



Rooting of Mini-Cuttings of Guava Cultivars (*Psidium guajava* L.) Treated with IBA under Misting Irrigation

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ABSTRACT

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Guava (*Psidium guajava* L.) is a plant whose fruit is very popular in Indonesia because it contains high vitamin C and its leaves can be used as herbal medicine. Orchard expansion of selected cultivars and fruit production of guava can be achieved vegetatively through propagation by using mini-cuttings with the use of Indole Butyric Acids (IBA). A study was conducted to compare rooting success of mini-cuttings of herbaceous stem of four guava cultivars with IBA treatment. Four guava cultivars tested were Bengkulu Red, Getas, Crystal, and Bangkok. Mini-cuttings were immersed in IBA solution at 0 ppm or 1000 ppm for 2 hours before planting. Cultivar and IBA treatments were arranged factorially in a randomized complete block design with 3 replications, each consisting of 25 cuttings, hence 600 cuttings were used. Variables observed included percentage of original-leaf shedding, percentage of emerging shoots, percentage of sprouting cuttings, new leaf number, root number, root length, and percent of rooted cuttings. The results demonstrated that cultivars affected rooting percentage, sprouted percentage, percentage of original-leaf fall, and root number. Getas had highest rooting and sprouting percentages, and showed lowest number of original-leaf shedding; whereas, Bangkok demonstrated greatest number of root. IBA treatment increased the number of root and the number of original-leaf shedding, but reducing rooting percentage of mini-cuttings. There was no interaction between cultivar and IBA treatment.

INTRODUCTION

Guava (*Psidium guajava* L.) is a tropical fruit crop belongs to family Myrtaceae. It evergreen plant quite popular among the people in Indonesia. In addition to be consumed in form of fresh or table fruit, guava fruit is also used as processed products, such as juice, ice cream, jelly, pasta or jam (Gould and Raga, 2002, Parvez *et al.* 2018). Guava fruit are used for promoting health, especially for digestion, curing infections, lowering cholesterol, antioxidants, and raising the platelets for

dengue fever patient (Bangun, 2004, Kafle *et al.* 2018). Guava fruit contains high vitamin C (Uzzaman *et al.* 2018).

In Indonesia, guava harvested area increased during four year period from 9.028 hectares in 2014 to 10.397 hectares in 2018. In The same period, guava production also increase, from 187405 ton in 2014 to 230.690 in 2018 (Kementerian Pertanian RI, 2019). Increasing guava production especially through extensive cultivation is often limited mainly by the availability of plant material from commercial varieties, such as Crystals, Bangkok, Getas Red

and Bengkulu Red. Although guava plant material is commonly propagated vegetatively through air layering, current technique using herbaceous stem mini-cuttings under intermittent irrigation system is feasible (Sakai and Subiakto, 2007).

Vegetative propagation by mini-cuttings is easy, simple, and economical, being able to produce new plants that have same genetic properties with their parent in large quantities and in a relatively short time (Sakai and Subiakto, 2007). The success of propagation through cuttings is influenced by the interaction of environmental and genetic factors. Environmental factors, including rooting media, humidity, temperature, and light intensity, will also determine the success of cutting propagation (Hartmann et al., 1997). Whereas, genetic factors include the content of the food reserves in the network cuttings, water availability, age of mother plants, cuttings endogenous hormones in the network, and the type of plant. Many varieties of guava are popular and planted in home garden for self consumption of family or in orchard for commercial purpose. In Deli Serdang, plants, leaves and fruit of several varieties, including Sari, Getas, Australia, Sari Putri, and Kristal has been characterized (Fadillah *et al.* 2018). In Bengkulu, four cultivars are most commonly found, i.e. Getas, Kristal, Apel Merah and Bangkok. However, their capacity for propagation through cutting has not been published.

Guava can be propagated generatively through seed or vegetatively through air layering (macro cutting) or cuttings. Comparing several cutting sources, Ullah *et al.* (2005) found that herbaceous/softwood stem cuttings is the only cutting material suitable for vegetative propagation, resulted in 37% rooting success. Khodijah (2019) used Rootone-F at 2000 ppm to promote rooting success of guava cuttings up to 87% rooting. Exogenous auxin, Indol Butyric Acid (IBA), has been used to stimulate rooting success of guava cutting, in which higher concentration will may reduce treatment length. Manan *et al.* (2002), reported that dipping guava cutting into 1000 ppm IBA for 2 hours resulted in rooting percentage of 40%. On the other hand, Shahzad *et al.* 2019 demonstrated that 1600 ppm IBA with 10 second dipping time yielded best results for softwood cuttings of

guava. The purpose of this study was to compare the response of the multiplication of softwood cuttings of four guava cultivars (Getas, Kristal, Bengkulu Red and Bangkok) with IBA treated with IBA.

MATERIALS AND METHODS

This study was conducted in the Greenhouse Faculty of Agriculture, Bengkulu University, Indonesia. Treatments included four guava cultivars (Bengkulu Red, Getas, Crystal, or Bangkok) and dipping in IBA solution (0 or 1000 ppm) for two hours. Treatments were arranged factorially in a Randomized Complete Block Design and repeated 3 times. A treatment unit was 25 cuttings, so the number of cuttings used in this study was 600 stem cuttings.

River sand was used as rooting media, sterilized sun sterilized through wrapping the media with a heat-resistant transparent plastic and exposed under the sun for 4 days. The media was poured into a rooting wooden box without a base with a size of 3.8 m x 1.5 m with a height of 20 cm. The sand media is inserted until 15 cm thick. Rooting tunnel was made from a curved transparent plastic with a height of 60 cm. Shading structure was made to give 60% shading by using paranet.

Cutting material were obtained from commercial orchard in Bengkulu City. Shoots containing herbaceous green stem were harvest early morning (before 9 AM) and subsequently put in shaded area and sprayed with Reverse Osmosed mineral water to keep freshness. Herbaceous/softwood cuttings with ± 0.5 cm diameter 12-15 cm length were taken from the shoot tip, leaving 3 leaves at the top of the cuttings. Leaf area was reduced in half.

Solution of 1000 ppm IBA was made by dissolving 1 g of IBA (Sigma Aldrich) using 300 ml of 95% ethanol through stirring, and distilled water was then added up to 1000 ml solution. Guava cuttings were immersed into 1000 ppm IBA solution up to 5 cm in the bottom of the cuttings for 2 hours before planting.

Herbaceous cuttings were planted in sterilized sand media in the wooden box at distance of 10 x 5 cm at a depth of 5 cm with assistant of planting holes to avoid friction of cuttings with the sand.

Irrigation was conducted by using intermittent misting system which was installed for intermittent watering to maintain a stable temperature and humidity inside rooting tunnel. Misting installation was done by using the 0,5 inch PVC pipe and misting nozles were fitted into the pipe at a distance of 1 m, and connected to a water pump with electrical power. Intermittent misting was conducted for 5 minutes on in every 30 minutes from 8.00 AM to 17.00 PM every day. The daily average minimum temperatures of the tunnels ranged from 22 °C to 25°C and maximum temperature ranged from 28°C to 32°C. Whereas, air humidity were maintained above 85%. Fertilization was done once a week using a foliar fertilizer (Ganasil D) with a concentration of 1 g L⁻¹ of surface whell water. Weed control is done by pulling the weeds manually.

Cutting growth were observed through percent of leaf sanescence, sprouted cutting, time to shoot emergence, number of new leaf, rooted cutting, root number and root length at 8 weeks following planting.

Data were analyzed statistically by analysis of variance (F test) level of 5%. When in a variable a factor showed a significant effect, treatment averages were separated by Duncan's Multiple Range Test (DMRT) at level of 5%.

RESULTS AND DISCUSSION

Results of variance analysis demonstrated that variations of leaf senescence, sprouted cutting, rooted cutting, and root number occurred among guava cultivars (Table 1). Immersion in 1000 pm IBA affected leaf senescence, rooted cutting, and root number. Meanwhile, there was no interaction between cultivars and IBA found in all growth variables observed (Table 1).

Cultivar Response

Growth of upper portion of guava herbaceous cuttings were presented in Table 2. Original leaf senescence and sprouted cutting differed among the tested cultivars. Getas Red demonstrated leaf senescence of 71%, lower than the other cultivars, ranging 85 – 87%. The highest percentage of cutting bearing

Table 1. Compilation of variance analysis result on the effect of cultivars, IBA or their interaction on growth of herbaceous cutting of guava

Cutting growth	Cultivar	IBA	Interac-tion
Leaf senescence (%)	7.53*	18.55*	0.80 ns
Sprouted cutting (%)	4.65*	1.57 ns	1.69 ns
Time of shoot emer-gence (WAP)	0.54 ns	0.05 ns	0.77 ns
Number of new leaves	2.72 ns	1.06 ns	0.43 ns
Rooted cutting (%)	10.12*	20.55*	0.73 ns
Root number	4.14 *	24.94*	0.39 ns
Root length (cm)	1.46 ns	0.49 ns	2.64 ns

Notes: Effect was significant when calculated F value was higher than value in table F at 0.05 (*); effect was not significant when calculated F was lower than table F at 0.05 (ns).

shoot was Bangkok cultivar (14%), followed by Getas Red (10%), Crystal (4%), and Bengkulu Red (1%). Overall, the percentages of cutting bearing shoot were low as the experiment was conducted during rooting stage. The buds emerged from cutting within 2 to 3 weeks after planting in rooting medium. After transplanting, herbaceous cuttings of guava produced axillary buds within three weeks.

Root growth characteristics, in term of rooted cutting percentage and root number, of herbaceous cutting varied among guava cultivars (Table 3). Getas Red demonstrated greatest capability to produce root (55%); whereas other cultivars showing similarly low capability to root, ranging 22 – 28%. On the other hand, Getas Red and Bengkulu Red produced least root number (6 and 8 roots, respectively), though being not lower than

Table 2. Effect of cultivars on Growth of upper portion of guava herbaceous cutting under intermittent misting system

Cultivar	Leaf se-nescence (%)	Sprouted cutting (%)	Time to bud emergence (week)
Bengkulu Red	87.3 a	1.3 c	1.83
Getas Red	70.9 b	10.0 ab	1.86
Crystal	85.3 a	4.0 bc	2.18
Bangkok	86.9 a	14.0 a	3.10

Notes: Effect was significant when calculated F was higher than table F at 0.05 (*); however, effect was not significant when caculated F was lower than table F at 0.05 (ns).

Table 3. Effect of cultivar on Growth of lower portion of guava herbaceous cutting under intermittent misting system at 8 weeks after panting.

Cultivar	Rooted cutting (%)	Root number	Root length (cm)
Bengkulu Red	22.00 b	6.2 b	10.5
Getas Red	54.67 a	8.2 b	13.03
Crystal	28.00 b	10.6 ab	13.21
Bangkok	24.00 b	16.2 a	12.50

Notes: Data in the same column followed by the same letter were not significantly different based on DMRT at α 5%

Crystal (11 roots), their root number were less than those in cv. Bangkok (16 roots). In term of root length, however, the four cultivars tested in this experiment demonstrated no significant difference in root length, root length average ranging between 10 – 13 cm.

This experiment demonstrated that the presence of leaf in the cutting was essential to promote rooting. Getas, which exhibited the lowest leaf senescence (71%), although the difference is only about 16% compared to others (ranging 85% - 87%), demonstrated the highest capacity to root (55%), being around doubled than those of the other tested cultivars (24% - 28%).

It is interesting to note that red-flesh fruit guava cultivars (Bengkulu Red and Getas Red) can not be differentiated with white-fleshed fruit cultivars (Crystal and Bangkok) in term on upper-cutting growth, as indicated by leaf senescence, sprouted cutting and time to shoot emergence. However, in term of root growth, white-fleshed fruit guava cultivars, especially Bangkok, produced profuse roots (16 roots), greater than those in red-fleshed cultivars Bengkulu Red and Getas Red, 6 and 8 roots, respectively.

IBA Effect

IBA is a plant hormone that has been widely used to promote rooting success in propagating plant through cuttings. Immersing herbaceous stem cuttings of guava into 1000 ppm IBA for 2 hours significantly affected the success of the multiplication of the cuttings in term of percentage of original leaf senescence, rooted

cutting, and root number (Table 1). Original leaf senescence of cuttings treated with 1000 ppm IBA (89%) was significantly higher as compared to those in control (76%) (Table 4). However, percent of rooted cuttings was half, and root number were tripled in the IBA treated plot. IBA treatment did not significantly affect the current emerging shoots, cuttings sprout percentage, the number of new leaves, and root length (Table 1, Table 4).

Guava plant (*Psidium guajava* L.) may be propagated with seeds, layering, grafting or budding, cuttings (roots or shoots) or by micro propagation. The guava propagation by seeds is conducted for the growing rootstocks and for growing plants to be detected in the early stages of the cultivation of guava trees. Whereas, the asexual propagation methods are used to clone chosen genotypes of resulted from breeding programs and to establish commercial fruit orchards, as they provide all the distinctiveness of each cultivar. In Indonesia, most orchards of guava are established from propagules obtained by using air layering or marcotting, as its success is very high, almost all marcotts bearing roots. However, this air layering is laborious and very slow rate of propagation. Therefore, the use of mini-cuttings from soft-wood has been developed in the last decades by using misting system (Ullah *et al.*, 2005).

There has been scanty information regarding rooting responses of different genetic to propagation using cuttings in guava. Debnath and Maiti (1990) investigated the effect of growth regulators, on the rooting behavior of

Table 4. Effect of IBA on the success of the multiplication of herbaceous stem cuttings of guava under intermittent misting system

Variabel yang diamati	IBA 0 ppm	IBA 1000 ppm
Leaf senescence (%)	76.44 b	88.77 a
Sprouted cutting (%)	9.00	5.67
Time to bud emergence (WAP)	2.34	2.15
Rooted cutting (%)	43.00 a	21.33 b
Root number	5.00 b	15.6 a
Root length (cm)	12.67	11.95

Notes: Data in the same row followed by the same alphabet was not significantly different based on DMRT at α 5%.

soft-wood cuttings of guava cultivars, namely Baruihur, Sardar and Harijha in India. They found that the responses of varieties varied, and Baruihur being the most responsive cultivar to the treatment with different growth regulators, followed by Sardar and Harijha. Growth regulators applied were auxin, including Indole butyric acid (IBA), Indole Acetic Acid (IAA) and Naphthalene Acetic Acid (NAA), all of which are incorporated in commercial rooting promotor - Rootone-F formulation.

In this experiment, Getas Red was the easiest to root cultivar with 54% rooting success, followed by Crystal, Bangkok and Bengkulu Red, which all rooting at similar level of rooting success (24 – 28%). Rooting capacity of a guava cultivars might be correlated with vigorness of the plant, as matured plant of Getas Red in the field was observed as being the most vigor as compared to the others. Khodijah (2019) also reported that Crystal Red, a vigor stature variety, has rooting success of 87%.

We found that the effect of IBA increased leaf shedding (senescence) and reduced percentage of rooted cutting; these were contradictory to those of some experiments (Manan *et al.*, 2002; Gusniwati *et al.*, 2007, Shofiana *et al.* 2013; Heriandi, 2015). This experiment tested IBA at concentration and time of cutting immersion according to those recommended by Manan *et al.* (2002). IBA concentrations that are too high can produce ethylene which can accelerate the aging of the leaves so that the leaves will fall (Shofiana *et al.*, 2013). The mechanism is that IBA hormone rise will increase the ACC synthase, which is an enzyme to convert a precursor of S-adenosyl-methionine (AdoMet) to 1-Aminocyclopropane-1-carboxylic acid (ACC), which subsequently became ethylene (Taiz and Zeiger, 2010). Experimenting IBA concentration in Crystal guava, Heriandi (2015) reported that at 2 hour immersion, the optimum concentration of IBA was achieved 617 ppm. At concentration of 1000 ppm, IBA did not demonstrated any effect.

CONCLUSIONS

The propagation experiment of guava using herbaceous mini-cuttings of four guava

cultivars: Bengkulu Red, Getas, Crystal, and Bangkok, and immersion for 2 hour in 0 ppm IBA or in control, we obtain several conclusion: (1) cultivars affected rooting percentage, sprouted percentage, percentage of original-leaf fall, and number of root. Getas had highest rooting and sprouting percentages, and showed lowest number of original-leaf shedding; whereas, Bangkok demonstrated greatest number of root. (2) IBA treatment increased the number of root and of original-leaf shedding, but reducing rooting percentage of mini-cutting. (3) There was no interaction between cultivar and IBA treatment.

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