LIGHT for LIFE Seminar Series 2023

LIGHT-BASED PROCESSES AND TECHNOLOGIES FOR SUSTAINABLE FUTURE

Sofia, Bulgaria, Bulgarian Academy of Sciences, 16.05.2023
(Acad. G Bonchev Str., Bl. 21, floor 2)

BOOK OF ABSTRACTS

Organizers:

- Institute of Biophysics and Biomedical Engineering
- Institute of Plant Physiology and Genetics

Main topics:

- Light-induced biological processes
- Light sources in life sciences, biomedicine and nanotechnology
- Advanced communication and optical technologies

https://biomed.bas.bg/en/light-for-life-2023/

Program Committee:

- Prof. Sashka Krumova, IBPhBME-BAS
- Prof. Anelia Dobrikova, IBPhBME-BAS
- Prof. Emilia Apostolova, IBPhBME-BAS
- Prof. Violeta Velikova, IPPG-BAS
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Invited speakers:

Prof. Lian Nedelchev – Institute of Optical Materials and Technologies “Acad. Jordan Malinowski”, Bulgarian Academy of Sciences

Prof. Natalia Krasteva – Institute of Biophysics and Biomedical Engineering, Bulgarian Academy of Sciences

Assoc. Prof. Kiril Mishev – Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences

Assoc. Prof. Margarita Kouzmanova – Faculty of Biology, Sofia University “St. Kliment Ohridski”

Assoc. Prof. Miroslava Zhiponova – Faculty of Biology, Sofia University “St. Kliment Ohridski”

Assoc. Prof. Radka Vladkova – Institute of Biophysics and Biomedical Engineering, Bulgarian Academy of Sciences

Sen. Assist. Charilaos Xenodochidis – Institute of Biophysics and Biomedical Engineering, Bulgarian Academy of Sciences

Sen. Assist. Kolyo Dankov – Faculty of Biology, Sofia University “St. Kliment Ohridski”

Sen. Assist. Momchil Paunov – Faculty of Biology, Sofia University “St. Kliment Ohridski”

Zhaneta Georgieva – Faculty of Biology, Sofia University “St. Kliment Ohridski”

Scientific program

13:00	**Opening** ***Sashka Krumova***
13:10 – 14:10	**Session I: Light-Induced Biological Processes** ***Session Chair: Emilia Apostolova***
13:10 – 13:30	"Photosynthetic Electron Transport Pathways. Exploring the Application of Delayed Chlorophyll Fluorescence for Assessing Stress Response of the Light-Driven Reactions in Plants" *Kolyo Dankov, Vasilij Goltsev*
13:30 – 13:50	"Hydrophobic Mismatch Molecular Model for Light State Transitions – Optimization of Photosynthetic Electron Transport under Light-Spectrum Changes" *Radka Vladkova*
13:50 – 14:10	"Light-Dependent Hormone Regulation and Hormone-Dependent Light Signaling Pathways in Plants" *Kiril Mishev*
14:10 – 16:00	**Session II: Light Sources in Life Sciences, Biomedicine and Nanotechnology** ***Session Chair: Anelia Dobrikova***
14:10 – 14:30	"How Quality of Light Affects the Bioactive Potential of Microalgae?" *Zhaneta Georgieva, Zornitsa Karcheva, Detelina Petrova, Miroslava Zhiponova, Ganka Chaneva*
14:30 – 14:50	"Effects of Light on the Medicinal Plant *Nepeta nuda*" *Miroslava Zhiponova, Grigor Zehirov, Adriana Krasteva, Uroš Gašić, Danijela Mišić, Krasimir Rusanov, Mila Rusanova, Zhenya Yordanova, Momchil Paunov, Ganka Chaneva, Valya Vassileva*
14:50 – 15:20	**Coffee Break**
15:20 – 15:40	"Light as Multifunctional Tool for Medical Purposes" *Charilaos Xenodochidis, Natalia Krasteva*
15:40 – 16:00	"Near-Infrared Light in Intelligent Nanoplatform for Synergistic Chemo- and Phototherapy of Cancer" *Kamelia Hristova-Panusheva, Trayana Kamenska, Zlatina Gospodinova, Milena Georgieva, Natalia Krasteva*
16:00 – 17:10	**Session III: Advanced Light Technologies** ***Session Chair: Violeta Velikova***
16:00 – 16:20	"Beyond Light: Cellular Effects of 2.45 GHz EMF. Preliminary Results on Yeasts" *Boyana Angelova, Meglena Kitanova, Momchil Paunov*
16:20 – 16:40	"Possible Effect of Electromagnetic Fields Emitted by Wireless Communication Devices on the Surface Charge of *Lactobacillus*" *Clio Gleridis, Gabriela Atanasova, Nikolai Atanasov, Svetla Danova, Dragomir Yankov, Margarita Kouzmanova*
16:40 – 17:10	"Polarized Light: Applications for Polarization Holography and Polarimetry" *Lian Nedelchev*
17:10	**Reception**

Photosynthetic Electron Transport Pathways. Exploring the Application of Delayed Chlorophyll Fluorescence for Assessing Stress Response of the Light-Driven Reactions in Plants

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Photosynthetic electron transport pathways or chains are the collection of the biochemical processes by which light energy is captured and converted into chemical energy during photosynthesis. These pathways involve a series of redox reactions that occur in the thylakoid membranes of chloroplasts. One important aspect of these pathways is the production of ATP and NADPH, which are crucial for the synthesis of carbohydrates during photosynthesis.

Delayed Chlorophyll Fluorescence (DCF) is a non-invasive biophysical tool that can be used to monitor the photosynthetic electron transport pathways. By measuring the light emitted by chlorophyll molecules in darkness after initial excitation, DCF can provide insights into the efficiency of these pathways and the overall health of plants. DCF measurements have been used to investigate plant stress responses to environmental factors, such as temperature, drought, and light intensity, and to evaluate the impact of different management practices on plant growth and development.

DCF measurements are based on the detection of the red and far-red fluorescence emitted by chlorophyll molecules after a period of darkness. This technique has been shown to be a sensitive indicator of changes in photosynthetic performance and energy transfer within the photosynthetic apparatus. In addition, DCF measurements can provide information on the activity of the electron transport chain, which is critical for plant growth and survival.

Recent advancements in DCF technology have enabled the development of new methods for the analysis of plant physiological state. Additionally, the use of DCF measurements in combination with other physiological and biochemical assays, such as gas exchange and enzyme activity assays, can provide a more comprehensive understanding of plant responses to environmental stressors.

In this talk, we will discuss the application of DCF for assessing physiological changes in plants. We will review the basic principles of DCF measurements and highlight recent advances in DCF technology. We will also present case studies that demonstrate the utility of DCF measurements for assessing plant responses to different environmental stressors, and discuss future directions for the development of DCF as a tool for plant physiology research. Overall, we hope to demonstrate the value of DCF as a powerful and versatile tool and encourage its wider adoption in plant science research.

Hydrophobic Mismatch Molecular Model for Light State Transitions – Optimization of Photosynthetic Electron Transport under Light-Spectrum Changes

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Oxygen-evolving (oxygenic) photosynthesis is the most important light-induced biological process because it converts sunlight energy into chemical energy, and directly or indirectly sustains all the aerobic life on Earth. Its primary light reactions and their regulation are performed in the thylakoid membranes of cyanobacteria, algae and plants. Given the vital significance of oxygenic photosynthesis and its potential to provide bioinspired solutions in diverse membrane-related fields, it is important to understand its specific regulatory mechanisms.

From a membrane biophysical viewpoint, the most attractive seems to be the short-term adaptive regulatory mechanism light State Transitions (STs). STs are activated when one of the two photosystems (PSII and PSI) is preferentially excited by light-spectrum changes. The STs optimize photosynthesis by balancing the excitation energy supply to PSII and PSI via membrane structural reorganization. STs are unique not only because they are specific for oxygenic photosynthesis, but also because STs are the only regulatory mechanism which (i) optimizes the photosynthetic electron transport at low light and in dynamic environments, (ii) is reversible and occurs without energy losses, (iii) has characteristics of a perpetual mobile for membrane reorganizations, and (iv) is the reason for the recently proposed completely new molecular mechanism for induction and realization of membrane reorganizations without analog among the known regulatory/signaling mechanisms of biological membranes. The proposed mechanism has been termed hydrophobic mismatch molecular model for cytochrome b_6f governed STs [Vladkova (2016) *J. Biomol. Struct. Dyn*. 34: 824-854; Vladkova (2023) submitted and under revision].

This presentation will describe our model and the current stage of its confirmation. The proposed model provides for the first time not only a unified description of entire state transitions process in cyanobacteria, algae and plants, but also proposes the driving force for the induction of membrane reorganizations upon state transitions.

Light-Dependent Hormone Regulation and Hormone-Dependent Light Signaling Pathways in Plants

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Plants use numerous signaling mechanisms and networks to regulate development, confer stress tolerance and ensure survival. Light quality and light quantity are key determinants that shape plant fitness in the adverse environment. Besides its role as an energy source for photosynthesis, light is implicated in a number of intertwined signal transduction pathways. The latter converge with plant hormone signaling at the level of common transcription factors to ensure finely tuned and coordinated transcriptional reprogramming. Here, I discuss the interplay of light and phytohormones with respect to photomorphogenesis and phototropism, with an emphasis on the role of auxins and brassinosteroids (BR) for these light-dependent processes. Recent studies have revealed important aspects of the signaling crosstalk that deal with the impact of light on hormone transport, hormone perception, and hormone-induced transcription. In particular, photoreceptor activation has been shown to induce changes in the polar localization of PIN-FORMED (PIN) auxin efflux transporters at the plasma membrane. Further on, light signaling affects the activity of BRI1-EMS-SUPPRESSOR 1 (BES1), a key transcription factor which is part of the BR signaling pathway. The importance of the intracellular localization of the BR receptor for photomorphogenesis will also be discussed by analyzing the involvement of the exocyst vesicle-tethering complex for plasma membrane protein trafficking. Altogether, the recent developments in light- and hormone-signaling research uncover novel possibilities for improvement of agronomically important plant traits based on modulation of the light regime.

How Quality of Light Affects the Bioactive Potential of Microalgae?

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The newly isolated Bulgarian strain *Coelastrella* sp. BGV (Chlorophyta) was investigated to determine the influence of light quality on its growth, metabolic composition, antimicrobial potential and antioxidative activity. For the purpose of that study, we have assembled a laboratory setup that allowed algal suspension to be intensively cultivated under 5 different LED light formulas – white/red (W/R); royal blue (Royal B); photo red; red/blue (R/B) and white light (W) as a control.

The highest growth of microalgal culture was observed under red light illumination – 1.3 mg ml^{-1} DW, followed by the white light – 1.2 mg ml^{-1} DW. All other light formulas showed a decrease in biomass accumulation – from 15% to 50%, especially in the R/B variant. No strict correlation was found between the changes in growth rate and those in protein and pigment contents. The highest amount of proteins was recorded in the variants R/B and W/R – 30% and 20% above the control. The most pronounced increase of pigment content was measured under the R/B light, where the amounts of chlorophyll *a* and carotenoids were about three times higher than the control, and the content of chlorophyll *b* was twice as high.

Further, for the preparation of microalgal extracts (ethanolic extracts, water extracts and cultural medium), R/B and Royal B variants were preferred, considering them the most promising. The ethanolic extracts obtained under R/B, Royal B and white light (as control), were characterized by the enhanced total antioxidant activity (TAA), as well as the highest phenolic and flavonoid content. The most pronounced antimicrobial activity was shown by the cultural medium grown under R/B and Royal B light conditions.

The present investigation confirmed the high bioactivity of *Coelastrella* sp. BGV, so more in-depth studies are needed to establish the mechanisms of action and beneficial effects of the extracts and products obtained from algal biomass. In our future research, several prokaryotic microalgal strains will be also included, in an attempt to investigate and assess their biotechnological potential.

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Effects of Light on the Medicinal Plant *Nepeta nuda*

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Light is a major factor controlling plant metabolism including the phytochemical composition. In our study we aimed to investigate the effect of different light spectra on the medicinal plant *Nepeta nuda* L. grown under controlled *in vitro* conditions. For our experiment three different light qualities and intensities were examined: white (W) fluorescent light as a control, and a combination of blue and red LED light with high intensity – BR, and with low intensity – BRS. In vitro internode explants were cultivated for 5 weeks under the three light variants, and the obtained plantlets were compared in respect to their metabolic and antioxidant parameters. In addition, since the applied blue and red light combination was designed to stimulate flowering, a comparison between the phytochemical profiles of *in vitro* plants and wild-growing plants was performed. The metabolic analyses (UHPLC/MS2 and GC-MS) revealed that the applied light conditions altered the content of bioactive compounds specific for *N. nuda*, such as phenolics, iridoids, and volatiles. The effect of BR light on in vitro *N. nuda* demonstrated closest relation to the metabolic profile in flower – with upregulation of the most abundant phenolics and iridoid glucoside, as well as volatiles. In comparison, in vitro plants under W and BRS lights had much lower content of the investigated phytochemicals, except for the 4a-α,7-β,7a-α-nepetalactone that was with higher level than the BR light variant and the wild-growing plants. These data could serve as a basis for regulation of *N. nuda* flowering by light modulation, as well as for directed biosynthesis of metabolites of interest. Further observation for oxidative stress in leaves of the *in vitro* variants showed an absence in the epidermis. In a following *ex vitro* adaptation it could be pointed out which light treatment would lead to most successful adaptation in nature and restoration of the bioactive potential.

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Light as a Multifunctional Tool for Medical Purposes

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Light as a whole, and visible light in particular, as an object for exploration, has existed since the days of the great Greek ancient philosophers Socrates, Plato and Aristotle. However, Euclid was the scientist who summarized for the first time all the accumulated knowledge. In the last century, physicists like Albert Einstein and Max Plank made suggestions against traditional and conservative beliefs on the nature of light. They proposed that light, except for an electromagnetic wave, could also be seen as a photon. Nowadays, scientific society classifies electromagnetic radiation (ER), namely light, into ionizing and non-ionizing. The main difference between those two is based on the energy of the emitted photons or particles: the greater energy, the greater penetration in the matter. However, ionizing radiation refers to the fact that the emitted photons/particles can interact in such a way with the matter, to be precise biological structures (i.e. tissues), that they can convert atoms into reactive ions. Notably, ionizing and non-ionizing radiation have been widely applied in medical and clinical practice over the last 6-7 decades to diagnose and treat various medical conditions. Modalities that use non-ionizing radiation have been magnetic resonance imaging, ultrasonography, low-level laser therapy or photobiomodulation, photodynamic therapy, and bright light therapy. Alternatively, modalities such as computed tomography, positron emission tomography, fluoroscopy, teletherapy, brachytherapy, gamma knife, etc., use ionizing radiation.

Meanwhile, technological evolution has led to greater integrity of those above therapeutic and diagnostic approaches in the healthcare sector. Consequently, we discuss the overall application of ER due to its beneficial impact on treating mental, neurodegenerative, dermatological and neurological disorders and various types of cancer.

Near-Infrared Light in Intelligent Nanoplatform for Synergistic Chemotherapy and Phototherapy of Cancer

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Cancer remains one of the most deadly diseases worldwide. Despite the encouraging progress in cancer therapy, an urgent need for new, more efficient cancer treatment methods still exists. Conventional treatments such as chemotherapy, radiotherapy, and surgery often are accompanied by many drawbacks, including side effects, severe pain, drug resistance, and weak effectiveness due to the instability and rapid clearance of drugs, resulting in unsatisfactory outcomes of anticancer therapy.

Drug delivery systems (DDSs) based on nanomaterials have shown great promise to overcome these limitations and improve cancer treatment efficacy. Nanomaterials due to their unique properties increase drug stability, reduce systemic toxicity, improve pharmacokinetics, elevate bioavailability, precise drug transportation capability, and control drug release. However, localizing the on-demand release of anticancer drugs in tumor tissues remains a great challenge. In this regard, photoresponsive DDSs that use light as an external stimulus can offer precise spatiotemporal control of drug release at desired sites. Most photoresponsive DDSs are only responsive to ultraviolet-visible light that shows phototoxicity and/or shallow tissue penetration depth, thereby greatly restricting their applications. Near-infrared (NIR) photoresponsive DDSs have been developed to address these issues. They can achieve on-demand drug release in tumors through photothermal, photodynamic, and photoconversion mechanisms, affording amplified therapeutic effects in synergy with phototherapy. Thus, the combination of near-infrared light, chemotherapy, and nanotechnology is a preposition for developing perspective and intelligent nanoplatforms for synergic cancer therapies.

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Beyond Light: Cellular Effects of 2.45 GHz EMF. Preliminary Results on Yeasts

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In recent years, the use of various wireless communication devices that generate electromagnetic fields (EMF) has significantly increased. Most of them are worn near or on the body (phones, fitness bracelets, etc.), while implanted devices gain more and more applications, especially in medicine. All this leads to high public concern regarding possible health risks. For a clearer understanding of the effects of EMF on the body, a thorough knowledge of the mechanisms of action at the cellular and subcellular level is necessary. The safety of wireless devices is evaluated mainly by the specific absorbed rate (SAR) which itself is defined by the heating of the tissues. However, in addition to thermal, non-thermal EMF effects on living systems have been reported such as increased oxidative stress, changes in cell viability, peroxidation of lipids, disruption of cell membrane integrity, even genetic disorders. There are just few studies of the biological effects of EMF acting near to its source.

The aim of our study was to define the effects of 2.45 GHz near-field EMF, widely used in commercial devices, on yeast cells as a model system. Baker's yeast suspensions were absorbing 20 W/kg SAR in the near-field EMF, generated by half-dipole antenna, during 40 minutes periods, repeating up to 8 hours. Cell membrane permeability was tested by following the release of nucleotides measuring the absorption at 260 nm of the extracellular medium. Cell antioxidant activity was determined by applying the trolox equivalent antioxidant capacity method. DNA damage was traced by alkaline comet assay. For discerning non-thermal from thermal effects, yeast suspensions, incubated at the maximal temperature reached during EMF exposure, were used as control. There were no statistically significant differences in the examined parameters between EMF-treated and control suspensions therefore non-thermal biological effects of EMF should not exist at the applied experimental conditions.

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Possible Effect of Electromagnetic Fields Emitted by Wireless Communication Devices on the Surface Charge of *Lactobacillus*

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With the rapidly increasing application of wireless technologies, the anxiety and speculation about microwave (MW) induced potential health hazards has been attracting more and more attention. Continuous exposure to electromagnetic fields (EMF) can lead to adverse effects on human health such as headaches, chronic fatigue, heart problems, nausea; to affect the central nervous, endocrine and immune systems, and many others. EMF exposure could also affect microorganisms in the human body and change their functions. Bacteria in the human body are more than 1000 species, they mainly live in the large intestine. The gut microbiota is very important for the overall health of the organism. For this reason, interest in the effects of MW on the beneficial bacteria inhabiting our gastrointestinal tract is arising.

There are few studies carried out on the subject in the available literature. The results of our preliminary experiments with a mixture of six species of *Lactobacillus* exposed to 2.41 GHz EMF at three intensities (20, 40 and 180 V/m) for 30 min showed alterations in the growing of lactic acid bacteria and in their ability to form biofilm. Biofilms are social communities of bacteria which are important for survival in their natural environments and protect them from the adverse factors. The surface charge is one of the major determinants of whether a bacterium colonizes a surface to establish a biofilm or not. The initial interactions between the bacterial cell and the surface depend largely on their respective surface properties. Zeta potential (ZP) measurements are appropriate technique for exploring surface processes in bacteria. ZP is a key factor in biofilm formation. There is also data about alterations in zeta potential and electrophoretic mobility of different cells under influence of high frequency EMF. There is no information about effects of EMF on bacterial surface charge. Our hypothesis is that the changes in *Lactobacillus* ability to form biofilm could be a result of alterations in zeta potential after EMF exposure.

Polarized Light: Applications for Polarization Holography and Polarimetry

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Polarization is an essential property of all electro-magnetic waves, including light. Together with amplitude, phase and wavelength it fully describes the light wave. Polarization can be linear, circular or in the general case elliptical. We will consider the polarization characteristics of light and in particular the Stokes parameters, that allow us to describe both fully and partially polarized light. The polarization state of light can be controlled and measured using polarizers, wave plates and polarimeters. The two-dimensional polarization distribution of a light field can also be recorded using the methods of polarization holography. It requires special media, sensitive to the polarization of light, namely photoanisotropic materials. Using polarization holography diffractive optical elements with unique polarization properties can be obtained.

Measurement and analysis of the polarization of light transmitted through or reflected from an object is known as polarimetry. It allows to determine the anisotropic properties of various materials – optical materials, biological tissues, etc. Polarimetry finds numerous applications in medicine, in particular in early-stage cancer diagnostics, Mueller microscopy, remote sensing and other fields.

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