

Abstracts of selected publications of Assoc. Prof. Anelia G. Dobrikova
participation in the competition for occupying the academic position of
Professor

1. Dobrikova A., Apostolova E., Hanć A., Yotsova E., Borisova P., Sperdouli I., Adamakis I.D.S., Moustakas M. Cadmium toxicity in *Salvia sclarea* L: An integrative response of element uptake, oxidative stress markers, leaf structure and photosynthesis. *Ecotoxicol. Environ. Saf.* 209 (2021) 111851. doi:10.1016/j.ecoenv.2020.111851 IF-4.872 (Q1 WebS)

The herbal plant *Salvia sclarea* L. (clary sage) is classified to cadmium (Cd) accumulators and considered as a potential plant for phytoremediation of heavy metal polluted soil. However, the effect of Cd only treatment on the function of the photosynthetic apparatus of *S. sclarea*, as well as the mechanisms involved in Cd tolerance have not yet been studied in detail. This study was conducted to examine the integrative responses of *S. sclarea* plants exposed to a high Cd supply (100 µM) for 3 and 8 days by investigating element nutrient uptake, oxidative stress markers, pigment composition, photosynthetic performance and leaf structure. Measurements of the functional activities of photosystem I (PSI, by P700 photooxidation), photosystem II (PSII, by chlorophyll fluorescence parameters), the oxygen-evolving complex (oxygen evolution by Joliot- and Clark-type electrodes), as well as the leaf pigment and phenolic contents, were used to evaluate the protective mechanisms of the photosynthetic apparatus under Cd stress. Data suggested that the molecular mechanisms included in the photosynthetic tolerance to Cd toxicity involve strongly increased phenolic and anthocyanin contents, as well as an increased non-photochemical quenching and accelerated cyclic electron transport around PSI up to 61%, which protect the function of the photosynthetic apparatus under stress. Furthermore, the tolerance of *S. sclarea* to Cd stress is also associated with increased accumulation of Fe in leaves by 25%. All the above, clearly suggest that *S. sclarea* plants employ several different mechanisms to protect the function of the photosynthetic apparatus against Cd stress, which are discussed here.

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2. Adamakis I.-D.S., Sperdouli I., Hanć A., Dobrikova A., Apostolova E., Moustakas M. (2020) Rapid hormetic responses of photosystem II photochemistry of clary sage to cadmium exposure. *Int. J. Mol. Sci.* 22(1) (2021) 41. doi:10.3390/ijms22010041 IF-4.556 (Q1 WebS)

Five-day exposure of clary sage (*Salvia sclarea* L.) to 100 µM cadmium (Cd) in hydroponics was sufficient to increase Cd concentrations significantly in roots and aboveground parts and affect negatively whole plant levels of calcium (Ca) and magnesium (Mg), since Cd competes for Ca channels, while reduced Mg concentrations are associated with increased Cd tolerance. Total zinc (Zn), copper (Cu), and iron (Fe) uptake increased but their translocation to the aboveground parts decreased. Despite the substantial levels of Cd in leaves, without any observed defects on chloroplast ultrastructure, an enhanced photosystem II (PSII) efficiency was observed, with a higher fraction of absorbed light energy to be directed to photochemistry (Φ PSII). The concomitant increase in the photoprotective mechanism of non-photochemical quenching of photosynthesis (NPQ) resulted in an important decrease in the dissipated non-regulated energy (Φ NO), modifying the homeostasis of reactive oxygen species (ROS), through a decreased singlet oxygen (1O_2) formation. A basal ROS level was detected in control plant leaves for optimal growth, while a low increased level of ROS under 5 days Cd exposure seemed to be beneficial for triggering defense responses, and a high level of ROS out of the boundaries (8 days Cd exposure), was harmful to plants. Thus, when clary sage was exposed to Cd for a short period, tolerance mechanisms were triggered. However, exposure to a combination of Cd and high light or to Cd alone (8 days) resulted in an inhibition of PSII functionality, indicating Cd toxicity. Thus, the rapid activation of PSII functionality at short time exposure and the inhibition at longer duration

suggests a hormetic response and describes these effects in terms of “adaptive response” and “toxicity”, respectively.

- 3. Dobrikova A., Apostolova E., Hanč A., Yotsova E., Borisova P., Sperdouli I., Adamakis I.D.S., Moustakas M. Tolerance mechanisms of the aromatic and medicinal plant *Salvia sclarea* L. to excess zinc. *Plants* 10 (2021) 194. doi: 10.3390/plants10020194 IF-2.762 (Q1 WebS)**

In recent years, due to the development of industrial and agricultural production, heavy metal contamination has attracted increasing attention. Aromatic and medicinal plant *Salvia sclarea* L. (clary sage) is classified to zinc (Zn) accumulators and considered as a potential plant for the phytoremediation of heavy metal polluted soils. In this study, an adaptation of clary sage to 900 μM (excess) Zn exposure for eight days in a hydroponic culture was investigated. The tolerance mechanisms under excess Zn exposure were assessed by evaluating changes in the nutrient uptake, leaf pigment and phenolic content, photosynthetic activity and leaf structural characteristics. The uptake and the distribution of Zn, as well as some essential elements such as: Ca, Mg, Fe, Mn and Cu, were examined by inductively coupled plasma mass spectrometry. The results revealed that *Salvia sclarea* is a Zn-accumulator plant that tolerates significantly high toxic levels of Zn in the leaves by increasing the leaf contents of Fe, Ca and Mn ions to protect the photosynthetic function and to stimulate the photosystem I (PSI) and photosystem II (PSII) activities. The exposure of clary sage to excess Zn significantly increased the synthesis of total phenolics and anthocyanins in the leaves; these play an important role in Zn detoxification and protection against oxidative stress. The lipid peroxidation and electrolyte leakage in leaves, used as clear indicators for heavy metal damage, were slightly increased. All these data highlight that *Salvia sclarea* is an economically interesting plant for the phytoextraction and/or phytostabilization of Zn-contaminated soils.

- 4. Yotsova E., Dobrikova A., Stefanov M., Misheva S., Bardáčová M., Matušíková I., Žideková L., Blehová A., Apostolova E. Effects of cadmium on two wheat cultivars depending on different nitrogen supply. *Plant Physiol. Biochem.* 155 (2020) 789-799. doi:10.1016/j.plaphy.2020.06.042. IF- 3.720 (Q1 WebS)**

Heavy metal pollution as well as improper fertilization management represent serious threats to a clean environment and healthy food. This study was conducted to investigate how nitrogen supply influences a plant's ability to cope with cadmium stress in the two wheat cultivars – the modern cv. Katya (carrier of the semidwarfing gene *Rht8*) and the old cv. Slomer. Here we examined the effects of 100 μM CdCl_2 on both wheat genotypes grown hydroponically under three different nutrition regimes of 5.5, 10 and 20 mM NO_3^- by investigating plant growth, pigment content and the functional activity of the photosynthetic apparatus through a combination of PAM chlorophyll fluorescence, P700 photooxidation, oxygen evolution and oxidative stress markers. Data showed that the different genetic background affects the different strategies for metal uptake and allocation, as well as abilities to deal with oxidative stress. The modern cv. Katya restricts the entry of the metal to the roots, but allows its translocation to the shoots. Nevertheless, the photosynthetic performance indicated better protection, possibly mediated by the *Rht8* allele. In contrast, the old cv. Slomer tolerates higher cadmium levels in roots and possesses efficient barriers against its transfer to the shoots, but still showed more impaired photosynthetic activity. In general, the impact of cadmium on the photosynthetic apparatus was most deleterious under the lowest nitrogen concentration which was applied, while the highest nitrogen supply alleviated the negative effects of cadmium. The data suggest that the modern breeding allele (*Rht8*), as well as a better nutrition might contribute to the tolerance to heavy metal stress in the wheat.

5. Moustakas M., Hanč A., Dobrikova A., Sperdouli I., Adamakis I.D.S., Apostolova E. **Spatial heterogeneity of cadmium effects on *Salvia sclarea* leaves revealed by Chlorophyll fluorescence imaging analysis and Laser ablation inductively coupled plasma mass spectrometry.** *Materials (MDPI)* 12(18) (2019) 2953. doi:10.3390/ma12182953. IF- 3.057 (Q2 WebS)

In this study, for a first time (according to our knowledge), we couple the methodologies of chlorophyll fluorescence imaging analysis (CF-IA) and laser ablation inductively coupled plasma mass spectrometry (LA-ICP-MS), in order to investigate the effects of cadmium (Cd) accumulation on photosystem II (PSII) photochemistry. We used as plant material *Salvia sclarea* that grew hydroponically with or without (control) 100 μM Cd for five days. The spatial heterogeneity of a decreased effective quantum yield of electron transport (ΦPSII) that was observed after exposure to Cd was linked to the spatial pattern of high Cd accumulation. However, the high increase of non-photochemical quenching (NPQ), at the leaf part with the high Cd accumulation, resulted in the decrease of the quantum yield of non-regulated energy loss (ΦNO) even more than that of control leaves. Thus, *S. sclarea* leaves exposed to 100 μM Cd exhibited lower reactive oxygen species (ROS) production as singlet oxygen ($^1\text{O}_2$). In addition, the increased photoprotective heat dissipation (NPQ) in the whole leaf under Cd exposure was sufficient enough to retain the same fraction of open reaction centers (qp) with control leaves. Our results demonstrated that CF-IA and LA-ICP-MS could be successfully combined to monitor heavy metal effects and plant tolerance mechanisms.

6. Maglovski M., Gregorová Z., Rybanský E., Bardáčová M., Moravčíková J., Bujdoš M., Dobrikova A., Apostolova E., Kraic J., Blehová A., Matušíková I. **Effects of nutrition on wheat photosynthetic pigment responses to arsenic stress.** *Polish J. Environ. Stud.* 28 (3) (2019) 1-9. doi:10.15244/pjoes/89584. ISI IF-1.383 (Q2 SJR)

Arsenic is a serious soil pollutant with toxic effects on biological systems. Elevated soil concentrations may negatively affect crop production and food safety. The impact of arsenic on plants depends on many factors, including nitrogen availability. Nitrogen (N) as an essential mineral affects overall energetics of plants, while its non-optimal doses have been shown to also impact plant performance and yield, as well as tolerance to environmental constraints. The combined effects of these two factors, however, have been rarely studied. Here we investigated the impact of sublethal doses of As^{3+} (5 mM) on wheat plants grown in hydropony, applying a set of 8 different N concentrations spanning from starvation (0 mM N in the media) through optimum (7.5 mM N) to excessive amounts (up to 35 mM N). The results showed that the content of photosynthetic pigments varies depending on N concentration and As^{3+} presence. The different energetic status of plants also affected the final As uptake. Establishing nutrition conditions might be important for limiting metal(loid) uptake from soil in contaminated areas.

7. Yotsova E., Dobrikova A., Stefanov M., Apostolova E. **Impact of salicylic acid on the growth and the activity of photosynthetic apparatus in rice under non-stress conditions.** *Comp. Rend. Acad. Bulg. Sci.* 71(3) (2018) 368-375. doi:10.7546/CRABS.2018.03.09. ISI IF-0.321 (Q2 SJR)

The impact of exogenous application of different concentrations of salicylic acid (10, 50 and 100 μM) through the rooting medium on the plant growth, the pigment content and the photochemical activities of both photosystem I and photosystem II was investigated. Data revealed that the observed alterations strongly depend on the concentration of applied salicylic acid, as 10 μM is the optimal concentration for the growth and the functional activity of photosynthetic apparatus of rice plants under non-stress conditions. In addition, the concentrations

of salicylic acid lower than 100 μM had no effect on the energy transfer between the chlorophyll-protein complexes in thylakoid membranes.

8. Yotsova E.K., Dobrikova A.G., Stefanov M.A., Kouzmanova M., Apostolova E.L. Improvement of the rice photosynthetic apparatus defence under cadmium stress modulated by salicylic acid supply to roots. *Theor. Exp. Plant Physiol.* 30(1) (2018) 57-70. doi:10.1007/s40626-018-0102-9. ISI IF- 1.532 (Q2 WebS)

The present study was conducted to investigate the effect of exogenous salicylic acid (SA) added to the nutrient solution on the growth parameters and the functions of the photosynthetic apparatus of rice plants under cadmium (Cd) stress. Our investigations have shown that 10 μM SA has an optimal effect in rice plants grown hydroponically. Pulse amplitude modulated chlorophyll fluorescence, low-temperature chlorophyll fluorescence, oxygen evolution (measured with Clark-type and Joliot-type electrodes) and P700 photo-oxidation measurements were carried out to assess the effect of SA on the activity of the photosynthetic apparatus. The levels of three important parameters associated with oxidative stress (hydrogen peroxide, lipid peroxidation and proline content) were measured. The application of low concentration of SA significantly decreased the levels of hydrogen peroxide, lipid peroxidation and proline under Cd stress. The results revealed that low concentration of SA, applied in plants exposed to 150 μM CdCl₂, significantly improves plant growth, photochemical activities of both photosystems, the electron flow from QA to plastoquinone, energetic distribution between pigment-protein complexes and the kinetic parameters of oxygen-evolving reactions. This study suggests that exogenous application of 10 μM SA through the rooting medium has a protective effect against Cd toxicity in rice plants. The possible molecular mechanisms involved in the defence effect of SA on the function of photosynthetic apparatus are discussed.

9. Jusovic M., Velitchkova M.Y., Misheva S.P., Börner A., Apostolova E.L., Dobrikova A.G. Photosynthetic responses of a wheat mutant (*Rht-B1c*) with altered DELLA proteins to salt stress. *J. Plant Growth Regul.* 37(2) (2018) 645-656. doi:10.1007/s00344-017-9764-9 ISI IF-2.179 (Q1 SJR)

Salinity increases in the world's land area and significantly affects the rate of photosynthesis and corresponding plant growth. In this study, the impact of salt stress (200 mM NaCl equivalent to an electrical conductivity of 18.6 mS cm⁻¹) on the photosynthetic apparatus and some growth parameters were investigated in wheat DELLA mutant (*Rht-B1c*) and wild-type (*Rht-B1a*) seedlings grown on a half-strength Hoagland solution. Results revealed that salt toxicity was alleviated in the *Rht-B1c* mutant compared to the *Rht-B1a* wild type, as manifested by less-reduced leaf pigment content, relative water content, and photochemical activity of photosystem II (PSII) and photosystem I (PSI) after a 9-day salt exposure of plants. Compared to the wildtype wheat, a higher capacity for PSI-dependent cyclic electron flow, preventing the photosynthetic apparatus from oxidative damage, was observed in the mutant plants before and after salt treatment. In addition, an increase of PsaB proteins was detected in the mutant plants after long-term salt stress unlike the wild type. The observed higher oxidation level of P700 (P700⁺) in the mutant was consistent with higher abundance of PSI-related protein complexes. The data demonstrated that alterations in thylakoid membrane proteins and/or their structural reorganization in wheat DELLA mutant (*Rht-B1c*) significantly contribute to the alleviation of salt-induced damage of the photosynthetic apparatus. Molecular mechanisms involved in the photosynthetic responses of wheat DELLA mutants to salt stress are discussed.

10. Dobrikova A.G., Yotsova E.K., Börner A., Landjeva S.P., Apostolova E.L. The wheat mutant DELLA-encoding gene (*Rht-B1c*) affects plant photosynthetic responses to cadmium stress. *Plant Physiol. Biochem.* 114 (2017) 10-18. doi: 10.1016/j.plaphy.2017.02.015. ISI IF- 2.718 (Q1 WebS)

The sensitivity to cadmium (Cd) stress of two near-isogenic wheat lines with differences at the *Rht-B1* locus, *Rht-B1a* (tall wild type, encoding DELLA proteins) and *Rht-B1c* (dwarf mutant, encoding modified DELLA proteins), was investigated. The effects of 100 μ M CdCl₂ on plant growth, pigment content and functional activity of the photosynthetic apparatus of wheat seedlings grown on a nutrient solution were evaluated through a combination of PAM chlorophyll fluorescence, oxygen evolution, oxidation-reduction kinetics of P700 and 77K fluorescence. The results showed that the wheat mutant (*Rht-B1c*) was more tolerant to Cd stress compared to the wild type (*Rht-B1a*), as evidenced by the lower reductions in plant growth and pigment content, lower inhibition of photosystem I (PSI) and photosystem II (PSII) photochemistry and of the oxygen evolution measured with Clark-type and Joliot-type electrodes. Furthermore, the enhanced Cd tolerance was accompanied by increased Cd accumulation within mutant plant tissues. The molecular mechanisms through which the *Rht-B1c* mutation improves plant tolerance to Cd stress involve structural alterations in the mutant photosynthetic membranes leading to better protection of the Mn cluster of oxygen-evolving complex and increased capacity for PSI cyclic electron transport, protecting photochemical activity of the photosynthetic apparatus under stress. This study suggests a role for the *Rht-B1c*-encoded DELLA proteins in protective mechanisms and tolerance of the photosynthetic apparatus in wheat plants exposed to heavy metals stress.

11. Yotsova E.K., Stefanov M.A., Dobrikova A.G., Apostolova E.L. Different sensitivities of photosystem II in green algae and cyanobacteria to phenylurea and phenol-type herbicides: effect on electron donor side. *Zeitschrift für Naturforschung C* 72(7-8) (2017) 315-324. doi: 10.1515/znc-2016-0089. ISI IF- 0.882 (Q3 SJR)

The effects of short-term treatment with phenylurea (DCMU, isoproturon) and phenol-type (ioxynil) herbicides on the green alga *Chlorella kessleri* and the cyanobacterium *Synechocystis salina* with different organizations of photosystem II (PSII) were investigated using pulse amplitude modulated (PAM) chlorophyll fluorescence and photosynthetic oxygen evolution measured by polarographic oxygen electrodes (Clark-type and Joliot-type). The photosynthetic oxygen evolution showed stronger inhibition than the PSII photochemistry. The effects of the studied herbicides on both algal and cyanobacterial cells decreased in the following order: DCMU > isoproturon > ioxynil. Furthermore, we observed that the number of blocked PSII centers increased significantly after DCMU treatment (204–250 times) and slightly after ioxynil treatment (19–35 times) in comparison with the control cells. This study suggests that the herbicides affect not only the acceptor side but also the donor side of PSII by modifications of the Mn cluster of the oxygen-evolving complex. We propose that one of the reasons for the different PSII inhibitions caused by herbicides is their influence, in different extents, on the kinetic parameters of the oxygen-evolving reactions (the initial S₀ – S₁ state distribution, the number of blocked centers SB, the turnover time of S_i states, misses and double hits). The relationship between the herbicide-induced inhibition and the changes in the kinetic parameters is discussed.

12. Dobrikova A.G., Apostolova E.L. Damage and protection of the photosynthetic apparatus from UV-B radiation. II. Effect of quercetin at different pH. *J. Plant Physiol.* 184 (2015) 98-105. doi:10.1016/j.jplph.2015.06.008. ISI IF- 2.971 (Q1 WebS)

The effect of the exogenously added quercetin against the UV-B inhibition of the photosystem II (PSII) functions in isolated pea thylakoid membranes suspended at different pH of the medium (6.5, 7.6 and 8.4) was investigated. The data revealed that the interaction of this flavonoid with the membranes depends on the pH and influences the initial S₀–S₁ state distribution of PSII in

the dark, the energy transfer between pigment-protein complexes of the photosynthetic apparatus and the membrane fluidity. Quercetin also displays a different UV-protective effect depending on its location in the membranes, as the effect is more pronounced at pH 8.4 when it is located at the membrane surface. The results suggest that quercetin induces structural changes in thylakoid membranes, one of the possible reasons for its protection of the photosynthetic apparatus.

- 13. Dobrikova A., Apostolova E. Protective effects of naringin on the photosynthetic apparatus against UV-B radiation. *Comp. Rend. Acad. Bulg. Sci.* 67 (5) (2014) 675-682. ISI IF - 0.284 (Q3 SJR)**

The effects of the flavonoid naringin against UV-B induced changes in the oxygen evolution and the energy transfer between pigment-protein complexes of the photosynthetic apparatus were studied. The exogenous application of naringin to thylakoid membranes results in a modification of the oxygen-evolving complex by influence on the S₀-S₁ state distribution in darkness, as well as influences the energy redistribution between the two photosystems during UV-B irradiation. Data also reveal that naringin diminishes UV-induced impairment of both the acceptor and the donor side of photosystem II, as the defence effect is more pronounced on the donor side.

- 14. Misra A.N., Vladkova R., Singh R., Misra M., Dobrikova A.G., Apostolova E.L. Action and target sites of nitric oxide in chloroplasts. Review. *Nitric Oxide* 39 (2014) 35-45. doi:10.1016/j.niox.2014.04.003. ISI IF-3.521 (Q2 WebS)**

Nitric oxide (NO) is an important signalling molecule in plants under physiological and stress conditions. Here we review the influence of NO on chloroplasts which can be directly induced by interaction with the photosynthetic apparatus by influencing photophosphorylation, electron transport activity and oxidoreduction state of the Mn clusters of the oxygen-evolving complex or by changes in gene expression. The influence of NO-induced changes in the photosynthetic apparatus on its functions and sensitivity to stress factors are discussed.

- 15. Dobrikova A.G., Vladkova R.S., Rashkov G.D., Todinova S.J., Krumova S.B., Apostolova E.L. Effects of exogenous 24-epibrassinolide on the photosynthetic membranes under non-stress conditions. *Plant Physiol. Biochem.* 80 (2014) 75-82. doi:org/10.1016/j.plaphy.2014.03.022. ISI IF- 2.756 (Q1 WebS)**

In the present work the effects of exogenous 24-epibrassinolide (EBR) on functional and structural characteristics of the thylakoid membranes under non-stress conditions were evaluated 48 h after spraying of pea plants with different concentrations of EBR (0.01, 0.1 and 1.0 mg.L⁻¹). The results show that the application of 0.1 mg.L⁻¹ EBR has the most pronounced effect on the studied characteristics of the photosynthetic membranes. The observed changes in 540 nm light scattering and in the calorimetric transitions suggest alterations in the structural organization of the thylakoid membranes after EBR treatment, which in turn influence the kinetics of oxygen evolution, accelerate the electron transport rate, increase the effective quantum yield of photosystem II and the photochemical quenching. The EBR-induced changes in the photosynthetic membranes are most probably involved in the stress tolerance of plants.

- 16. Dobrikova A.G., V. Krasteva, E.L. Apostolova, Damage and protection of the photosynthetic apparatus from UV-B radiation. I. Effect of ascorbate. *J. Plant Physiol.* 170 (3) (2013) 251-257. doi:10.1016/j.jplph.2012.10.002. IF- 2.770 (Q1 WebS)**

In this work, the effect of the exogenously added ascorbate (Asc) against the UV-B inhibition of the photosystem II (PSII) functions in isolated pea thylakoid membranes was studied. The results reveal that Asc decreases the UV-B induced damage of the donor and the acceptor side of

PSII during short treatment up to 60 min. The exogenous Asc exhibits a different UV-protective effect on PSII centers in grana and stroma lamellae, as the effect is more pronounced on the PSII β centers in comparison to PSII α centers. Data also suggest that one of the possible protective roles of the Asc in photosynthetic membranes is the modification of the oxygen-evolving complex by influence on the initial S₀–S₁ state distribution in the dark.

- 17. Dobrikova A.G., Domonkos I., Sözer Ö., Laczkó-Dobos H., Kis M., Párducz Á., Gombos Z., Apostolova E.L. Effect of partial or complete elimination of light-harvesting complexes on the surface electric properties and the functions of cyanobacterial photosynthetic membranes. *Physiologia Plantarum* 147 (2) (2013) 248-260. doi: 10.1111/j.1399-3054.2012.01648.x. IF- 3.262 (Q1 WebS)**

Influence of the modification of the cyanobacterial light-harvesting complex [i.e. phycobilisomes (PBS)] on the surface electric properties and the functions of photosynthetic membranes was investigated. We used four PBS mutant strains of *Synechocystis* sp. PCC6803 as follows: PAL (PBS-less), CK (phycocyanin-less), BE (PSII-PBS-less) and PSI-less/apcE⁻ (PSI-less with detached PBS). Modifications of the PBS content lead to changes in the cell morphology and surface electric properties of the thylakoid membranes as well as in their functions, such as photosynthetic oxygen-evolving activity, P700 kinetics and energy transfer between the pigment–protein complexes. Data reveal that the complete elimination of PBS in the PAL mutant causes a slight decrease in the electric dipole moments of the thylakoid membranes, whereas significant perturbations of the surface charges were registered in the membranes without assembled PBS–PSII macrocomplex (BE mutant) or PSI complex (PSI-less mutant). These observations correlate with the detected alterations in the membrane structural organization. Using a polarographic oxygen rate electrode, we showed that the ratio of the fast to the slow oxygen-evolving PSII centers depends on the partial or complete elimination of light-harvesting complexes, as the slow operating PSII centers dominate in the PBS-less mutant and in the mutant with detached PBS.

- 18. Rashkov G.D., Dobrikova A.G., Pouneva I.D., Misra A.N., Apostolova E.L. Sensitivity of *Chlorella vulgaris* to herbicides. Possibility of using it as a biological receptor in biosensors. *Sensors & Actuators: B* 161 (1) (2012) 151-155. doi: 10.1016/j.snb.2011.09.088. ISI IF- 3.535 (Q1 WebS)**

In the present study the sensitivity of *Chlorella vulgaris* to herbicides was investigated using polarographic oxygen rate electrode and the Pulse-Amplitude-Modulated (PAM) chlorophyll fluorescence measurements. Data reveal: (i) higher sensitivity of parameters of photosynthetic oxygen evolution (flash induced oxygen yields and oxygen burst under continuous illumination) in comparison to the widely used parameters of the chlorophyll fluorescence; (ii) higher sensitivity of oxygen evolution parameters of *Chlorella* cells to QB-binding herbicides in comparison to the pea thylakoid membranes; (iii) similar sensitivity of the PAM parameters to herbicides for both *Chlorella* cells and thylakoid membranes from higher plants. The relationship between the herbicide sensitivity and the kinetic parameters of the oxygen evolution of green algae and higher plants are discussed.

- 19. Apostolova E.L., Dobrikova A.G., Rashkov G.D., Dankov K.G., Vladkova R.S., Misra A.N. Prolonged sensitivity of immobilized thylakoid membranes in cross-linked matrix to atrazine. *Sensors & Actuators: B* 156 (2011) 140-146. doi:10.1016/j.snb.2011.04.002. ISI IF- 3.898 (Q1 WebS)**

Freshly prepared pea thylakoid membranes were immobilized in bovine serum albumin–glutaraldehyde cross-linked matrix (BSA–GA matrix) and their stability under long term storage was analyzed by PulseAmplitude-Modulated (PAM) chlorophyll fluorescence and photosynthetic oxygen evolution measured by oxygen rate electrode. The thylakoid membranes stored at 4°C showed prolonged stability in BSA–GA matrix and additional adsorption on nitrocellulose

membrane filters gave them more stability. The sensitivity of the parameters of the oxygen evolution of thylakoid membranes to atrazine increased with immobilization. The half-inhibition time for oxygen evolution and quantum efficiency of photosynthesis could be prolonged to more than 15 days. These results suggest that the immobilized thylakoid membranes in BSA–GA matrix can be used as biological receptor in biosensors for a long period of time (up to 25 days) applying the proposed new method for atrazine detection by using polarographic oxygen rate electrode. This method is more sensitive, faster and easier to use than other methods for detection of herbicides based on determination of the photochemical activity of photosystem II.

- 20. Vladkova R., Dobrikova A.G., Singh R., Misra A.N., Apostolova E. Photoelectron transport ability of chloroplast thylakoid membranes treated with NO donor SNP: Changes in flash oxygen evolution and chlorophyll fluorescence. *Nitric Oxide* 24 (2011) 84-90. doi:10.1016/j.niox.2010.12.003. ISI IF -3.548 (Q1 SJR)**

The nitric oxide (NO) donor sodium nitroprusside (SNP) is frequently used in plant science *in vivo*. The present *in vitro* study reveals its effects on the photosynthetic oxygen evolution and the chlorophyll fluorescence directly on isolated pea thylakoid membranes. It was found that even at very low amounts of SNP (chlorophyll/SNP molar ratio 77:1), the SNP-donated NO stimulates with more than 50% the overall photosystem II electron transport rate and diminishes the evolution of molecular oxygen. It was also found that the target site for SNP-donated NO is the donor side of photosystem II. Compared with other NO-donors used in plant science, SNP seems to be the only one exhibiting stimulation of electron transport through photosystem II.

- 21. Dankov K.G., Dobrikova A.G., Ughy B., Bogos B., Gombos Z., Apostolova E.L. LHCII organization and thylakoid lipids affect the sensitivity of the photosynthetic apparatus to high-light treatment. *Plant Physiol. Biochem.* 49 (2011) 629-635. doi: 10.1016/j.plaphy.2011.02.019. ISI IF -2.832 (Q1 WebS)**

Pulse-amplitude-modulated (PAM) chlorophyll fluorescence and photosynthetic oxygen evolution were used to investigate the role of the different amount and organization of light-harvesting complexes of photosystem II (LHCII) in four pea species on the susceptibility of the photosynthetic apparatus to highlight treatment. In this work we analyzed the thylakoid membrane lipid composition of the studied pea plants. A relationship between the structural organization of LHCII proteins, the amount of the main lipid classes and the sensitivity of the photosynthetic apparatus to high-light treatment was found. The results reveal that the photosynthetic apparatus, enriched in oligomeric forms of LHCII concomitant with decreased amount of anionic lipids and increased content of the monogalactosyldiacylglycerol (MGDG), is less sensitive to high light. Our data also suggest that the degree of LHCII oligomerization, as well as the lipid composition do not influence the degree of recovery of the PSII photochemistry after excess light exposure.
