

## ATTEMPTION OF APPLYING LASER PHOTOSTIMULATION TO THE ROOTING OF SHOOT CUTTINGS OF JUNIPER SPECIES

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**Abstract.** Propagation of juniper from cuttings is the primary method used in the nursery. Process of rhizogenesis in these plants is long and not always allows to obtain a good quality of rooted cuttings. In the experiment there was investigated the effect of laser light doses (semiconductor laser, model CTL 1106MX, power 200 mW, wave length 670  $\mu\text{m}$ ) and the footing agent AB on percentage of rooted cuttings, plants height, length and the number of roots. The examined juniper taxa proved diverse response to the variants applied (combination of laser radiation and the rooting agent AB). The increase in the number of rooted cuttings after application of the rooting agent AB, as well as three times introduced irradiation in Eastern redcedar 'Blue Arrow'. This form also produced longer roots for the variant combining twelve times applied irradiation and the effect of rooting agent. In the cuttings of Chinese juniper 'Stricta', exposed to laser radiation in six – (D6) and twelve times applied doses (D12), there was observed significant stimulation of root length. Introduced treatment variants of shoot cuttings of Common juniper 'Schneeverdinger Goldmachangel' brought about considerable root elongation (except for D3 dose). The dose providing for significant increase in the percentage of rooted cuttings in all the examined juniper varieties proved to be D3 dose.

**Key words:** juniper, semiconductor laser light, seedlings, rooting

### INTRODUCTION

Varieties of juniper species (*Juniperus* sp.) bushes are propagated from half – hardy shoot cuttings during autumn and winter dormancy. In these plants the process of rhizogenesis lasts relatively long and developing adventitious roots are not always of good quality, which is expressed by a small number of poorly branched adventitious roots. Young plants, with appropriately shaped root system, show better development in

further production stages and nursery material obtained features high quality [Dirr and Heuser 1987].

In the process of adventitious roots emergence a superior role is played by endogenous auxins, whose concentration in tissues is a decisive factor in most of species, as far as the mentioned process is concerned [Tipton 1990]. Auxins concentration in shoots subjected to cuttings collection does not show even values, for their occurrence in plants depends on a number of factors. Therefore, there are commonly used exogenous auxins like IBA and NAA, which considerably affect the process of root system regeneration [Copes 2000]. Another method of the improvement in the quality of shoot cuttings rooting is spraying mother plants with auxins of different concentrations, as it took place in mother plants of garden hydrangea, whose cuttings rooted in a significantly higher percentage [Bailey and Clark 1997]. Some juniper taxa, however, cause cultivation problems connected with shoot cuttings rooting. In research involving Shore juniper 'Blue Pacific', whose cuttings were placed in discs of peat imbibed with auxins, the results regarding rooting ranged 53–93% [Blythe et al. 2004a, b].

In the literature there can be found only a few works describing application of semiconductor laser beams to stimulate plant material [Aladjadjian 2007, Ashrafijou et al. 2010, Hernandez et al. 2010, Michtchenko and Hernandez 2010, Vasilevski 2003]. In nursery practice, there have been made efforts to find new, more efficient methods of improving the quality of root system, especially in plants characterizing long and difficult rooting. One of such methods can be cuttings stimulation with laser light [Kralik and Rauscherowa 1990]. Śliwka and Jakubiak [2007] proved that stimulation with laser beams applied to cuttings of willow (*Salix viminalis* L. 'Turbo'), provided for the fact that higher number of branched roots were obtained. Research conducted on *Isatis indogotica* species [Yi-Ping et al. 2005, Chen et al. 2005] indicate that laser radiation cause not only temporary biological changes, such as alterations in seeds energy, but also long-term effects resulting in acceleration of growth and development of seedlings.

The aim of research was determination of the effect of semiconductor laser beams stimulation on the process of adventitious roots emergence in three juniper species: Common juniper (*Juniperus communis* L.) 'Schneverdinge Goldmachangel', Eastern redcedar (*J. virginiana* L.) 'Blue Arrow' and Chinese juniper (*J. chinensis* L.) 'Stricta'.

## MATERIAL AND METHODS

The experiment was carried out in the years 2009 and 2010, in a propagating hot-house in Research – Development Station of Vegetables and Ornamental Plants. Plant material was subjected to irradiation in laser laboratory at Department of Genetics, Plant Breeding and Seed Production at Wrocław University of Environmental and Life Sciences.

Material for examination were shoot cuttings 6 cm long, collected from 5-year – old mother bushes, on 15<sup>th</sup> September 2009 and 2010, from three juniper species taxa: Common juniper (*Juniperus communis*) 'Schneverdinge Goldmachangel', Eastern redcedar (*J. virginiana*) 'Blue Arrow' and Chinese juniper (*J. chinensis*) 'Stricta'. Before placing them in multipot palette 40 × 30 cm, filled with medium consisting of

highmoor peat and perlite in a volume ratio 1:1, the cuttings underwent three – (D3), six – (D6) and twelve – times applied (D12) irradiation with a basic dose ( $2.5 \times 10^{-1} \text{J} \cdot \text{cm}^{-2}$ ) of semiconductor laser beams (model CTL 1106MX, power 200 mW, wave length 670  $\mu\text{m}$ ). In the experiment there was used the rooting agent AB of the following composition NAA -0.3%, IBA 0.05% and activated carbon -5.0%. A part of the cuttings was also subjected to combinations of laser light doses mentioned above with the rooting agent AB (D3 + AB, D6 + AB, D12 + AB). Control (C) involved cuttings treated with neither laser light nor the rooting agent. The experiment was established in three replications, with 10 cuttings in every replication. After 6 months there were assessed the following parameters: percentage of rooted cuttings, length and number of produced roots, as well as the height of young plants. Obtained results underwent statistical analysis with the use of Duncan test, at significance level  $\alpha = 0,05$  there were formed homogenous groups.

## RESULTS

In order to root cuttings of Common juniper of ‘Schneeverdinger Goldmachangel’ variety, there were created two homogenous groups. The first one, characterizing significantly higher degree of rooting in comparison to control cuttings (44%), included the cuttings treated with AB rooting agent (55.2%), D3 dose of laser radiation (52.3%), as well as combination of D3, D6 and D12 doses with the rooting agent (from 51.3 to 53.2%). The applied combinations resulted in the increased value of the examined trait by 11.2 to 7.3% respectively, in relation to control (30.3%). Chinese juniper ‘Stricta’ variety featured the best rooting when subjected to threefold irradiation with semiconductor laser beams and AB rooting agent. There was obtained 57% of rooted cuttings, which provided for the increase ranging 26.7%. Additionally, introduced combinations

Table 1. Influence of Ukorzeniacz AB and laser radiation for percent of rooted cuttings of juniper selected taxa

Combination	Juniper taxa		
	<i>Juniperus communis</i> L ‘Schneeverdinger ‘Goldmachangel’	<i>Juniperus chinensis</i> ‘Stricta’	<i>Juniperus virginiana</i> L ‘Blue Arrow’
C	44.0 b	30.3 c	40.0 b. c
AB	55.2 a	44.7 a. b. c	52.3 a
D3	52.3 a	51.7 a. b	51.7 a
D3+AB	51.3 a	57.0 a	39.8 b. c
D6	43.0 b	42.3 a. b. c	32.7 c
D6+AB	53.0 a	53.0 a. b	33.3 b. c
D12	41.0 b	41.0 b. c	40.3 b
D12+AB	53.2 a	55.0 a. b	40.0 b. c
LSD $\alpha=0.05$	5.3	13.4	6.8

of D6 and D12 doses with AB rooting agent and D3 dose allowed to obtain stimulation involving the number of rooted cuttings of Chinese juniper, from 22.7 to 24.7%. The cuttings of Eastern redcedar ‘Blue Arrow’ did significantly respond to the use of AB rooting agent (52.3%) and D3 dose (51.7%). Percentage of rooted cuttings in both cases belonged to the homogenous group characterized by significantly highest values, indicating 12.3–11.7% stimulation (tab. 1).

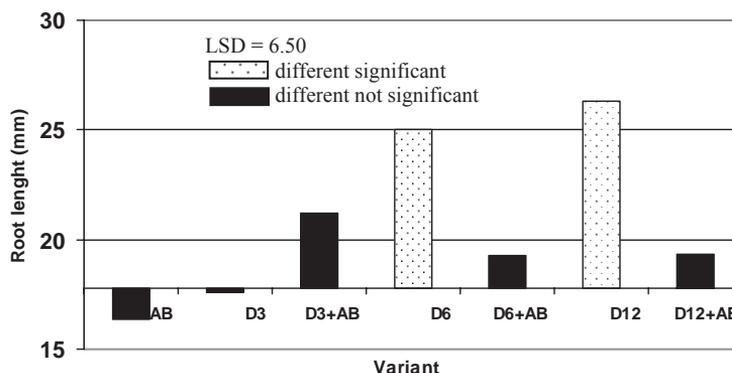


Fig. 1. Influence of Ukorzeniacz AB and laser radiation for root length seedlings of *Juniperus chinensis* ‘Stricta’ with control (C = 17.80 mm)

As far as Chinese juniper was concerned, there was observed considerable effect of laser radiation exclusively on the length of roots. Six – (D6) and twelfefold (D12) irradiation brought about stimulation of this trait value, resulting in root elongation by 7.2 cm, i.e. by 40.4% and by 8.5 cm, i.e. by 47.7% respectively. There was not recorded any influence of the applied AB rooting agent on the examined traits (fig. 1)

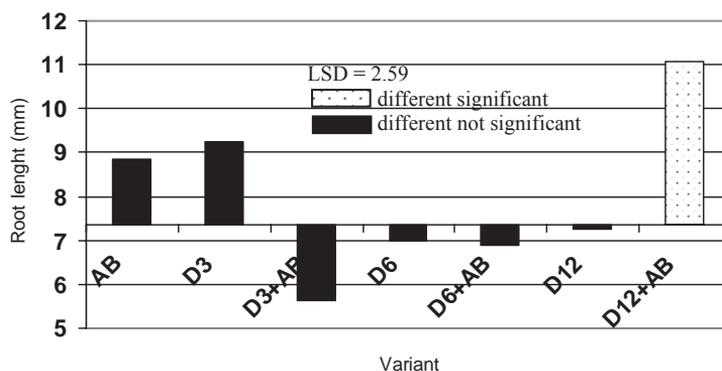


Fig. 2. Influence of Ukorzeniacz AB and laser radiation for root length seedlings of *Juniperus virginiana* ‘Blue Arrow’ with control (C = 7.35 mm)

The roots of Eastern redcedar became significantly elongated after application of combined twelvefold (D12) irradiation with the rooting agent (fig. 2). The combination of two factors allowed to obtain over 50% increase in roots length of cuttings expose to laser light. The plant height and the number of roots proved to be the traits whose values did not undergo alteration under the influence of the factors applied.

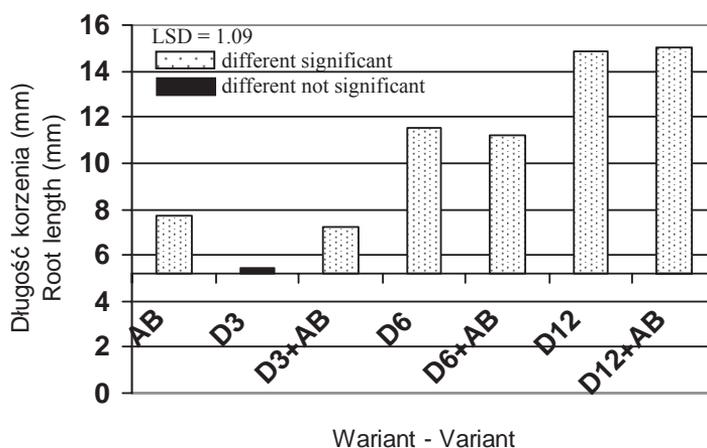


Fig. 3. Influence of Ukorzeniacz AB and laser radiation for root length seedlings of *Juniperus communis* L. ‘Schneverdinge’ ‘Goldmachangel’ with control (C = 5.23 mm)

Common juniper occurred to be the most sensitive taxon, among the examined species, to the treatment of cuttings with laser light, as well as to combined use of irradiation and the rooting agent. The application of the rooting agent alone provided for the development of cuttings roots longer by 2.5 cm, i.e., stimulation by 47.8% in relation to control cuttings, not treated with laser light. In the case of D6 dose, the use of semiconductor laser radiation caused the increase by 6.3 cm, i.e. by 120.5% and 9.67 cm, i.e. by 184.9% – for D12 dose as compared to control which produced roots only 5.23 cm long. Combined application of those two factors resulted in elongation of root length by 37.7% in the case of D3 + rooting agent and up to 186.8% after the introduction of twelvefold irradiation and AB rooting agent. Only D3 dose did not result in any significant stimulation of roots length (fig. 3).

## DISCUSSION

Seed lot, also including cuttings, is one of the most important means of production and its quality is decisive when profitability of particular cultivation is taken into account. Currently, the market forces enterprises to introduce a number of different treatments improving the value of seed lot. There are used both chemical – for example

preparations stimulating rooting as well as physical methods, among others, laser irradiation. Physical methods are safe to the environment, as they do not cause its pollution, and the effect of their application is higher vigor, as well as higher yield of better quality.

The exemplary effect of pre – sowing laser stimulation of seeds are the changes in chemical composition and the content of sugar in white beets. There was also recorded increased concentration of nitrogen, potassium, sodium and magnesium, as well as positive effect on sucrose content [Prośba-Białczyk et al. 2012]. Moreover, obtained results indicate that laser radiation modifies physiological processes taking place in the course of plants development, among others, it influences the content of chlorophyll and carotenoids [Prośba-Białczyk et al. 2011, Sacala et al. 2012].

In a nursery production there is a tendency to increase and accelerate the process of rhizogenesis through application of different preparations contributing to improved quality of rooted cuttings [Świstowska and Kozak 2004, Pacholczak et al. 2012]. In research with the use of Actisil, conducted on cuttings of Shore juniper (*Juniperus conferta* Parl.) ‘Schlagier’ and Singleseed juniper (*Juniperus squamata* Buch.-Ham. ex Lamb.) ‘Blue Star’, there was proved beneficial effect of that preparation on the length and the number of roots, as well as on the height of rooted cuttings. As a result, there were obtained young plants of a good quality which can be used in further stages of plants production [Bąbelewski 2008]. Another finding involved the fact that the quality of rooted softwood cuttings of roses is affected by the size of assimilation surface of leaves [Costa and Challa 2002].

Most commonly are used preparations containing synthetic auxins which have a significant impact on intensification of rhizogenes. Long-term research proved participation of plant hormones, including IAA, in regulation of plants growth and development from germination to reproduction. The activity of those compounds is dependent on their cellular concentration, as well as on tissue sensitivity. The mechanism of regulation, maintaining optimal level of hormones, is governed by both external and internal factors [Jakubowska 2004]. Biostimulation with laser beams and the effect of the rooting agent containing auxin, indole-3-butyric acid (IBA) and 1-naphthyl-acetic acid (NAA) can probably be external factors influencing the mechanism of hormones level regulation. In research by Copes [2000] on application of six IBA concentrations and four NAA concentrations, regarding their ability to strengthen rhizogenesis process of hard to root cuttings of Douglas fir. Diverse doses of IBA and NAA improved the quality of root system in comparison to control. In research by the author, one of the introduced factors was AB rooting agent, containing both NAA and IBA. The cuttings subjected to its effect rooted in significantly higher percentage than control cuttings, which confirmed the results obtained by Copes. Six independent investigations, involving misting of selected Douglas fir clones were made. It was possible to state that different Douglas fir clones, stimulated by auxins, produced higher number of roots and higher percentage of rooted plants. It occurred difficult to determine which concentration of auxin evidently stimulated the development of best-quality root system because both NAA and IBA affected strengthening of cuttings root system. In research carried out by Szajsner [2009] on the influence of laser radiation on caryopses of wheat, rye and triticale, there was proved significant increase in the content of indole-3-acetic acid (IAA)

in irradiated material. It can be supposed that laser stimulation of cuttings brings about similar alterations in IAA content, which can result in their better rooting. The experiment by the author proved significant, stimulatory effect of semiconductor laser radiation on increased percentage of rooted cuttings of juniper species. Moreover, laser radiation applied to irradiation of plant material provides for increased accumulation of macro- and microelements in plants [Dobrowolski and Rózanowski 1998].

In research on Eastern redcedar (*Juniperus scopulorum* Sarg.) rooting, conducted by Bielenin [2003], as well as on eastern arborvitae (*Thuja occidentalis* L.), cuttings bases were treated with powder preparation containing potassium salt of indole-3-acetic acid in concentrations 0.01, 0.2, 0.6 and 0.9%. It was proved that according to the increase in auxin concentration percentage of rooted cuttings did significantly increase. Another observation involved its advantageous effect on the quality of root system, especially regarding the length and number of roots. During rooting of cuttings of eastern arborvitae and Eastern redcedar taxa, there was investigated intensity of cuttings photosynthesis and breathing, to determine if different auxin concentrations affect the intensity of the examined physiological processes. Research results spoke for the fact that the treatment of cuttings with indole-3-acetic acid did not significantly influence on the intensity of photosynthesis and breathing of cuttings undergoing the rooting process.

In research conducted by Jakubiak and Śliwka [2007] on willow cuttings (*Salix viminalis* 'Turbo') stimulation with laser light, there was obtained both higher number and more numerous root branches, which confirms positive effect of laser radiation on the development of root system. Kralik and Rauscherowa [1990] in experiments on laser stimulation of common privet cuttings (*Ligustrum vulgare* L.) reported significant positive influence of low doses of laser light on rhizogenesis. The cuttings exposed to irradiation produced longer and more numerous adventitious roots. That effect became visible, first of all, in the initial stages of roots development. In research by the author it was possible to observe that biostimulation of juniper cuttings with laser radiation allowed to obtain elongated roots in Chinese juniper, Eastern redcedar and Common juniper. The most considerable effects were recorded for Common juniper, which proved to be the species most susceptible to laser light treatment. In the case of Chinese juniper, application of laser radiation occurred more beneficial than the use of AB rooting agent, which did not provide any effects. Combined application of both factors, i.e. laser radiation and the rooting agent, proved to be advantageous for Eastern redcedar and Common juniper, where combination of the highest dose of laser light and rooting agent resulted in the most considerable stimulation of root length.

In the literature there can be also found descriptions of experiments dealing with the use of laser radiation to increase plant biomass. The most significant increase in biomass of yellow iris (*Iris pseudoacorus* L.) was reported for plants irradiated with argon laser (irradiation time – three times for 30 seconds) – [Śliwka and Jakubiak 2007]. In the experiments by the authors there was neither observed any considerable effect of both examined factors, nor their combined application on the height of young plants of the examined taxa from juniper.

## CONCLUSIONS

1. The response of the examined taxa to applied variants (combination of laser radiation and the rooting agent AB) was diverse.

2. Eastern redcedar ‘Blue Arrow’ proved to be the most sensitive taxon, responding with the increase in the number of rooted cuttings after application of the rooting agent AB, as well as three times irradiation and with roots elongation after introduction the variant with twelve times irradiation and the rooting agent.

3. In Chinese juniper ‘Stricta’ there was observed the effect of laser radiation on roots length, while the doses of six – (D6) and twelve times applied (D12) irradiation caused considerable stimulation of this trait value.

4. Common juniper ‘Schneeverdinger Goldmangel’ occurred to be the form susceptible to the variants applied, responding with significant roots elongation (except for D3 dose).

5. Application of D3 dose resulted in significant stimulating effect on cuttings rooting in all the examined juniper species.

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## PRÓBY ZASTOSOWANIA FOTOSTYMULACJI LASEROWEJ DO UKORZENIANIA SADZONEK PĘDOWYCH RODZAJU JAŁOWIEC

**Streszczenie.** Rozmnażanie jałowców z sadzonek jest podstawową metodą stosowaną w szkółkarstwie. Proces rizogenezy u tych roślin jest długotrwały i nie zawsze pozwala na otrzymanie ukorzenionych sadzonek dobrej jakości. W doświadczeniu badano wpływ da-

wiek światła laserowego i Ukorzeniacza AB na procent ukorzenionych sadzonek, wysokość roślin, długość i liczbę korzeni. Badane taksony jałowca wykazały zróżnicowaną reakcję na zastosowane warianty (połączenie promieniowania laserowego i Ukorzeniacza AB). Zwiększenie liczby ukorzenionych sadzonek po zastosowaniu Ukorzeniacza AB jak również trzykrotnego naświetlania obserwowano u jałowca skalnego 'Blue Arrow'. Forma ta wykształciła również istotnie dłuższe korzenie dla wariantu łączącego dwunastokrotne naświetlanie i działanie ukorzeniacza. U sadzonek jałowca chińskiego 'Stricta' pod wpływem promieniowania laserowego – dawka sześćo- (D6) i dwunastokrotna (D12) – obserwowano istotną stymulację długości korzeni. Zastosowane warianty traktowania sadzonek pędowych jałowca pospolitego 'Schneverdingen Goldmachangel' spowodowały istotne wydłużenie korzeni (z wyjątkiem dawki D3). Dawką powodującą istotne zwiększenie procentu ukorzenionych sadzonek u wszystkich badanych odmian jałowców była dawka D3.

**Słowa kluczowe:** jałowiec, laser półprzewodnikowy, sadzonki, ukorzenianie

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