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EFFECT OF ROOTING HORMONE TREATMENTS ON PROPAGATION OF *Actinidia* sp. BY HARDWOOD CUTTINGS

Adrian George PETICILĂ, Roxana Maria MADJAR, Gina VASILE SCĂEȚEANU, Adrian ASĂNICĂ

University of Agronomic Sciences and Veterinary Medicine of Bucharest, 59 Mărăști Blvd, District 1, Bucharest, Romania

Corresponding author email: rmadjar@yahoo.com

Abstract

*Recently, kiwifruit consumption increased significantly, having in view its high nutritive and medicinal values. As consequence, the development and expansion of the kiwifruit industry has led to an increasing need for propagation material. The kiwifruit can be efficiently propagated under certain circumstances (proper substrate, hormonal rooting treatments) by hardwood cuttings, the most accessible methods for vegetative propagation for many horticultural crops. Our study presents the results of investigations on the effects of some rhizogene treatments (Radistim, naphtylacetic acid (**NAA**) 2000 ppm, beta-indolyl butyric acid (**IBA**) 2000 ppm and a mixture of **NAA+IBA** 1000 ppm) on root performance of hardwood cuttings for Actinidia deliciosa (AD20, Hayward, Katiuscia, Kramer, Tomuri) and Actinidia arguta (Francesca, Jumbo) species grown on a substrate composed from sand and perlite. It was developed a bifactorial experiment where "**a**" factor was kiwi cultivar/hybrid and "**b**" factor was the applied treatment represented by different rhizogene compounds. The efficiency of hardwood cutting under various hormonal rooting treatments was quantified by rooting percentage, number of formed roots, roots lengths (cm/cutting). The best rooting parameters were found when **NAA+IBA** 1000 ppm solution was used for Actinidia deliciosa (Hayward): rooting percentage was 73.1%, number of formed roots 17.2 and root length was evaluated as 19.3 cm/cutting.*

Key words*: Actinidia* sp.*, hardwood cutting, IBA, NAA, rooting.*

INTRODUCTION

Originally grown in mountainous regions of China and known as *mihoutau* (Singletary, 2012), kiwi belongs to *Actinidia* genus and it is derived from a deciduous, woody fruiting vine. *Actinidia* species are perennial, present vigorous growth and has climbing and strangling characteristics (Cangi et al., 2006). Species with fruits commonly consumed are kiwifruit/fuzzyfruit (*Actinidia deliciosa*), golden kiwifruit (*Actinidia chinensis*), baby kiwifruit (*Actinidia arguta*), Arctic kiwifruit (*Actinidia kolomikta*), red kiwifruit (*Actinidia melanandra*), silver vine (*Actinidia polygama*), purple kiwifruit (*Actinidia purpurea*) (Gjeloshi et al., 2014; Lee, 1990).

In ancient times, in China, kiwifruit was a remedy for digestive disturbances, rheumatism, dyspepsia, diminution of skin disorders and lately has gained worldwide popularity since consumers associated the consumption of kiwi fruits with potential health effects (Singletary, 2012). These fruits are a great source of antioxidants (ascorbic acid, α-tocopherol, flavones) (Singletary, 2012), minerals (Na, K, Ca, Mg, Mn, Fe, Cu, Zn) (Çeliket al., 2006; Jesion et al., 2013) and organic hydroxiacids (mainly citric acid) that are involved in the acid-base balance of the body.

Kiwifruit represents an important horticultural crop in New Zealand (Ferguson, 1990) where kiwi plant seeds were brought in the early 20th century (Singletary, 2012). Since then, commercial growth of this fruit has spread in many countries such as United States, Japan, Greece (Singletary, 2012), China (Huang, 2001), Korea (Koh et al., 2005), Italy (D'Evoli et al., 2015; Testolin et al., 2009), Turkey (Zenginbal et al., 2014) and lately, there are interest regarding this culture in Romania, for instance the mild microclimates where peach, apricot and almond trees grow (Peticilă et al., 2012; 2016). *Actinidia arguta* it has been proved to be more resistant at low temperatures and due to this behaviour, may be cultivated in almost all the regions of Romania (Peticilă et al., 2012; 2016).

Accordingly, the importance of kiwi culture, the development and expansion of the kiwifruit industry has led to an increasing need for propagation material.

The kiwifruit can be propagated by sexual reproduction (Ono et al., 2000) and by vegetative methods (green or hardwood cuttings, grafting, budding) the latter being preferred (Karkurt et al., 2009).

It is known that kiwifruit cuttings are more difficult to root than cuttings of many other species (Atak et al., 2015). *Actinidia arguta* it is commonly propagated by hardwood cuttings with different rates of success (Beyl et al., 1995).

For *Actinidia deliciosa* and *Actinidia arguta* propagation could be used softwood/green cuttings (Atak et al., 2015; Peticilă et al., 2016) or hardwood cuttings (Atak et al., 2015), the main problem, especially for hardwood cuttings, being represented by the tendency for cuttings to form excessive callus and few roots (Beyl et al., 1995). Although present this disadvantage, hardwood cuttings are the most accessible methods (least expensive and easiest) for vegetative propagation for many horticultural crops (Karkurt et al., 2009).

Rooting from the cutting it was demonstrated that is influenced by fruit specie, planting time, cultivar and rooting hormones (Kumar et al., 2002).

According to literature, the efficiency of various substrates for rooting process has been investigated: sand, perlite, sand+perlite, peat, peat+perlite, cocopeat, cocopeat+perlite (Atak et al., 2015; Peticilă et al., 2016). Some researchers (Atak et al., 2015) found that for hardwood cuttings of *Actinidia deliciosa* 'Hayward', perlite was not proper for rooting, meanwhile for rooting of softwood cuttings perlite and cocopeat were found to be suitable rooting media.

Contrariwise, a mixture (1:1) of sand and perlite it proved its superiority over other substrates (sand, sand+azocompost) regarding influence of rooting for semi-hardwood cuttings of *Actinidia chinensis* (Miri-Nargesi et al., 2015).

Other factor extensively studied and involved in rooting of cuttings is represented by different synthetic auxin-like hormones. So far, beta-indolyl butyric acid (**IBA**) (Alam et al., 2007; Peticila et al., 2016; Üçleret al., 2004; Zenginbal et al., 2014) and naphtylacetic acid (**NAA**) (Ono et al., 2000; Peticilă et al., 2016) used separately or together has proven the efficiency on rooting capacity of kiwi cuttings. Notwithstanding, some investigations have revealed that the application of auxins had no significant impact on the root number and rooting percentage for semi-hardwood cuttings of *Actinidia chinensis* (Miri-Nargesi et al., 2015; Ono et al., 2000).

The aim of the current research was to investigate the manner in which hormonal treatments determine the rooting of hardwood cutting for *Actinidia deliciosa* and *Actinidia arguta* species grown on a substrate composed from sand and perlite.

The present study is a continuation of our previously reported researches (Peticilă et al., 2016), were it was analyzed the influence of hormonal treatments on rooting for the same kiwi species, in the same experimental conditions (substrate composition, hormonal treatments) the main difference being the type of cutting (green cutting).

It was developed a bifactorial experiment where "**a**" factor was kiwi cultivar/hybrid and "**b**" factor was the applied treatment represented by different rhizogene compounds. It was evaluated the influence of the cultivar/hybrid and applied treatment on rooting percentage, number of formed roots, roots lengths (cm/cutting).

MATERIALS AND METHODS

In the experiment located at UASMV Bucharest greenhouse was used lignified cuttings of 15-20 cm length (3-4 internodes) that were obtained from species *Actinidia deliciosa* and *Actinidia arguta*. For rooting, was used a bench that allowed to maintain the substrate temperature of 18-20^{o}C. As air temperature in the greenhouse began to increase, it appeared the risk of buds start growing, so it was turned on the misting system, which succeeded to prevent the sudden increase of air temperature.

According to different studies (Alam et al., 2007; Kumar et al., 2002; Ono et al., 2000; Peticilă et al., 2016; Üçleret al., 2004; Zenginbal et al., 2014), rooting from the cutting is strongly influenced by the rooting hormones and accordingly, to enhance the success of rooting the cuttings were dipped for 10 seconds in different rhizogene compounds (Table 1).

Statistical analysis was performed using Tukey's multiple range test at p≤0.05. Tukey's test compares the means of every treatment to the means of every other treatment.

Table 1. Experimental design

Actinidia species	*Actinidia deliciosa*: Kramer, Hayward, Katiuscia cultivars, Tomuri and AD20 hybrids *Actinidia arguta*: AA5 (Jumbo), AA2 (Francesca) cultivars
Substrate	sand and perlite (1:1, volumetric ratio)
Experimental variants	V1 - control V2 - **Radistim** V3 - **NAA** 2000 ppm (NAA = napthyl acetic acid) V4 - **IBA** 2000 ppm (IBA = beta-indolyl butyric acid) V5 - **NAA** + **IBA** 1000 ppm

Table 2. Bifactorial experiment for hardwood cutting at *Actinidia* sp.

a Factor = **cultivar/hybrid**	b Factor = **treatment applied**
a1 = AA2 (Francesca) a2 = AA5 (Jumbo) a3 = AD20 a4 = Hayward a5 = Katiuscia a6 = Kramer a7 = Tomuri	b1 = untreated (control) b2 = **Radistim** b3 = **NAA** 2000 ppm b4 = **IBA** 2000 ppm b5 = **NAA** + **IBA** 1000 ppm

RESULTS AND DISCUSSIONS

1. *Influence of the cultivar/hybrid (a factor) and treatment applied (b factor) on the rooting percentage (%) at Actinidia* sp. *grown on substrate of sand and perlite*

The variance analysis indicates significant differences given by the cultivar/hybrid for the same rooting hormonal treatment. The highest rooting percentage (73.1%) was found for *Actinidia deliciosa* (Hayward) as against other tested cultivars/hybrids for rooting using sand and perlite mix substrate (Table 3). There are no significant differences on rooting percentage given by the treatment with **IBA** 2000 ppm (V4) between AA2 (Francesca), Katiuscia and Kramer. In the case of V5 treated with **NAA+IBA** 1000 ppm there are no significant differences on rooting percentage between Hayward cultivar and Tomuri hybrid.

Investigations on rooting process for the same cultivar/hybrid with different stimulating treatments led to significant differences, the best results for rooting percentage being observed after **NAA+IBA** 1000 ppm treatment, followed by **IBA** 2000 ppm and **NAA** 2000 ppm. In the case of *Actinidia* species, Radistim application for rooting stimulation generated results that it ranks it on last position in comparison with other treatments. The best rooting percentage after Radistim treatment was obtained for AA5 (Jumbo), this value allowing the ranking on the second position after **NAA+IBA** 1000 ppm treatment.

The lowest value for rooting percentage was obtained for control (untreated) variant regardless cultivar or variety.

In the issue, it is recommended to use rooting hormonal treatments for good results of rooting percentage and the most efficient experimental variant it has been encountered for **NAA+ IBA** 1000 ppm. The best result for rooting percentage irrespective of applied treatment was found for Hayward.

2. *Influence of the cultivar/hybrid (a factor) and treatment applied (b factor) on number of formed roots at Actinidia* sp. *grown on substrate of sand and perlite*

Variance analysis indicates significant differences given by the cultivar or hybrid for the same rooting hormonal treatment. The highest number of formed roots is found in the case of *Actinidia deliciosa* (Hayward) in comparison with other cultivars and varieties tested for rooting process on sand+perlite substrate (Table 4). For control (untreated) variant (V1) appear significant differences given by the cultivar/hybrid for Hayward and Kramer as against AA2 (Francesca), AD20, Katiuscia. There are no significant differences given by the cultivar/hybrid when Radistim treatment is used for AA5 (Jumbo), Kramer and Tomuri and AA2 (Francesca), AD20 and Katiuscia, respectively, but very significant differences between those above exemplified and Hayward. Also, there are no significant differences given by the cultivar/hybrid when **NAA** 2000 ppm (V3) treatment is applied between Hayward and Kramer, AA5 (Jumbo) and Tomuri, AA2 (Francesca), AD20 and Katiuscia, respectively, but very significant between the mentioned three resulted categories due to the difference in the number of formed roots.

Investigation on the influence of the same cultivar/hybrid treated with different rooting stimulating products on the number of formed roots conducted to significant differences. The best response objectified as number of formed roots is obtained for *Actinidia deliciosa* in the case of **NAA+IBA** 1000 ppm treatment, followed by **IBA** 2000 ppm and **NAA** 2000 ppm, the last ranked being Radistim treatment. Treatments on *Actinidia arguta* indicate no significant differences for Radistim, **NAA** 2000 ppm and **IBA** 2000 ppm for AA2 (Francesca), respectively between Radistim, **NAA** 2000 ppm and **NAA+IBA** 1000 ppm for AA5 (Jumbo).

As conclusion, for *Actinidia deliciosa* it is recommended **NAA+IBA** 1000 ppm treatment to obtain large number of formed roots, meanwhile for *Actinidia arguta* all applied treatments provide similar response, with no significant differences. The cultivar/hybrid that recorded the highest number of formed roots regardless the used treatment is Hayward in the case of which the highest number of formed roots was 17.2 when **NAA+IBA** 1000 ppm was applied.

Table 3. Influence of the cultivar/hybrid (a factor) and treatment applied (b factor) on the rooting percentage (%) at *Actinidia* sp. grown on substrate of sand and perlite

Cultivar/ hybrid	a\b	Rooting percentage (%)				
		Variants				
		V1 Control untreated	**V2** Radistim	**V3** NAA 2000 ppm	**V4** IBA 2000 ppm	**V5** NAA+IBA 1000 ppm
Actinidia arguta	AA2 (Francesca)	b44.8d	d52.1c	e51.7c	d55.7a	f53.9b
	AA5 (Jumbo)	c39.5e	f47.5b	f43.9c	e41.8d	e56.1a
Actinidia deliciosa	AD20	d35.1d	c55.1c	c56.1c	c58.7b	b61.3a
	Hayward	a50.9e	a64.5d	a66.1c	a69.7b	a73.1a
	Katiuscia	f30.0e	e51.0d	e52.2c	d56.5b	c60.1a
	Kramer	e33.1e	d52.1d	d53.8c	d55.9b	d58.1a
	Tomuri	b45.1e	b63.1d	b64.5c	b67.2b	a72.1a
[1]B constant A variable: LSD 5%=1.00*% ; LSD 1%=1.34%; LSD 0.1%=1.77% [2]A constant B variable: LSD 5%=1.06*%; LSD 1%=1.41%; LSD 0.1%=1.84%						

There were made interpretations by DL 5% indicated in the table by *
[1]Means with different letters in a column (in front of data) are significant different.
[2]Means with different letters in a row (in back of data) are significant different.

Table 4. Influence of the cultivar/hybrid (a factor) and treatment applied (b factor) on number of formed roots at *Actinidia* sp. grown on substrate of sand and perlite

Cultivar/ hybrid	a\b	No. of formed roots				
		Variants				
		V1 Control untreated	**V2** Radistim	**V3** NAA 2000 ppm	**V4** IBA 2000 ppm	**V5** NAA+IBA 1000 ppm
Actinidia arguta	AA2 (Francesca)	c6.9b	c8.3a	c7.9a	d8.9a	f7.1b
	AA5 (Jumbo)	d6.3c	b9.9a	b9.8a	d8.5b	e10.1a
Actinidia deliciosa	AD20	c7.9b	c7.3c	c8.5b	c9.5a	e9.7a
	Hayward	a11.0d	a12.9c	a12.1c	a14.8b	a17.2a
	Katiuscia	c7.1d	c7.5c	c8.3c	c10.2b	d11.5a
	Kramer	a10.2c	b10.9c	a11.8b	b12.6b	b15.5a
	Tomuri	b9.1d	b9.9c	b10.7c	b11.9b	c13.9a
[1]B constant A variable: LSD 5%=1.11*% ; LSD 1%=1.51%; LSD 0.1%=2.05% [2]A constant B variable: LSD 5%=1.00*%; LSD 1%=1.33%; LSD 0.1%=1.74%						

There were made interpretations by DL 5% indicated in the table by *
[1]Means with different letters in a column (in front of data) are significant different.
[2]Means with different letters in a row (in back of data) are significant different.

Table 5. Influence of the cultivar/hybrid (a factor) and treatment applied (b factor) on roots length (cm/cutting) at *Actinidia* sp. grown on substrate of sand and perlite

Cultivar/ hybrid	a\b	Root length (cm/cutting)				
		Variants				
		V1 Control untreated	**V2** Radistim	**V3** NAA 2000 ppm	**V4** IBA 2000 ppm	**V5** NAA+IBA 1000 ppm
Actinidia arguta	AA2 (Francesca)	d6.8b	e6.7c	e7.6b	e8.8a	f6.9b
	AA5 (Jumbo)	c8.0d	d8.2d	d9.3c	d10.5b	d11.7a
Actinidia deliciosa	AD20	c7.1c	c9.3a	d8.5b	e9.1a	e9.8a
	Hayward	a10.9d	a19.7a	a18.4b	a17.0c	a19.3a
	Katiuscia	a10.1c	b11.9b	c10.3c	c12.1a	c12.9a
	Kramer	b8.9d	b11.5c	b12.9b	c11.3c	b14.3a
	Tomuri	b9.0d	b11.1c	b12.8b	b13.5b	b14.8a
[1]B constant A variable: LSD 5%=0.99*% ; LSD 1%=1.36%; LSD 0.1%=1.86% [2]A constant B variable: LSD 5%=0.83*%; LSD 1%=1.10%; LSD 0.1%=1.44%						

There were made interpretations by DL 5% indicated in the table by *
[1]Means with different letters in a column (in front of data) are significant different.
[2]Means with different letters in a row (in back of data) are significant different.

3. *Influence of the cultivar/hybrid (a factor) and treatment applied (b factor) on roots length (cm/cutting) at Actinidia* sp. *grown on substrate of sand and perlite*

The variance analysis indicates significant differences given by the cultivar/hybrid at the same treatment for root length stimulation. The highest root length (cm/cutting) is found in the case of Hayward, followed by Tomuri, Kramer and Katiuscia from *Actinidia deliciosa* in comparison with other cultivars and varieties tested on sand and perlite mix substrate (table 5). In the case of Radistim treatment (V2) there are no significant differences given by the cultivar/hybrid for Katiuscia, Kramer and Tomuri but appear very significant differences for Hayward in the case of which was found the highest root length (19.7 cm/cutting). When **NAA** 2000 ppm (V3) and **NAA**+**IBA** 1000 ppm (V5) were applied there are no significant differences given by the cultivar/hybrid for Kramer and Tomuri but appear very significant differences for Hayward when it was found the highest root lengths, 18.4 cm/cutting and 19.3 cm/cutting, respectively.

Analyzing the influence of the same cultivar/hybrid tested with hormonal rooting treatments appear significant differences, the highest root lengths being obtained when **NAA** + **IBA** 1000 ppm treatment is applied. In the case of AD20, the best results are obtained when Radistim, **IBA** 2000 ppm and **NAA**+**IBA** 1000 ppm treatments are applied meanwhile, for Katiuscia are recommended treatments with **IBA** 2000 ppm or **NAA**+**IBA** 1000 ppm.

Finally after all investigations, for root length stimulation it is recommended to use treatments with **NAA**+**IBA** 1000 ppm but also with Radistim and **IBA** 2000 ppm. The Hayward cultivar offered the best response as highest root lengths irrespective of applied treatment.

In other circumstances, solution **NAA+IBA** 1000 ppm also proved its efficiency on rooting ability of green cuttings for *Actinidia arguta* (Peticilă et al., 2016). Use of **IBA** 6000 ppm and **NAA** 8000 ppm in the case of semi-hardwood cuttings for *Actinidia deliciosa* provided similar effects on rooting (Üçler et al., 2014). Some studies (Gjeloshi et al., 2014) indicated that use of a solution **IBA** 3000 ppm produced the highest rooting percentage (80%) of *Actinidia deliciosa* cv. Hayward hardwood cuttings. Also, the highest rooting percentage (63%) of semi-hardwood cuttings of *Actinidia deliciosa* cv. Hayward treated with solution **IBA** 3000 ppm were obtained on perlite-peat mix substrate (60:40) (Gjeloshi et al., 2014).

CONCLUSIONS

The experiment presented in this paper was designed to investigate the efficiency of some hormonal treatments on the rooting performance of hardwood cutting for *Actinidia deliciosa* and *Actinidia arguta* species. Based upon the findings of this study, the main conclusions are presented below.

1. For good results of rooting percentage it is recommended to use hormonal treatments, the most efficient experimented variant being **NAA+IBA** 1000 ppm.

2. The best result for rooting percentage irrespective of applied treatment was found for Hayward.

3. Concerning the number of the formed roots, the best response is obtained for *Actinidia deliciosa* in the case of **NAA+IBA** 1000 ppm treatment, followed by **IBA** 2000 ppm and **NAA** 2000 ppm, the last ranked being Radistim treatment.

4. For *Actinidia deliciosa* it is recommended **NAA+IBA** 1000 ppm treatment to obtain large number of formed roots, meanwhile for *Actinidia arguta* all applied treatments provide similar response, with no significant differences.

5. For root length stimulation it is recommended to use treatments with **NAA+IBA** 1000 ppm but also with Radistim and **IBA** 2000 ppm. The Hayward cultivar offered the best response as highest root lengths irrespective of applied treatment.

6. As final conclusion, for rooting stimulation it is recommended treatment **NAA+IBA** 1000 ppm; the best response concerning rooting percentage, number of formed roots and root length stimulation was found in the case of *Actinidia deliciosa* (Hayward). This conclusion is consistent with our previously reported results (Peticilă et al., 2016), the main difference being the propagation technique by hardwood cuttings.

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