Journal of Ecological Engineering
Volume 15, No. 2, April 2014, pp. 112–118
DOI: 10.12911/22998993.1094987

Research Article

ASSESSMENT OF IMPACT OF COHERENT LIGHT ON RESISTANCE OF PLANTS GROWING IN UNFAVOURABLE ENVIRONMENTAL CONDITIONS

Małgorzata Śliwka[1]

[1] Department of Environmental Engineering and Mineral Processing, Faculty of Mining and Geoengineering, AGH University of Science and Technology in Krakow, 30 Al. Mickiewicza Ave., 30-059 Krakow, Poland, e-mail: sliwka@agh.edu.pl

Received: 2014.02.03
Accepted: 2014.03.17
Published: 2014.04.04

ABSTRACT
The results of experiments on the effect of the coherent light emitted by lasers on plant material show that properly selected laser stimulation parameters, such as: wavelength, power, time and type of exposure, allow to obtain a greater growth of plant biomass, changes in the content of elements in the biomass and increasing plant resistance to unfavorable environmental conditions. The aim of this study was to determine the effect of laser stimulation on selected plant species (*Iris pseudoacorus* L., *Lemna minor* L.) to increase their resistance to low temperatures and the ability to adapt to an environment polluted by mining activities (*Phelum pratense* L.). Plants from experimental groups (*Iris pseudoacorus* L., *Phelum pratense* L., *Lemna minor* L.) were stimulated with coherent light with specific characteristics. To irradiate plants from experimental groups different algorithms of stimulation parameters, differentiating the method and time of exposure were used. Plants group without the stimulation, were the reference group. The article discusses the results of preliminary experiments carried out on a laboratory scale and pot experiments.

Keywords: laser stimulation, biostimulation, plants resistance, stress factors, ecological valence.

INTRODUCTION

Visible light is responsible for the stimulation of important physiological and biochemical processes and biophysical phenomena such as: photosynthesis, phototropism, photomorhogenesis, which are based on a series of photo-biological reactions induced by quantas energy of electromagnetic radiation in the visible range 380–780 nm. The results of scientific research [Inyushin et al. 1981, Suppan 1997, Kopcewicz, Lewak 2007] on the laser stimulation of biological material confirm a significant effect of radiation on bioenergetic processes of cells. Depending on the irradiation parameters: the amount of energy supplied, the method of exposure and the type of biological material, you can get different effects of exposure: a phototermel effect, a photochemical effect, photoionization or effect of biostimulation. The effect of stimulation is the effect of laser irradiation, accompanied by a local increase of temperature (0.5-1 °C), and the changes on the cellular level does not respond to the stress. Laser radiation is absorbed by the biologically active compounds or cellular organelles, and can lead to changes in the metabolism of the cells, rather than to its destruction [Inyushin 1977, Bryszewska 1997, Dobrowolski 1999, Drozd, Szajsner 2001, Drozd et al. 2003, Popp 2006, Szajsner et al. 2008]. Scientific research [Inyushin 1977] also showed that the effect of stimulation is closely related to the properties of coherent light, such as coherence and polarization, which allow better penetration of the radiation into the tissue and cause photochemical reactions through resonant absorption and provide energy charge corresponding to a specific wavelength. Incoherent light does not show comparable to laser effects on bioenergetic processes [Karlander 1968, Friedman, Lubart 1993, Cenian et al. 2005, Liedtke, Popp 2006].

The practical use of the effect of laser stimulation of biological material were used, among other things in medicine [Inyushin et al. 1981, Dobrowolski 1993, Sieroń, Cieślar 1994, Dobrowolski, Różanowski 1995, Fiedor 1995]. It was found the impact of coherent light on bone marrow cells and blood and the activity of some enzymes. These studies have been used in the elimination of cancer cells, stimulation of the bone marrow and tissue regeneration.

There are also known studies [Gosh, Bupp 1992, Wilczek et al. 2006, Aladjadjiyan 2007, Wilczek, Ćwintal 2009, Ćwintal 2010] relating to stimulation crops before their sowing, in order to improve their germination, growth and yield, as well as alternative preparations of seed dressing against fungal diseases. An interesting direction of the use of laser stimulation is to use this technology in the practice of environmental engineering, primarily to increase the efficiency and effectiveness of biological reclamation of bio- and phytoremediation, enhance biological wastewater treatment, accelerate the growth of energy crops and increase the resistance of plants growing on soils contaminated with heavy metals and saline soils [Śliwka 2007, Śliwka, Jakubiak 2009, Śliwka, Jakubiak 2010, Dobrowolski et al. 2012, Jakubiak 2012]. Research on the proecological use of laser stimulation has already proposed in the 70's of last century by Dobrowolski.

The objective of these experiments was to determine the effect of the laser simulation of selected plant species for their ability to adapt to unfavorable environmental conditions, such as climate and pollution.

METHODS

Selected plant species were chosen as an experimental material: duckweed (*Lemna minor* L.) and yellow iris (*Iris pseudoacorus* L.) used in the wastewater treatment plant and species of grass *Phleum pratense* L. recommended for use in the reclamation treatments: fallow, contaminated land, landfills of waste and ash [Góral 2001].

In the following experiments was used a coherent light source:

- a laser diode emitting light with a wavelength corresponding to red color (660 nm) with a power of 20 mW,
- a laser diode emitting light with a wavelength corresponding to blue color (473 nm) with a power of 20 mW,
- an argon laser emitting light with a wavelength corresponding to celadon color (514 nm) with a power of 21 mW,
- impulse medical laser emitting light with a wavelength corresponding to red color (670 nm) with a power of 20 mW.

The article discusses two preliminary experiments.

Experiment I (2005–2007) was conducted in the field conditions. In this experiment were used duckweed (*Lemna minor* L.) and the yellow iris (*Iris pseudoacorus* L.) as experimental material.

In the case of duckweed were irradiated whole plants. In each experimental group were 90 plants. After this, plants were placed in three ponds with the same volume (160 dm^3) filled with wastewater from a municipal sewage treatment Kraków Płaszów, diluted (1:1) with a marked chemical composition [Śliwka 2007]. Irradiated rhizomes of yellow iris (about the same weight: 400 g) were also placed in each of the ponds.

The rhizomes of yellow iris and duckweed (whole plants) were irradiated using an argon laser (514 nm) and laser diode (660 nm). For irradiation was used parameters of stimulation, which have been selected under laboratory conditions (wavelength, exposure time and method of exposure), in terms of the expected stimulation effect: increased biomass growth, the resistance of plants [Śliwka 2007].

For duckweed were used the following parameters: wavelength 514 nm exposure time 3 times 3 seconds and a wavelength of 660 nm at the time of exposure 3 times 3 seconds. For yellow iris were selected the following parameters: wavelength 514 nm exposure time 3 times 30 seconds and a wavelength of 660 nm at the time of exposure 3 times 30 seconds. In order to compare the results prepared unexposed control groups of plants. Plants were irradiated with perpendicular light beam from a distance of 20 cm.

Experiment II was conducted in laboratory conditions.

Timothy grass (*Phleum pratense* L.) was sown in Petri dishes on the soil collected from the heaps area of KWK Jankowice (0–20 cm depth), contaminated with Cd (38 $mg \cdot kg^{-1}$

d.m.) [Rozporządzenie... 2002] according to the scheme: 30 seeds × 3 repetitions for each of the irradiation algorithm. In the case of timothy grass as an optimal (to increase the capacity of germination, germination rate) on the basis of preliminary experiments were chosen following parameters: wavelength of 473 nm for the second time 3×1 and the wavelength of 670 nm for 3×9 seconds of exposure time. Additionally, three control groups of unexposed plant (30 seeds) as control groups were prepared. Observations continued for 14 days. Plants were irradiated with perpendicular light beam from a distance of 20 cm.

Following parameters were measured: biomass growth and plant condition at the end of each of the three growing seasons (2005–2007) for duckweed (*Lemna minor*) and for yellow iris (*Iris pseudoacorus*) and ability to germinate, germination rate by Pieper method, growth of roots and shoots of *Phleum pratense*.

The programs: Statistica 7.1 and MS Excel were used for statistical evaluation of the results. The Student t-test and ANOVA ranks Kruskal-Wallis were used to assess the statistical significance of the results and also used in the analysis of the test of normality the Shapiro-Wilk.

Discussed below relate to the results of preliminary observations and will be prerequisite for the continuation of these studies.

RESULTS AND DISCUSSION

Results of experiments and preliminary observations confirm the significant impact of the coherent light on biological material. It was found, among other things, a positive and statistically significant effect of laser stimulation on the growth of plant biomass and increasing ecological valence unfavorable environmental factors.

Pot experiments on duckweed (*Lemna minor*) showed a significant effect of laser stimulation on the growth of biomass. As the most optimal algorithm of irradiation (wavelength: 660 nm and exposure time: 3 times 3 seconds), at the end of the first growing season 300% greater increase of biomass in compared to the control group of unirradiated plants was obtained a [Śliwka 2010, Śliwka 2011, Śliwka 2012]. Similar effects were noted in case of *Salix viminalis* [Jakubiak, Gdowska 2013].

It is worth noting that the effect of stimulation was maintained during the following observations in the next growing seasons, which indicates its durability. These conclusions apply only to plant reproducing vegetatively, and the effect of stimulation can carry on the progeny plants. In the case of plants which reproduce generatively, a similar relationship was not found.

Duckweed (*Lamna minor*) exposed to laser diode 660 nm were characterized by a higher resistance to temperature drop, also exhibited a higher viability (in winter) in subsequent years of observation (preliminary observations) (Figure 1).

Significant differences in the condition of individual plants of the experimental groups were found (Figure 2).

Experiments concerning the influence of coherent light on the growth of yellow iris (*Iris pseudoacorus*) showed significant differences in the increase of the biomass of leaves for groups irradiated with argon laser (514 nm) with an exposure time 3 times 30 seconds. These groups of plants were characterized as more abundant and flowered earlier (by observation). For the group irradiated with diode laser (660 mn) re-growth of leaves after collecting the biomass at the end of the vegetation season was found (end of October).

The results of laboratory experiments on the influence of coherent light on seed germination

Figure 1. Comparison of plants condition from each experimental groups of duckweed (*Lemna minor*) after ponds frozen a) control group, b) plants irradiated with laser diode (660 nm), irradiation time: 3 times 3 seconds, c) plants irradiated with argon laser (514 nm), irradiation time: 3 times 3 seconds

Figure 2. Comparison of plants condition from each experimental groups of duckweed (*Lemna minor*) a) control group, b) plants irradiated with laser diode (660 nm), irradiation time: 3 times 3 seconds, c) plants irradiated with argon laser (514 nm), irradiation time: 3 times 3 seconds

of *Phelum pratense* and plants growth in soil contaminated with Cd. Cadmium significantly impairs metabolic processes in plants (photosynthesis, transpiration, metabolism of nitrogen compounds, changes in the permeability of cell membranes and DNA structures).

Differences were found in the germination of plant and the condition of plants from each experimental group. The average length of coleoptile, after 14 days of growth, was 3.4 cm for the plants in the control group, 3.2 cm for the plant irradiated with laser diode (473 nm) and 3.7 cm for plant irradiated with impulse laser (670 nm) (Figure 3).

On the other hand average root length, measured after 14 days of growth, was 2.1 cm for the plants in the control group, 1.6 cm for the plant irradiated with laser diode (473 nm) and 1.8 cm for plant irradiated with impulse laser (670 nm) (Figure 4).

Assessment of growth of coleoptile and roots in the experimental groups of *Phelum pratense* requires repeating with regard to extending the irradiation time to increase the differences in growth of experimental plants.

The germination capacity, calculated after 10 days of duration of the experiment, indicated a beneficial effect the red laser diode (670 nm) for germination capacity (Figure 5), as well as shorten the time of germination (calculated by Pieper) as a result of irradiation of the material with blue laser diode (473 nm) (Figure 6).

The results of preliminary observations showed the impact of coherent light on the germination of plants: increase of germination and shortening of time of germination, which may be crucial for efficiency of soil reclamation. The described experiment requires a continuation, which will allow to confirm the assumed thesis.

Positive effects of laser light stimulation on the morphological and physiological features at the molecular level of other species (kidney bean, cabbage, grass pea, soybean, spring durum wheat, sugar beet) were also observed [Kacharava et al. 2009, Khalifa, El Ghandoor 2011, Prośba-

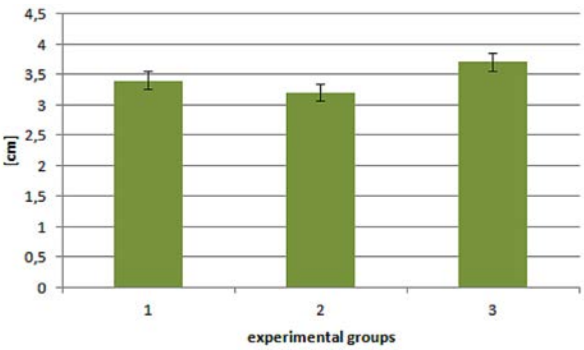

Figure 3. Comparison of growth of coleoptile [cm] *Phelum pratense* in experimental groups: 1 – control group, 2 – plants irradiated with laser diode (473 nm), 3 – plants irradiated with impuls laser (670 nm) (on the graph was marked bars bugs with the standard error)

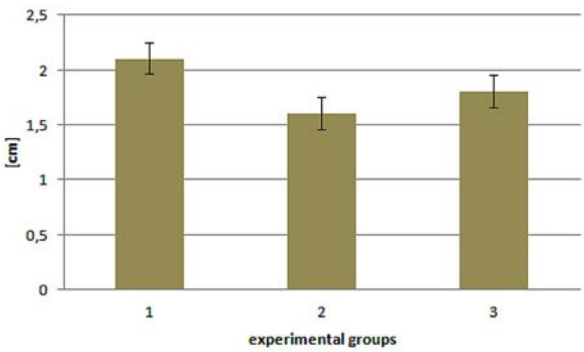

Figure 4. Comparison of growth of roots [cm] *Phelum pratense* in experimental groups: 1 – control group, 2 – plants irradiated with laser diode (473 nm), 3 – plants irradiated with impuls laser (670 nm) (on the graph was marked bars bugs with the standard error)

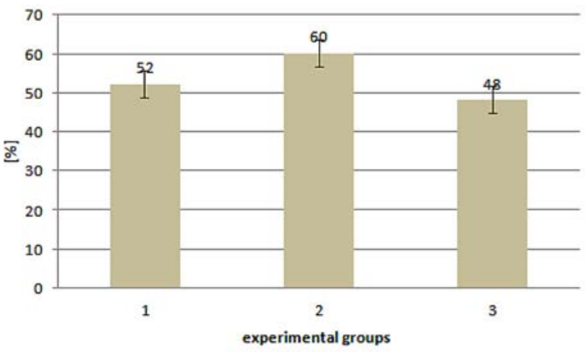

Figure 5. Comparison of ability of germination of *Phelum pratense* in experimental groups: 1 – control group, 2 – plants irradiated with laser diode (473 nm), 3 – plants irradiated with impuls laser (670 nm) (on the graph was marked bars bugs with the standard error)

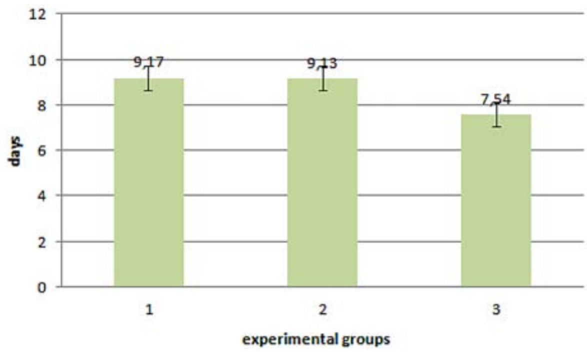

Figure 6. Comparison of germination rate of *Phelum pratense* in experimental groups: 1 – control group, 2 – plants irradiated with laser diode (473 nm), 3 – plants irradiated with impuls laser (670 nm) (on the graph was marked bars bugs with the standard error)

Białczyk et al. 2013, Ritambhara, Girjesh 2013, Zare et al. 2014]. Irradiation of sugar beet seeds resulted in significantly bigger length of the hypocotyl and root of seedlings, in a higher dry matter, chlorophylls and carotenoids content in leaves and roots during vegetation than in non-stimulated seeds, as well as favourably modified the chlorophyll *a* to *b* ratio. It also significantly increased yield and the saccharose content, and reduced sodium and potassium content [Prośba-Białczyk et al. 2013]. Changes of the antioxidants content in leaves of kidney bean, cabbage and beet was observed as well [Kacharava et al. 2009]. Biostimulation may be used for enhancing productivity of the *Lathyrus sativus* L. plants [Ritambhara, Girjesh 2013]. Pre-sowing laser biostimulation of seeds caused increase crops of maize by 10–20%, spring wheat by 20–30%, spring barley by 20-25% and sugar beets by 10–35% [Koper 1994]. The additional effects are stronger plant seedlings, higher resistance to cold, shortening the vegetation period of corn by about 10 days, earlier plant maturation, the better plants quality and possible diminishing of nitrogen fertilizers application without essential crops decrease, as well as changes in protein banding patterns and large subunit of Rubis CO [Koper 1994, Khalifa, El Ghandoor 2011]. Laser irradiation of spring durum wheat (*Triticum turgidum* L.) under salinity stress induced considerable biological changes in plant metabolism e.g. significantly increases free proline accumulation in leaves and salt tolerance [Zare et al. 2014], and may induce an increase of plants resistance to environmental pollutions and unfavorable environmental factors, possibility of higher efficient of sewage treatment and soil reclamation processes and increase of bioremediation abilities as well [Jakubiak, Gdowska 2013]. The results of the discussed experiments allow to conclude that it is possible to increase plant resistance to unfavorable environmental conditions e.g. a low temperature. Irradiation of plants with a coherent light source may significantly extended the growing season of plants (like *Lemna minor, Iris pseudoacorus*) used in hydrobotanical treatment plants [Śliwka 2005, Śliwka, Jakubiak 2007].

The observed effect of laser stimulation on the ability of germination and germinate rate of *Phelum pratense*, may contribute to an increase of biomass growth, and thus, the efficiency of biological remediation of soils contaminated with heavy metals, by increasing the resistance of plants to pollutants that cause phototoxic effects.

Researches carried out by Dobrowolski et al. [1995] and Zielinska-Loek [2003], related to the assessment of the impact of coherent light on different species of willows (*Salix viminalis, Salix acutifolia* and *Salix varieties* Rapp), showed a significant effect of stimulation on accelerate the growth and increase of plants resistance cultivated on lands belonging to ZGH – Mining and Metallurgical Factory "Bolesław" in Bukowno, Cement Plant "Chełm" and in the areas of express roads.

CONCLUSION

The implementation of laser stimulation of plants used in phytoremediation technologies (reclamation, wastewater treatment plants), should significantly contribute to increasing the efficiency of biological reclamation and revitalization of degraded areas.

Acknowledgments

The research was prepared within the statutory research for the Faculty of Mining and Geoengineering AGH-UST, Department of Environmental Engineering and Mineral Processing No. 11.11.100.482 (2013).

REFERENCES

1. Aladjadjiyan A. 2007. The use of physical methods for plant growing stimulation in Bulgaria. J. Cent. Eur. Agric., 8(3): 369–380.
2. Bryszewska M., Leyko W. 1997. Biofizyka dla biologów. Wyd. PWN Warszawa.
3. Ćwintal M. 2010. Wpływ zapraw nasiennych i stymulacji laserowej nasion na wschody oraz strukturę łanu i plonowanie koniczyny czerwonej w roku siewu. Ann. UMCS, Sec. EE, LXV(4): 84–93.
4. Dobrowolski J.W. 2002. Zastosowanie biostymulacji laserowej w ekoinżynierii i ekorozwoju. Inż. Ekol., 6: 194–196.
5. Dobrowolski J.W., Różanowski B. 1998. The influence of laser light on accumulation of selected macro, trace and ultra trace elements by some plants. Microelements and trace elements. [In:] Mengen- und Spurenelemente, Arbeitstagung 18, Verlag Harald Schubert, Leipzig, 147–156.
6. Dobrowolski J.W., Różanowski B., Zielińska A., Budzyński M., Walczak P. 1995. Próby zastosowania biostymulacji laserowej w celu przyspieszenia wzrostu niektórych gatunków roślin i rekultywacji terenów silnie skażonych. II Kraj. Konf. Nauk. Las-Drewno-Ekologia, 20–22 czerwca, PAN, Poznań, 25–29.
7. Dobrowolski J.W., Śliwka M., Mazur R. 2012. Laser biotechnology for more efficient bioremediation, protection of aquatic ecosystems and reclamation of contaminated areas. J. Chem. Technol. Biot. B, 87(9): 1354–1359.
8. Drozd D., Szajsner H. 2001. Promienie lasera jako czynnik fizyczny stymulujący wartość użytkową nasion. Acta Agrophys., 58: 71–79.
9. Drozd D., Szajsner H., Bielecki K. 2003. Wpływ światła lasera na aktywność alfa-amylazy w ziarniakach różnych genotypów pszenżyta. Biul. IHAR, 226/227(1): 177–180.
10. Friedman H., Lubart R. 1993. Nonlinear photostimulation: The mechanism of visible and infrared laser-induced stimulation and reduction of neural excitability and growth. Laser Therapy, 5(1): 30–42.
11. Góral S. 2001. Roślinność zielna w ochronie i rekultywacji gruntów. Inż. Ekol., 3: 161–178.
12. Gosh S., Bupp S. 1992. Stimulation of biological uptake of heavy metals. Water Sci. Technol., 26: 227–236.
13. Inyushin W.M. 1977. Laser technology in agricultural services. An attempt of using a new optic-quantum generator. Nowe Rolnictwo, 21/22: 21–26 (in Polish).
14. Inyushin W.M., Iliasov G.U., Fedorowa N.N. 1981. Laser light and crop. Kainar Publication, Ałma-Ata. 1981, 210: 165–168 (in Russian).
15. Jakubiak M. 2012. Degraded lands biological reclamation using modern methods of stimulating the growth of selected varieties of willows (*Salix* sp.). Pol. J. Environ. Stud., 21(5A): 129–133.
16. Jakubiak M., Gdowska K. 2013. Innovative envirommental technology applications of laser light stimulation. En***e***r***g***etika ***i a***wt***o***m***a***tik***a***, 3: 14–21.
17. Kabata-Pendias A., Pendias H. 1999. Biogeochemia pierwiastków śladowych. Wyd. PWN, Warszawa.
18. Kacharava N., Chanishvili S., Badridze G., Chkhubianishvili E., Janukashvili N. 2009. Effect of seed irradiation on the content of antioxidants in leaves of kidney bean, cabbage and beet cultivars. Australian J. Crop Sci., 3(3): 137–145.
19. Karlander E.P., Krauss R.W. 1968. The laser as a light source for the photosynthesis and growth of *Chlorella vannielii*. Biochim. Biophys. Acta, 153(1): 312–314.
20. Khalifa N.S., El Ghandoor H. 2011. Investigate the effect of Nd-Yag Laser Beam on soybean (*Glycine max*) leaves at the protein level. Intern. J. Biology, 3(2): 135–144.
21. Kopcewicz J., Lewak S. 2007. Fizjologa roślin. PWN, Warszawa.
22. Koper R. 1994. Pre-sowing laser biostymulation of seeds of cultivated plants and its results in agrotechnics. Int. Agrophys., 8(4): 593–596.
23. Prośba-Białczyk U., Szajsner H., Grzyś E., Demczuk A., Sacała E., Bąk K. 2013. Effect of seed stimulation on germination and sugar beet yield. Int. Agrophys., 27: 195–201.
24. Ritambhara S., Girjesh K. 2013. Biostimulating effect of laser beam on the cytomorphological aspects of *Lathyrus sativus* L. Annals Plant Sci., 02(05): 141–148.
25. Rozporządzenie Ministra Środowiska z dnia 9 września 2002 r. w sprawie standardów jakości gleby oraz standardów jakości ziemi, które określa dopuszczalne wartości stężeń zanieczyszczeń w glebie lub ziemi. Dz.U. 2002, Nr 165, poz. 1359.
26. Śliwka M. 2005. Wykorzystanie biostymulacji laserowej roślin do zwiększenia przyrostu ich biomasy oraz zdolności bioremediacyjnych. [In:] Obieg pierwiastków w przyrodzie, Monografia, 3: 637–640.
27. Śliwka M. 2007. Zastosowanie stymulacji laserowej wybranych gatunków hydrofitów do zwiększenia ich zdolności bioremediacyjnych. Roz-

prawa doktorska, Wydz. GGiIŚ, AGH Kraków, 242 p. (online: http://winntbg.bg.agh.edu.pl/rozprawy/9951/full9951.pdf)

28. Śliwka M., Jakubiak M. 2007. Wpływ stymulacji laserowej na zwiększenie przyrostu biomasy oraz zdolności bioremediacyjnych roślin wykorzystywanych w hydrofitowych oczyszczalniach ścieków. Ochrona Środowiska i Zasobów Naturalnych, 33: 103–107.
29. Śliwka M., Jakubiak M. 2009. The application of the innovative biotechnology in hydrobotanical wastewater treatment plants. Pol. J. Environ. Stud., 18(3A): 445–449.
30. Śliwka M., Jakubiak M. 2010. Application of laser biotechnology for more efficient phytoremediation of biogenic elements. Ecol. Chem. Eng. A, 17(2–3): 297–303.
31. Śliwka M., Jakubiak M. 2010. Application of laser stimulation of some hydrophytes species for more efficient biogenic elements phytoremediation. Proceedings of Ecopole, 4(1): 205–211.
32. Szajsner H., Drozd D., Błażewicz J. 2008. Wpływ stymulacji laserowej nasion na kształtowanie cech morfologicznych siewek i wielkość siły diastatycznej form żyta i pszenżyta. Biuletyn Instytutu Hodowli i Aklimatyzacji Roślin, 250: 177–187.
33. Wilczek M., Ćwintal M. 2009. Ocena możliwości poprawy wartości siewnej nasion koniczyny czerwonej poprzez zastosowanie przedsiewnej stymulacji laserowej. Acta Agrophys., 14(1): 221–231.
34. Wilczek M., Ćwintal M., Kornas-Czuczwar B., Koper R. 2006. Wpływ laserowej stymulacji nasion na plonowanie di- i tetraploidalnej koniczyny czerwonej w roku siewu. Acta Agrophys., 8(2): 527–536.
35. Zare N., Sadat Noori S.A., Kholgh Sima N.A.K., Mortazavian S.M.M. 2014. Effect of laser priming on accumulation of free proline in spring durum wheat (*Triticum turgidum* L.) under salinity stress. Int. Trans. J. Eng. Manag. Sci. Tech. 5(2): 119-130.
36. Zielińska-Loek A. 2002. Ocena możliwości wykorzystania fotostymulacji laserowej roślin do proekologicznego zagospodarowania rejonów dróg. Praca doktorska, Wydz. GGiIŚ, AGH Kraków, 173 p.