



ORIGINAL ARTICLE

The Effect of Natural and Chemical Compounds on Rooting Traits of *Bougainvillea* (*Bougainvillea spectabilis* L.)

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(Received: 26 April 2017 Accepted: 11 July 2017)

KEYWORDS

IBA;
Rooting;
Grape syrup;
Carbohydrate;
Bougainvillea

ABSTRACT: *Bougainvillea* (*Bougainvillea spectabilis* L.) is an evergreen, difficult-to-root ornamental plant with plentiful applications in green spaces. One of the effective methods in plants propagation is the use of hormonal compounds such as indole butyric acid. Despite the positive effects of these hormones on rooting process, their chemical nature causing environmental risks. Thus, the use of alternative natural compounds with favorable influence to create environmental health and living creatures is important. This experiment was conducted to examine the impact of natural carbohydrate compounds and chemical hormonal compounds on the rooting traits of bougainvillea in the research greenhouse of Gorgan Agricultural Research Center and in the Horticulture Laboratory of Department of Plant Production, Gorgan University of Agriculture Science and Natural Resources, Iran in 2015-2016. The study was based on a Completely Randomized Design with six treatments (indole butyric acid (IBA), aspirin pills, LD pills, natural honey, grape syrup, and gum Arabic), each at four rates, and a control with five replications, each one including five cuttings. The natural and chemical treatments influenced rooting traits including rooting percentage, the number of primary and secondary roots, primary root length, root fresh and dry weight, and survival rate of the cuttings significantly at the 1% level. The treatments of 4000 ppm IBA and 10% grape syrup exhibited significant differences with other studied treatments, control, and displayed the best quantity and quality of the rooting. However, 20% gum Arabic and honey, 4 pills L⁻¹ (320 mg L⁻¹) aspirin, and 4 pills L⁻¹ (0.72 mg L⁻¹) LD were related to the best rooting of the cuttings as compared to other treatments and control. Given the effectiveness of natural compounds like grape syrup on the rooting of bougainvillea cuttings, because of the natural and non-chemical nature, their cost-effectiveness, organic and friendly environmentally it is recommended to use them.

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INTRODUCTION

Flowers and ornamental plants have economic significance in addition to their environmental and decorative importance. These plants are the crops grown in most parts of Iran. They have high potential to generate foreign currency; also, they can be exported as a major non-oil product.

Bougainvillea (*Bougainvillea spectabilis*) is an ever-green ornamental plant native to hot and semi-hot regions of South America from Nyctaginaceae family. This ornamental plant has a special place due to its dense bracts and diverse coloration [1]. Bougainvillea is generally propagated by hardwood and semi-hardwood cuttings. The difficult-to-root cultivar can be propagated by layering or grafting onto high-growth rootstocks [2]. The difficult-to-root bougainvillea is widely grown in green spaces. Therefore, their propagation is economically important. Thus, the selection of the methods and treatments to accelerate the rooting of this commercial plant should be prioritized.

Plants are propagated sexually and asexually. Among various propagation techniques, cutting is the easiest, fastest and most inexpensive asexual technique. It produces more uniform plants that are completely similar to maternal plant [3]. Various factors influence the rooting of the plants including physiological and environmental conditions of the maternal plant, the tissue of the cutting, the wood prepared for cutting, the time of cutting preparation, rooting hormones and stimulators, environmental conditions of the cutting storage etc., whose understanding is critical in successful commercial propagation of these plants [3].

One significant determinant of rooting is the plant hormones, among which auxin is particularly important. One of the best and common auxin compounds used in the rooting of the cuttings is indole butyric acid (IBA) [4]. Presently, the division of the first root initiating cells is triggered by the presence of indigenous or exogenously applied auxin. Auxin has diverse impacts on a

plant's growth and morphology. In addition, this growth hormone improves the rooting rate and percentage, root number and quality, and the uniform rooting of the cuttings [3]. In addition to auxin, some other compounds are used to stimulate and accelerate the rooting of the plants. We used aspirin (acetylsalicylic acid) because of its salicylic acid content as quasi-hormonal phenolic compounds and LD pills due to having estrogen and progesterone steroid hormones that play an effective role in plant growth traits including rooting. Positive results have been reported about their application [5-9].

Another determinant of rooting is carbohydrates, which are of significance in plant rooting as energy sources [10, 11]. Numerous studies have demonstrated the favorable influence of carbohydrates on plant roots. The natural carbohydrate-containing compounds can be exemplified as honey, grape syrup and gums used due to their high content of sugar compounds [10, 12-14]. Despite the favorable impact of some chemical compounds such as hormonal compounds on different processes including rooting, these created risks in terms of environmental and health. Moreover, the entry of these compounds into soil and water, and then the life cycles of living organisms will have abnormal consequences. Thus, the use of compounds with natural origin with influence on processes such as rooting, within the economic savings, will be a significant role in product safety, soil, water and living organisms.

The objective of the present study was to examine the rooting of difficult-to-root cuttings of bougainvillea by their treatment with chemical hormones including IBA, aspirin and LD pills, and natural carbohydrate compounds like honey, grape syrup, and gum Arabic.

MATERIALS AND METHODS

The impact of natural carbohydrate compounds and chemical hormonal compounds were studied on the rooting traits of bougainvillea in the research green-

house of Gorgan Agricultural Research Center and in the Horticulture Laboratory of Department of Plant Production, Gorgan University of Agriculture Science and Natural Resources, Iran in 2015-2016. The study was based on a Completely Randomized Design with six treatments at four rates and a control with five replications, each one including five cuttings (25 observations). First, the uniform and disease-free semi-hardwood cuttings were taken from the maternal plants in Gorgan Research Center. The cuttings were 10-15 cm long and their terminal leaves were removed to avoid their rotting. Then, they were immediately transferred to the greenhouse. The treatments included indole butyric acid (1000, 2000, 3000 and 4000 ppm), ASA pill (0.5, 1, 2 and 4 pills per 1 L water equivalent to 40, 80, 160 and 320 mg L⁻¹), LD pill (0.5, 1, 2 and 4 pills per 1 L water equivalent to 0.09, 0.18, 0.36 and 0.72 mg L⁻¹), natural honey (10, 20, 40 and 60%), grape syrup (5, 10, 15 and 20%), and gum Arabic (5, 10, 15 and 20%) in a comparison with distilled water (control). After the treatment of the cuttings (their placement in bunches in the prepared solutions), they were planted in disinfected cocopeat + perlite substrate in the greenhouse. The greenhouse temperature was set to 25-30 °C with 85%-90% humidity. The greenhouse was equipped with a fogging system that irrigated the cuttings by spraying for two minutes once an hour. Two months after the treatment, the cuttings were slowly taken out of the rooting substrate and

after the removal of the foreign materials their traits were measured. All traits were measured at the time of their removal from substrate except the survival percentage. The rooting percentage was calculated as the ratio of the rooted cuttings to total cuttings. The number of primary and secondary roots was counted and their length was measured with a caliper. The roots were weighed with a digital scale (to get their fresh weight) after their clipping and cleaning. Then, they were oven-dried at 70 °C for 48 h to read their dry weight. The remaining rooted cuttings were labeled and were placed in pots containing a mixture of leaf mold, sand, and garden soil in equal ratios in the greenhouse. Then, they were regularly irrigated for one month and next, their survival percentage was calculated as the ratio of the survived cuttings to total cuttings. In the present study, the means comparison of the data was carried out by LSD test at the five percent probability level using SAS Statistical Software Package and MS-Excel Package drew the graphs.

RESULTS AND DISCUSSION

The results of analysis of variance (ANOVA) in Table 1 revealed that the applied treatments significantly influenced rooting percentage, the number of primary and secondary roots, the length of primary root, fresh and dry weight, and survival percentage of bougainvillea cuttings at the 1% level.

Table 1. Analysis of variance of the effect of natural and chemical treatments on rooting traits of Bougainvillea

Mean squares								
Sources of changes	df	Rooting percent	Number of primary roots	Number of secondary roots	Length of primary root	Fresh weight of root	Dry weight of root	Survival percent
Treatment	24	1848.333 **	7.554 **	77.537 **	8.345 **	0.034 **	0.001 **	36.739 **
Error	100	0.02	0.02	0.001	0.0008	0	0	0
Coefficient of variation	-	0.346	10.045	0.607	1.247	0.368	1.149	0

** , * , n.s. :Significant in 1%, 5% and no significant

Rooting percentage

The highest rooting percentage of bougainvillea was 78% observed in those treated with 4000 ppm IBA and differed with other treatments and control significantly (Figure 1). The second and third highest rooting percentages of 72% and 66% were obtained from the application of 3000 ppm hormone and 10% grape syrup, respectively. The lowest rooting percentage (0%) was related to cuttings treated with 10% and 15% gum. The second lowest one was associated with control in which 16% rooting was observed. In addition, 20% gum and honey, and aspirin and LD at the rates of 4 and 1 pill L⁻¹, respectively, exhibited higher rooting than other concentrations and control.

Rooting is a complicated process influenced by numerous factors like the concentration of the base medium, carbohydrate-containing nutrients, light, darkness, temperature, and the presence of phenolic compounds [15]. Plant hormones, especially auxins, play a crucial role in the regulation of plant growth and development [16]. IBA is one of the best and most commonly used auxin compounds used to stimulate the rooting of the cuttings of a wide range of plant species [17]. The positive impact of auxins on rooting is associated with their effective role in stimulating cell division, stimulating the initiation of root initiators accelerating root initiation, and inducing uniform rooting [3]. In a study on bou

gainvillea rooting, IBA and the position of cutting influenced the rooting of the cuttings significantly and that rooting was increased with IBA concentration [18]. Five rates of IBA were applied on hardwood cuttings of *Jatropha curcas* and were found the hormone changed rooting significantly so it improved as the hormone rate was increased. These results confirm our findings of the effect of IBA on bougainvillea. Rooting needs energy [19]. Carbohydrates supply the energy requirements of the cuttings maybe through EMP-TCA pathway and/or pentose phosphate pathway. Many researchers have reported the positive relationship between carbohydrates and root initiation [20]. Exogenous application of sucrose (in the presence or absence of auxin) is known to be effective on rooting of many herbaceous and woody species [11]. Cuttings treated with 30 g L⁻¹ sucrose was showed the highest rooting percentage of 56.2% without a significant difference to that obtained from sucrose rate of 45 g L⁻¹. [21]. The translocation of carbohydrates to the rooting region and the stimulation of cell division in these regions have stimulated the rooting [22]. The application of date syrup as a carbohydrate-containing compound, particularly at 75% rate, considerably influenced the rooting of ornamental beriberi's cuttings. We found a similar effect of grape syrup on bougainvillea cuttings [23].

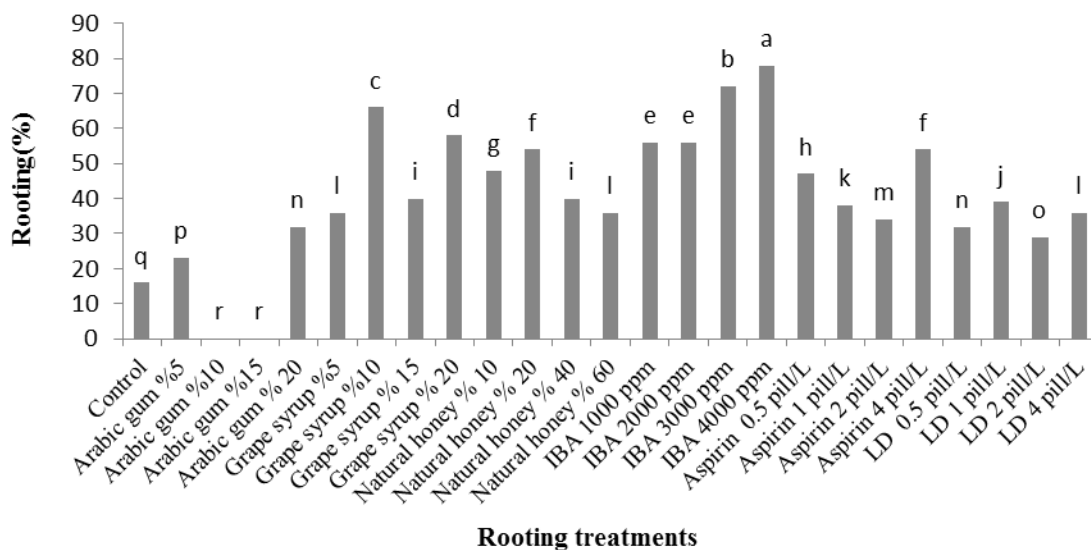


Figure 1. Effect of natural and chemical treatments on rooting percent of Bougainvillea

Number of primary and secondary roots

The highest number of primary roots (4.165, on average) grew on cuttings treated with 4000 ppm IBA but it did not differ with that of 10% grape syrup significantly. Other concentrations of IBA and grape syrup also played a considerable role in the formation of primary roots. The cuttings treated with 10% and 15% gum did not produce primary roots at all and did not show significant differences with 60% honey in this respect. Control had a few primary roots (0.4 roots) (Figure 2). A look at the other treatments indicated that 5% gum, 20% honey, and 4 pills L^{-1} aspirin and LD played a significant role in enhancing the number of primary roots in the cuttings of bougainvillea with a significant difference with that of control.

The highest number of secondary roots (15 roots, on average) was observed in cuttings treated with 10% grape syrup (Figure 3). The treatment of the cuttings with 4000 ppm IBA was related to the generation of 13.75 secondary roots in the cuttings of bougainvillea with significant differences with that of other treatments. The least number of secondary roots (no root, indeed) was associated with those treated with 10% and 15% gum followed with control (in which 1.5 roots

were observed). Among treatments, 5% gum, 20% honey, and 0.5 pills L^{-1} aspirin and LD were related to the development of more secondary roots, showing significant differences to control.

The desirable effect of auxin on the root initiation and the development of primary and secondary roots are obvious, as it has been reported in many reports including our study. The highest number of the roots in the cuttings of *Callistemon viminalis* was observed in those treated with 4000 $mg L^{-1}$ IBA [22]. Higher IBA rates resulted in the development of more roots in the cuttings of rosemary and that the highest number of roots (11.67 roots) was developed in cuttings treated with 4000 ppm IBA [24]. Rooting percentage and root number of semi-hardwood cuttings of *C. lanceolatus* were enhanced as IBA rate was increased from 1000 ppm to 4000 ppm. Higher IBA rates were related to fewer secondary roots in the cuttings of this ornamental plant [25]. Rooting percentage and root number in olive cuttings were higher in carbohydrate treatment with sucrose than control [26]. In which various sources of carbohydrate-containing compounds were applied, root number and primary root formation were higher in treatment with

glucose than with other carbon sources [27]. The impact of natural carbohydrate treatments on the rooting of the cuttings in ornamental berberis, when the cuttings were prepared in Feb-Mar, the highest root number (9.6 roots) was obtained from the treatment of 100% natural honey without significant differences with those treated with

75% honey and 50% and 75% date syrup, but over the time, the number of the roots started to decline [23]. The positive influence of carbohydrate compounds of grape syrup on the formation of roots on the studied cuttings is confirmed by the aforementioned research.

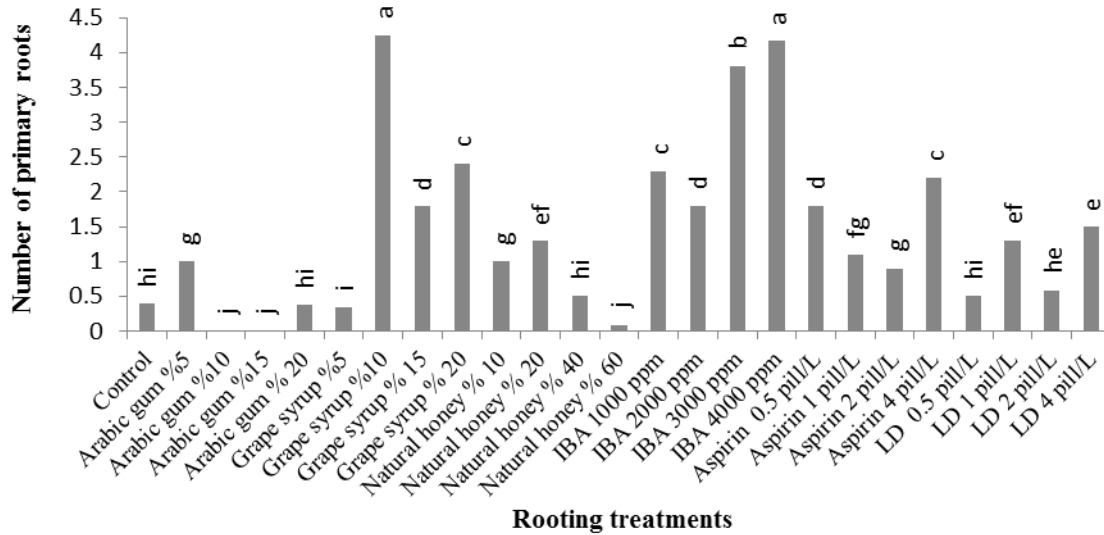


Figure 2. Effect of natural and chemical treatments on the number of primary roots of Bougainvillea

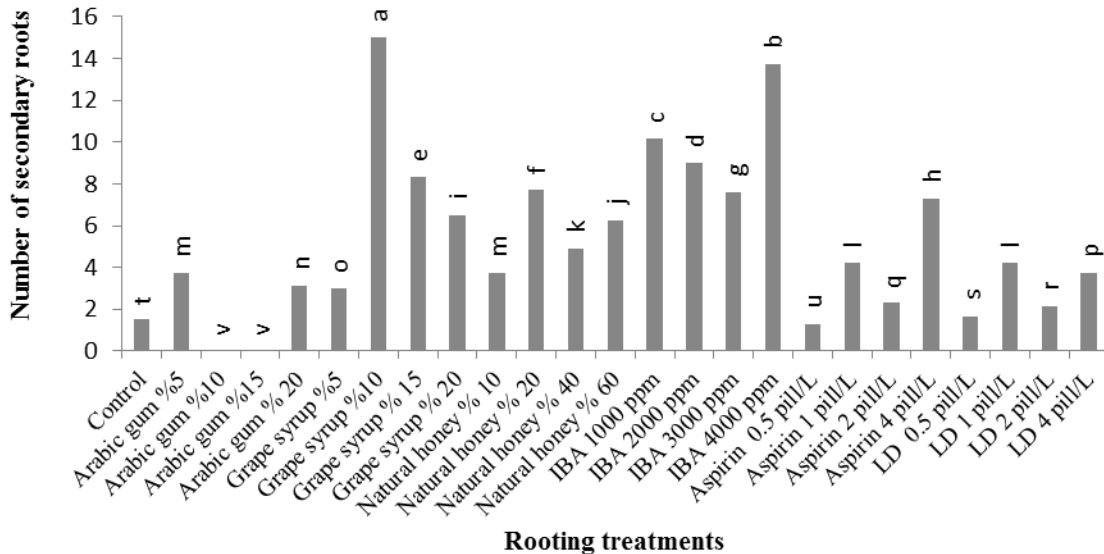


Figure 3. Effect of natural and chemical treatments on the number of secondary roots of Bougainvillea

Length of primary roots

The longest primary roots (4.507 cm, on average) were produced by cuttings treated with 4000 ppm IBA that did not differ with 10% honey significantly (Figure 4). The shortest primary roots were observed in 10% and 15% gum (0 cm) followed by control (0.575 cm). Among various concentrations of the studied compounds, 20% gum Arabic, 15% grape syrup and 4 pills L⁻¹ aspirin and LD were related to the longest primary roots and showed considerable differences with control.

One crucial parameter to evaluate the rooted cuttings is the root length [28] because root length in soil reflects plant's potential to uptake water and nutrients [29]. Total root length of cuttings is a function of root number in addition to root size. Therefore, the best hormone rate is the one in that the product of root number × mean root length is the highest with a significant difference in other rates. Like bougainvillea, the longest roots in the cuttings of *C. viminalis* were observed in those treated with 4000 mg L⁻¹ IBA [22]. In another study, higher IBA rate

resulted in the development of longer roots and the longest roots (40 cm, on average) were observed in cuttings treated with 20000 mg L⁻¹ IBA 80 d after their planting [30]. The application of IBA on the cuttings of bougainvillea was influenced root length significantly so that mean root length was 9.77 cm when the hormone was applied and it had a significant difference with control (6.88 cm) [31].

The effect of carbohydrate compounds is obvious as an energy supplier and the stimulator of root formation and then, its elongation. The cuttings of Persian walnuts were treated with 45 g L⁻¹ sucrose produced the longest roots (2.88 cm, on average) [21]. The highest root length of 10.39 cm in cuttings treated with 75% date syrup in Feb-Mar but they reported that it did not exhibit a significant difference with those obtained from 100% date syrup treatment. In addition, 75% date syrup applied in May-Jun was associated with high root length that is consistent with the favorable effect of grape syrup observed in the present study [23].

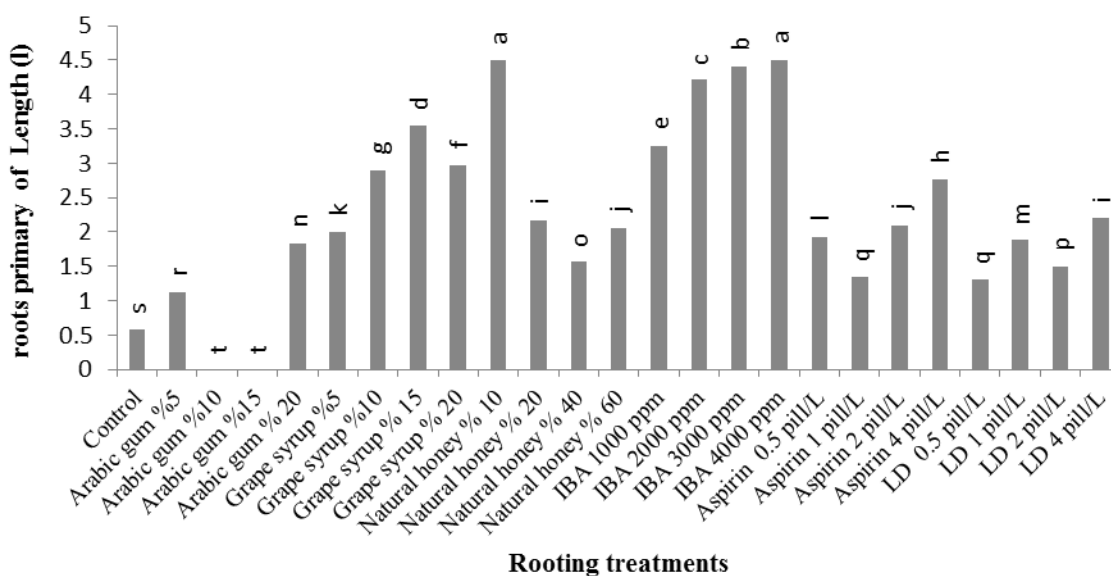


Figure 4. Effect of natural and chemical treatments on the length of primary roots of Bougainvillea.

Root fresh and dry weight

The highest and lowest fresh and dry weights were related to cuttings treated with IBA and as IBA rate was increased, root fresh weight was increased so that 400 ppm IBA resulted in the highest root fresh and dry weight of 0.329 and 0.080 g, respectively (Figure 5 and 6). In addition, 20% gum Arabic and natural honey, 10% grape syrup, and 4 pills L⁻¹ aspirin and LD had higher fresh and dry weight than other concentrations and control. The lowest fresh and dry weights (0 g) were related to the roots of the cuttings of plants treated with 10% and 15% gum. Control was in the next ranks of the lowest fresh and dry weight of 0.022 and 0.040 g, respectively.

The fresh and dry weights of the roots were increased with rooting percentage [32]. Higher IBA rate was increased fresh and dry weight [30]. The highest fresh weight (4.43 g) and dry weight (0.56 g) at 3000-4000 ppm improved the dry weight of oleander roots [33]. These results confirm our findings of bougainvillea. Auxin induces rooting and thereby, results in the translocation of more carbohydrates towards rooting zone and increased dry matter. The treatment of bougainvillea cuttings with IBA influenced fresh and dry weight significantly so that fresh and dry weights of 6.88 and 0.5 g were obtained from IBA treatment that showed significant difference with control [31].

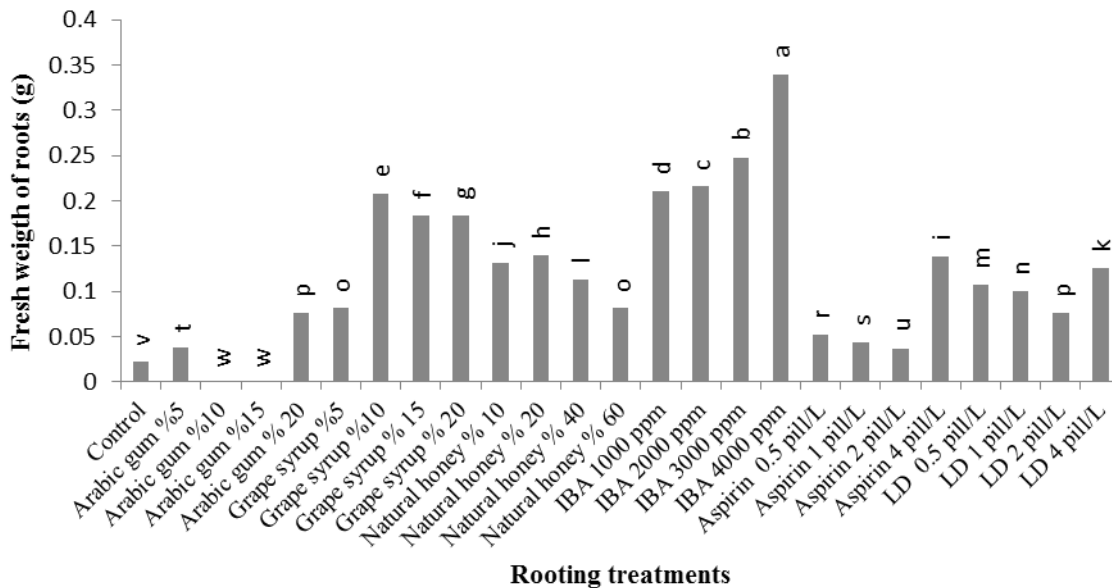


Figure 5. Effect of natural and chemical treatments on the fresh weight of roots of Bougainvillea

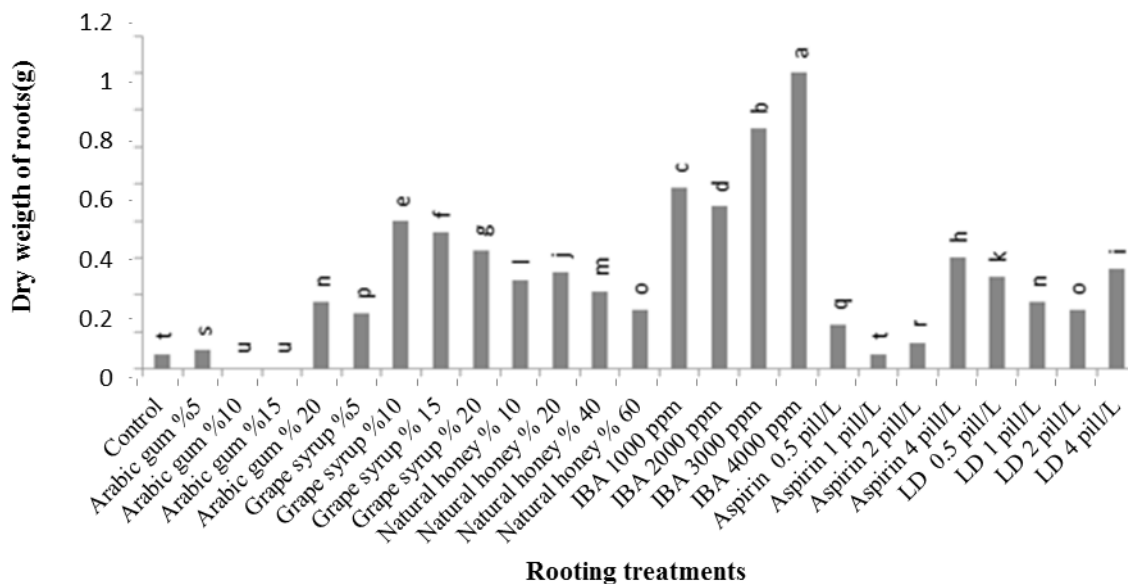


Figure 6. Effect of natural and chemical treatments on the dry weight of roots of Bougainvillea

Survival percentage

In the present study, except the treatment of 10 and 15% gum Arabic in which no rooting occurred, the other rooted cuttings were healthy after movement to pots, showed a perfect survival (100%), and kept growing. The lowest survival rate was 87% related to control. The other treatments did not show significant differences in survival rate (Figure 7).

Various studies have focused on the effect of hormone treatments, planting date, planting substrate, etc. on the survival of rooted cuttings of different species. In a study on the effect of IBA on the survival rate of semi-hardwood cuttings of *Sophora japonica*, when the hormone was applied at the rate of 10000 mg L⁻¹, the cuttings showed 83.33% survival rate [30]. The transfer of rooted stems of *Rosa damascene* Mill to the pots containing a 1:1:1 mixture of garden soil, sand and peat in the greenhouse and irrigated conditions resulted in the survival of 90% of the rooted stems [34]. In cuttings of

ornamental beriberi's in Feb-Mar, most natural treatments of date syrup, coconut syrup, and natural honey as well as 1000 mg L⁻¹ IBA were associated with high survival rate. Hormone treatment in Apr-May and Oct-Nov had similar results with previous treatments, showing the highest survival percentage [23]. In the present study, all natural and hormonal treatments had desirable influences on the survival of cuttings and it was only declined in the control cuttings due to the generation of fewer and weaker roots. Appropriate soil substrate (proper physical and chemical characteristics, adequate water availability to meet water requirement of plant growth), the sound transfer from rooting substrate to main substrate, and the placement of rooted plants in an appropriate environment are among the determinants of cutting survival. These considerations could have been the reason for 100% survival of the rooted cuttings of bougainvillea.

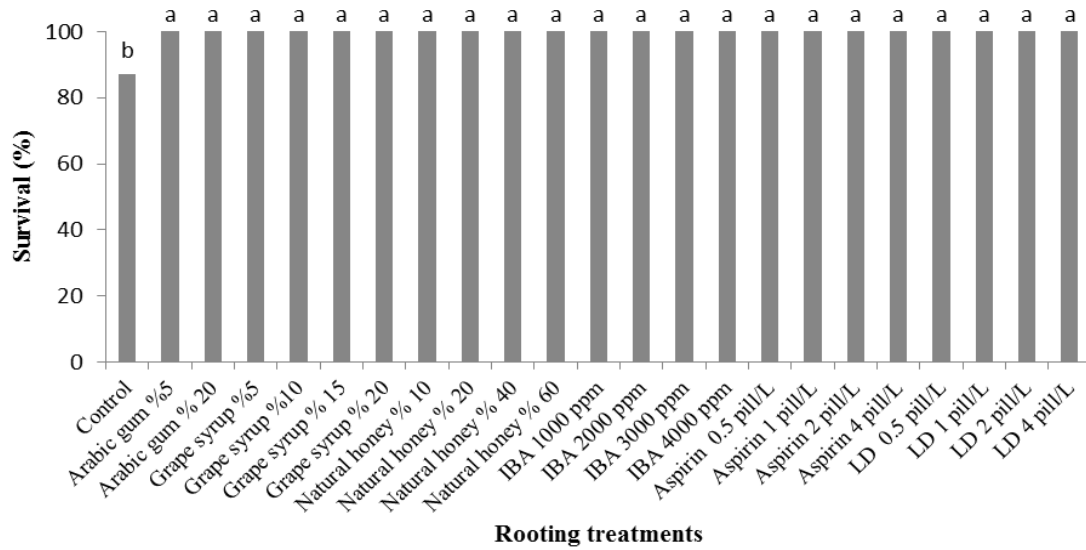


Figure 7. Effect of natural and chemical treatments on the survival percentage of rooted cuttings of Bougainvillea

In Figure 8, the best treatments in rooting it have been shown in compared with control.



Figure 8. Rooted cuttings of Bougainvillea

CONCLUSIONS

The difficult-to-root cuttings of bougainvillea exhibited the optimum results when they were treated with 4000 ppm IBA and 10% grape syrup sugar mixture. The examination of individual treatments revealed that 20% gum Arabic and honey, 10% grape syrup, 4000 ppm IBA, 4 pills L⁻¹ (320 mg L⁻¹) aspirin and 4 pills L⁻¹ (0.72 mg L⁻¹) LD were related to the best rooting of the cuttings. IBA and grape syrup at proper concentrations had optimum effects on the rooting of bougainvillea cuttings. However, given the economic and health advantages of natural compounds and their critical role in organic agriculture, the use of natural carbohydrate-

containing compounds like grape syrup that induce rooting is in priority and is recommended for the propagation of ornamental plants.

ACKNOWLEDGMENTS

The authors declare that there is no conflict of interests.

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