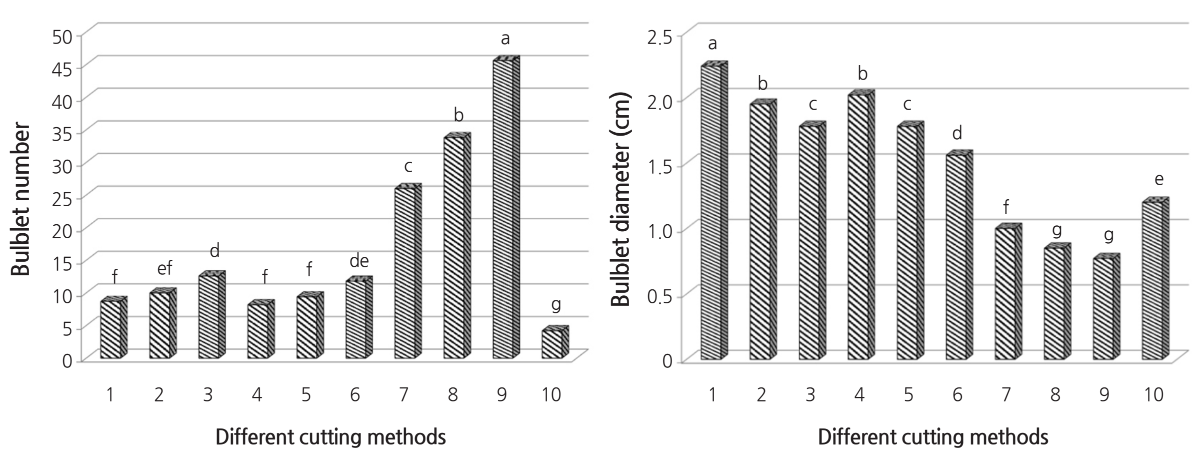

Fig. 2. Effect of different cutting methods on bulblet number and diameter (after five months).
(1, 2, and 3: sectioning the bulb into 8, 12, and 16 sections, respectively. 4, 5, and 6: notching the bulb into 8, 12, and 16 sections, respectively. 7, 8, and 9: cutting the bulb into 48, 72, and 96 twin scales, respectively. 10: scooping.). Means with the same letters had no significant difference based on an LSD test with a 5% significance threshold.

The weight of the propagated bulblets also differed between the various cutting methods. Increasing the number of cuttings in the sectioning, notching, and twin scaling methods reduced the weight of the bulblets produced. The highest and lowest weights were obtained when the mother bulbs were cut into 8 sections and 96 twin scales, respectively. These results also show that the weight of bulblets produced by the scooping method was similar to those produced by the twin scaling method (Fig. 3).

The percentage of rotting mother scales is an important factor in determining the best method for propagating bulbous plants, but it has only rarely been mentioned in previous studies. The different cutting methods had a significant effect on the rate of rotten scales (Fig. 4). Among the different cutting methods, the twin scaling method resulted in the highest percentage of rotten scales, with the highest percentage observed when bulbs were cut into 96 twin scales (39%). Other methods resulted in less than 10% rotten scales. There was no sign of rotting when the mother bulb was sectioned into 8 sections. In the sectioning, notching, and twin scaling methods, the percentage of rotten scales increased as the mother bulb was cut into smaller segments (Fig. 3).

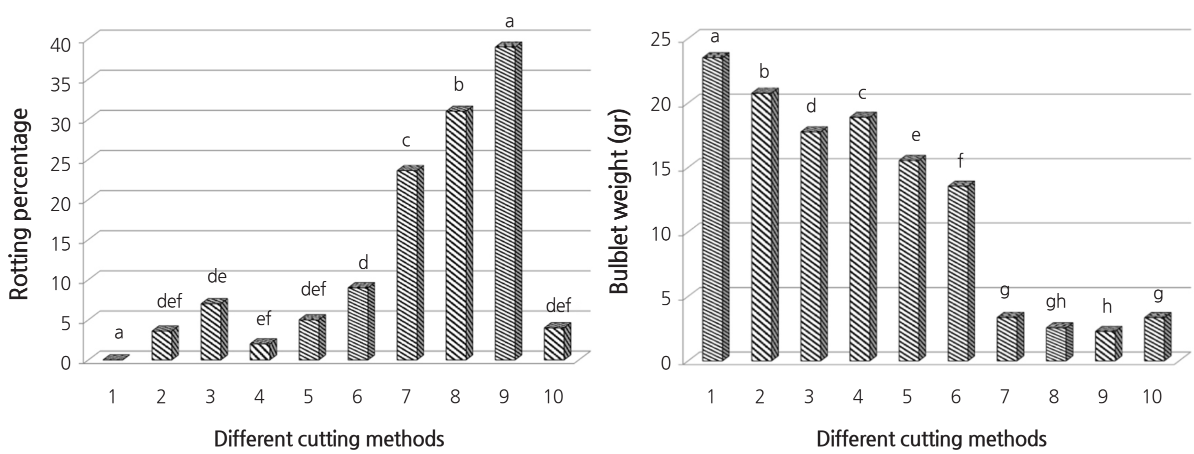

Fig. 3. Effect of different cutting methods on bulblet scale rotting percentage and weight (after five months).
(1, 2, and 3: sectioning the bulb into 8, 12, and 16 sections, respectively. 4, 5, and 6: notching the bulb into 8, 12, and 16 sections, respectively. 7, 8, and 9: cutting the bulb into 48, 72, and 96 twin scales, respectively. 10: scooping.). Means with the same letters had no significant difference based on an LSD test with a 5% significance threshold.

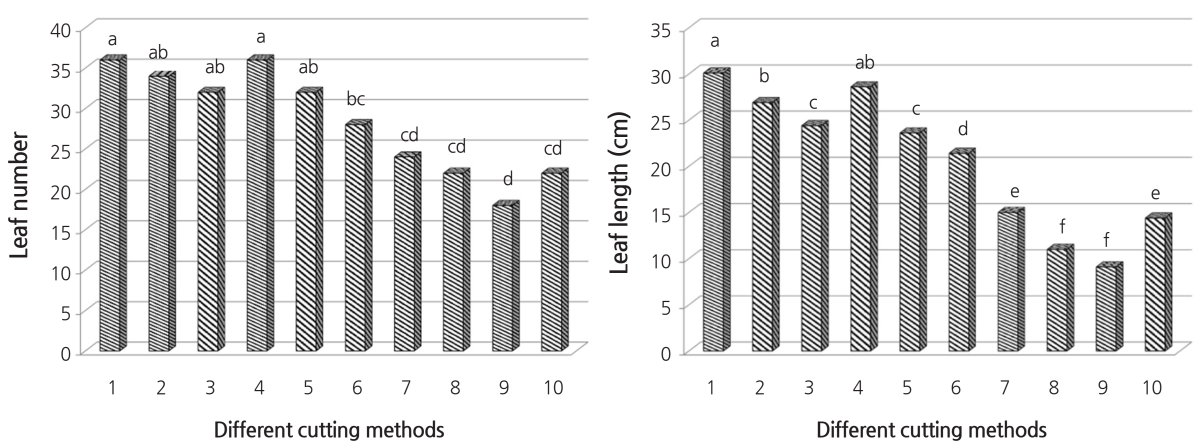

Fig. 4. Effect of different cutting methods on leaf number and length (after five months).
(1, 2, and 3: sectioning the bulb into 8, 12, and 16 sections, respectively. 4, 5, and 6: notching the bulb into 8, 12, and 16 sections, respectively. 7, 8, and 9: cutting the bulb into 48, 72, and 96 twin scales, respectively. 10: scooping.). Means with the same letters had no significant difference based on an LSD test with a 5% significance threshold.

The number of leaves produced was affected by the different cutting methods (Fig. 4). The maximum number of leaves were produced when the mother bulbs were cut into 8 sections or 8 notches. There were no significant differences between the sectioning methods (8, 12, and 16 sections) in terms of leaf number. The lowest number of leaves was produced when the mother bulb was cut into 96 twin scales. The number of leaves produced by the scooping method was similar to the twin scaling method (Fig. 4).

Comparison of the mean leaf length showed that the different cutting methods had an influence on this parameter. The longest leaves were produced when the bulbs were cut into 8 sections. There was no significant difference between notching the bulb into 8 segments and sectioning the bulb into 8 sections. Cutting the bulb into 96 twin scales resulted in the lowest mean leaf length (Fig. 5).

Root length and number varied significantly across the different cutting methods. The maximum root length and number was achieved when mother bulbs were cut into 8 sections. Similar to leaf length, the difference between bulbs which were sectioned into 8 segments or notched into 8 segments was not significant. The lowest root length and number was observed when mother bulbs were cut into 96 twin scales. Although roots produced by the scooping method were longer than those produced by the 96 twin scales method, root number was equal between these two methods (Fig. 6).

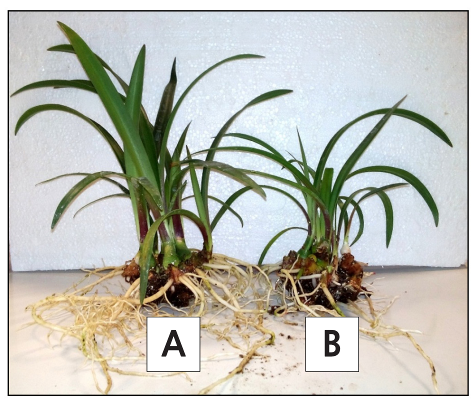

Fig. 5. Comparison of two different notching methods (A: notching into 8 segments and B: notching into 16 segments).

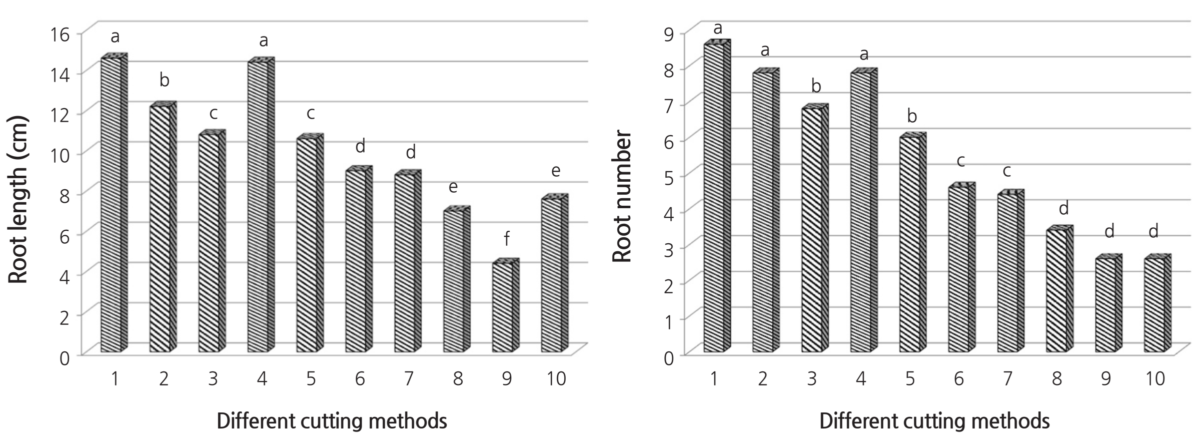

Fig. 6. Effect of different cutting methods on root length and number (after five months).
(1, 2, and 3: sectioning the bulb into 8, 12, and 16 sections, respectively. 4, 5, and 6: notching the bulb into 8, 12, and 16 sections, respectively. 7, 8, and 9: cutting the bulb into 48, 72, and 96 twin scales, respectively. 10: scooping.). Means with the same letters had no significant difference based on an LSD test with a 5% significance threshold.

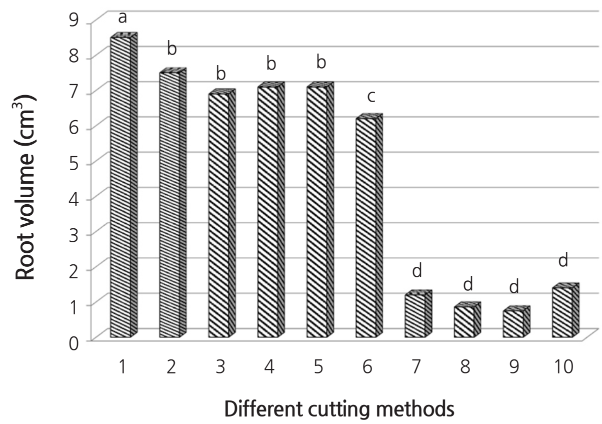

Fig. 7. Effect of different cutting methods on root volume (after five months).
(1, 2, and 3: sectioning the bulb into 8, 12, and 16 sections, respectively. 4, 5, and 6: notching the bulb into 8, 12, and 16 sections, respectively. 7, 8, and 9: cutting the bulb into 48, 72, and 96 twin scales, respectively. 10: scooping.). Means with the same letters had no significant difference based on an LSD test with a 5% significance threshold.

The different cutting methods had a significant effect on root volume. The largest volume of roots was observed when the mother bulb was cut into 8 sections while the smallest volume was observed when bulbs were cut into 96 twin scales. There was no significant difference between the scooping and twin scaling methods in terms of root volume (Fig. 7).

Discussion

During the propagation of bulbs, the growth of bulblets is affected by the nutritional reserves in the mother bulb (Andrade-Rodríguez et al., 2015). In Amaryllis, starch serves as the nutritional reserve used in the initial growth phase of bulblets before they begin to photosynthesize (Stancato et al., 1995). During the propagation of mother bulbs, the number of cuttings into which the bulbs are divided is critically important because increasing the number of sections leads to a reduction in bulblet weight (Witomska et al., 2005). In the twin scaling method, the nutritional reserves of each individual bulblet is less than in other methods (chippind and notching). Consequently, bulblets produced by the twin scaling method had a lower weight than other methods (Zhu et al., 2005). Our results corroborate the conclusion that the average weight of bulblets decreases as the number of sections into which the bulbs are divided increases, and this occurs across the various cutting methods. In fact, increasing the number of sections resulted in a smaller quantity of nutrients available in the cuttings, and the time required to produce a mature bulb increased (Ephrath et al., 2001).

In terms of the number of leaves produced, Zhu et al. (2005) showed that daughter bulbs produced by chipping and notching (12 segments) had more leaves in comparison with the twin scaling method (48 twin scales). Furthermore, the leaves produced by notching and chipping were longer. However, Andrade-Rodríguez et al. (2015) reported that most bulblets had no leaves at the end of the experiment, and those that did have only one or two. Their findings are in contrast with Zhu et al. (2005) and the current study.

Both the number and diameter of daughter bulbs is also affected by the different cutting methods (Andrade-Rodríguez et al., 2015; Zhu et al, 2005; Ephrath et al., 2001). The growth of bulblets, assayed by diameter and weight, was shown to depend on the size of the section used. The larger the sections, the higher the growth of the bulblet. Conversely, the smaller the size of cutting, the lower the growth (Andrade-Rodríguez et al., 2015; Jamil et al., 2014). Zhu et al. (2005) indicate that bulblet diameter was lower in the twin scaling method compared to the chipping and notching methods, which is in accordance with our results. Ephrath et al. (2001) reported that increasing the number of sections resulted in a larger number of bulblets, but the circumference of the bulblets decreased. Their results showed that sectioning the bulb into 8 sections was the best method for Amaryllis bulb propagation. Application of the twin scaling method was recommended by Andrade-Rodríguez et al. (2015). However, since the bulblets produced by the twin scaling method are neither vigorous nor large, it does not seem appropriate to choose this method for Amaryllis bulbs production.

The amount of rotten scales is another crucial factor in choosing the best method of propagation. Ephrath et al., (2001) found that when the number of sections into which the bulb was divided increased, the probability of obtaining a new bulblet from those sections decreased. The results of another study showed that the small size of the bulblets produced by the twin scaling method could lead to crop losses in the field (Andrade-Rodríguez et al., 2015). Our results indicate that when mother bulbs were cut into increasingly smaller segments, the amount of rotten scales increased.

Producing roots can increase the chances that bulblets will survive. Andrade-Rodríguez et al. (2015) reported a significant effect of the different cutting methods on root numbers. In their study, the lowest number of roots (3) was obtained when a basal section of the bulb was used and cut in half. The highest number of roots was achieved by the twin scaling method (44) and by sectioning the bulb into 8 sections (43.5). However, in this study, we observed significant difference in root number between the twin scaling and sectioning methods. The number of roots produced by sectioning was two times larger than the number produced by the twin scaling method. This contradiction may be due to difference in the number of cuttings. The results of Zhang et al. (2013) are in line with our findings. They reported that when mother bulbs are cut into 36 twin scales, 1 - 2 roots are produced. It is clear that the stronger bulbs produced by the sectioning method have a higher rooting potential compared to those produced by the twin scaling method.

Conclusion

The results of this study show that the largest number of bulblets was produced by the twin scaling method, but they take longer to grow to a final saleable size, a serious disadvantage for bulb production. The sectioning and notching methods resulted in larger but fewer bulblets than the twin-scaling method. Since the sectioning method provides more space for bulblets to grow compared to the notching method, this method resulted in better plant growth. Increasing the number of sections into which the bulb was cut resulted in a larger number of bulblets, but the diameter of bulblets produced decreased. Taken together, we find that sectioning the bulb into 8 segments is the best method for vegetative propagation of Amaryllis.

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