

## Original Research Article

# The Survival Rate and Growth of Grafted-Seedling of Durian (*Durio zibethinus* Murr.) Using Various Age of Rootstocks and Different Numbers of Scion Bud

This experiment was performed to determine the survival rate of grafted-seedling growth of durian using various age of rootstocks and different numbers of scion bud. The study was arranged in a two-factorial randomized block design with three replications. The first factor was rootstock age (4, 6, and 8 weeks) and the second factor was number of buds on scion (1, 2, and 3 buds). The percentage of the successful grafting and several growth parameters, i.e., time of budbreak, length and diameter of new shoots were evaluated for three months. Data were analysed using analysis of variance and continued with the post-hoc Tukey's test. The results showed that the age of the rootstock used had no effect on the percentage of successful grafting, time of budburst, shoot length and shoot diameter. Likewise, the number of shoots did not affect the percentage of successful grafting, shoot length, and shoot diameter, but had a significant effect on bud bursting time ( $P < 0.05$ ). Scion that had three buds resulted in the fastest time of budbreak (28.45 days). The highest percentage of successful graft union was found in the 4 weeks-old rootstock and scion with 2 and 3 buds (100%), the maximal successful also obtained in the six weeks-old rootstocks with 3 buds. The four weeks-old rootstocks also gave the best result in the new shoot length and diameter. Nevertheless, there was no interaction between rootstock age and the number of buds on scion in the successful or survival rate of grafted seedling of durian.

**Comment [MM1]:** There is no background to explain reason for carrying out the experiment

**Comment [MM2]:** Abstract should be concise

*Keywords: Durian, grafting, shoot, budbreak*

## 1. INTRODUCTION

Vegetative propagation is one of the options to obtain quality seeds with the same superior characteristics as those of the parents and fast production. Plants that are propagated vegetatively can generally bear fruit at the age of 3-5 years after planting [1]. Vegetative propagation can be done through chip budding (bud grafting), grafting, feeding, and cuttings, as well as tissue culture. However, chip budding and grafting techniques for durian plants are more recommended.

**Comment [MM3]:** Seeds or seedlings?

Propagation through chip budding and grafting beside to preserve the nature of the parent tree, it also resulted in plants that are is more superior than to the parent tree. Grafting is one of the efforts to improve the quality of plants because the resulting plants come from combining a scion with desired superior characteristics, such as fruit density, large size, and nutritional content with a specific rootstock having the ability to survive the biotic and abiotic components of the environment. The combination of plant parts that are put together will develop to form a new plant by bringing with the properties of the two plant parts that have

**Comment [MM4]:** Reference

~~been combined.~~ The desired superior properties of the parent tree as a source of scion, such as the quality and quantity of fruit combined with a rootstock that have a good root system therefore ~~become~~ resilient to non-mountainous soil conditions. The purpose of propagation through grafting is to maintain clone traits, obtain strong plants, improve unwanted plant species, accelerate tree growth and stem straightness, and accelerate fruiting plants [2][3]. Grafting has been used as a propagation method to increase vigor, uniformity, and robustness to biotic and abiotic stresses of vegetatively regenerated plants [4][5].

The propagation of durian seedlings through chip budding is constrained by the length of time to obtain rootstocks ~~that are ready to be grafted.~~ Propagation through grafting can overcome these obstacles ~~due to using~~ a rootstock ~~that~~ is ready to be connected at a relatively young age. In addition, the grafted plants produced by grafting perform a vigorous growth because the scion used in the grafting can have many bud sources compared to the chip budding that containing a single bud, more buds will produce more shoots expressed by more branches and leaves [6].

Shoot tip grafting is ~~one of the a~~ grafting techniques that does not take long in providing the rootstock because grafting can be done at an early age of rootstock ~~to obtain seedling accordingly that seedlings can be obtained~~ in a short time. The best percentage of successful grafting of three durian cultivars was obtained at six weeks old rootstock [7], however, even two-week-old rootstock provided the highest survival rate on early bud grafting of five cocoa clones [8]. Hence, recognition of apposite stage of rootstock with appropriate morphological and anatomical features is essential to identify its influence on growth of scion, grafting success and plant survival [9]. Therefore, it is necessary to determine the age of the rootstock which is most suitable to be grafted to the desired scion of a superior trait.

In the current study, various ages of durian seedlings were tested as rootstocks and grafted to the scions with various numbers of buds. The study was aimed to obtain a younger rootstock age to be grafted for a shoot tip grafting technique which can provide a high percentage of successful union. The ~~investigation objectives were was conducted to~~ 1) determine the percentage of successful grafting of durian shoots from various ages of rootstock with different number of scion buds; 2) ~~to~~ determine the age of rootstock and the number of buds that are most effective for grafting, and 3) ~~to~~ find out whether there is an interaction between rootstock age and number of buds in successful grafting.

## 2. MATERIAL AND METHODS

This experiment was conducted for five months (May to September) including two months of rootstock preparation. The study was a two-factor ~~ef~~-factorial experiment arranged in a randomized block design with three replications. The first factor was rootstock aged 4 weeks (W4), 6 weeks (W6), and 8 weeks (W8). The second factor was the number of scion buds consisting of one bud (B1), two buds (B2), and three buds (B3). There were 9 treatment combinations which were ~~repeated-replicated~~ three times and each experimental unit consisted of 2 plants so that there were 54 experimental units.

Observation parameters ~~were are~~ percentage of successful grafting (%), age of bud break (days), length of new buds (cm), and bud diameter (cm). Data were analyzed using analysis of variance (Anova) and a significantly different of the treatments was subjected using the Tukey's test ( $P = .05$ ).

### 3. RESULTS AND DISCUSSION

#### 3.1 Percentage of Successful Grafting

The grafting was successful if the connection between the entrees and the rootstock was well unified and it is marked by the bud burst in the scions about three weeks after grafting. Some of the leaves that are left on entrees remain fresh and some fall then followed by growth of new shoots from the buds. The grafted plants that are still alive are indicated by green and fresh scion leaves and the color of the stem does not change [10]. The unification of the scion and rootstock including the cambium of the scion and rootstock must be attached to each other, the cambium of the scion and rootstock produces parenchyma cells that form a callus between the cambium layer of the rootstock and scion to form a new cambium, and new cambium cells produce new vascular tissue, inward to form xylem and outward to form phloem [11].

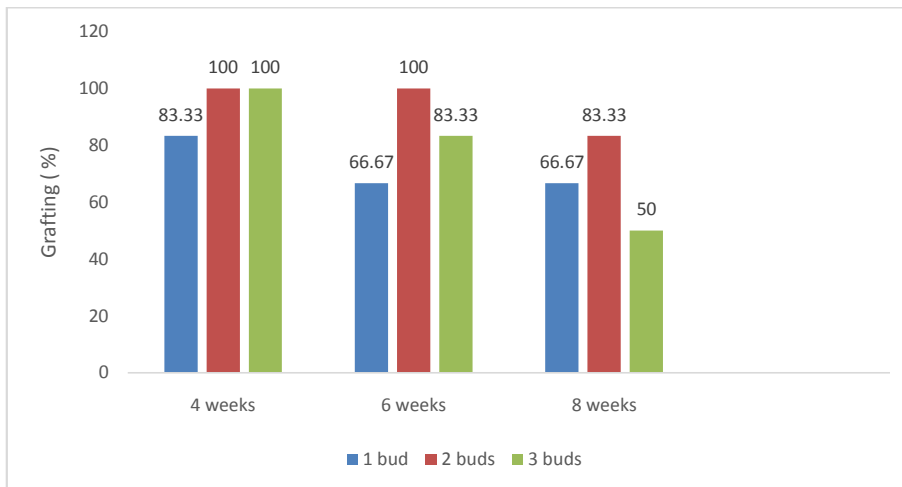
**Comment [MM5]:** Start with results before discussion

The results of the analysis (Fig 1) showed that the age of rootstock and the number of buds of entries did not affect the percentage of successful grafting. However, the four-week treatment tended to be better than other treatments and resulted in 94% of successful live connections. The condition of the plant also determined the success of the connection, especially the age of the rootstock [12]. The effect of rootstock age is important for the survival and vigor of grafts morphological and anatomical affinity between scion and rootstock, for gene expression patterns, and for reproductive development of the grafts [13][14]. In the current study, rootstock age did not influence graft success significantly, while the number of days for bud breaking was affected ( $P \leq .05$ ; Table 1). Younger rootstocks, 4 and 6 weeks old, improved grafting success, 94.44% and 83.33%, respectively. In this case, it was supposed that the younger durian stock made the cambium cells linking between the scion and rootstock was faster due to the meristematic cells that actively dividing. Therefore, the selection of stock was related to the growth of the rootstock plants that were in an active growing condition; the plant is still young hence, cells divided rapidly.

**Comment [MM6]:** Begin with this paragraph with results

The success of grafted union was determined by a tightly adhering of the cambium between the rootstock and the scion. The unification is achieved at a time when the rootstock and scion are proficient of producing callus and other wound-response tissues allocating water and nutrients to transport from the stockroom to the scion [15].

**Comment [MM7]:** Part of methods



**Fig.1.**Percentage of successful live connections 4 weeks after grafting

Observations in the field showed that the higher number of successfully grafted was the treatment of four weeks of rootstock. The successful graft union was represented by the unification of vascular tissue and cambial layers of the scion and rootstock cut. The present results indicated that the relatively young rootstocks (less than five weeks) were adequate to perform linking process with scions and resulted in a new growth.

### 3.2Bud Break

The time of bud break is when the shoots begin to sprout and develop the first golden yellow primordial leaf. The analysis of variant showed that the age of the rootstock used in the grafting did not significantly affect the time of bud break (Table 1), but the treatment of the number of buds had a significant effect on the time of bud breakage resulting from the grafting of durian plants. The scion with three buds showed the fastest bud break time (28 days in average). The successful grafting had led to the initial growth of the grafted durian plants as indicated by the bud breaking. Based on the result that the scion with three buds grew faster than the scions with a lower number of buds, indicated a positive effect on subsequent growth and development as the grafted had many bud sources will-which produced more shoots preceded. The amount of food storage in the scions is another important factor which can affect grafting success. Generally, the scion with three buds has more reserved foods such as carbohydrates which is essential for callus formation in grafting union. Stored carbohydrates are hydrolysed to simple ones and transported to the grafting union [16].

**Table 1. Average age of bud break 4 weeks after grafting**

Rootstock Age	Number of scion buds			Average
	1	2	3	
4 weeks	29.50 <sup>ab</sup>	29.83 <sup>ab</sup>	28.17 <sup>ab</sup>	29.50
6 weeks	31.00 <sup>ad</sup>	29.33 <sup>ab</sup>	29.17 <sup>ad</sup>	29.83
8 weeks	33.33 <sup>bc</sup>	29.50 <sup>ab</sup>	28.00 <sup>a</sup>	28.17
Average	31.28	29.55	28.45	

\*Values followed by the same letter are not significantly different ( $P = .05$ )

The time needed for bud break on durian rootstocks aged eight weeks was faster, which ranged between (28 - 29 days) compared to relatively younger rootstocks. Older rootstocks having a well-developed root system for absorption of water and nutrients that play a significant role in graft success. The rootstock age produced most of the callus on the graft joining, implying the role of a rootstock age [17]. The successful grafted is revealed by bud sprouting that occurs after the scions are linked to the rootstocks, resulting in the smooth distribution of nutrients from the rootstocks to the scions through tissues that have been perfectly attached and therefore the nutrients are used for callus formation [18]. The earlier the wound healing and callus formation on the rootstock and scion, the sooner the bud breaks. Accordingly joining up of the vascular system is required to facilitate water uptake and to enable nutrient transport to the graft junction [19]. This suggests that the linkage between the cambium of the scion and rootstock will cause the process of shoot and leaf formation to take place more quickly, hence led to optimal seedling growth [20].

Taken together, the ontogenic age of durian rootstock and the number of scion buds affected the bud take and graft success, therefore, the success of grafting in durian can be optimized by taking a relatively young rootstock with two or three scion buds.

**Comment [MM8]:** Which age, point unclear

**Comment [MM9]:** Relate this to root stock age

**Comment [MM10]:** What age according to your findings



**Fig.2.**The grafted durian plants using two and three buds

### 3.3 Shoot Length

The length of shoot was not significantly affected by different number of scion and rootstock ages used (Fig 3). The shoot length was recorded maximum in W4B3 (1.27 cm) while, lowest in W8B1. The results showed higher value for the youngest rootstock in combination with the highest number of buds possibly due to rapid and strong formation of union between the rootstock and scion. After the vascular tissue of the scion and the rootstock were connected, the transport of phloem and xylem will function to support plant growth successively influencing greater absorption of nutrients by sprouted shoots.

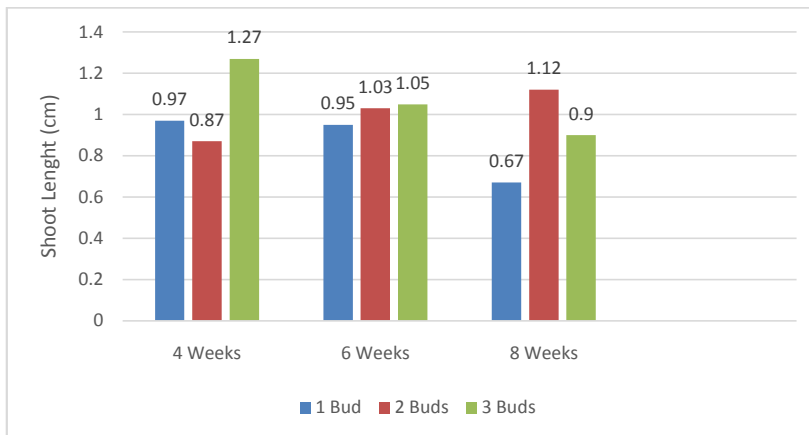
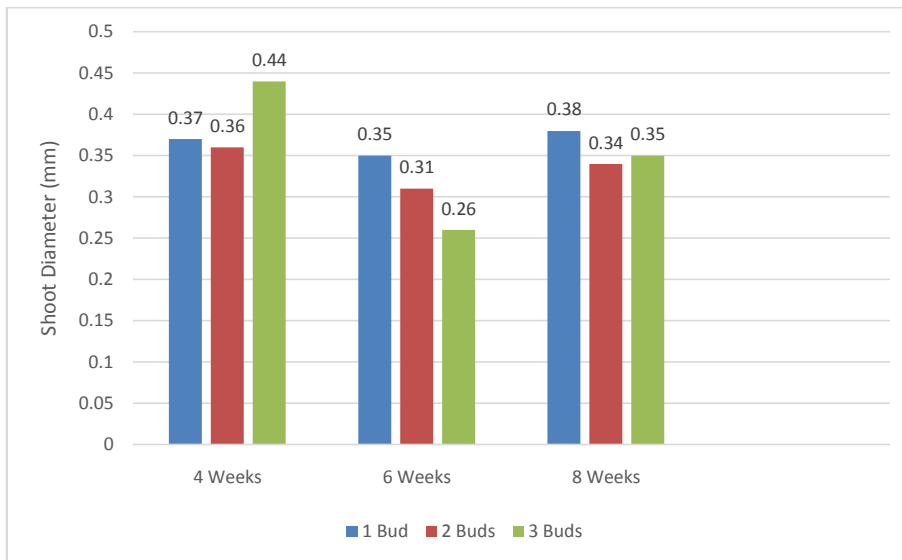


Fig.3.Average shoot length 12 weeks after grafting

### 3.4 Shoot Diameter

~~Another growth parameter Shoot diameter after 4 weeks (shoot diameter) was not significantly affected also by different both rootstock ages and number of buds or a combination of both. The diameter was measured after 12 weeks of grafting. As the four-week-old rootstock in combination with three buds (W4B3) showed the highest average of shoot length, the maximum shoot diameter also found at the combination treatment (Fig 4).~~

**Comment [MM11]:** Is this not in the methods, if so it is repetition.



**Fig.4. Average shoot diameter 12 weeks after grafting**

One of the factors that determine the bud growth was cambium activity which was influenced by hormonal balance at the junction of the scion and rootstock and the fusion will induce bud growth through the hormonal activity and accumulation. The graft union development also involves cell to cell communication between scion and rootstock for endogenous growth regulator accumulation in triggering shoots in the scion [21]. The bud growth relationship was a homeostatic mechanism (the plant body's efforts to maintain physiological balance, so that the body's organs can function normally) and in an effort to maintain a balance shoot growth was highly dependent on conditions related to metabolism such as hormones; cytokinin stimulated cell division by increasing the rate of protein synthesis, while auxin stimulated the elongation of cells which causes stem elongation [22]. In addition, the auxins acting for plant growth initiation is produced in a shoot apical meristem (SAM), which is part of the scion of the grafted plant [23], therefore it will support the growth of new shoots from the buds in the scion.

The endogen growth hormone such as cytokinin also has a significantly role to stimulate the shoot induction, shoot multiplication and play an important role in enhancing cell division, cell proliferation and radial cell growth during the initial stage of shoot formation [24] [25]. The enlargement cells will affect the size of the shoots.

#### 4. CONCLUSION

Based on the observations, it can be concluded that the age of rootstock used did not affect the percentage of successful connections, bud rupture time, bud length and bud diameter. Similarly, the treatment of the number of buds did not affect the percentage of successful grafting activity that live, bud length, and bud diameter, but did have a significant effect on bud break time. Scion of three buds gave the effect of the fastest bud break time (28.45 days). The highest percentage of successful live grafting was found in rootstocks aged 4 weeks and scion of 2 buds (94.4%). Meanwhile, the age rootstock at 4 weeks the results

were better than other treatments seen from the percentage of successful grafting, bud length, and bud diameter. However, there was no interaction between rootstock age treatment and the number of buds in the success of grafting durian seedlings.

**Comment [MM12]:** Is this not contradicting the first part of the conclusion which indicates that there is effect from root stock age

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