



12th International Conference
Photosynthesis and Hydrogen Energy Research for
Sustainability

in honor of
John F. Allen (UK), Eva-Mari Aro (FIN), Ibrahim Dincer (CAN),
Kazunari Domen (JPN), Elisabeth Gantt (USA), Andrey B. Rubin (RUS)

Oct 13 – 19, 2024
BAHÇEŞEHİR UNIVERSITY SOUTH CAMPUS
Istanbul

ABSTRACTS AND PROGRAMME

International Conference “Photosynthesis and Hydrogen Energy Research for Sustainability-2024: in honor of John F. Allen (UK), Eva-Mari Aro (FIN), Ibrahim Dincer (CAN), Kazunari Domen (JPN), Elisabeth Gantt (USA), Andrey B. Rubin (RUS)

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The volume contains abstracts of the lectures and poster presentations at 12th International Meeting Photosynthesis and Hydrogen Energy Research for Sustainability – 2024 which was held on Oct 13 – 19, 2024, in Istanbul, at Bahçeşehir University.

The conference delves into both experimental and theoretical research across a broad spectrum of topics related to photosynthesis and (bio)hydrogen. Discussions range from the fundamental processes of electron transfer and energy conversion to the physiological aspects of photosynthesis and the practical applications of hydrogen production. Significant emphasis is placed on the structural organization of photosynthetic reaction centers, the structure and function of photosystems, artificial photosynthesis, and hydrogen production mechanisms. This volume will be valuable to researchers and students engaged in the study of photosynthesis and (bio)hydrogen production.

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Committee's Note

We are excited to invite you to the 12th International Conference on "Photosynthesis and Hydrogen Energy Research for Sustainability-2024," honoring the remarkable contributions of leading figures in the field. This prestigious event will take place at the Bahçeşehir University in Istanbul, Türkiye.

Organized by the Faculty of Engineering and Natural Sciences, this conference serves as a vital platform for scholars, researchers, and industry professionals from around the world to gather and share their insights. This year's conference will continue to stimulate engaging discussions on past, present, and future research related to photosynthesis and hydrogen energy, encompassing a broad spectrum of topics from molecular dynamics to global sustainability initiatives.

We will explore the critical field of (bio)hydrogen production, addressing both fundamental research and practical applications. Attendees will have the unique opportunity to engage with distinguished researchers dedicated to advancing our understanding of photosynthesis and hydrogen energy, fostering an environment conducive to intellectual exchange and collaboration.

This conference is designed for students, postdoctoral fellows, and scientists from diverse backgrounds, promoting professional growth and the establishment of meaningful connections. Participants will benefit from a dynamic program featuring a wide array of topics organized into two primary areas: photosynthesis and (bio)hydrogen.

In the photosynthesis section, we will delve into fundamental processes, exploring the complexities of photosynthetic apparatus structure, function, and biogenesis. Topics will include insights into photosystem I and II, mechanisms of water oxidation, artificial photosynthesis, and the regulation of photosynthesis under environmental stress.

The (bio)hydrogen section will highlight significant advancements in biological hydrogen production, examining the roles of hydrogenases and the potential of artificial photosynthesis. We will also discuss key issues related to hydrogen purification and storage, the evolving dynamics of the hydrogen economy, and the importance of education in fostering hydrogen energy solutions.

With tens of captivating presentations, including lectures and poster sessions, this interdisciplinary conference will facilitate rich participant interactions, enhancing collaboration and knowledge sharing.

As we prepare for this momentous gathering, we look forward to a week filled with inspiring presentations, thought-provoking discussions, and invaluable insights into the fields of photosynthesis and hydrogen energy research. Join us as we embark on this exciting journey toward a sustainable future together!

Topics

1. Photosynthesis Research for Sustainability:

- 1.1. Primary Processes of Photosynthesis
- 1.2. Structure, Function and Biogenesis of the Photosynthetic Apparatus
- 1.3. Photosystem I and Bacterial Photosynthesis
- 1.4. Photosystem II and Water Oxidation Mechanism
- 1.5. Energy Transfer and Trapping in Photosystems
- 1.6. Plant Development and Growth Regulation
- 1.7. Carbon Fixation (C3 and C4) and Photorespiration
- 1.8. Artificial and Applied aspects of Photosynthesis including Nanotechnology
- 1.9. Regulation of Photosynthesis, ROS production and Environmental Stress
- 1.10. Photosynthesis under Extreme Environments: Arctic, Antarctic and Deserts Research
- 1.11. Environmental Control of Photosynthetic Sustainability: Global Aspects and Challenges
- 1.12. Systems Biology of Photosynthesis: Integration of Genomic, Proteomic, Metabolomic and Bioinformatic Studies
- 1.13. Plant Mineral Nutrients and Photosynthetic Capacity
- 1.14. Photosynthesis Education and Emerging Techniques for Studying Photosynthesis including Neutron Scattering

2. Hydrogen Energy Research for Sustainability:

- 2.1. Energy for the Future – Hydrogen economy
- 2.2. Elevating Climate Change
- 2.3. Biological Hydrogen Production
- 2.4. Waste-to-Hydrogen Scenarios
- 2.5. Hydrogen production from renewable feedstock
- 2.6. Hydrogenases
- 2.7. Proton Reduction Catalysts
- 2.8. Reduction of Carbon Dioxide
- 2.9. Artificial Photosynthesis for Hydrogen energy
- 2.10. Fuel cell and electrolyzes
- 2.11. Nanomaterials for Hydrogen Production
- 2.12. Hydrogen Energy Education and Emerging Techniques for Studying of Hydrogen Energy
- 2.13. Hydrogen storage
- 2.14. Hydrogen policy and strategies
- 2.15. Solar Hydrogen Panels
- 2.16. Photovoltaic Solar Cells

12th International Conference of Photosynthesis and Hydrogen Energy Research for Sustainability Conference Program

14 October	BAU SOUTH CAMPUS
09:00 - 17:00	REGISTRATION Conference registration at registration desk
14-18 October	BAU SOUTH CAMPUS
POSTERS	
Chairs	
Jan Kern (USA), Gert Schansker (Germany), Marek Zivcak (Slovakia), Alessandro Agostini (Italy), Ryouichi Tanaka (Japan), Shailendra P. Singh (India)	
14 October	BAU SOUTH CAMPUS
10:00 - 10:30	OPENING CEREMONY Esra Hatipoğlu (Türkiye) <i>Rector (BAU)</i> Ahmet Arif Ergin (Türkiye) <i>Dean (Faculty of Engineering & Natural Sciences, BAU)</i> Julian J. Eaton-Rye (New Zealand) <i>Chairman</i> Barry Bruce (USA) <i>co-Chairman</i> Tatsuya Tomo (Japan) <i>Secretary</i> Suleyman I. Allakhverdiev (Russia/Türkiye) <i>Coordinator</i> Gyozo Garab (Hungary) <i>Advisor</i>
Plenary Lectures	
Session Chairs	
Gyozo Garab (Hungary) Suleyman Allakhverdiev (Russia/Türkiye)	
10:30 - 11:00	Kazunari Domen (Japan) Photocatalytic water splitting to produce green hydrogen and other fuels on a large scale
11:00 - 11:30	Andrey B. Rubin (Russia) Primary photosynthesis and problems in physical-chemical biology
11:30 - 12:00	COFFEE BREAK

14 October	BAU SOUTH CAMPUS
Plenary Lectures Session Chairs Barry Bruce (USA) Tatsuya Tomo (Japan)	
12:00 - 12:30	John Allen (UK) Why bioenergetic organelles contain their own genomes and genetic systems (<i>online</i>)
12:30 - 13:00	Elisabeth Gantt (USA) Accessory Pigments: Phycobiliproteins in Three Algal Classes (<i>online</i>)
13:00 - 14:30	MIDDAY BREAK
14:30 - 15:00	Ibrahim Dincer (Canada) (<i>online</i>) Innovative Energy Systems for Sustainable Future
15:00 - 15:30	Eva-Mari Aro (Finland) Photodamage and Regulation of Photosynthetic Light Reactions Arjun Tiwari (Finland) A critical role of photosystemI photoinhibition and subsequent recovery, in regulation photosynthetic electron flow between linear and alternate electron transport
Invited Lectures	
15:30 - 15:50	Győző Garab (Czech Republic/Hungary) The lipid polymorphism and the energization of plant thylakoid membranes. facts and hypotheses
15:50 - 16:20	COFFEE BREAK
Invited Lectures Session Chairs: Gyozo Garab (Hungary) Can Erkey(Türkiye)	
16:20 - 16:40	Jan Kern (USA/Germany) X-ray studies of photosynthetic proteins: What we can learn about their inner workings from diffraction and spectroscopy experiments at room temperature
16:40 - 17:00	Seiji Akimoto (Japan) Responses of light-harvesting and energy-transfer processes in typical cyanobacteria to light qualities

14 October	BAU SOUTH CAMPUS
17:00 - 17:20	Yuu Hirose (Japan) Understanding the photosensing mechanism of chromatic acclimation in cyanobacteria
17:20 - 17:40	Deniz Uner (Türkiye) (Artificial) Photosynthesis is an Oxygen Management Problem
18:00 - 19:00	Award Ceremony
19:00 - 21:00	Welcome Party
15 October	BAU SOUTH CAMPUS
Invited Lectures Session Chairs: Xiaochun Qin (China) Deniz Uner (Türkiye)	
10:00 - 10:20	Cheryl Kerfeld (USA) Carboxysome-Inspired Compartmentalization of Catalysis
10:20 - 10:40	Yusuke Matsuda (Japan) CO ₂ -evolving machinery in the pyrenoid - Structure and function to optimize CO ₂ assimilation efficacy and light energy use in marine diatoms
10:40 - 11:00	Yuichiro Kashiya (Japan) Maintenance and control of carbon fixation in photosynthesis in a kleptoplasty
11:00 - 11:30	COFFEE BREAK
Invited Lectures Session Chairs: Jan Kern (USA) Arvi Freiberg (Estonya)	
11:30 - 11:50	Xiaochun Qin (China) Structure of the red-shifted <i>Fittonia albivenis</i> photosystem I
11:50 - 12:10	Guangye Han (China) Structural insights into the assembly of photosystem II
12:10 - 12:30	Alessandro Agostini (Italy) Red-shifted chlorophyll <i>a</i> absorption in algal Light-Harvesting Complexes: insights from opto-magnetic spectroscopies

15 October	BAU SOUTH CAMPUS
12:30 - 12:50	Keisuke Kawakami (Japan) Structure of the far-red light utilizing photosystem II interacting with chlorophyll-binding protein (CBP) from <i>Acaryochloris marina</i> at 2.4 Å resolution
12:50 - 14:20	MIDDAY BREAK
Invited Lectures Session Chairs: Tatsuya Tomo (Japan) Dmitry Dunikov (Russia)	
14:20 - 14:40	Min Yu (China) The Role of Hydrogen-Rich Water in Enhancing Salt Tolerance in Rice Seedlings
14:40 - 15:00	Bekzhan Kossalbayev (Kazakhstan/China) Immobilization of cyanobacterium cells in alginate gels to enhance hydrogen production efficiency
15:00 - 15:20	Fatemeh Khosravitar (Sweden) A Straightforward Single-Phase Protocol for Initiating and sustaining Algal Hydrogen Production
15:20 - 15:40	COFFEE BREAK
Invited Lectures Session Chairs: Min Yu (China) Keisuke Saito (Japan)	
15:40 - 16:00	Tatsuya Tomo (Japan) Protective effects of hydrogen against light-induced oxidative stress in <i>Synechocystis sp.</i> PCC 6803
16:00 - 16:20	Can Erkey (Türkiye) PEM Electrolyzers for Green Hydrogen Production: Current Status and Research Needs
16:20 - 16:40	Dmitry Dunikov (Russia) Metal hydrides for hydrogen storage, purification and compression

16 October	BOSPHORUS
All day Cultural Tour	
09:30	Meeting in front of the Fazıl Say Conference Hall (Fazıl Say atrium)
09:45	Meeting the tour guide in front of the Bahçeşehir University Faculty of Engineering and Natural Sciences
10:00	Beşiktaş Pier Ferry departure and start of the Bosphorus Tour
12:00	Beşiktaş Pier Ferry arrival and end of the Bosphorus Tour
12:00 – 17:00	Free time
17 October	BAU SOUTH CAMPUS
Invited Lectures Session Chairs: Barry Bruce (USA) Guangye Han (China)	
10:00 - 10:20	Yusuke Tsukatani (Japan) Unraveling 3.5 billion years of co-evolution between photosynthesis and bacteria
10:20 - 10:40	Kentaro Ifuku (Japan) Evolution and function of the light-harvesting complex in red-lineage algae
10:40 - 11:00	Toru Kondo (Japan) Flexible and heterogeneous regulation of photosynthetic light harvesting revealed by single-molecule spectroscopy
11:00 - 11:30	COFFEE BREAK
Invited Lectures Session Chairs: Kentaro Ifuku (Japan) Rajagopal Subramanyam (India)	
11:30 - 11:50	Keisuke Saito (Japan) A Role of Protein Matrix in Superexchange Electron Transfer in Charge Separation
11:50 - 12:10	Daisuke Takagi (Japan) Oxygen sensitivity of photosystem I photoinhibition in land plants: land plants activate protective mechanisms on PSI photoinhibition depending on the growth light environment
12:10 - 12:30	Barry D. Bruce (USA) PSI-SMALP: All the Fat but Hold the Soap

17 October	BAU SOUTH CAMPUS
12:30 - 12:50	Francesco Francia (Italy) Effect of trehalose on the electron transfer chain of photosynthetic bacteria
12:50 - 14:20	MIDDAY BREAK
Invited Lectures Session Chairs: Francesco Francia (Italy) Yusuke Tsukatani (Japan)	
14:20 - 14:40	Marek Zivcak (Slovakia) Acclimation of photosynthetic processes in wheat leaves to high temperature
14:40 - 15:00	Rajagopal Subramanyam (India) Characterization of the possible acclimation strategies and remodeling of photosystems under long-term hypersaline conditions in <i>Dunaliella salina</i>
15:00 - 15:20	Ryouichi Tanaka (Japan) Towards understanding sustained thermal dissipation mechanisms in overwintering evergreen leaves
15:20 - 15:40	COFFEE BREAK
Invited Lectures Session Chairs: Ryouichi Tanaka (Japan) Marek Zivcak (Slovakia)	
15:40 - 16:00	Agepati Raghavendra (India) Restriction of photorespiration raises ROS levels and their relationship depends on the robustness of redox regulation in leaves
16:00 - 16:20	Shailendra P. Singh (India) Chromatic acclimation and photosynthetic fitness in cyanobacteria
16:20 - 16:40	Jörg Pieper (Estonia) Protein dynamics mediates the structural adaptation of the orange carotenoid protein in cyanobacterial photoprotection: a neutron scattering study
16:40 - 18:00	Report/Decision of the Posters Chairs Awards Ceremony for Young Researchers
18:30 - 21:00	GALA DINNER

18 October	BAU SOUTH CAMPUS
Invited Lectures Session Chairs: Gert Schanske (Germany) Seiji Akimoto (Japan)	
10:00 - 10:20	Mohammad Mahdi Najafpour (Iran) How Manganese Compounds Facilitate Water Oxidation in Artificial Photosystems (<i>online</i>)
10:20 - 10:40	Alexander N. Tikhonov (Russia) Regulation of the Intersystem Electron Transport in Plant Chloroplasts <i>in Situ</i> and <i>in Silico</i> (<i>online</i>)
10:40 - 11:00	Iftach Yacoby (Israel) Algal Hydrogen & Biomass Farming (<i>online</i>)
11:00 - 11:30	COFFEE BREAK
Invited Lectures Session Chairs: Agepati Raghavendra (India) Rahul Kumar (India)	
11:30 - 11:50	Gert Schansker (Germany) Pulses and flashes to probe the photosynthetic electron transport chain and the nature of fluorescence
11:50 - 12:10	Arvi Freiberg (Estonia) Current Understanding of Color-Tuning in Bacterial Photosynthesis
12:10 - 12:30	Petar Lambrev (Hungary) Inter-subunit energy transfer in photosynthetic supercomplexes observed by two-dimensional electronic spectroscopy
12:30 - 14:00	MIDDAY BREAK
Invited Lectures Session Chairs: Bekzhan Kossalbayev (China/Kazakhstan) Petar Lambrev (Hungary)	
14:00 - 14:20	Irada Huseynova (Azerbaijan) Study of water-soluble sugars and phosphoenolpyruvate carboxylase in photosynthetic and non-photosynthetic organs of wheat cultivars with contrasting drought tolerance

18 October	BAU SOUTH CAMPUS
14:20 - 14:40	Rahul Kumar (India) Cryo-milled nano-DAP fertilizer application promotes photosynthetic performance and plant growth in tomato and rice
14:40 - 15:00	Rupal Singh Tomar, Prabha Rai-Kalal, Anjana Jajoo (India) Boosting Growth and Bioremediation Power in <i>Chlorella vulgaris</i> Using Silicon Nanoparticles
15:00 - 15:20	Yogesh Mishra (India) How does LexA regulate the tolerance of <i>Anabaena</i> sp. PCC 7120 to cadmium stress through regulation of photosynthetic responses?
15:20 - 15:40	COFFEE BREAK
Invited Lectures Session Chairs: Jörg Pieper (Estonia) Rajagopal Subramanyam (India)	
15:40 - 16:00	Mohammad Yusuf Zamal (India) Stability of photosynthetic apparatus, transcriptome, and metabolome of <i>Rhodobacter alkalitolerans</i> strain JA916T, response to alkaline along with high light environment
16:00 - 16:20	Nuria K. Koteyeva (Russia) Biochemical and Structural Diversification of C ₄ Photosynthesis in <i>Tribe Zoysieae (Poaceae)</i>
16:20 - 16:40	Talha Kuru (Türkiye) Photocatalytic hydrogen evolution by CoAl ₂ O ₄ photocatalyst under solar irradiation
16:40 - 17:00	Yiğit Osman Akyıldız (Türkiye) Size-dependent electrocatalytic hydrogen evolution by MoS ₂ micro- and nanoparticles
17:00 - 17:20	Suleyman I. Allakhverdiev (Russia) Exogenous carbon substrate for sustainable hydrogen production by cyanobacteria
19 October	BAU SOUTH CAMPUS
Workshop and General Discussion	

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Photocatalytic Water Splitting to Produce Green Hydrogen and Fuels on a Large Scale



Kazunari Domen

Special Contract Professor / University Professor

Shinshu University / The University of Tokyo

2007; Catalysis Society of Japan Awards

2011; The Chemical Society of Japan Awards

2019; Advance of Catalysis Award of APACS (Asia-Pacific Association of Catalysis Societies)

2022; Award the EIC Horizon Prize for “Fuel from the Sun: Artificial Photosynthesis”

2024; Heinz Heinemann Award of IACS (International Association of Catalysis Societies)

2024; Clarivate Citation Laureates (Chemistry)

Kazunari Domen received B.S. (1976), M.S. (1979), and Ph.D. (1982) honors in chemistry from the University of Tokyo. Dr. Domen joined Chemical Resources Laboratory, Tokyo Institute of Technology in 1982 as Assistant Professor and was subsequently promoted to Associate Professor in 1990 and Professor in 1996. Moving to the University of Tokyo as Professor in 2004, and Cross appointment with Shinshu University as Special Contract Professor in 2017. University Professor of the University of Tokyo in 2019. Domen is interested in heterogeneous catalysis, especially in photocatalysts for water splitting. He has been developing extremely high quantum efficiency photocatalysts, visible light responsive photocatalysts, Z-scheme photocatalysts and so on. He also developed a pilot scale solar hydrogen production system with 100 m² light receiving area and hydrogen separation system. His major is physical chemistry, heterogeneous catalysis and surface chemistry. He also worked on infrared spectroscopy of adsorbed species on heterogeneous catalysts, sum-frequency generation (SFG) spectroscopy of adsorbed species. Domen also developed various solid acid catalysts and microporous materials.

Abstract

Sunlight-driven water splitting using particulate photocatalysts has been attracting growing interest as a means of producing renewable solar hydrogen on a large scale¹. A solar hydrogen production system based on 100-m² arrayed photocatalytic water splitting panels and an oxyhydrogen gas-separation module was built, and its performance and system characteristics including safety issues were reported recently². Nevertheless, it is essential to radically improve the solar-to-hydrogen energy conversion efficiency (STH) of particulate photocatalysts and develop suitable reaction systems³. In my talk, the history of water splitting photocatalysts and recent progress in photocatalytic materials and reaction systems for solar fuel production will be presented. In addition to water splitting reaction to form green hydrogen, some other green fuels production will be discussed. Especially, CH₄ formation with CO₂ methanation and NH₃ formation will be discussed⁴.

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Primary Photosynthesis and Problems in Physical-Chemical Biology



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Academician of Russian Academy of Sciences (2022), Chairman of National Committee of Russian Biophysicists, Head of Scientific Council on Biophysics of Russian Academy of Sciences, The member of RAS Council on the Space Biology and Biological Membranes, Laureate of the MV Prize Lomonosov for pedagogical activities of the teachers at Moscow University for 2018, Laureate of the K. A. Timiryazev Prize 2016, Distinguished Professor of Moscow State University (2001), Honored Worker of the

Higher School of the Russian Federation (1997), Laureate of the A. A. Krasnovsky Prize in the field of photochemistry and photosynthesis (1996), Laureate of the Lomonosov Prize of Moscow State University (1992), Laureate of the State Prize (1988). He is the author of more than 900 papers and 46 books.

Abstract

Traditional notions such as concentration, mass action law, temperature dependence of reaction rate constants, stochastic collisions and random distributions of reagents in a reaction volume are not generally valid in heterogeneous internal structures of living cells. We shall discuss some data on the mechanisms and regulation of primary electron transfer in photosynthesis as related to these problems.

Why Bioenergetic Organelles Contain Their Own Genomes and Genetic Systems



John F. Allen

Honorary Professor

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John Allen obtained his PhD at King's College London in 1975. In 1973 he found that superoxide is the initial product of photosynthetic reduction of oxygen in chloroplasts. In 1975 he found that ferredoxin reduces oxygen to superoxide, and superoxide to peroxide. In 1980, as a postdoc at Warwick University and the University of Illinois, he discovered that light-dependent phospho-

rylation of the major chloroplast light-harvesting protein results from activation of its protein kinase by the reduced form of plastoquinone. This redox control provided an explanation for state transitions – how absorbed excitation energy becomes redistributed between the reaction centres of photosystem I and photosystem II. This discovery opened a whole field of research. In the universities of Oxford, Leeds, Oslo, Lund, and Queen Mary University of London, Allen made a number of original contributions, including redox control of chloroplast transcription; its role in organelle genome function; and the origin and evolution of oxygenic photosynthesis. In 2009 Allen was elected a Fellow of the Linnean Society of London.

Abstract

Chloroplasts and mitochondria are subcellular bioenergetic organelles that contain their own genomes and genetic systems. DNA replication and its transmission to daughter organelles produces cytoplasmic inheritance of characters associated with primary events in photosynthesis and respiration. The prokaryotic ancestors of chloroplasts and mitochondria were endosymbionts whose genes became copied to the genomes of their cellular hosts. Those copies that survive are now nuclear, chromosomal genes that encode either cytosolic proteins or precursor proteins imported into the organelle into which the endosymbiont evolved. What accounts for the retention of genes for the complete synthesis within chloroplasts and mitochondria of only a tiny minority of their protein subunits? One hypothesis is that expression of genes for components of electron transport chains must respond to physical environmental change by means of a direct and unconditional regulatory control—control exerted by change in the redox state of the corresponding gene product¹. This hypothesis proposes that, to preserve function, an entire redox regulatory system must be retained within its original membrane-bound compartment together with the DNA upon which it acts. Co-location of gene and gene product for Redox Regulation of gene expression (CoRR)² is an hypothesis in agreement with the results of a variety of experiments designed to test it and that seem to have no other satisfactory explanation³. I present evidence relating to the CoRR hypothesis, and describe a two-component mechanism by which transcription of genes for reaction centre apoproteins is coupled to photosynthetic electron transport in chloroplasts.

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Accessory Pigments: Phycobiliproteins in Three Algal Classes



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Formerly: Smithsonian Inst., RBL, Rockville, MD

President: Phycological Society of America

President: American Society of Plant Physiologists

S. Hales award from ASPP

Fellow of the American Association for the Advancement of Science

Kettering award from ASPP

National Academy of Sciences member 1996

Elisabeth Gantt, a retired university professor, is best known for her work on the discovery and structural elucidation of phycobilisomes in red algae and cyanobacteria. Much of the work was carried out while she was still at the Smithsonian Institution but followed up at the Univ. of Maryland. She has also been active in many other ways serving numerous plant and algal societies, organizing meetings, serving as officers in recruiting new members and serving as an effective female member. She also participated as lecturer in high school science classes and trained some high school students during summer vacation breaks. Gantt's activities further included a semester at Cornell University, by accepting an NSF sponsored program demonstrating successful examples of mature female scientist in male-dominated departments. As a professor in the plant biology at the University of Maryland she was recognized as a very effective teacher and honored by a College of Life Sciences Service Award.

Abstract

The demonstration of phycobilisomes in a red alga (*Porphyridium cruentum*) consisting of a core of allophycocyanin, attached to phycocyanin, and externally to phycoerythrin was consistent with the expected energy transfer of the phycobilin absorption. Although in practice, the arrangement required, and was done by immunoelectron microscopy, on antibodies to purified pigments. In cyanobacteria the basic pigment arrangement turned out to be the same, with the phycobilisomes occurring on the stromal side of thylakoids. In contrast to the cyanobacteria and red algae, the phycobiliproteins in the cryptophyte algae we showed to be in the intra-thylakoidal space.

Innovative Energy Systems for Sustainable Future



İbrahim Dincer

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Ibrahim Dincer is a full professor of Mechanical Engineering at Ontario Tech. University. Renowned for his pioneering works in the area of sustainable energy technologies he has authored/co-authored many books and book chapters, along with many refereed journal and conference papers. Dr. Dincer has chaired many national and international conferences, symposia, workshops and technical meetings. Dr. Dincer has delivered many keynotes and invited lectures. Dr. Dincer is an active member of various international scientific organizations and societies, and serves as editor-in-chief, associate editor, regional editor, and editorial board member on various prestigious international journals. Dr. Dincer currently serves as President for Hydrogen Technologies Association in Türkiye and Chair for Energy Working Group in Turkish Academy of Sciences. Dr. Dincer is a recipient of several research, teaching and service awards, including the Premier's research excellence award in Ontario, Canada. During the past 10 years he has been recognized by Thomson Reuters as one of the Most Influential Scientific Minds in Engineering and one of the most highly cited researchers. During the past 25 years Dr. Dincer's research and activities have been diverse and primarily focussed on sustainable energy solutions, sustainable communities and cities, district energy systems, green buildings, renewable energy technologies, energy storage technologies, hydrogen energy technologies, and waste to energy technologies. His group has developed various novel technologies for commercialization. He is known for his engineering education related talks as a committed educator.

Abstract

Increasing rate of local and global energetic and environmental problems, it has brought us to a level that we need to change the course of action and approach more innovatively to the development of energy solutions in a clean and sustainable manner. These innovative solutions are categorized as renewable energy technologies, alternative fuel technologies, hydrogen energy technologies, energy storage technologies, nuclear energy, waste to energy technologies and integrated energy systems. The presentation will introduce these solutions and provide challenges, opportunities and future directions specific to each of these. The presentation will also dwell on sustainability aspects where energy, exergy, engineering, environment, economy, education and ethics are introduced as key dimensions. These will further be linked to hydrogen age where its ecosystem will be described along with the requirements. A clear outline will then be given about what specific technologies will play a crucial role in local and global economies.

Photodamage and Regulation of Photosynthetic Light Reactions

Eva-Mari Aro

Professor University of Turku, Finland



Eva-Mari Aro is a research director and Professor (Emerita) of Molecular Plant Biology in the Department of Life Technologies, University of Turku, Finland. She was honored as an Academician of Science in 2017 and Commander First Class of the Order of the Lion of Finland in 2019. She was elected a member of the Finnish Academy of Science and Letters in 2002 and Finnish Technical Sciences Academy in 2012. In 2018 Aro was elected as Foreign Associate of the US National Academy of Sciences (NAS), and in 2023 she was awarded a Foreign Fellowship of the Royal Society (LDN).

Abstract

Eva-Mari Aro is known for her contributions to international photosynthesis research on plants and cyanobacteria. She has particularly focused on the diversity of regulation mechanisms that protect the photosynthetic light reactions against inherent, but potentially hazardous, generation of reactive oxygen species. Data accumulated from prokaryotic cyanobacteria to eukaryotic algae, mosses, conifers and flowering plants, together with development of SynBio approaches, is providing us with a toolbox to enhance photosynthesis in living “cell factories” and thereby boost the development of truly sustainable production platforms for chemicals that currently rely on fossil fuels.

A Critical Role of Photosystem I Photoinhibition Under High Light and Subsequent Recovery in Regulating Photosynthetic Electron Flow in Higher Plants

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Photosystem I (PSI) primarily supports linear electron transport (LET), with some electron flow directed to alternative (AET) and cyclic electron transport (CET). While the electron transfer from P700 to ferredoxin via phylloquinone and FeS clusters is well understood, their regulatory role under stress is unclear. We have shown that electron flow to PSI is essential for H₂O₂ accumulation during the photosynthetic electron transport chain, potentially increasing mitochondrial respiration and CO₂ release[1]. H₂O₂ from other sites is rapidly eliminated, limiting its signaling role. Sequential damage to PSI FeS clusters under high light in *Arabidopsis thaliana* is followed by recovery under low light. In wild-type plants, photodamaged FeS_{A/B} clusters do not affect P700 oxidation or CO₂ fixation. However, in *pgr5* mutants, damage extends to FeS_X clusters, impairing P700 oxidation and causing ferredoxin release. PSI FeS cluster damage is regulated under high light, favoring LET and preventing the formation of reactive oxygen species.

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The Lipid Polymorphism and the Energization of Plant Thylakoid Membranes. Facts and Hypotheses

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It is now well established that functional plant thylakoid membranes (TMs), besides the bilayer, contain non-bilayer lipid phases, which mediate the fusion of membranes, secure the activity of the luminal photoprotective enzyme, VDE, and originate from association of lipid molecules with (a) stroma-side protein(s) (Garab et al. 2022 Progr Lipid Res, Dlouhý et al. 2022 Cells, Böde et al. 2024 Photosyn Res). The polymorphic phase behavior of TMs is evidently linked to TMs' major (50%) lipid species, MGDG, which belongs to the class of non-bilayer lipids. Because of the strong segregation propensity of the bulk lipid molecules, the bilayer phase of TMs is comprised of small-sized 'inclusions' between photosynthetic supercomplexes. It has also been shown that the presence of MGDG substantially increases the permeability of the bilayer (Fehér et al. 2023 Photosynthetica). Based on these data, it is inferred that the efficiency of photophosphorylation in TMs depends more on the integrity of protein networks, safeguarded by MGDG, than on a strict impermeability of the bilayer. In line with models advocating for the operation of proton conduction pathways at the membrane interfaces in energy converting biological membranes, and the so-called protet model (Kell 2024 BBA), in particular, we pose the following experimentally testable hypotheses: (i) non-bilayer lipid phases responsible for TM fusions constitute barriers for the proton-conduction pathways along the membrane surface; this may lead to an energetic autonomy of each granum-stroma unit; and (ii) that protons on the luminal side, which according to the protet model, are stored in and can be drained from the protein networks, modulate the quantum yield of chlorophyll-a fluorescence emission of Photosystem II in plant TMs.

X-ray Studies of Photosynthetic Proteins: What We Can Learn About Their Inner Workings from Diffraction and Spectroscopy Experiments at Room Temperature

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Utilizing femtosecond X-ray pulses from XFELs it is possible to obtain “undamaged” spectroscopic and structural snapshots of proteins involved in photosynthetic processes at room temperature. Given adequate reaction triggering options by controlled light flashes these can be collated to a “movie” that shows the sequence of events at the catalytic site necessary for the reaction to take place. We used this approach to record data from different systems, including the purple bacterial reaction center (PBRC), Photosystem I (PS I) and Photosystem II (PS II). In this presentation I will cover recent results on the PBRC and PSI, where we obtained new room temperature structures with the goal of better understanding the factors that govern electron transfer branching in these (pseudo)symmetrical systems. I will also describe our extensive studies on the light driven water oxidation reaction in PS II. We obtained structures at around 2 Å resolution for the four stable states (S_0 to S_3) [1] as well as for time points in the S_2 - S_3 transition [2,3] and the S_3 - S_0 transition [4]. We also performed molecular dynamics simulations to better understand the role of the large network of solvent channels within the PS II structure [5]. In addition we collected X-ray absorption (XAS) and X-ray emission (XES) data for different points in the reaction. The XES data provide kinetic information about the Mn redox changes that can be correlated with structural observations at different time points in the reaction cycle. In addition XAS data can be used to obtain more detailed information about changes in the electronic structure of the metal site. Our combined data indicate the presence of an observable reaction intermediate at around 500 to 1200 μs into the S_3 - S_0 transitions.

We acknowledge funding by NIH NIGMS and DOE BES

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Responses of Light-Harvesting and Energy-Transfer Processes in Typical Cyanobacteria to Light Qualities

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Light-harvesting and energy-transfer systems in photosynthetic organisms are modified depending on the ambient light conditions. In the present study, the modifications in two typical cyanobacteria, *Synechocystis* sp. PCC 6803 and *Synechococcus* sp. PCC 7942, were examined, by measuring steady-state absorption, steady-state fluorescence, and time-resolved fluorescence for the cyanobacterial cells grown under different colored LEDs. The time-resolved fluorescence spectra of *Synechocystis* cells grown under white and red LEDs were very similar to each other, whereas those of *Synechococcus* cells were quite different. From the time evolution of the spectra, it was suggested that *Synechococcus* cells may be less likely to form photosystem I–photosystem II megacomplexes under the red LED. For further discussion, the time-resolved fluorescence spectra were globally analyzed using time constants common to two cyanobacteria, and fluorescence decay-associated spectra were constructed. The result confirmed that the formation of the photosystem I–photosystem II megacomplex was reduced in the *Synechococcus* cells grown under red LED, compared to the cells grown under different colored LEDs. In this talk, responses common and different between the two cyanobacteria will be discussed.

Understanding the Photosensing Mechanism of Chromatic Acclimation in Cyanobacteria

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Cyanobacteria are prokaryotes that perform oxygen-evolving photosynthesis. They utilize a light-harvesting supercomplex, phycobilisome, to transfer the solar energy to the photosystems. Certain cyanobacteria can alter the absorbing wavelength of the phycobilisome in response to green and red light, a process called complementary chromatic acclimation. This phenomenon is controlled by the photosensor protein RcaE, which belongs to a phytochrome-related photosensor family called cyanobacteriochromes (1). Phytochromes bind a linear tetrapyrrole (bilin) chromophore and photoconvert between red-absorbing (Pr) and far-red-absorbing states. Like phytochrome, RcaE binds a bilin but photoconverts between green-absorbing (Pg) and Pr states (2). Using X-ray crystallography and 1D NMR experiments, we revealed the structural basis of the unique green/red light-sensing mechanism in RcaE (3, 4). The bilin chromophore adopts C15-*Z,anti* in Pg and C15-*E,syn* in Pr. This structural change leads to the formation and deformation of the water channel that provides solvent access to the bilin. The desolvation in Pg leads to the deprotonation of the B-ring nitrogen, which leads to the decrease in the bilin p-conjugated system. In phytochromes and other cyanobacteriochromes, the four bilin pyrrole nitrogens are fully protonated. Thus, we revealed a novel absorption tuning mechanism in the “protochromic” green/red photocycle in the chromatic acclimation sensor protein.

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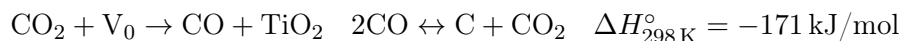
(Artificial) Photosynthesis Is an Oxygen Management Problem

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Oxygen management strategies of the natural and artificial photosynthesis have striking similarities. In the natural photosynthesis, photon absorption is mainly involved in water splitting steps of the reaction producing oxygen as a toxic byproduct. Photogenerated protons and electrons are transported through thylakoid membrane to participate in CO₂ reduction reactions proceeding in dark. We demonstrated that CO₂ to CH₄ reaction can proceed in dark, as long as hydrogen is available in a dissociated form¹, photo-reduced Pt/TiO₂ surfaces can readily adsorb CO₂, and coke formation ensued as a result of the exergonic Boudouard disproportionation reaction readily taking place at room temperature, according to the following reaction scheme:



The spontaneity of the second reaction is confirmed by the appreciable amounts of methane that could be synthesized through the addition of gas phase hydrogen to the reaction chamber via the exergonic methanation reaction $\text{C} + 2\text{H}_2 \rightarrow \text{CH}_4$.

In the presence of a precious metal, oxygen evolution is favored², and the oxygen vacancy density is important in determining the hydrogen evolution yields³. The oxygen evolution characteristics of bulk oxides of Mn (material for oxygen evolution in natural photosynthesis) by solar thermal energy was also investigated⁴. Similar to the natural systems, we reported evidence for the thermal energy that could be generated when charge recombination ensues⁵. These results lead us to investigate the thermodynamic constraints that limits the product yields of artificial photosynthesis. The strategies to overcome kinetic and thermodynamic bottlenecks will be presented.

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Carboxysome-Inspired Compartmentalization of Catalysis

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Bacterial microcompartments (BMCs) such as carboxysomes represent biological modularity in the form of a multienzyme-containing proteinaceous organelle. Bioinformatic analyses have revealed the widespread occurrence of BMCs across the Bacterial Kingdom. The generalized structure of BMCs establishes catalyst proximity and spatial control of local reactant and substrate concentrations, sequesters volatile or reactive intermediates, and controls metabolite and gas exchange with the surrounding environment. Accordingly, BMCs can be viewed as a biological paradigm for spatially confined chemistry. In addition to fundamental studies of the structure and function of BMCs, recent advances in programming and assembling BMCs in vivo and in vitro poise this biological architecture to become a platform for the study spatially confined chemistry.

CO₂-evolving Machinery in the pyrenoid - Structure and Function to Optimize CO₂ Assimilation Efficacy and Light Energy Use in Marine Diatoms

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Marine diatom is a group of eukaryotic algae that accounts for up to 20% of annual global primary production. This massive productivity by diatoms is sustained by the function of their uniquely structured secondary chloroplast. Diatom chloroplast possess globe shaped thylakoid membranes heavily layered at the interior of the chloroplast envelope, which is comprised of outer-most girdle lamellae and inner stroma thylakoid membranes. At the central part of these layered thylakoid system, there is an endo-plastidic organelle, pyrenoid. Pyrenoid is known to be a phase-separated body of Rubisco enzyme interlinked by intrinsically disordered proteins. As unique features of diatom pyrenoid, a protein shell (PyShell) shapes the pyrenoid structure and a couple of layers of thylakoid membranes traverse along the axis of the pyrenoid core, that is denoted as the pyrenoid-penetrating thylakoid (PPT). The PPT lumen possesses a specific θ -Type carbonic anhydrase (CA) and the genome editing disruption of this luminal θ -CA genes resulted in a null CCM phenotype in both marine diatoms, *Phaeodactylum tricornerutum* and *Thalassiosira pseudonana*; that is, these Δ Pt θ CA and Δ Tp θ CA strains exhibited an extremely inefficient photosynthesis that only could saturate at around 10 mM HCO₃⁻, indicating that PPT with luminal θ -CA plays an essential role as a “CO₂ -evolving machinery” for Rubisco condensate. One of unsolved problems of the CO₂ -evolving machinery was the system to provide HCO₃⁻ into the PPT lumen. For this candidate, we discovered multiple chloroplastic transporters belonging to bestrophin (BST) family. These BSTs were localized at either outer stroma thylakoid or peripheral pyrenoid areas. Of these, genome editing KO of outer stroma type PtBST1 resulted in a phenotype of suppressed CCM, indicating a role of the outer stroma thylakoid with BST as a HCO₃⁻ canal towards the CO₂-evolving machinery. Δ PtBST1 also showed a phenotype of moderately enhanced NPQ compared to wild type cells, strongly suggesting that HCO₃⁻ entry to the lumen of the stroma thylakoid mitigates NPQ, presumably by interacting non-enzymatically with luminal proton. These results suggest that the CO₂-evolving machinery in the pyrenoid coordinates CO₂ and light utilization efficiency.

Maintenance and Control of Carbon Fixation in Photosynthesis in a Kleptoplasty

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The euglenozoan flagellate *Rapaza viridis* does not have its inherent chloroplast, but instead exploits a transient organelloid called a "kleptoplast", which is formed by modifying and functionalizing chloroplasts acquired from the green alga *Tetraselmis* spp. It has been shown that the nuclear genome of *R. viridis* has acquired various protein genes related to chloroplast function by horizontal transfer, and it is expected that they are actually expressed (Karnkowska et al., 2023). Therefore, we established a CRISPR/Cas9 genome editing experimental system in *R. viridis* NIES-4477 to verify the function of these proteins biochemically. In particular, we have focused on the RuBisCO enzyme, which includes the large and small RuBisCO subunits (RbcL and RbcS) in addition to the RuBisCO activase (Rca) that is generally required for its function. The kleptoplast initially carries all of these proteins upon establishment in the *R. viridis* cell, but orthogonal RbcS and Rca are no longer expressed because the nuclear genome of *Tetraselmis*, from which the organelloid was originally derived, was eliminated by *R. viridis* at the outset. In the present work, we demonstrate instead that homologous protein genes found in the nuclear genome of *R. viridis* are indeed expressed and targeted to the stroma of the kleptoplast. The kleptoplast initially carries all of these proteins upon establishment in the *R. viridis* cell, but orthogonal RbcS and Rca are no longer expressed because the nuclear genome of *Tetraselmis*, from which the organelloid was originally derived, was initially eliminated by *R. viridis*. In the present work, we show instead that homologous protein genes found in the nuclear genome of *R. viridis* are indeed expressed and targeted to the stroma of the kleptoplast, where the RbcS homologue in particular is critical for photosynthesis by the kleptoplast.

Structure of the Red-shifted *Fittonia Albivenis* Photosystem I

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Photosystem I-light harvesting complex I (LHCI) supercomplex is the basis for higher plants to absorb and utilize far-red light, and the search for higher plants that strongly absorb and utilize far-red light is an urgent problem. In this study, we detected a series of plants which grow in undercanopy or shady environment, where far-red light is abundant, and found that *Fittonia albivenis* has strongly red-shifted fluorescence emission spectrum with the main peak longer than 750 nm, which is red-shifted nearly 30 nm than that of *A. thaliana*. We confirmed that the red-shift of the plant comes from PSI-LHCI, and then separated the PSI-LHCI supercomplex from *Fittonia albivenis*. We solved the structure of PSI-LHCI supercomplex from *F. albivenis* at 2.46-Å resolution using cryo-electron microscopy. The supercomplex contains a core complex of 14 subunits (PsaA-L, PsaN and PsaO) and an LHCI belt with four antenna subunits (Lhca1–4) similar to previously reported angiosperm PSI-LHCI structures; however, Lhca3 differs in three regions surrounding a dimer of low-energy chlorophylls (Chls) termed red Chls, which absorb far-red beyond visible light. The unique amino acid sequences within these regions are exclusively shared by plants with strongly red-shifted fluorescence emission, most of which are belong to the Acanthaceae family, suggesting candidate structural elements for regulating the energy state of red Chls. These results provide a structural basis for unravelling the mechanisms of light harvest and transfer in PSI-LHCI of under canopy plants and for designing Lhc to harness longer-wavelength light in the far-red spectral range.

Keywords: far-red light, LHCI, Lhca3, PSI-LHCI, photosynthesis

Structural Insights into the Assembly of Photosystem II

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Photosystem II (PSII) is a protein machine performing the oxygen evolving reaction in oxygenic photosynthetic organisms. The mature PSII contains a core complex and external light harvesting complexes (LHCs). The PSII core is structurally conserved and assembly of PSII core is a stepwise process involved in different intermediate complexes. On the other hand, the LHCs and supramolecular organization between LHCs and PSII cores are variable, suggesting different photosynthetic organisms have regulated their light harvesting systems to live in different habitats. Here, we isolated PSII assembly intermediates Psb27-PSII and Psb28-PSII from *Thermosynechococcus vulcanus* (*T. vulcanus*), and analyzed their cryo-electron microscopy (cryo-EM) structures. The structures of these two PSII intermediates reveal the binding features of Psb27 and Psb28, which illustrate the functional mechanism of Psb27 and Psb28 in the process of PSII biogenesis. In addition, we solve the structures of PSII supercomplex from cyanobacterium *Acaryochloris marina* (*A. marina*), cryptophyte alga *Chroomonas placoidea* (*C. placoidea*), and the green alga *Chlamydomonas reinhardtii* (*C. reinhardtii*) using single-particle cryo-EM. The PSII from *A. marina* exhibits a tetramer organization including two PSII core dimers associated by eight Pcb proteins. The structure of cryptophyte PSII is organized as a dimer consisting of two PSII core monomers surrounded by six ACPII antenna subunits. The PSII cores of these three photosynthetic organisms are conserved whereas Pcb or ACPII antenna subunits exhibits a unique structural organization. Our structures reveal different supramolecular assembly of core and antenna systems in PSII of these organisms.

Keywords:Photosystem II, Psb27-PSII, Psb28-PSII *Acaryochloris marina*; *Chroomonas placoidea*

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Red-Shifted Chlorophyll *a* Absorption in Algal Light-Harvesting Complexes: Insights from Opto-Magnetic Spectroscopies

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In eukaryotes, light-harvesting is primarily accomplished by a class of light-harvesting complexes (LHCs) exhibiting remarkable sequence and structural homology. In environments where visible light is scarce, organisms have evolved strategies to harvest the residual light, enriched in the far-red portion. While cyanobacteria achieve this through the synthesis of specialized red-shifted chlorophylls (Chls *d* and *f*), some eukaryotes accomplish far-red light absorption using solely Chl *a*, in red-shifted LHCs [1-6]. Since the underlying mechanism by which these red-shifted antennae form far-red forms based solely on Chls *a* remain largely unexplored, we performed a combined electron paramagnetic resonance (EPR) and optically detected magnetic resonance (ODMR) investigation aimed at the characterization of these red-adapted LHCs.

As model systems for these adaptations, we selected the freshwater eustigmatophyte alga *Trachydiscus minutus* and the endozoic alveolate alga *Chromera velia*. In the latter, we found that the pigments responsible for red light adaptation form an excitonic cluster of chlorophylls *a* located at the periphery of the complex. In contrast, in *T. minutus*, we assigned their intense far-red absorption to a excitonic chlorophyll *a* cluster at the core of the complex, near the central carotenoids in the highly conserved L1/L2 sites. We propose that the correct configuration of the interacting Chls *a* in *T. minutus* far-red LHC is obtained thanks to the His-to-Asn substitution in the magnesium-ligating residue of the Chl *a*603 (following the nomenclature of Liu et al. [7]) and to the presence of a small amino acid at the i-4 position from the ligating residue [8]. Phylogenetic analysis of various eukaryotic far-red LHCs identified potential organisms that could share this tuning mechanism based on a conserved [G/A]xxx[N] chlorophyll *a*603 binding motif [8], such as the antenna Lhcf15 in the diatom *Pheodactylum tricornerutum* [9-10].

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Structure of the Far-red Light Utilizing Photosystem II Interacting with Chlorophyll-binding Protein (CBP) from *Acaryochloris Marina* at 2.4 Å Resolution

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Photosynthetic organisms, such as plants and algae, absorb solar energy and convert it into chemical energy by utilizing numerous pigments in the photosynthetic membrane protein complexes (photosystems I and II; PSI and PSII). *Acaryochloris marina* MBIC 11017 (*A. marina*), one of the cyanobacteria, uses chlorophyll (Chl) *d* to absorb far-red light to carry out photochemical reactions [1]. In contrast, many oxygen-evolving photosynthetic organisms utilize Chl *a* to conduct photochemical reactions. Recently, Shen and co-workers reported the structure of *A. marina* PSII interacting with prochlorophyte Chl-binding (Pcb) tetrameric complex at 3.3 Å resolution using cryo-electron microscopy (cryo-EM), and its overall structure and the arrangement of numerous ligands such as Chl *d*, carotenoids, and lipids were identified [2]. However, due to resolution limitations, ligands with weak cryo-EM map signal intensities, such as water molecules in the structure, could not be identified. To investigate the detailed structure and function of the *A. marina* PSII, we analyzed the *A. marina* PSII dimeric structure interacted with light-harvesting complexes at 2.4 Å resolution. We also changed the name of the light-harvesting complex from Pcb to CBP, as proposed by Chen et al. [3]. In this conference, we will discuss the detailed structure and its function, such as the hydrogen bonding networks of Chl *d* and water molecules of the *A. marina* PSII dimer interacting with the eight CBPs.

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The Role of Hydrogen-Rich Water in Enhancing Salt Tolerance in Rice Seedlings

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Salt stress is a major environmental challenge in global agricultural production, severely limiting the growth and yield of important food crops such as rice and wheat. As the area of land affected by salinization continues to expand, finding effective strategies to alleviate salt stress and enhance crop salt tolerance has become a national necessity to ensure food security. In recent years, hydrogen-rich water, due to its unique antioxidant properties and physiological regulatory functions, has been considered a potential new cultivation method to alleviate plant stress. Although numerous studies have reported the beneficial effects of hydrogen-rich water on plant stress, the mechanisms by which it mitigates salt stress in rice are still unclear. Therefore, this study selected rice varieties with different salt tolerances as materials to comprehensively examine their agronomic traits, photosynthesis, and physiological indices related to salt stress, and conducted transcriptomic analysis. Hydrogen-rich water at a concentration of 480 ppb has a significant mitigating effect on rice under salt stress, mainly reflected in rice growth and photosynthesis. The results show that 480 ppb hydrogen-rich water significantly improves the plant growth and dry weight of rice under salt stress, and enhances the seedlings' photosynthetic capacity by increasing chlorophyll content and chlorophyll fluorescence parameters, transpiration rate, and stomatal conductance. In osmotic adjustment, hydrogen-rich water increases the content of soluble sugars and proline, significantly reducing the cell osmotic potential under salt stress. In ion balance, hydrogen-rich water changes the transport and distribution of Na^+ and K^+ in various tissue parts of the seedlings, significantly lowering the Na^+/K^+ ratio in the leaves under salt stress. In oxidative stress, hydrogen-rich water increases the activity of antioxidant enzymes POD, SOD, CAT, and GR, the content of antioxidants GSH and AsA, and the GSH/GSSG ratio, thereby enhancing the antioxidant capacity of rice seedlings under salt stress, while significantly reducing the MDA content. In summary, under salt stress, hydrogen-rich water enhances the physiological regulation of rice by regulating the expression of genes related to osmotic adjustment, ion absorption, and oxidative stress at the transcriptional level, thereby mitigating the inhibitory effect of salt stress on the growth and photosynthesis of rice seedlings.

Keywords: Salt stress; Hydrogen-rich water; Rice; Osmotic regulation; Ion balance.

Immobilization of Cyanobacterium Cells in Alginate Gels to Enhance Hydrogen Production Efficiency

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The extensive use of fossil fuels like petroleum and coal has significantly disrupted global ecological systems, with a notable average annual temperature increase of 1.5°C over the past century. Addressing this challenge requires innovative technologies that harness renewable energy sources. Cyanobacteria, particularly *Synechocystis* sp. PCC6803, have emerged as a viable candidate for sustainable hydrogen production through photosynthesis. However, the efficiency of this process is hampered by the sensitivity of the hydrogenase enzyme to oxygen. To address this limitation, we developed a two-stage cultivation method. This approach combines carbohydrate accumulation under nitrogen-limited conditions with hydrogen production in an anaerobic environment. Additionally, we immobilized cyanobacterial cells in sodium alginate gels to increase cell density, minimize mechanical stress, and improve hydrogen yield. Our study found that immobilized cells achieved a peak hydrogen production rate of 113 nM/mg chl a/h at 264 hours, compared to 96.6 nM/mg chl a/h in non-immobilized cells. Even after 336 hours, immobilized cells maintained a production rate of 46.6 nM/mg chl a/h, demonstrating a more sustained hydrogen evolution. These findings underscore the potential of cyanobacterial immobilization for prolonged hydrogen production, which could be pivotal for the advancement of renewable energy technologies.

Keywords: hydrogen production, cyanobacteria, immobilization, alginate gel, renewable energy

A Straightforward Single-Phase Protocol for Initiating and Sustaining Algal Hydrogen Production

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Green microalgae like *Chlamydomonas reinhardtii* hold a great potential for producing green hydrogen (H₂) using only water and sunlight, with zero carbon emissions. However, to achieve sustainable and high-yield H₂ production, it is essential to effectively manage the hydrogenase enzyme's sensitivity to oxygen (O₂) and optimize the allocation of electrons to this enzyme.

Several well-established methods for prolonging H₂ production include oxygen uptake-based strategies, pulse illumination protocols, substrate limitation, and ambient protocols. In many of these approaches, an initial dark incubation period (typically around 1 hour) before light exposure is considered essential for creating hypoxia and initiating H₂ production. In this study, we employed real-time monitoring of dissolved O₂ (DO₂) in *C. reinhardtii* suspension cultures using a contactless O₂ sensor.

Contrary to common belief, we found that preliminary dark incubation may not be the most effective method for inducing anoxia and initiating H₂ production. Our results show that dark incubation significantly intensifies photosystem II electron transport, leading to a rapid increase in DO₂ levels upon light exposure. This dark adaptation also appears to reinforce photosynthetic control mechanisms.

As an alternative, we demonstrated that anoxia can be established in the suspension culture by applying a brief N₂ flushing of the headspace followed by incubation at 34°C under continuous light at 120 μmol photons m⁻²s⁻¹ with mixing properly. This method established zero DO₂ in the culture (despite over 5% O₂ in the headspace) and maintained it for 10 hours after light onset. Using this protocol, the *pgr5* mutant produced approximately 100 ± 30 mL/L of H₂ within the first 10 hours of light exposure without requiring additional treatments.

Using a continuous gas discharge system under these incubation conditions is anticipated to extend hypoxia in the culture, which may enable a longer duration of H₂ production. Additionally, this effective method for initiating and maintaining H₂ production could be integrated with other established techniques to further improve the duration and efficiency of H₂ production period.

Protective Effects of Hydrogen Against Light-Induced Oxidative Stress in *Synechocystis* SP. PCC6803

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Reactive oxygen species (ROS) are known to cause oxidative damage to cellular components, yet organisms have evolved protective mechanisms to eliminate ROS. However, under intense light exposure, the generation of ROS increases, potentially overwhelming these protective systems, leading to damage of key cellular molecules. Certain cyanobacteria, such as *Synechocystis* sp. PCC6803, produce molecular hydrogen under anaerobic conditions, although its physiological role remains largely unknown. In this study, we investigated the effects of hydrogen water on ROS generation and scavenging in *Synechocystis* sp. PCC6803. Thylakoid membranes and isolated photosystem I complexes were exposed to high light intensity in both hydrogen water and control conditions lacking hydrogen. Additionally, ROS-modulating reagents were introduced to assess the effects of hydrogen on oxidative stress. Spectral analysis post-illumination revealed that chlorophyll degradation was significantly suppressed in hydrogen water compared to the control. Furthermore, the presence of hydrogen was associated with reduced ROS generation as detected by ROS-specific reagents. This presentation will discuss the role of hydrogen in protecting against ROS-induced oxidative damage during high-light stress.

PEM Electrolyzers for Green Hydrogen Production: Current Status and Research Needs

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Proton Exchange Membrane electrolyzer (PEMEL) is a class of electrolyzers which is expected to meet a significant fraction of the growing hydrogen demand in the future. These electrolyzers that use a proton exchange membrane have many advantages over conventional alkaline electrolyzers. The electrolyzer cells consist of an ionomer layer sandwiched between two catalyst layers. The so-called catalyst coated membrane (CCM) is placed between two gas diffusion layers for enabling efficient transport of hydrogen, oxygen and water. Improvements in both ionomer properties including ionic conductivity, mechanical properties, durability, gas permeability and cost are needed. PEMELs are currently more expensive to construct due to the PGM metals including Ir, Ru and Pt used in catalyst layers. The research efforts are directed towards reducing the catalyst loadings and developing non-PGM catalysts. The latter for the anode layer is difficult due the corrosive environment. The GDL properties such as porosity, thickness and tortuosity are also very important in obtaining high operation efficiencies. Efforts are also directed towards increasing the size of the CCMs as much as possible which also increases the diameter of the stacks. With further developments in the areas mentioned above, these electrolyzers will gain market share from alkaline electrolyzers in the future.

Metal Hydrides for Hydrogen Storage, Purification and Compression

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Hydrogen can be used for energy storage and transportation and it is widely considered as a prospective energy carrier and useful tool for development of low-carbon energy and diminishing anthropogenic impact on Earth's climate. The most environmentally friendly hydrogen production methods include water electrolysis on the base of nuclear and hydro power stations and renewable sources, and biomass conversion, including biohydrogen production. In low-emission energy systems hydrogen could be used in mixtures with methane (energy mixtures) and/or carbon dioxide (biohydrogen) with low concentration (less than % H_2). Extraction of hydrogen from these mixtures, compression and storage are complex and unsolved problems, since the state-of-the-art commercial hydrogen purification systems are optimized for separation of hydrogen from mixtures with high concentration (more than 70% H_2) and lose efficiency if hydrogen is the minor component. Metal hydrides selectively absorb hydrogen and can be used for hydrogen storage, purification and compression. We present results of our project, aimed to investigate and develop fundamental basis for sustainable production of purified low-carbon hydrogen from mixtures with methane and/or carbon dioxide including biologically produced mixtures by metal hydrides. Pressure of the feed should be less than 1 MPa with partial hydrogen pressure less than 0.2 MPa. Hydrogen is purified by metal hydrides with recovery higher than 70% and stored in metal hydrides with the use of low potential (less than 100 °C) heat. The product hydrogen should be pure and ready-to-use in PEM fuel cells with pressure higher than 1 MPa to serve as a first stage of further hydrogen compression up to 35-70 MPa.

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Unraveling 3.5 Billion Years of Co-Evolution Between Photosynthesis and Bacteria

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The origin and evolution of photosynthesis have transformed the relationship between life and Earth. This leads to a flux of organic matter and oxidants to the Earth's surface and forms a biosphere dominated by heterotrophy and respiration. However, how life facilitated this transition - that is, the evolutionary process of photosynthetic organisms themselves - is still unclear. In this study, we used a large amount of sequence information (covering more than 10,000 genomes) to conduct a systematic comparison of bacteria and all photosynthesis-related proteins, and identified evolutionary relationships between organisms (bacteria) and chlorophototrophy, thereby clarifying the intertwined evolutionary path of photosynthesis. We identified the Last Phototroph Common Ancestor (LPCA) of all extant photosynthetic organisms and found a gap between the origin of the oxygen-evolving ability and the origin of cyanobacteria. We demonstrated that photosynthetic ability was acquired through vertical inheritance from the LPCA in the Terrabacteria lineage, and through horizontal transmission in other lineages. The results of the analysis also showed that photosynthesis had already emerged before the LPCA, that is, at the stage of the common ancestor of Bacteria. Taken together, it is clear that a detailed view of the coevolution of photosynthesis and Bacteria is consistent with the geological record[1].

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Evolution and Function of the Light-Harvesting Complex in Red-Lineage Algae

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Red-lineage algae, which have acquired plastids through successive endosymbioses from red algae, include cryptophytes, heterokonts (photosynthetic stramenopiles such as diatoms), haptophytes, and dinoflagellates. These taxa account for the majority of primary production in modern oceans. Red-lineage algae consistently use the three-helix light-harvesting complex (LHC) family proteins as the major light-harvesting antenna for photosystem (PS) I, while they have used LHCs for PSII after the endosymbiosis event. Recent advances in cryo-electron microscopy have revealed the structures of PSI–LHCI and PSII–LHCII supercomplexes in red-lineage algae. We have performed both taxon-rich and sequence-rich molecular phylogenetic analyses of LHCs using recently reported genomic and transcriptomic data. By integrating these analyses with structural models, the molecular evolution of LHCs in red algae and red-lineage algae can be estimated [1]. We propose LHCs in red-lineage algae are classified into the distinct subfamilies of Lhcr, Lhcz, Lhcq, Lhcf, Lhcx, the homolog of *Chaetoceros gracilis* Lhcr9, and RedCLH [2]. The molecular function of LHCs will be presented with a special focus on LHCs in diatoms.

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Flexible and Heterogeneous Regulation of Photosynthetic Light Harvesting Revealed by Single-Molecule Spectroscopy

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Photosynthetic reactions are driven by sunlight. Light capture occurs in light-harvesting antenna complexes, followed by photoelectric conversion in reaction center (RC) proteins. The near-unity quantum yield of the photoreaction process is highly attractive, prompting extensive studies on its regulatory mechanisms. Structural information provided by X-ray crystallography and cryogenic electron microscopy enables the interpretation of spectroscopic data and the construction of theoretical models based on precise architecture. This has led to a general consensus that the conformation of biological systems responsible for the photoreaction is highly optimized at the angstrom level. However, nuclear coordinates are not static but thermodynamically perturbed at physiological temperatures, as visualized by molecular dynamics simulations. Additionally, the conformation exhibits significant heterogeneity. These factors raise new questions about how dynamic and heterogeneous properties contribute to and optimize the photoreaction. To address this question, we have applied single-molecule spectroscopy, which allows the identification of small but significant conformational variations through spectroscopic inhomogeneity and temporal spectral changes. Furthermore, we have developed single-molecule transient absorption spectroscopy to directly investigate how conformational heterogeneity and dynamics perturb the ultrafast photoreaction process, which is inaccessible with conventional fluorescence-based single-molecule spectroscopy. Our approaches provide an opportunity to understand the importance of flexibility and heterogeneity in the photosynthetic light-harvesting function.

A Role of Protein Matrix in Superexchange Electron Transfer in Charge Separation

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In type II reaction centers, such as photosystem II (PSII) and purple bacterial reaction centers (PbRC), the process of light-induced charge separation involves electron transfer from pheophytin (Pheo) to quinone (Q_A). This electron transfer occurs close to a conserved tryptophan residue, D2-Trp253 in PSII and Trp-M252 in PbRC, which is positioned near both Pheo and Q_A in the active branch. As the tryptophan likely forms a $\pi - \pi$ stacking interaction with Q_A , it was proposed that the tryptophan residue is electronically coupled to the electron donor site and may mediate “superexchange electron transfer” between Pheo and Q_A [1]. In this study, we examined the electron transfer pathway from Pheo to Q_A , with a focus on the superexchange interaction within the PSII protein matrix, using quantum mechanical/molecular mechanical approaches [2]. Our results show that the calculated superexchange coupling for the Pheo to Q_A electron transfer, mediated by the unoccupied molecular orbitals of D2-Trp253 (with a $[\text{Trp}]^{\bullet}$ -like intermediate state), is substantially larger than that of the direct electron transfer without a superexchange mediation. This indicates that superexchange is the primary mechanism driving the electron transfer in the PSII protein matrix. We also discuss the role of the protein matrix in the superexchange electron transfer.

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Oxygen Sensitivity of Photosystem I Photoinhibition in Land Plants: Land Plants Activate Protective Mechanisms on PSI Photoinhibition Depending on the Growth Light Environment

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Light energy is an essential driving force for photosynthesis in land plants. However, the excess light energy can cause harmful effects by the generation of reactive oxygen species (ROS), especially on photosystems embedded in the thylakoid membranes, named photoinhibition. Toward excessive light energy, photosynthetic organisms activate several types of protective mechanisms such as non-photochemical quenching or optimization of light-harvesting systems qualitatively and quantitatively. However, these protective mechanisms are often referred to in photosystem II. On the other hand, it remains elusive whether ROS-mediated PSI photoinhibition can be activated by sensing the light intensity under growing conditions, and whether the protective mechanisms can protect PSI from its photoinhibition.

Here, we investigated the dependency of the O₂-dependent PSI photoinhibition by using intact rice leaves grown under different growing light conditions. We found that the degree of O₂-dependent PSI photoinhibition linearly increased in response to the increase in O₂ partial pressure, but the rice plants grown under the higher photon flux density showed higher robustness to the PSI photoinhibition. These results suggested that rice plants can activate the photoprotective mechanisms of ROS-dependent PSI photoinhibition by light acclimatory responses.

PSI-SMALP: All the Fat But Hold the Soap

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Since Dowhan, Wickner, and Kennedy's pioneering work in the 1960s/1970s utilizing nonionic detergents from the industrial sector (Cutscum, Rohm and Haas) to extract membrane-bound enzymes for lipid biosynthesis, membrane biochemists have continuously improved detergent methods for membrane purification. Dodecyl maltoside (DDM) has become the standard detergent for isolating membrane proteins, yielding structures predominantly determined in DDM micelles. However, in recent years, Styrene Maleic Acid/Lipid Particles (SMALPs) have emerged as a novel method for membrane protein isolation. Unlike nanodiscs, SMALPs employ an amphipathic polymer to directly extract proteins from native membranes, preserving the lipid environment. While SMALP isolation has been applied to phospholipid-rich membranes, research on galactolipid-rich membranes like thylakoids has been limited. Over the past five years, our group has developed novel polymers enabling high-yield isolation of PSI and other thylakoid membrane proteins, synthesizing over 60 polymers to explore structure-function relationships in polymer properties and protein solubilization. We have successfully isolated PSI-SMALPs from various thermophilic cyanobacteria, characterized by approximately 1200 lipids per PSI trimer, notably enriched in anionic SQDG. Femtosecond spectroscopy reveals ultrafast (<100 fs) charge separation in detergent-free PSI-SMALPs, hinting at an ultrafast pathway potentially disrupted during detergent isolation. This research explores how the SMA method can enhance our understanding of photosynthetic proteins in a native context.

Effect of Trehalose on the Electron Transfer Chain of Photosynthetic Bacteria

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It has long been known that disaccharides used as co-solutes are efficient stabilizing agents of biological macromolecules. Among these, trehalose, a disaccharide formed by two glucose units joined by 1-1 glycosidic bond, has proved in many cases particularly effective. In this work we studied the effect of trehalose on the electron transfer reactions that occur in the cytochrome (cyt) bc_1 complex of the photosynthetic bacterium *Rhodobacter sphaeroides*. The study was performed using chromatophores, sealed photosynthetic membrane vesicles that are easily isolated from the bacterium through mechanical fractionation. In the presence of 0.8 M trehalose, following activation of the primary photochemistry via a single turnover flash of light, a notable slowdown of the electrogenic reactions related to the photo-induced activity of the cyt bc_1 complex is observable. In particular, the kinetics of the third phase of the electrochromic carotenoid shift, due to the electrogenic events linked to the reduction of cyt b_H heme via the low potential arm of the cyt bc_1 complex and its re-oxidation by quinone molecules at the Q_i site of the complex is about 4 times slower in the presence of trehalose. In parallel, the re-reduction of oxidized cytochromes c_2 is strongly slowed down, as well as the reduction of cyt b_H heme of the cyt bc_1 complex in the presence of the inhibitor antimycin. Preliminary data indicate that trehalose at high concentrations permeates the membrane of chromatophores. These observations suggest that the disaccharide at high concentrations interferes with the electron transfer reactions of the high-potential arm of the bc_1 complex. The possible mechanism by which trehalose exerts its effect is discussed in terms of the propensity of trehalose to form strong hydrogen bonds with its surroundings.

Acclimation of Photosynthetic Processes in Wheat Leaves to High Temperature

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High temperature represents an important stress factor with increasing severity due to climate change. Several components of photosynthetic machinery were found to be heat-sensitive, such as photosystem II or Calvin cycle enzymes. However, the responses to heat stress are very diverse and specific for different plants. Moreover, exposure to high temperatures may lead to acclimation at various levels, resulting in enhanced heat tolerance. In a series of experiments with diverse wheat genotypes of different origins, we examined the photosynthetic rate, carboxylation efficiency, regulation of electron transport, redox states of the PSI and PSII, and PSI inactivation in parallel with other photosynthetic parameters in non-stressed and heat-stressed conditions, including analyses of acclimation resulting from long-term exposure to elevated temperatures. Heat stress led to changes in redox states of the PSI donor and acceptor side, associated with alterations in transthylakoid proton gradient (ECS_t), proton conductance (gH^+), and non-photochemical quenching (NPQ). We observed acclimation at the level of PSII thermostability, with significant genotypic differences associated with the area of origin. In addition to PSII, we also found damage at the PSI level. The period of high temperatures resulted in decreased PSI activity in sensitive but not in tolerant genotypes. The genotypes also differed in their recovery after heat stress relief; a poor recovery was associated with an overly reduced acceptor side of photosystem I and a high membrane potential in the chloroplast. In conclusion, our results confirmed the capacity of crop species to acclimate the photosynthetic processes and structures to heat stress. We identified a key role of regulation of electron transport and PSI protection in heat stress acclimation and recovery. In wheat germplasm, we identified significant genotypic variability in responses to evaluated heat stress scenarios, including differences in photoprotective mechanisms employed by different genotypes or groups of genotypes.

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Characterization of the possible acclimation strategies and remodeling of photosystems under long-term hypersaline conditions in *Dunaliella salina*

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Dunaliella (D.) salina is a representative of highly salt-tolerant green algae and is well known to show superior tolerance against abiotic stresses. Hence it is an effective model organism to study various acclimatization mechanisms adapted by photosynthetic organisms under high salinity. Apart from salt tolerance and photosynthetic efficiency, *D. salina* is a promising microalga worldwide for commercially producing an enormous amount of β -carotene. Transcriptomic study of long-term acclimated cells given the idea of a possible remodeling at the supercomplex level with the addition of chloroplast TIDI, flavodoxin IsiB, and CBR proteins. Also, the transcriptomic study revealed the upregulation of the tetrapyrrole biosynthesis pathway (TPB) and identified the presence of a negative regulator of this pathway, called the s-FLP splicing variant. These observations point towards the accumulation of TPB pathway intermediates PROTO-IX, Mg-PROTO-IX, and P-Chlide, those earlier reported as retrograde signaling molecules. These two major findings from the RNA seq study suggested a possible remodeling of the photosynthetic apparatus and an efficient retrograde signaling mechanism in the chloroplast under hypersaline conditions for acclimation. Hence, we have fetched out the remodeling and other acclimation strategies at the photosystem supercomplex level, especially emphasizing chloroplastic protein expression and photosystem supercomplex characterization in hypersaline conditions compared to control. To study the involvement TPB pathway we analyzed Chl a fluorescence after aminolaevulinic acid (ALA) treatment and showed a significant increase in photosynthetic efficiency (Fv/Fm) after 6 hrs of treatment with 1mM ALA in hypersaline grown cells. However, there was no significant change in photosynthetic efficiency in control cells after treatment. This suggests that the enhancement in the TPB pathway in hypersaline conditions leads to positive regulation of photosynthesis. Photosystem supercomplex characterization has been done using BN-PAGE analysis and sucrose density gradient separation of complexes. BN-PAGE analysis of thylakoid sample isolated from 1.5M, 2M, and 3M NaCl conditions on day 3 and day 4 showed the stable photosystem supercomplexes in hypersaline conditions and this is correlating with the enhanced photosynthetic efficiency observed in chlorophyll fluorescence analysis. Sucrose density gradient separation of photosystem supercomplexes using the thylakoid sample followed by SDS-PAGE analysis of fractions and Western blot analysis of D1, PsaA, Lhcb2, and CP47 proteins from different fractions showed the effects of hypersalinity on LHC proteins is more significant than that of the core proteins. However, the supercomplex studies suggested that *Dunaliella salina* maintains stable photosystems even in hypersaline conditions as compared to optimum salt conditions.

Keywords: *Dunaliella salina*, Chloroplast retrograde signaling, TPB pathway, Photosystems.

Towards Understanding Sustained Thermal Dissipation Mechanisms in Overwintering Evergreen Leaves

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Plant photosystems are generally sensitive to low temperatures. However, evergreen species in the cryosphere have evolved mechanisms to avoid damage and maintain photosystem function even in cold conditions. This resilience is largely due to a significant decrease in photosystem II (PSII) activity during winter, a phenomenon known as “sustained thermal dissipation,” which allows most absorbed light energy to be dissipated as heat. To elucidate the molecular mechanisms behind this phenomenon, we investigated annual changes in the fluorescence yield of PSII (YII), transcriptomes, photosystem complexes, and photosynthetic pigments in an evergreen conifer and an evergreen dicotyledonous plant: *Taxus cuspidata* (yew) and *Euonymus fortunei*. We observed a massive induction of early-light induced protein (ELIP) in winter in both species, with their levels comparable to those of light-harvesting complexes. Concurrently, time-resolved chlorophyll fluorescence analysis revealed rapid heat dissipation at the antenna of PSII during winter. In yew, the accumulation of PSII was only slightly reduced in winter, while in *E. fortunei*, it was markedly reduced. These results suggest two possible molecular mechanisms for the decrease in PSII activity in winter: a reduction in the PSII level and an increase in sustained thermal dissipation. By purifying ELIP from *E. fortunei*, we confirmed that this protein binds chlorophyll *a* and a few carotenoids. Time-resolved chlorophyll fluorescence analysis showed that ELIP is capable of dissipating excitation energy from chlorophyll. The possible roles of ELIP in sustained thermal dissipation will be discussed in this presentation.

Restriction of Photorespiration Raises ROS Levels and Their Relationship Depends on the Robustness of Redox Regulation in Leaves

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Photorespiratory metabolism is upregulated under oxidative stress due to increased elevated ROS levels and in turn restricts ROS levels. We point out photorespiration's inverse relationship with leaf ROS levels. The patterns of key photorespiratory enzymes and ROS levels in *Arabidopsis* leaves under low O₂ or treatment with and aminooxy acetic acid (AOA, a photorespiratory inhibitor) and exposure to dark, medium or high light (HL). The photorespiratory enzymes, such as glycolate oxidase, catalase, and phosphoglycolate phosphatase, were downregulated. An increase in ROS levels (superoxide and H₂O₂) reflected the induction of oxidative stress. We emphasize that photorespiration helps to minimize ROS levels, while restricted photorespiration leads to an increase in ROS and oxidative stress.

We also tried to assess the consequences of oxidative and photo-oxidative stress induced by menadione and HL) on the photorespiratory metabolism in mutants deficient in redox-balancing components of *Arabidopsis thaliana*. Maximum increase in ROS levels and a decrease in ascorbate was noticed in *vtc* mutant, which may be due to the deficiency of ascorbate. The extent of oxidative damage was minimal in the *nadp-mdh* and *aox1a* mutants, which seem to be primed against oxidative stress. The photorespiratory enzyme components in the WT and mutants increased concurrently with ROS levels under HL. We propose that the modulation of photorespiration under stress depends on the robustness of redox-balancing components in leaves.

Chromatic Acclimation and Photosynthetic Fitness in Cyanobacteria

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Cyanobacteria are photoautotrophic organisms that use light and water as a source of energy and electrons, respectively, to fix atmospheric carbon dioxide and release oxygen as a by-product during photosynthesis. However, photosynthesis and fitness of organisms are challenged by seasonal and diurnal fluctuations in light environments. Also, the distribution of cyanobacteria in a water column is subjected to changes in the light regime. The quality and quantity of light change significantly in low and bright light environments that either limit photochemistry or result in photoinhibition due to an excess amount of light reaching reaction centers. Therefore, cyanobacteria have to adjust their light-harvesting machinery and cell morphology for the optimal harvesting of light. This adjustment of light-harvesting involves remodeling of the light-harvesting complex called phycobilisome or incorporation of chlorophyll molecules such as chlorophyll *d* and *f* into their light-harvesting machinery. Thus, photoacclimation responses of cyanobacteria at the level of pigment composition and cell morphology maximize their photosynthetic fitness under a dynamic light environment. Cyanobacteria exhibit different photoacclimation responses commonly known as chromatic acclimation (CA). *Fremyella diplosiphon* is a well-known model organism to study the process of CA which is mediated by the phytochrome-related photosensor protein RcaE. *F. diplosiphon* can alter the pigment composition of its major light-harvesting complex, i.e., phycobilisomes, together with cellular morphology in response to changes in quality and quantity of light. In red light (RL) enriched environment, which prevails in bright light at the surface of the aquatic system, the phycobilisomes consist of RL-absorbing pigment, i.e., phycocyanin, and the organism possesses spherical cellular morphology. However, in a green light (GL) enriched environment, which prevails in low light conditions/shady places at a lower depth in an aquatic system, the phycobilisomes consist of GL-absorbing pigment, i.e., phycoerythrin, and the organism possess extended rod shape cellular morphology. This alteration in cellular morphology and reshuffling in pigment composition of phycobilisomes allows *F. diplosiphon* to efficiently harvest the available wavelengths of RL and GL to increase its photosynthetic fitness. Recent advances in the mechanistic insight of cell shape alteration have indicated that photosensor RcaE is equally important together with cytoskeleton proteins. My presentation will focus on the molecular mechanism of well-known type 3 CA. I will include other aspects of type 3 CA that have been recently studied at a molecular level and highlight the importance of morphogenes, cytoskeleton, and carboxysome proteins. Similarly, *S. elongatus* alters its cellular morphology in response to different colors of light; GL and blue light (BL) cause cell elongation while RL induces shortening of cells. *Oscillatoria sp.* Malviya1 can use GL and blue light (BL) to drive photosynthesis, and it is a unique feature among cyanobacteria as these organisms are known for their inability to use BL. In summary, my presentation will include recent developments on the mechanistic insight of the light-dependent developmental process that increase cyanobacterial fitness in a given light condition.

Protein Dynamics Mediates the Structural Adaptation of the Orange Carotenoid Protein in Cyanobacterial Photoprotection: A Neutron Scattering Study

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The orange carotenoid protein (OCP) bears great importance in the photoprotection of cyanobacterial light-harvesting antennae and is known to undergo a structural change from its dark-adapted to the active state upon photoactivation. Here, we report a solution structure of the active state differing significantly from that observed upon binding to the phycobilisome antenna and, thus, requires a structural rearrangement of OCP. Here, we focus on the role of internal protein dynamics in enabling the latter structural transition and, thus, to ensure the functionality of OCP. We use quasielastic neutron scattering to directly probe the protein dynamics of OCP in a wide observation time range from ~ 750 picoseconds to ~ 100 nanoseconds. Measurements in the dark and using actinic blue light reveal the dynamics in the dark-adapted and active states of OCP, respectively. It is shown that the internal protein dynamics is enhanced upon photoactivation, while the global diffusion of OCP is slowed down. Molecular dynamics simulations permit a residue-resolved understanding of the experimental data indicating that the β -sheets of the C-terminal domain remain the most rigid part of the OCP in its active state. We assume that this stability is essential for the dimerization of the protein, which occurs via interaction between the β -sheet surfaces. In contrast, the increased flexibility of other parts of OCP can be a crucial prerequisite for the structural adaptation necessary for binding to the phycobilisomes.

How Manganese Compounds Facilitate Water Oxidation in Artificial Photosystems

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The water-oxidation reaction (WOR) is a key challenge and a slow process in water splitting [1]. Manganese oxides have shown promise for WOR due to their stability, low cost, and eco-friendly nature. Notably, plants, algae, and cyanobacteria utilize a CaMn_4O_5 cluster for WOR [2,3]. This cluster resides within Photosystem II, a membrane-protein complex that acts as a light-driven water oxidase in oxygenic photosynthesis, effectively oxidizing water at low overpotentials. As a result, manganese compounds are of significant interest as potential catalysts for WOR in artificial photosynthetic systems [4,5]. Recent advances in characterization techniques and spectroscopic methods have provided deeper insights into the mechanisms of WOR in the presence of manganese compounds. In this study, we present our findings on the transformation of manganese compounds into active catalysts during WOR. We demonstrate that many manganese compounds undergo conversion into manganese-oxide-based catalysts during or before WOR, which serve as the true catalysts for the reaction. Additionally, some biomimetic models of the CaMn_4O_5 cluster decompose during WOR. For many manganese compounds, the leaching of Mn(II) or Mn(III) into the electrolyte, followed by deposition of these ions, leads to the formation of MnO_x , the active catalyst.

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Regulation of the Intersystem Electron Transport in Plant Chloroplasts *in Situ* and *in Silico*

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In this study, we investigated the intersystem electron transport in chloroplasts *in situ* in the leaves of shade-tolerant and light-loving plant species, *Tradescantia* (*T. fluminensis* and *T. sil-lamontana*) and *Cucumis* (*C. sativus* and *C. melo*) genus. Plants were acclimated to moderate light (ML, 50–125 μmole) or high light (HL, 850–1000 $\mu\text{mole photons m}^{-2} \text{ s}^{-1}$) conditions. Photosynthetic activity of the chloroplast electron transport chain (ETC) was assayed by measuring chlorophyll (Chl) *a* fluorescence and redox transients of photosystem I reaction centers (P₇₀₀). Acclimation of plants to ML or HL irradiation influenced the Chl(*a/b*) levels and expression of the PsbS and Lhcb1 proteins. Shade-tolerant species of both genus revealed a high sensitivity to variations of light intensity during acclimation. In HL-leaves, we also observed a lag-phase in the kinetics of P₇₀₀ photooxidation that was attributed to cyclic electron transport around PSI. Enhanced expression of PsbS and rapid response HL-leaves to dark-to-light transitions provide a high sustainability of their photosynthetic apparatus under fluctuating light. Other structure–function relationships associated with regulation of oxygenic photosynthesis are briefly outlined: *a*) pH-dependent control of electron transport, *b*) a balance between the linear and cyclic ETCs, and *c*) species-dependent variability of photosynthetic electron transport upon plant acclimation to high and low light conditions.

In the context of structure-function regulation of electron transport in chloroplasts, we have modelled, using DFT calculations, the oxidation of PQH₂ by the Cyt b₆f complex. This bifurcated (two-electron) reaction is the rate-limiting step in the chain of the intersystem ETC. Our theoretical analysis has demonstrated that the overall rate of PQH₂ turnover is determined by the first step of PQH₂ oxidation at the catalytic site Q_o - the uphill electron transfer from PQH₂ to the Fe₂S₂ cluster of the high-potential Rieske protein. The second step of PQH₂ oxidation is associated with a rapid downhill (exergonic) plastoquinone (PQH•) oxidation by the low-potential heme of the Cyt b₆.

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Algal Hydrogen & Biomass Farming

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In view of the increasing demand for biofuels and sustainable chemical feedstocks, we develop a new approach in agriculture, aiming to harness microalgae. In stark contrast to land plants, these microbes can be harvested daily and produce hydrogen and nutritious super food biomass. Photobiological Hydrogen (H_2) production from green microalgae holds great promise for sustainable production of a clean, zero carbon footprint fuel. However, in nature, the process of H_2 production is temporary, lasting for 2 minutes. Thereafter, it ceases due to electron loss to competing processes, mainly the Calvin cycle, and later on, due to an accumulation of inhibitory concentrations of oxygen. Recently, we found that a *Chlamydomonas* mutant in the Proton-Gradient-Regulation-Protein-5 (*pgr5*) gene harbors faster respiration and a slower Calvin cycle allowing scalable (culture volumes of 10L) continuous production of H_2 under ambient mixotrophic conditions for a duration up to 20 days. This achievement allows engineering studies which focus on scale up of the process. However, while the *pgr5* bypasses the oxygen sensitivity and electron loss to competing processes. Our recent discovery reveals that in anoxic cultures of green microalgae, the main limitation for maximizing hydrogen production rates lies in an anoxic switch occurring in photosystem II. Upon a 20 seconds of light exposure, we observed a significant three-fold reduction in the photosynthetic electron flow, which was not recovered to its original rate. We found that a notable alteration in the activity of photosystem II, leading to a three-fold decrease in electron output. This discovery represents a significant paradigm shift, challenging the prevailing notion that the rate-limiting control of photosynthesis is determined by changes in electron transfer rate within the Cytochrome b_6f complex. This down-regulation process, potentially involving the photoreduction of O_2 at the acceptor site of PSII and an alternative conformation of its acceptor site residue arrangement, ultimately translates into a remarkable reduction in H_2 production. Consequently, this sets a new challenge and engineering target PSII itself, for further exploration. Overall, this research not only challenges the existing paradigm but also opens up new avenues for investigation and engineering interventions to enhance hydrogen production in green algae.

Pulses and Flashes to Probe the Photosynthetic Electron Transport Chain and the Nature of Fluorescence

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Light Curves can show us how Chl *a* fluorescence and the redox states of PC, P700 and Fd change as a function of the light intensity. Considering the step length as well, information is obtained on the response time of the electron flow to changes in the light intensity. At the same time, the electron flow through all photosystem (PS) IIs as derived from fluorescence measurements should under steady state conditions equate the electron flow through all PS Is. However, ETR(I) (electron transport rate through PS I) is almost always higher than ETR(II). If we reject cyclic electron transport (CET) as explanation, how can we explain this difference then? And if we cannot derive information on CET from Light Curves, what can far-red Light Curves tell us in that respect? Light Curves give, at higher light intensities, a linear correlation between the parameter qP (photochemical quenching) and the redox state of P700. What does this correlation tell us about the meaning of the parameter qP? Looking at time resolved saturation pulses under different actinic light conditions, is the quenching then homogeneous, affecting the photochemical and photothermal phase to the same extent? And what happens with O-I₁-I₂-P transients if the electron flow between cyt b₆f and PS I is affected by either phosphor or copper deficiency? With such measurements it is possible to get some grip on the nature of the thermal phase, without knowing its exact nature. If the pulses are replaced by μ s-flashes, is it then easier to obtain information on this topic? Making use of the measurement of time-resolved fluorescence induced by μ s flashes, this point will be discussed.

Current Understanding of Color-tuning in Bacterial Photosynthesis

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Photosynthesis is a fundamental process that captures solar energy, sustaining life across Earth's ecosystems. A crucial aspect of this process is color adaptation, which allows photosynthetic organisms to thrive in a wide range of ecological niches. While various factors have been implicated in the spectral tuning of photosynthetic chromophores—such as protein electrostatics, macrocycle ring deformation, the orientation and dynamics of macrocycle functional groups, axial ligation, hydrogen bonding, and exciton effects—a comprehensive understanding of this phenomenon remains elusive.

In this study, we provide an overview of color adaptation in a specific group of phototrophic organisms: photosynthetic purple bacteria, which exhibit an impressive 160 nm tuning of their absorption spectra in the near-infrared region. Using a basic disordered exciton model, we demonstrate that this spectral tuning arises from the combined effects of exciton shifts and site energy shifts. These findings provide a broader understanding of the mechanisms driving bacterial color adaptation, offering potential inspiration for the development of sustainable energy solutions and innovative technologies.

Inter-Subunit Energy Transfer in Photosynthetic Supercomplexes Observed by Two-Dimensional Electronic Spectroscopy

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Photosynthesis as a fundamental biological process employs a conserved biochemical machinery including the photosystem I and photosystem II reaction centres that share a high degree of homology among all oxygenic phototrophs from cyanobacteria to higher plants. Nevertheless, photosynthetic organisms have evolved remarkably diverse light-harvesting antenna systems adapted and tailored to the specific living conditions in each possible habitat. The light-harvesting complexes increase the absorption cross-section of the photosystems and actively and dynamically regulate the excitation flow to the reaction centres balancing efficient energy capture and photoprotection. In recent years, the structures of many different antenna-reaction centre supercomplexes have been determined potentially revealing different strategies and mechanisms ensuring efficient light harvesting¹. Ultrafast time-resolved spectroscopy and more specifically two-dimensional electronic spectroscopy (2DES) is a powerful tool to probe the excitation energy transfer from the antenna to the reaction centre². Here we have employed 2DES to investigate the dynamics of excitation energy transfer in several types of multisubunit antenna and antenna-reaction centre supercomplexes isolated from plants³, cyanobacteria⁴ and algae⁵. A comparison between supercomplexes from plants and the diatom *Thalassiosira pseudonana* revealed markedly different antenna organizations and energy transfer dynamics. Rapid energy transfer on timescales of a few picoseconds was measured between the fucoxanthin-chlorophyll protein (FCP) complexes and both photosystems II and I, suggesting a previously underappreciated functional aspect of the FCPs antenna system. Despite structural and dynamic differences, a universal cross-species strategy for light harvesting is proposed as a blueprint for artificial energy-converting systems.

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Study of Water-Soluble Sugars and Phosphoenolpyruvate Carboxylase in Photosynthetic and Non-Photosynthetic Organs of Wheat Cultivars With Contrasting Drought Tolerance

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Abiotic stressors that impair photosynthetic electron transport and photophosphorylation, such as drought and high temperatures, have a negative impact on the metabolic processes of photosynthesis. These factors pose a threat to global food security, and understanding metabolic pathways is crucial to the creation of stress-tolerant cultivars. Consequently, one of the key elements that significantly influences plant metabolism is the metabolism of sugars. Outside of the Krebs cycle, organic acids generated in the presence of phosphoenolpyruvate carboxylase (PEPC) play a different role. By promoting the conversion of PEP to malate and other organic acids, PEPC also has a physiological role in the redox balance of the NAD(P)H/NAD (P)⁺ ratio.

In photosynthetic and non-photosynthetic organs of drought-exposed durum and bread wheat genotypes with contrasting drought tolerance during different developmental phases of the grain (milk ripeness, wax ripeness, and full ripeness), a comparative study of water-soluble sugars, PEPC, as well as proline has been conducted. In well-watered variations of the drought-tolerant Barakatli 95 and Gobustan genotypes, the level of soluble sugars was comparable to values seen in variants subjected to drought during all investigated developmental phases. In the milk ripeness and wax ripeness phases, the amounts of soluble sugars in both variants of the drought-sensitive Garagylchyg 2 and Tale 38 genotypes were similar. However, in the full ripeness phase, this parameter was higher in the drought-exposed variants compared to the watered variants, and this increase was 17.5%, 37.14%, 21.7%, 42.3%, and 33.6% in the leaf, sheath, stem, awn, and grain, respectively. The amount of soluble sugars in the stem was greater than in the sheath and awn in the full ripeness phase of the grain. Tolerant wheat genotypes (Barakatli 95 and Gobustan) showed reduced PEPC enzyme activity in drought-exposed variants compared to watered variants. Whereas, the enzyme activity was often higher in the drought-exposed variants of the sensitive genotypes that were watered. Sugars, which are frequently the substrate or outcome of chemical events, are crucial for cell metabolism, according to the findings of earlier examinations of biochemical parameters. In this research, depending on the stages of grain maturation in durum and bread wheat genotypes, water-soluble sugars and the enzyme PEPC were investigated under both well-watered and drought conditions.

According to the results of the research, proline content increased in the leaves of both normally irrigated and drought-stressed durum (Barakatli 95 and Garagylchyg 2) and bread wheat genotypes (Tale 38 and Gobustan) towards the end of the grain ripening period. The content of proline in the drought-sensitive Garagylchyg 2 and Tale 38 genotypes exposed to water stress during the wax ripeness stage of the grain increased slightly compared to the drought-tolerant Barakatli 95 and Gobustan 2 genotypes. Based on the results, a positive correlation was observed between the amount of proline and tolerance to water stress. The objective of this study was to identify wheat genotypes with enhanced resistance that can be utilized in future breeding programs aimed at developing drought-tolerant wheat varieties.

Keywords: wheat, soluble sugars, phosphoenolpyruvate carboxylase, proline, drought

Cryo-Milled Nano-DAP Fertilizer Application Promotes Photosynthetic Performance and Plant Growth in Tomato and Rice

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Phosphorus (P), a macronutrient, is indispensable for plant growth and development. It is taken up by roots in its inorganic form, i.e., orthophosphate (Pi). In a eukaryotic cell, P is an important component of ATP, DNA, lipids, and cell signaling machinery. Due to its highly reactive nature, Pi is often fixed into the soil solution and becomes unavailable for plants. The Pi shortage in the soil is routinely mitigated by the application of chemical Pi fertilizers. However, the use of such chemicals in bulk is expensive and their effectiveness is compromised by the poor nutrient use efficiency of conventional Pi fertilizers. Further, due to the non-renewable nature of natural P reserves and their asymmetric distribution globally, there is a demand to cut down on using Pi fertilizers in agriculture. Herein, we used a cryo-milling approach to prepare nano Diammonium Phosphate fertilizers (n-DAP) without altering DAP's bonding structure. Cryo-milling led to a significant reduction in particle size of n-DAP, which decreased ~ 5000 times with a specific surface area ~ 14000 times greater than that of c-DAP. n-DAP supplementation was found to enhance the growth of rice and tomato seedlings due to improved bioavailability of Pi even for a far lower input than c-DAP. The n-DAP supplemented plants also exhibited better photosynthetic efficiency and more stomata per leaf. At the molecular level, applying n-DAP seems to activate the TOR pathway to improve the plant's growth. The soil-based experiments further revealed significantly enhanced yield potential of tomato plants supplemented with n-DAP. Overall, our results demonstrate the enhanced efficacy of cryo-milled n-DAP over c-DAP for P-use-efficiency and plant yield, making it an interesting product for agriculture.

Keywords: Nano-Agri Input: Pi-fertilizer: Plant Growth and Development: Nutrient-use-efficiency: Sustainable agriculture

Boosting Growth and Bioremediation Power in *Chlorella vulgaris* Using Silicon Nanoparticles

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Photosynthetic microalgae can utilize organic compounds as carbon sources, enabling their growth in diverse environmental conditions and making them effective agents for bioremediation. Recent advancements have shown that nanoparticles (NPs) can enhance microalgae's growth in stress and bioremediation potential for various pollutants. This study investigates the impact of silicon oxide nanoparticles (SiO₂ NPs) on the photosynthesis and bioremediation capacity of *C. vulgaris* for the polycyclic aromatic hydrocarbon (PAH) pyrene (PYR). Our findings reveal that SiO₂ NPs at a concentration of 1 mg/L mitigated the toxicity of PYR (5 mg/L), enhancing photosynthetic efficiency. This was evidenced by a significant increase in Y(II) and ETR(II), alongside a decrease in Y(NPQ) and Y(NO). Notably, while superoxide dismutase (SOD) activity remained unaffected by PYR exposure, peroxidase (POD) and catalase (CAT) activities were elevated in the presence of PYR. The addition of SiO₂ NPs further enhanced POD activity and reduced ROS and MDA content under PYR-induced stress. Bioremediation of PYR by *C. vulgaris* was enhanced by 31% with the presence of SiO₂ NPs over 5 days. Catechol 2,3-dioxygenase activity was identified as a key player in PYR degradation, while dehydrogenase activity showed no significant change. These results suggest that SiO₂ NPs enhance PYR biodegradation in *C. vulgaris* by boosting PSII activity and activating the dioxygenase pathway.

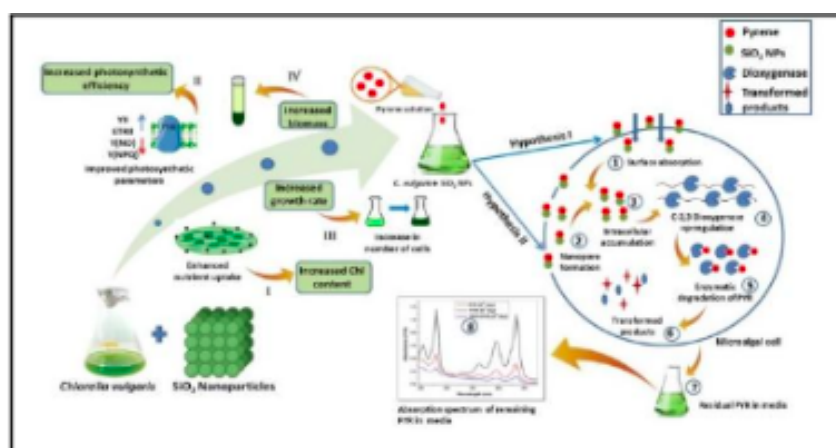


Figure 1: Schematic representation of impact of SiO₂ nanoparticles (SNPs) on growth, photosynthesis and bioremediation potential of *C. vulgaris*.

How Does LexA Regulate the Tolerance of *Anabaena* sp. PCC 7120 to Cadmium Stress Through Regulation of Photosynthetic Responses?

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Abiotic stresses are known to disrupt the redox poise of photosynthetic electron transport chain (pETC). Cyanobacteria have developed several poisoning strategies, but the regulatory network involved is not well understood. Here, we attempted to answer this question by using Cd as an abiotic stressor, since it disrupts pETC redox poise, and a LexA-overexpressing recombinant *Anabaena* strain (*AnlexA⁺*), since LexA has been identified as a global regulator in *Anabaena*. Assessment of the photosynthetic responses of *AnlexA⁺* and AnpAM (vector control), under unstressed and Cd-stressed conditions using TEM, chlorophyll fluorescence, and BN-PAGE, indicated that some pETC redox poisoning responses, including PSII photodamage, energy dissipation and NDH-mediated cyclic electron flow were decreased in *AnlexA⁺* under unstressed conditions. After Cd-exposure, *AnlexA⁺* elicited these responses similar to AnpAM, albeit at a slower rate, indicting a physiological role for LexA in optimizing pETC redox poisoning responses under stressful conditions. The presence of AnLexA-box upstream of over 70 of the 90 pETC component genes supports the regulation of LexA in pETC redox poisoning. LexA regulation on few redox-controlled pETC component genes was confirmed using qRT-PCR and EMSA. To summarise, the expression of a limited set of redox-controlled photosynthetic genes is regulated by LexA, which simplifies pETC redox-poisoning responses to abiotic stress.

Keywords: *Anabaena*, Blue native-PAGE, chlorophyll *a* fluorescence, LexA, PSII photophysiology, Redox-controlled pETC component genes, Transcription regulator, Transmission electron microscopy

Stability of Photosynthetic Apparatus, Transcriptome, and Metabolome of *Rhodobacter Alkalitolerans* Strain JA916^T: Response to Alkaline Along with High Light Environment

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Photosynthesis in purple bacteria is performed by well-oriented and densely packed pigment-protein complexes embedded in membrane lipid bilayer, which are known as light-harvesting antenna 2 (LH2), and reaction center light-harvesting antenna 1(RC-LH1). These pigment-protein complexes are accommodated in a spherical lipid bilayer membranous structure known as intracytoplasmic membranes. RC-LH1 and LH2 are present in a unique ratio and change depending on the physiological status of the cell and abiotic stress conditions. One of the species of purple bacteria, *Rhodobacter (R.) alkalitolerans*, can grow in neutral to alkaline conditions. Here we studied the effect of high light on the photosynthetic apparatus of the *R. alkalitolerans* in relation to its high pH tolerance ability by using absorbance and circular dichroism spectroscopy, LP- BN, P515, and reactive oxygen species-antioxidant activities. We found that in alkaline conditions, high light had a lesser effect on the photosystems than culture grown at neutral pH conditions. Since membrane lipids are the anchoring place for membrane protein complexes, make sure they are stable and sufficiently fluidic to facilitate electron transfer. We found a few membrane lipids, phosphatidylcholine, cardiolipin, and phosphatidylglycerol in relatively slight abundance in alkaline conditions. This study is further complemented by transcriptome and metabolome, where expression of several photosystem genes like *pucA*, *pucB*, *pufM*, *pufL*, *puha* and *pufX*, was differentially expressed in response to high light. In addition, several metabolites, including putrescine, proline, phytol, 2,3 butanediol, important in osmo-protection, reactive oxygen scavenging, redox balance, and terpene biosynthesis, were found to have major interacting patterns. Therefore, this study gives clues as to how the anaerobic photosynthetic organisms acclimatize to the different environmental stresses. Further, the specific genes can be manipulated to increase the biomass in order to obtain several bioactive compounds and transfer specific genes to the land plants to improve the crop productivity.

Keywords: Light-harvesting complexes (LH1 and LH2), high light, intracytoplasmic membranes, reaction center-light-harvesting complex (RC-LH1), *Rhodobacter alkalitolerans* strain JA916T membrane lipid, metabolites.

Biochemical and Structural Diversification of C₄ Photosynthesis in *Tribe Zoysieae (Poaceae)*

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C₄ photosynthesis has evolved independently multiple times in grass lineages with nine anatomical and three biochemical subtypes. *Chloridoideae* represents one of the separate events and contains species of two biochemical subtypes, NAD-ME and PEP-CK. Assessment of C₄ photosynthesis diversification is limited by species sampling. In this study, the biochemical subtypes together with anatomical leaf traits were analyzed in 19 species to reveal the evolutionary scenario for diversification of C₄ photosynthesis in tribe *Zoysieae (Chloridoideae)*. The effect of habitat on anatomical and biochemical diversification was also evaluated. The results for the 19 species studied indicate that 11 species have only NAD-ME as a decarboxylating enzyme, while eight species belong to the PEP-CK subtype. Leaf anatomy corresponds to the biochemical subtype. Analysis of *Zoysieae* phylogeny indicates multiple switches between PEP-CK and NAD-ME photosynthetic subtypes, with PEP-CK most likely as the ancestral subtype, and with multiple independent PEP-CK decarboxylase losses and its secondary acquisition. A strong correlation was detected between C₄ biochemical subtypes studied and habitat annual precipitation wherein NAD-ME species are confined to drier habitats, while PEP-CK species prefer humid areas. Structural adaptations to arid climate include increases in leaf thickness and interveinal distance. Our analysis suggests that multiple loss of PEP-CK decarboxylase could have been driven by climate aridization followed by continued adaptive changes in leaf anatomy.

Keywords: C₄ photosynthesis; decarboxylases; evolution; Kranz anatomy; Poaceae

Photocatalytic Hydrogen Evolution by CoAl_2O_4 Photocatalyst Under Solar Irradiation

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In recent years, spinel photocatalysts have been frequently used in various photocatalytic reactions such as photocatalytic hydrogen production due to their optical and electrochemical properties. Spinel is a material in the cubic crystal system, with the general formula AB_2O_4 , where A and B represent different transition metals[1]. High stability, surface area, abundance of active sites, and narrow band gap that can absorb visible light make spinel catalysts stand out. Among spinel photocatalysts, CoAl_2O_4 has excellent light absorption capability under visible light due to its 1.80 eV band gap. Since the conduction band of CoAl_2O_4 is close to 0 eV, it cannot meet the H^+/H_2 redox potential (H^+/H_2 is 0 V vs. NHE), and the photo-excited charges undergo recombination, reducing the photocatalytic hydrogen production efficiency of the catalyst[2]. In this study, it was aimed to create active sites for hydrogen evolution reaction (HER) by increasing the charge separation and migration ability of CoAl_2O_4 photocatalyst by using co-catalyst. Although charge recombination can be reduced by using metals such as platinum, gold, silver and palladium as co-catalysts, the use of these metals is not a sustainable approach due to high cost. [3]. MoS_2 was preferred as co-catalyst in this study because it has similar catalytic properties to noble metals such as platinum and is more cost-effective than these metals. In this study, CoAl_2O_4 photocatalyst was synthesized by co-precipitation method using precursor salts and calcined at 900 °C. The synthesized CoAl_2O_4 was characterized by X-ray photoelectron spectroscopy (XPS) and transmission electron microscope (TEM) techniques. Herein, CoAl_2O_4 photocatalyst with a spinel structure was investigated for photocatalytic hydrogen evolution under solar light irradiation in the presence of eosin-Y as a photosensitizer dye. Then, the effect of a co-catalyst on HER was examined by performing in-situ MoS_2 photodeposition from $(\text{NH}_4)_2\text{MoS}_4$ precursor salt on the surface of the CoAl_2O_4 spinel photocatalyst to enhance photocatalytic hydrogen evolution.

Keywords: photocatalysis, hydrogen evolution, molybdenum sulfide

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Size-dependent electrocatalytic hydrogen evolution by MoS₂ micro- and nanoparticles

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Hydrogen is a crucial energy carrier and an alternative to fossil fuels due to its high energy density, ability to be produced from sustainable sources, and lack of carbon emissions. Hydrogen production *via* electrocatalytic water splitting is considered more straightforward and sustainable compared to other methods[1]. However, reaction kinetics pose a limiting factor. Therefore, the process requires the use of cost-effective and widely available catalysts. In previous studies, the use of noble metal catalysts (such as Pt, Pd) has been widespread in electrocatalytic hydrogen evolution reaction (HER). However, numerous studies have demonstrated that transition metal dichalcogenide (TMD) catalysts, owing to their high catalytic activity, could serve as alternatives to noble metal catalysts[2]. MoS₂, a member of the TMD group, has recently attracted attention as a potential alternative to Pt-group metals, due to its low cost, high catalytic activity, and stability. Nevertheless, a limiting factor is that bulk MoS₂ structures have a reduced active surface area, negatively impacting catalytic activity[3]. In this study, the catalytic performances of MoS₂ catalysts of different sizes in hydrogen evolution reactions were investigated using linear sweep voltammetry (LSV), cyclic voltammetry (CV), and electrochemical impedance spectroscopy (EIS). The LSV method was employed to understand the overpotentials and reaction kinetics (Tafel plot) of both bulk and nano-sized MoS₂ catalysts. Additionally, the capacitances of the catalysts were calculated using the CV method, and the resistances of bulk and nano-sized MoS₂ catalysts were analyzed via EIS. These results displayed that nano-sized MoS₂ catalyst has more active in electrocatalytic HER due to enhanced active surface area and active sites.

Keywords: Electrocatalysis, Hydrogen evolution, TMD, Molybdenum Sulfide

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Exogenous Carbon Substrate for Sustainable Hydrogen Production by Cyanobacteria

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This study is aimed to investigate the impact of an exogenous carbon source, glycerol, on the rate and duration of hydrogen (H₂) release by cyanobacterial strains *Dolichospermum sp.* IPPAS B-1213, *Sodalinema gerasimenkoae* IPPAS B-353 and *Synechocystis sp.* PCC 6803 GT-L. The addition of glycerol leads to a significant increase in H₂ production by *Dolichospermum sp.* and *Synechocystis sp.* strains. It also led to light-dependent hydrogen release by the *S. gerasimenkoae* IPPAS B-353, which was not the case in any of the other experiments when photosynthesis inhibitors were added to this culture, or when incubated under anaerobic conditions without additives.

The best hydrogen-producing activity was observed in the nitrogen-fixing cyanobacterium *Dolichospermum sp.* IPPAS B-1213. After 216 hours of incubation under light exposure and anaerobic conditions, in the presence of 350 mM glycerol, this strain achieved a maximum hydrogen generation rate of 132.3 $\mu\text{mol H}_2/\text{mg Chl } a/\text{h}$. Additionally, after 264 hours of incubation the concentration of H₂ in the gas phase reached up to 67%. The optimal range of glycerol concentrations for efficient hydrogen production by this strain was 200-400 mM.

In the future, we plan to carry out a comparative analysis of the effect of different external carbon sources on the rate of hydrogen production in different nitrogen-fixing cyanobacteria.

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POSTERS

Electrochemical Control of Electron Transport in Thylakoid Membranes as Studied for Microalga and Cyanobacterium with Modeling State Transitions 2-1 During Light Induction

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The closed systems of chloroplast and cyanobacterial thylakoid membranes (TM) support light powered electron / proton transfer (ET / PT) via the trans-membrane complexes [1]. The thylakoid membrane model (TM model) was developed as a system of ordinary differential equations [2-3] being modified in the present work to consider the state 2-1 transitions that participate in the balance of light harvesting between photosystems II and I (PSII and PSI). As a result, quantitative interpretations were obtained for signals detected upon light induction prolonged to a few minutes: the fluorescence induction (FI) curves of *Scenedesmus obliquus* exposed to light during 5 min, as well as FI and P700 redox transitions of *Synechocystis* PCC 6803 recorded in parallel for 100 s intervals. Fitting the TM model to induction data allowed us to calculate the time-dependent concentrations of components for the ET/PT via PSII - PSI reaction centers mediated by PQ/PQH₂ pool, cytochrome *b₆f* complex and plastocyanin or cyt *c₆*, while reducing equivalents Fdr and NADPH are generated. So, in silico, linear ET operation is considered along with the cyclic one, while charge fluxes between lumen/stroma depend on passive charge (H⁺, counterions) redistribution (V_{leak}) and active energy consumption in the reversible H⁺-ATPase (V_{ATP}). Fitting the parameter sets on different kinetic patterns for *Scenedesmus*, *Synechocystis* indicated the two TM types for chloroplasts/ cyanobacteria distinctive in PSI:PSII stoichiometry [1]. Also, values V_{leak} and V_{ATP} were found different for *Scenedesmus* and *Scenedesmus* TM systems, while the fast and slow FI kinetics OJIPSMT might be explained according to TM energization stages coordinated to proton motive force generation upon light induction. Modifications of V_{leak} , V_{ATP} parameters were tested as factors influencing the OJIPSMT dynamics. Results showed that operation of chloroplast and cyanobacterial TM can be modeled and compared.

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Comparative Chlorophyll *a* Fluorescence and Thylakoid System Ultrastructure in Chloroplasts of Two Italian *Cichorium Intybus* L. Cultivars

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Red chicory (*Cichorium intybus* L., Asteraceae) is a prominent and widely grown winter crop in Northern Italy, with Chioggia Precoce and Treviso Precoce as two main varieties cultivated along the eastern coast of Emilia Romagna region. The strong varietal selection has led to different leaf arrangement and orientation in the two cultivars, i.e., mostly plagiotropic in cv. Chioggia and orthotropic in cv. Treviso. These variations influence the angle of solar irradiation of the leaves and, therefore, could hint at possible morpho-functional specializations in the chloroplasts as a side effect of the varietal selection. To verify whether this was the case, we combined chloroplast ultrastructural, chlorophyll *a* fluorescence and the $\delta^{13}\text{C}$ isotopic analyses of the two aforementioned varieties. Plants grown in neighbouring fields in Ferrara coast were sampled between December 2023 and January 2024.

Transmission electron microscopy revealed that, in mature newly expanded leaves, the chloroplasts in cv. Chioggia were typically elliptical, with a well-developed thylakoid system. In contrast, chloroplasts in cv. Treviso were more globular, exhibiting an extremely heterogeneous internal structure, even within the same cell. The thylakoid system in cv. Treviso was characterized by long single thylakoid arrays and grana of highly variable size, along with small 'thylakoid circles.' The morphometric analysis of grana showed that the granum height (h) in cv. Treviso was greater than in cv. Chioggia, owing to more thylakoids per granum and higher stacking repeat distance (SRD) (Mazur et al., 2021). The cv. Chioggia showed, instead, a higher granum cross-sectional irregularity (GSI), i.e., a prominent tendency of thylakoid disks to slide apart on the lateral plane. Despite such structural features, in-field analysis using a Handy-PEA fluorimeter and a MultispeQ spectrometer, along with modulated chlorophyll *a* fluorescence quenching, showed no signs of impaired photosynthetic functionality in cv. Treviso. Remarkably, cv. Treviso appeared somehow even more efficient than cv. Chioggia. To support independently this inference, the $\delta^{13}\text{C}$ isotopic ratio was examined across roots, stems, and leaves, confirming more favourable values in cv. Treviso compared to cv. Chioggia (More et al., 2022).

Overall, the chloroplast structural abnormalities observed in cv. Treviso leaves do not seem to impact negatively the plant's photosynthetic performance. Instead, these changes may reflect a specific adaptation that arose during the cultivar selection process (Barcaccia et al., 2016).

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Effect of Compatible Osmolytes on the Efficiency of Photosynthesis Inhibitors in Thylakoid Membranes of *Spinacia Oleracea*

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Compatible osmolytes such as sucrose, trehalose, and glycine-betaine are able to stabilize isolated photosynthetic proteins. These compounds are able to work not only at the level of individual isolated protein globules, but also at the level of whole thylakoid membranes.

The integral mechanism of action of osmolytes on thylakoid membranes containing photosystems 1 and 2 as well as the b6f complex is not completely known. It is possible to evaluate the effect of osmolytes on the three-dimensional structure of photosystems and to identify the main targets for osmolytes in membranes using inhibitor assays. The classical inhibitors of photosynthesis, specifically DCMU and DBMIB, act on different sites of the photosynthetic electron-transport chain. DCMU acts on the acceptor side of photosystem 2, while DBMIB acts on the donor side of the b6f complex. Analyzing how osmolytes modify the inhibitory effect of these compounds will allow us to understand how much osmolytes affect the molecular environment of the binding sites of these inhibitors.

In this study, polarographic and fluorometric measurements of the activity of primary photosynthetic processes in spinach thylakoid membranes in the presence of inhibitors (DCMU and DBMIB) and osmolytes (sucrose, trehalose, and glycine-betaine) were performed to analyze how the presence of osmolytes alters inhibitor activity.

DBMIB-induced change in OJIP parameters F_v/F_m and V_i for thylakoids suspended with 1M GB differed from those for membranes suspended with disaccharides, as shown by two-way analysis of variance of these parameters. While the maximum quantum yield of PSII (F_v/F_m) was more susceptible to DBMIB in the presence of GB, the relative fluorescence values at the I-stage (V_i) were less susceptible. This result suggests that PSII complexes are more resistant to DBMIB inhibition in the presence of disaccharides, while another part of the ETC, including the b6f complex and/or the PSI complex, is more stable in the presence of GB. This may be due to the different mechanism of membrane interaction with these osmolytes.

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Biochemical and Spectroscopic Insights into the PSI-LHCI Supercomplex of the Diatom *Chaetoceros Gracilis* Lacking a Single LHCI

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During evolution from cyanobacteria to various photosynthetic organisms by primary and serial endosymbiosis, the photosystems (PS) are highly conserved, whereas an astonishing diversity of light-harvesting complexes (LHCs) has evolved. Generally, the serial endosymbionts (e.g., Cryptophytes, Ochrophytes, Alveolates) possess more LHCI s than that in primary endosymbionts. Especially, a diatom, *Chaetoceros gracilis* (*C. gracilis*), harbors up to 24 LHCI s around PSI forming the largest PSI antenna system in the PSI-LHCI structures that have been reported to date. These 24 LHCI s form sophisticated antenna belts, orchestrating photosynthetic light absorption under ever-changing light conditions. However, the physiological function and regulation of the PSI-LHCI supercomplex in this diatom, an outstanding representative of serial endosymbionts, characterized by a larger antenna size compared to PSI-LHCI in other primary endosymbionts, remain unclear at present.

To gain further insight into the function of the larger antenna system, we have independently knocked out two different *Lhc* genes encoding LHCI s that likely function as a “bridge” between the peripheral antenna and the PSI core in *C. gracilis*. The integrated analysis, which employed Chl fluorescence, P700 redox states, time-resolved fluorescence, and CN-PAGE, provided valuable information about electron transport, excitation energy transfer, and the putative antenna assembly in mutant strains with a truncated PSI-LHCI supercomplex. In the light of these observations, the importance of the huge PSI antenna in *C. gracilis* as well as the putative antenna detachment architecture in mutant strains will be discussed.

Salt-Induced Changes in the Protein Composition and Ultrastructure of Chloroplasts from Maize

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Plants belonging to the C₄ photosynthesis pathway are distinguished by the efficiency of CO₂ assimilation and the high level of specialization of the photosynthesis apparatus. The leaves of C₄ plants contain two distinct types of photosynthetic cells: mesophyll (M) and bundle sheath (BS), which are organized differently, both structurally and functionally. The objective of the presented work was to conduct a comparative study of M and BS chloroplasts isolated from the maize (*Zea mays* L.) plant subjected to different concentrations of NaCl. The polypeptide composition of thylakoid membranes of M and BS chloroplasts was analyzed by the gradient SDS polyacrylamide gel electrophoresis (SDS-PAGE). At high salt concentrations, the synthesis of the 68 kDa apoprotein belonging to the core of PSI and the α - and β - subunits of the catalytic CF1 domain of the ATP synthase complex were inhibited in the M thylakoid membranes. No significant changes were found in the thylakoid membrane systems of BS chloroplasts. According to the ultrastructural studies it was found that at relatively low salt concentrations, both M and BS cells accumulated starch granules, whereas at high salt concentrations, their size and quantity decreased. At the same time, salinity caused an increase in granal stacking of BS chloroplasts while that in M chloroplasts was damaged. These results suggest that chloroplasts of M cells are more sensitive to salt stress than BS cells. In NaCl-treated plants, the formation of granal stacking in BS chloroplasts may contribute to an increase in PSII activity of BS cells compared to that in the control plants.

Electron Transfer Between the Terminal Iron-Sulphur Cluster Acceptors of Photosystem I: Insights from Kinetic Modelling Simulations

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Electron transfer reactions in Photosystem I take place across two electron transfer chains that share both the terminal electron acceptors, a series of three 4Fe-4S clusters known as F_X , F_A and F_B and the terminal donor, F_{700} . The two electron transfer chains show kinetic differences, which are mainly attributable to the tuning of the physical-chemical reactivity of the bound cofactors, exerted by the protein surroundings. The factors controlling the rate of electron transfer between the terminal Iron-Sulphur clusters are still not fully understood. This is mainly due to the difficulties of monitoring these electron transfer events directly by spectroscopic methods. In an attempt to derive further information concerning the energetics of electron transfer reactions between F_X and F_A as well as between F_A and F_B an approach relying on the tunnelling-based description of the reaction rates coupled to the kinetic modelling of the forward and the recombination reactions is applied. The kinetic model also comprises the electron transfer steps upstream to F_X , that lead to its reduction by the two phyllo(semi)quinones A_{1A}^- and A_{1B}^- . Two energetic schemes for A_1^- oxidation are considered. One in which A_1^- oxidation is thermodynamically favourable on both electron transfer chains, the other in which the oxidation of A_{1B}^- is favourable but that of A_{1A}^- is energetically uphill. Both energetic configurations were already shown to describe satisfactorily A_1^- oxidation, at least on a semi-quantitative basis. Particular attention is here dedicated to exploring the driving forces associated to Iron-Sulphur centre involving electron transfer reactions, as well as the impact of the value of the reorganisation energy on these reaction rates. It is concluded that the reorganisation energy for F_X^- oxidation shall be lower than 1 eV. Moreover, it is suggested that the analysis of mutants with altered F_A redox properties can provide useful information concerning the upstream, phylloquinone, cofactor energetics. The kinetic simulations indicate that the modulation of the F_A redox potential produces markedly different perturbation of the upstream electron transfer kinetics which are dependent on the specific energetic model employed to describe the phyllo(semi)quinone oxidation by the Iron-Sulphur cluster F_X .

Efficient Light Harvesting by Fucoxanthin-Chlorophyll Proteins in Photosystem I of the Diatom *Thalassiosira pseudonana*

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Oxygenic photosynthetic organisms have evolved diverse peripheral light-harvesting complexes that enhance the absorption cross-section of their photosystems and optimize photosynthetic efficiency. In diatoms, which are one of the main groups of primary biomass producers of the oceans, the light-harvesting function is delegated to fucoxanthin–chlorophyll a/c binding proteins (FCPs). FCPs extend the wavelength range of light useable for photosynthesis toward the blue-green part of the spectrum. The FCP family itself is diverse, with different FCPs binding to each photosystem core and playing specific structural, light-harvesting and photoprotection roles. High-resolution structures determined recently for several diatom PSI and PSII supercomplexes have revealed significant differences in their organization compared to green algae and higher plants[1,2]. The specific structure and composition of diatom supercomplexes can have a significant impact on the pathways and efficiency of excitation energy transfer[3]. In this study, we explored the dynamics of energy transfer in the isolated FCPI and PSI-FCPI supercomplex from the diatom *Thalassiosira pseudonana* using time-resolved optical spectroscopy. In addition to time-resolved fluorescence measurements at room temperature and 77 K, we employed two-dimensional electronic spectroscopy (2DES) – a powerful technique for probing the excitation energy transfer in complex systems[3,5]. The fluorescence decay kinetics of PSI-FCPI was markedly different compared to higher plants PSI-LHCI, which is partly because of the lack of red-shifted chlorophylls in FCPI. The main trapping time at room temperature was about 40 ps, suggesting that the presence of FCPI slows down the overall trapping by about 20 ps, similar to PSI of *Chaetoceros gracilis*[4]. Global analysis of the 2DES data revealed multiple energy equilibration components in PSI-FCPI with lifetimes from 150 fs to about 6 ps at 77 K. Apart from a minor equilibration with red Chls (presumably in the PSI core) with a 35 ps lifetime, all components attributable to energy transfer between FCPI and PSI have lifetimes below 10 ps. These results, taken together with recent data on energy transfer from FCPII to PSII in the same organism, suggest that the FCP antenna system is optimized for remarkably fast energy transfer and can serve as a model for efficient light-harvesting.

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Utilizing UV Light for Photosynthesis via Fluorescent Protein-Mediated Energy Transfer

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Ultraviolet (UV) light is an abundant high-energy resource on Earth and in space, presenting a potential opportunity for energy conversion. However, photosynthetic organisms have not been observed to directly utilize UV light for photosynthesis. In this study, we propose a novel approach to enable photosynthetic systems to utilize UV light for energy conversion. We propose that mTurquoise2 (mTQ2), a fluorescent protein capable of absorbing UV light and emitting visible light, can be coupled with photosynthetic proteins to facilitate photosynthetic processes through fluorescence emission and/or Förster energy transfer. To test this proposal, we isolated and purified photosystem II (PSII) from *Thermosynechococcus elongatus* (equivalent to *vestitus*) and coupled it with mTQ2, expressed in *Escherichia coli*, via click chemistry. The absorption and low-temperature fluorescence spectra of the hybrid complexes were measured, revealing efficient energy transfer between mTQ2 and the PSII complex. Based on these results, the potential for utilizing UV light in photosynthesis through mTQ2 was systematically evaluated and discussed.

Signaling of Far-Red Regulates Photosynthetic Electron Transfer and Carbon Fixation

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Far-red (FR) light has been shown to impact leaf photosynthesis and photoprotection under high light conditions, but the underlying mechanisms of FR signaling in photosynthesis are still not fully understood. To investigate the role of FR signaling through phytochrome A (PHYA) in the regulation of responses to high light, we exposed tomato wild type (WT) and phytochrome A mutants (*phya*) to high-intensity white light ($1000 \mu\text{mol m}^{-2} \text{s}^{-1}$) with or without supplementary FR light ($250 \mu\text{mol m}^{-2} \text{s}^{-1}$). We analysed the influence of FR on electron transport and the Calvin-Benson cycle in both WT and *phya* plants. Our results demonstrated that exposure to FR in the *phya* mutants led to an increase in the quantum yield of non-photochemical quenching (Φ_{NPQ}) and a reduction in the quantum yield of non-regulated energy dissipation (Φ_{NO}). This redistribution of energy helped to prevent a marked decrease in the quantum yield of photosystem II (Φ_{PSII}). Furthermore, the increased energy dissipation via NPQ in FR-exposed (*phya*) mutants was linked to higher donor-side limitation (Φ_{ND}), a decrease in the effective quantum yield of photosystem I (Φ_{PSI}), and increased oxidation of P700. FR light also affected the Calvin-Benson cycle. The FR-induced increase in the maximum carboxylation rate of Rubisco ($V_{c_{max}}$) was more pronounced in (*phya*) compared to WT. However, increases in the maximum electron transport rate (J_{max}) and triose phosphate utilization (TPU) seen in WT were not observed in (*phya*). In conclusion, tomato plants exhibit different response to high light depending on the presence or absence of PHYA, highlighting a crucial role for FR signalling in modulating plant response to high light.

Keywords: Energy-partitioning, Far-red, Phytochrome, Photoprotection

A Light-Harvesting Protein for Adaptation in Specific Light and CO₂ Conditions in the Marine Diatom *Chaetoceros Gracilis*

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In aquatic environments, the quality and intensity of light change dynamically. Microalgae respond to these changes to maintain efficient photosynthesis. Diatoms, which are photosynthetic microalgae found in marine and freshwater environments, use fucoxanthin chlorophyll *a/c*-binding proteins (FCPs) as their light-harvesting complexes (LHCs) for photosystems. Our previous studies have shown that the marine diatom *Chaetoceros gracilis* has 46 genes encoding FCPs (Kumazawa et al., *Physiol. Plant.* 174: e13598, 2022). It has also been reported that under certain growth conditions, such as white LED illumination with elevated CO₂ levels or red LED illumination with air bubbling, a novel band of FCPs appears in the clear-native PAGE gel separating detergent-solubilized thylakoid membrane complexes (Ueno et al., *J. Phys. Chem. Lett.* 10: 5148–5152, 2019; Nagao et al., *Photosynth. Res.* 146: 189–95, 2020). In this study, we further investigate this particular FCP, which exhibits unique accumulation patterns compared to other FCPs. These findings provide new insights into the environmental adaptation of diatom photosynthesis through FCP remodeling.

Rhodobacter Capsulatus as a Tool to Investigate Green Chemicals Interaction with Native Membranes

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Ionic liquids (ILs) are a supposedly greener alternative to canonical volatile organic compounds used in many industries. We demonstrated the ability of some ILs, containing the TFSI anion, to collapse the membrane electrical potential at micromolar concentrations, when tested on chromatophores isolated from the photosynthetic bacterium *Rhodobacter (Rb.) capsulatus*. Curiously, while they do not seemingly inhibit any photoinduced electron transfer reaction, the cytochrome bc_1 heme b_H reduction appears to be accelerated. To assess whether such behavior translates to a toxicity for the whole organism and, if so, to what extent, we developed a system to grow *Rb.capsulatus* photosynthetically in 96-wells plates, monitoring the optical density of the cultures over 48 hours in the presence of growing ILs concentrations. Here, we found that growth is impaired at concentrations that are 10 times higher than those effectively collapsing chromatophores membrane electrical potential.

Photocurrent Generation by Thylakoid Membranes with Different Electron Mediators

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Biohybrid systems generating light current or voltage using components of the photosynthetic apparatus are of interest both as prototypes of light-to-electricity converters and as analytical devices for studying redox processes in photosynthetic membranes. In these biohybrid devices, two different methods of electron transfer between the photosynthetic electron transport chain (pETC) and an inorganic electrode can be implemented: direct electron transfer (DET) and mediated electron transfer (MET). DET is implemented through direct contact of photosynthetic membranes or pigment-protein complexes with electrodes and does not require additional connections. MET uses intermediate exogenous or endogenous charge carriers that transfer electrons between the pETC and electrodes. There are many reports in the literature that the efficiency of MET exceeds that of DET. Thus, MET studies and a search for optimal mediators are necessary for the development of an efficient biohybrid device.

Various classical Hill reagents can be used to redirect photosynthetic electron transfer to the electrode. Different exogenous electron mediators require different electrochemical measurement conditions and imply different electron transfer efficiencies. An electron donor must be used with an electron acceptor if ETC without an oxygen evolving complex is used, and the choice of an electron donor for a given electron acceptor and pETC moiety is also complex.

We analyzed the photoelectrochemical properties of a three-electrode circuit based on thylakoid membranes with potassium ferricyanide, dichlorobenzoquinone, and dichlorophenolindophenol as electron acceptors. The PSII Mn_4CaO_5 cluster is known very unstable and vulnerable to stresses and that will determine the stability of the biohybrid device. To test whether it is possible to generate photocurrent in the BD independently of the Mn cluster we investigated the photocurrent generation in Mn-free Mn_4CaO_5 cluster deprived thylakoids (apo-PSII thylakoids) in the presence of the exogenous electron donors diphenyl carbazide and $MnCl_2$. We showed that membranes with oxygen activity generate higher photocurrent and for a longer time than membranes without oxygen activity but in the presence of an electron donor, as well as that generation of photocurrent in biohybrid devices is possible and with apo-PSII membranes.

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Overexpression of cyanobacterial Glutaredoxin increase the fitness of *Synechococcus elongatus* PCC 7942 and *Escherichia coli*

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Cyanobacteria are photosynthetic photoautotrophs that produce natural antioxidants to combat reactive oxygen species (ROS), which can cause oxidative damage and affect their fitness under dynamic environmental conditions of light, temperature, and nutrient availability. Glutaredoxins (Grxs) protein plays a crucial role in maintaining redox homeostasis and protecting proteins from oxidative stresses. Grxs are widely present across different life forms and are involved in regulating redox states, facilitating glutathionylation/deglutathionylation, and serving as electron donors for various reductase enzymes. In higher plants, it is known that Grxs regulate the rubisco activase activity, and therefore, can affect the carbon fixation process. Cyanobacteria possess two types of Grxs, such as dithiol glutaredoxin (Grx3) and monothiol glutaredoxin (Grx4). Despite the extensive studies of Grxs in other organisms, limited studies are available for their physiological importance in cyanobacteria. We overexpressed the gene encoding Grx3 of *Synechococcus elongatus* PCC 7942 (*S. elongatus*) in *Escherichia coli* (*E. coli*), as well as in *S. elongatus* (under the control of *apc* promoter) to decipher its physiological relevance. The phenotypic characterization (growth, cell biomass, ROS levels, photosynthetic pigments and performance, morphology, antioxidant activity, etc.) of overexpression and wild-type (WT) strains was performed under different environmental conditions to study the impact of higher accumulation of Grx3. The Grx3 protein was successfully accumulated in *E. coli*. The overexpression strains of *S. elongatus* and *E. coli* were found to perform better in the presence of H₂O₂ and NaCl stress, in comparison to their WT strains. In *S. elongatus*, the overexpression strains were found to have better growth and cell biomass production, higher photosynthetic yield, low ROS levels, and higher antioxidant activity than WT. In summary, overexpression of the *grx3* gene can improve the fitness of the organisms under different environmental conditions.

Keywords: Cyanobacteria, Glutaredoxins, Photosynthesis, Reactive oxygen species

Anti-Radiation Properties of *Salvia Officinalis* L. Extract in Wheat Sprouts

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Sage is a well-known medicinal plant and is often used in medical practice in different countries of the world. Due to its rich chemical composition, sage extract is widely used in all areas of medicine as an antioxidant, immunomodulatory, anti-inflammatory agent. It seems interesting to study the anti-radiation properties of the extract of sage (*Salvia officinalis* L.), growing in Azerbaijan in irradiated wheat sprouts. The leaves of common sage collected in the Gek-Gelsky district were used for the research. In the experiments with plants, the seeds of wheat of the "Bereketli-95" variety were used. The seeds were irradiated using the URI (K-25) installation at a dose rate of 13.4 rad/sec, the source was ^{60}Co , the dose was 150 Gray. Before irradiation, the wheat seeds were treated with extract solutions and experiments were carried out in laboratory conditions. The following groups of plants were analyzed: control, irradiated at a dose of 150 Gy, without treatment and treated with 0.1%, 0.01% and 0.001% concentrations of the extract. The morphological parameters and growth dynamics of wheat seedlings were measured for 40 days. The amount of chlorophyll pigments, carotenoids, malonic dialdehyde was measured using a Multiscan GO spectrophotometer. Chlorophyll fluorescence in leaves was measured using a MINI-PAM device. At the initial stages of development, the irradiated variants sprouts developed poorly, and the sprouts treated with 0.1 and 0.001 percent solution of the extract were normal compared to the irradiated ones. At the end of the month, the sprouts obtained from gamma-irradiated seeds, treated with a 0.01% solution of sage showed better results in growth and development and in the amount of photosynthetic pigments. At the final stage of the experiments, the length of the sprouts was almost equal to the control variant.

It was found that a 0.01% solution of *Salvia officinalis* extract, eliminating the negative effect of gamma radiation, normalizes plant growth, the synthesis of photosynthetic pigments in the leaves, and the work of photosystem II in chloroplasts.

Interactive Effects of Sodium Nitroprusside and UV Stress on Chlorophyll Fluorescence and Antioxidant Responses in Lettuce

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Studying UV-B radiation stress is crucial due to its harmful effects on plant growth, leading to oxidative damage and reduced productivity. Understanding sodium nitroprusside (SNP) role as a stress mitigator is crucial due to increasing environmental challenges. This research explores the impact of UV stress and SNP on lettuce, emphasizing chlorophyll fluorescence and antioxidant responses. A controlled pot experiment was conducted with two factors: SNP application (0 and 200 μM), under both UV-B stress and normal light conditions. Changes in the structure and function of photosystem II (PSII) were analysed using the fluorescence parameters derived from fast fluorescence kinetics (OJIP transient). The results showed that UV stress negatively impacted photochemistry and chlorophyll parameters, but SNP treatment significantly enhanced plant growth and UV tolerance. This was achieved by mitigating the adverse effects on performance index (PI(abs)) and PSII's maximum quantum yield (Fv/Fm), through improving reaction centers (RC/ABS) and electron transport (ET0/RC) efficiency. Furthermore, SNP-treated plants exhibited better protection against oxidative damage under UV conditions, as indicated by increased antioxidant activities, including elevated levels of flavonoids (FLAV) and anthocyanins (ANTH). Correlation analysis showed that Fv/Fm positively correlated with Area, PSIIrea, RC/ABS, ET0/RC, PI(abs), and SFR, while significant negatively correlating with DI0/RC, ANTH, and FLAV. These findings suggest SNP as a promising agent to counteract UV stress in plants, improving photosynthetic efficiency and antioxidant defenses, and providing a valuable strategy for enhancing crop yield and quality in environments subject to high UV radiation.

Keywords: Antioxidants; chlorophyll fluorescence; lettuce; sodium nitroprusside; UV stress.

Fluoresce-Based Analysis of Wheat Physiological Responses to Water Deficit

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As a result of adverse climate change and a growing global population, there is a great need for more productive, stress-resistant crops. From the physiological viewpoint, it is crucial to deal with species-specific responses and properties of the photosynthetic apparatus of plants exposed to climatic extremes. It is also necessary to dispose with physiological criteria that enable a more effective and faster selection of varieties and their tolerance to environmental stress, including long-term soil water deficit. Since adverse factors' frequency, length and intensity are unpredictable, plants must have a certain adaptation potential representing a complete complex of mechanisms. Considering that photosynthetic parameters are part of one of the main processes in plant metabolism, such as photosynthesis, they can be affected by water deficit, demonstrated by the decrease of photosynthetic parameters and metabolic limitations. In our experiments, we combined the methods based on the fast kinetics of chlorophyll fluorescence (measured by Handy PEA, Hansatech, GB), with the spectrophotometric determination of free proline and pigment contents along with the leaf relative water content (RWC) to record the effects of drought stress simulated in pot experiments with winter wheat genotypes, including several chlorophyll-less mutants. The results confirmed the sensitivity of the fluorescence kinetic analysis to drought, enabling recognition of the sensitive and tolerant wheat varieties associated with different responses to drought stress identified at the level of osmolyte accumulation (proline) and pigment contents. The effect of drought was demonstrated by a significant decrease in photosynthetic parameters (RC/ABS, Ψ_{reo} , PIabs) in the varieties with chlorophyll mutations and RWC below 70%, especially in the ANDW7B and ANBW-4B mutant of wheat. The most tolerant wheat varieties were the San Pastore (originating in Italy) and the Thesee (France), in which we observed only a slight decrease in photosynthetic parameters while RWC was above 70%. The parameter characterizing the maximum photochemical efficiency of PSII (Fv/Fm) was not significantly affected, which indicated a limited capacity of the parameter to detect drought stress.

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The Effect of Heat and Drought on Chlorophyll Fluorescence Parameters in Wheat Varieties

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Drought and high temperatures are considered the most natural environmental limits that result in water deficit or temperature stress. They are also important limiting factors affecting crop yield and quality. The interaction of both stress factors causes significant damage to photochemical processes. The effect of high temperature in fully hydrated plants is associated with maintaining of optimal water balance, whereas the effect of high temperature under insufficient hydration leads to significant damage. Identification of varietal differences in sensitivity or tolerance based on responses of photosynthetic parameters to combined stress may help recognize valuable genetic resources that are useful for crop improvement programs. Therefore, we analyzed the responses to combined stress in a collection of wheat varieties pre-selected based on phenotypic and leaf traits using mainly non-destructive methods. The fast chlorophyll fluorescence transient analysis characterizing the structural and functional state of photosystem 2 (Handy Pea, Hansatech, GB) was measured in addition to the leaf photosynthetic rate by an open gas exchange system (LI-6400, Licor, Lincoln, NE, USA). To determine the leaf water status, relative water content (RWC) was measured. Moreover, spectrophotometric determination of the content of free proline and the photosynthetic pigments was realized. The results confirmed the varietal differences in responses, in addition to the significant effects of the environment. The combined effect of heat stress and drought was associated with a significant reduction in chlorophyll fluorescence parameters (F_v/F_m , RC/ABS , PI_{abs}), indicating a decrease of PSII quantum efficiency and related functions, as well as an increase in the W_k parameter indicating damage to the oxygen-evolving complex. The effects on PSII photochemistry were associated also with an overall decrease of photosynthetic rate (A_{CO_2}), caused by both stomatal limitation and non-stomatal inhibition of carbon assimilation caused by stress. The varietal effects were observed in most of the examined parameters.

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Fibrillin Regulates Carotenoid Biosynthesis and Photoprotection in Green Algae *Chlamydomonas Reinhardtii*

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Photosynthetic cells grow in an ever-changing environment, usually experience considerable fluctuation in light availability. Excess light can damage the photosynthetic apparatus by providing more energy than can effectively be utilized. Plastoquinone-9 (PQ-9) is a vital component of photosynthetic electron transfer (PET) that carries electrons in the linear and alternative electron transport chains. The redox state of PQ-9 regulates gene expression, enzymatic activity, and luminal pH which further triggers photoprotective response. PLAP6 is a gene in the fibrillin protein family that encodes the plastid-lipid associated protein (PLAP). PLAP6 is involved in PQ-9 biosynthesis, thereby playing a crucial role in determining the PQ pool size and the efficiency of PET. Here, we demonstrated that *Chlamydomonas reinhardtii* cells with a mutation in Fibrillin ($\Delta plap6$) were defective in carotenoid biosynthesis when grown in mixotrophic medium. Additionally, $\Delta plap6$ cells showed a drop in the rate of electron transfer (rETR) compared to wild type (WT) with it being more profound in mixotrophic conditions. Additionally, pH-dependent energy quenching (qE) was not efficient in $\Delta plap6$ cells in either the mixotrophic or autotrophic condition. We observed that, $\Delta plap6$ cells adopted different photoprotective strategies depending on the culture medium composition. Mutant cells grown in mixotrophic conditions relied on long-distance electron transfer from chloroplast to mitochondria depicted by elevated expression levels of ubiquinones. In contrast, in phototrophic conditions mutant cells relied on the chloroplast alternative electron transfer pathway mediated by the plastoquinone terminal oxidase (PTOX) activity. This study has enabled us to elucidate the functional significance of fibrillin in carotenoid biosynthesis and its consequential impact on photoprotective responses. Additionally, it sheds light on the photoprotective strategies the cells adapt in the presence or absence of acetate.

Reversible Induction of Photosynthetic Complexes to the Salt Stress from *Chlamydomonas Reinhardtii*

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Here, the photosynthetic algae *Chlamydomonas reinhardtii* is used as a model organism to understand the effect of salt stress and recovery on the photosynthetic apparatus. In this study, we would like to understand the role of cyclic electron transport in the acclimation process to high salt, a wild type, and cyclic electron transport-dependent proton gradient-related mutant's *pgrl1* and *pgr5* and their recovery process. As a primary response, there is a decrease in growth and chlorophyll fluorescence induction, OJIP transient under salt stress, indicating the reduction of photochemical efficiency. Further, there was a decrease in chlorophyll and an increase in carotenoid content in salt stress, which was more prominent in *pgr5*, and these alternations could arise from ROS generation. The mutants were more sensitive to ROS and showed more carotenoid content than the wild type. The circular dichroism data showed that the salt stress caused changes in pigment-pigment and pigment-protein interactions. When cells are exposed to high salt, there is a dynamic change in the core proteins, but LHCs and antenna proteins of PSI and PSII are not much affected. Further, the cell morphology and thylakoid membrane organization changes are also observed in high salt stress. These results show that the partitioning of electrons could also be mediated via PSI remodeling processes: decreased steady-state proton motive force and increased damage of PSI in *pgr5*.

However, when the salt-induced cells were grown in the normal medium, the cells were grown like control. Also, the photosynthetic efficiency reached the control level. Our results indicate that cyclic electron transport is critical in the acclimation process of high salt.

The Effect of UV-B Radiation on Photosynthetic Apparatus of Lettuce Genotypes Differing in Anthocyanin Content

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One crucial environmental component that affects how plants grow and develop is UV-B radiation. Depending on the level and duration of exposure, UV-B radiation may function as a stressor that impairs photosynthetic structures directly or disturbs plant homeostasis. Lower doses of UV-B help plants grow by inducing photomorphogenic reactions that modified plant growth and development by inhibition of stomatal movement, leaf expansion, and stem elongation. In the previous period it was described that the increase of UV-B-absorbing flavonols is an important plant response to UV-B radiation, but other chemicals, such as anthocyanins, are likely to play a protective function. The experimental work aimed to study the impact of different doses of UV-B radiation on photosynthetic apparatus and monitor varietal differences in the responses of two varieties of lettuce plants (*Lactuca sativa L.*) differing in anthocyanin content. Both whole plant chlorophyll fluorescence imaging, which allowed for the detection of UV-induced leaf damage, and fast fluorescence transient analysis, which offered in-depth knowledge of PSII photochemistry alterations, were utilized to evaluate the photosynthetic functions. Furthermore, growth intensity were evaluated and the accumulation of UV-absorbing substances, such as flavonols and anthocyanins, was determined. In cultivation systems using LED lighting, the presence or lack of UV-B radiation had an impact on almost all growth, content, and physiological indicators that were being tracked. Both positive and negative impacts of UV-B radiation were noted, depending on the strength of the added UV-B radiation. Significant genotypic differences in responses to UV-B radiation were observed. Additionally, the maximal yield of PSII photochemistry (Fv/Fm) decreased as a result of UV irradiation, and this reduction was linked to a decrease in the number of active reaction centers (RC/ABS). As anticipated, even in the red genotype of lettuce, UV radiation induced a fast rise in the flavonoid content of the leaves, with some genotype-specific variations. However, it did not result in an increased build-up of anthocyanins. Finally, the hypothesis that red-leaved genotypes are more resistant to UV-B radiation's damaging effects due to anthocyanins' protective role in tolerance to UV-B radiation was not supported.

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Comparative Analysis of Photosynthetic Complexes Under High Temperature Stress in *Chlamydomonas Reinhardtii*, *Dunaliella Salina*, and *Haematococcus Pluvialis*

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The photosynthetic organization plays a crucial role in the stabilization of the photosynthetic complexes. In our proposed study, a comparative assessment of the impact of high-temperature stress (i.e., 30°C, 35°C, 40°C and 45°C) on growth dynamics, cellular morphology, and Characterization of photosynthetic complexes of *Chlamydomonas reinhardtii*, *Haematococcus pluvialis*, and *Dunaliella salina*. Extreme heat stress adversely affects the photosynthetic efficiency. As a primary response, there is a decrease in the growth and quantum yield of Photosystem II (PS-II) through the Fv/Fm Ratio. Further, there was a reduction in the OJIP transient, which reflects modifications in PSII reaction centers or adaptations in the electron transport chain, indicating the photochemical activity of PSII is greatly affected by increased temperature. The radar plot data showed that when cells are exposed to 30°C the photosynthetic efficiency has been decreased. The cells showed increased non-photochemical quenching after 1hr of treatment. At 35°C, the cells showed an enormous increase in reaction centers per cross-section with little increase in absorption after 24 hours of treatment. This increase in absorption with an increase in temperature is further validated when the absorption index (PI (ABS)) peaks after 24 hr of 40°C treatment. With the further increment in the temperature, i.e., 45°C, the dissipation energy flux per RC has been profoundly increased, indicating that cells are sensitive to 45°C. Based on the results, it is observed that *C. reinhardtii* exhibited a remarkable ability to sustain growth at a temperature of 40°C, demonstrating its thermotolerance. In contrast, *Haematococcus pluvialis* and *D. salina* did not sustain well under high temperatures at 40°C. *Haematococcus* and *Dunaliella* cells exhibited a substantial reduction in growth capacity, indicating a compromised ability to sustain proper developmental processes. Simultaneously, the efficiency of PS II, a major component of photosynthesis, was adversely impacted by the high temperature in all species. These observations highlight the differing temperature sensitivities and responses between these algae.

Redox State of Electron Carriers in *Hordeum Vulgare* Seedlings Under Combined Influence of Elevated Temperature and *Fusarium Culmorum* Infection

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Global climate change causes a significant increase in the number and intensity of various stress factors. When plants adapt to the effects of combined stress factors, unique responses may occur along with non-specific ones. Abiotic stressors can alter the interaction of plants and pests, increase the susceptibility of the host plant to pathogens. On the other hand, abiotic stress factors can affect the emergence and spread of pathogens. It is obvious that only systematic studies are crucial for understanding the mechanisms of the combined action of abiotic and biotic stress factors on plant productivity.

In this research, we studied the effect of elevated temperature (40° C for 3 hours daily for four days), *Fusarium culmorum* infection and their combined effect on the photochemical activity of photosystem II (PSII) and PSI, the redox state of plastoquinones, ferredoxin and plastocyanin. It was found that exposure to elevated temperature (40° C for 3 h) decreased the level of reduced plastoquinone molecules, decreased the size of photoactive and increased the non-photoactive pool of plastoquinones in 7-day-old seedlings of *Hordeum vulgare* L. Heat treatment caused accumulation of reduced ferredoxin and activation of alternative electron flows from ferredoxin with the participation of plastoquinones. The decrease in cyclic and linear electron flow caused by heating was compensated by activation of electron transport catalyzed by NADH dehydrogenase-like complex (NDH). Elevated temperature significantly accelerated mycelial development and sporulation of *Fusarium culmorum* *in vitro* and *in vivo*. The combined effect of elevated temperature (40°C for 3 h) and *Fusarium culmorum* infection accelerated the development of Fusarium wilt in *Hordeum vulgare* L. seedlings and suppressed photosynthetic activity. In this case, a decrease in the rate of CO₂ assimilation, a drop in the effective quantum yield of photochemical reactions of PSII, suppression of non-photochemical quenching of chlorophyll fluorescence due to its light-induced component and an increase in unregulated non-photochemical quenching were observed. These processes were accompanied by suppression of photoinduced oxidation of P700, activation of non-photochemical energy dissipation on both the acceptor and donor sides of PSI. At the same time, the suppression of the linear and FQR-dependent cyclic electron flow induced by Fusarium wilt and elevated temperature was compensated by activation of NDH-dependent electron transport.

Limited Proteolysis of Chloroplast Proteins of Higher and Lower Plants as a Source of Short Antimicrobial Peptides

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Plants have a growing interest as sources for a variety of biologically active compounds, mainly attributed to secondary metabolites. However, we should not overlook the diverse roles that plant proteins perform, among which the functioning of photosystems I and II, as the foundation of photosynthesis, is highlighted. The sequencing of chloroplast genomes from numerous higher and lower plant species allows for the prediction of the structures of essential proteins involved in this procedure. Interestingly, many of these proteins are appealing as providers of short antimicrobial peptides produced by their limited proteolysis as a result of proteasome-independent degradation. Typically, these peptides lack an ordered secondary and spatial structure and the presence of specific molecular targets is not a defining characteristic. Their antimicrobial action tends to be mainly synergistic, where the combination of several peptides (peptide fragments) exhibits a strong inhibitory effect against a broad range of phytopathogenic fungi, oomycetes, and bacteria. Earlier in our research, we carried out a structural and functional analysis of the products produced by the limited hydrolysis of proteins from the green biomass of plants like greater celandine (*Chelidonium majus*), PE-Cm; elecampane (*Inula helertium*), PE-Ih; common horsetail (*Equisetum arvense*), PE-Eqi; sweet bay (*Laurus nobilis*), PE-Ln; green tea (*Camellia sinensis*), PE-Cs; touch-and-heal (*Hypericum perforatum*)¹; and red algae (*Galdieria sulphuraria*). We identified active peptides, which were the hydrolysis products of prominent chloroplast proteins such as Ribulose-1,5-bisphosphate carboxylase/oxygenase, chloroplast protein RF21, photosystem II proteins, as well as C-phycoyanin and allophycocyanin. These findings guide new directions in the research of the so-called auxiliary system of plant innate immunity against biotic environmental stress factors.

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Transcriptomic Analysis of Cyclic Electron Transport Mutants in *Chlamydomonas Reinhardtii* Under High Light Stress

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Chlamydomonas reinhardtii is a single-celled green alga frequently used as a model organism for photosynthesis research due to its well-studied genome and amenability to genetic manipulation. *pgr5* and *pgr5* are two genes involved in cyclic electron transport (CET) in the light reaction of photosynthesis. PGR5 is a protein kinase that phosphorylates PGRL1, a subunit of the cytochrome b6f complex (cytb6f), which is responsible for transferring electrons from water to plastoquinone during the light reaction of photosynthesis. In this study, we used transcriptomic analysis to compare the gene expression profiles of wild-type and two mutant strains of *Chlamydomonas reinhardtii* (*pgr5* and *pgrl1*) under low and high light conditions. Our analysis revealed 16,609 transcripts in the *pgrl1* mutant and 16,165 transcripts in the *pgr5* mutant under high light, compared to 16,703 transcripts in the wild type (WT). The RNA-seq data demonstrated significant differential expression of photosynthetic core proteins in the mutants compared to the WT. The photosystem (PS)II subunits (*psbA*, *psbB*) were upregulated under high-light conditions across all samples. Conversely, the oxygen-evolving complex (OEC) transcripts (*psbO*, *psbP*, *psbQ*) were downregulated in WT and *pgr5* under high light but upregulated in *pgrl1*. The PSI reaction center proteins (*psaA*, *psaB*, *psaC*, *psaD*) were consistently downregulated under high light stress. We also confirmed these transcripts by real-time PCR, where RNA-seq data is in agreement with RT-PCR data. The results indicate that high light significantly impacts gene expression, with responses varying between CET mutant strains. Identified differentially expressed genes are involved in various biological processes, including photosynthesis. These findings highlight strain-specific responses to light stress, underscoring the need for further research to elucidate the underlying molecular mechanisms.

Autophagy-mediated Protein Degradation in Lower Leaves contributes to the Formation of Vertical Leaf Nitrogen Gradient in the Rice Plant Population grown in the Paddy Field

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Much of the nitrogen (N) in the plant is allocated to chloroplasts and mainly used for photosynthesis. In a dense plant population, the light intensity declines vertically. N per leaf area (N_{area}) also declines as light extinction, resulting in a vertical leaf N gradient. Light and N both decline exponentially; therefore, it becomes the theoretically optimal N distribution to maximize photosynthesis of whole plant population when the coefficients of Light extinction and N gradient are similar. The N gradient of most plant populations is more uniform than optimal, implying that there is room for improvement. In rice plants grown in pot culture, autophagy contributes to the recycling of photosynthetic proteins as an N source for newly emerging leaves. In this study, we analyzed the role of autophagy for the formation of an N gradient in the rice plant population grown in the paddy field. Rice (*Oryza sativa* L.) autophagy-deficient mutant (*atg7-1*) and control plants, including wild type (*ATG7 +/+*) and its parental cultivar (Nipponbare), were grown in the paddy field with 8.8 g N m⁻² fertilized and 22.2 hills m⁻² density twice in 2022 and 2023. Relative light intensity and N_{area} in different cumulative leaf area index (LAI) were measured by the stratified clipping method at heading stage. As a result, the N_{area} of *atg7-1* did not change in the population, even though the light intensity decreased, whereas N_{area} of control plants declined exponentially as light decreased. More N and Rubisco remained in the lower leaves located in the lower layers of the plant population, where limited light is available, in *atg7-1* compared to control plants. NBR1, one of the specific substrates of autophagy, was accumulated in leaves of *atg7-1*, especially in the lower leaves. Those results suggested that autophagy contributes to the formation of vertical leaf N gradient in the rice plant population by degrading proteins in the lower leaves.

Investigation of the Nitrate Assimilation Pathway in the Kleptoplastic Organism *Rapaza Viridis*

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The euglenozoan protist *Rapaza viridis* (NIES-4477) lacks its own chloroplasts but uses temporary kleptoplasts acquired from the green alga *Tetraselmis sp.* (NIES-4478) for photosynthesis and photoautotrophic growth. Like plants and algae, *R. viridis* synthesizes organic molecules by utilizing inorganic ions in the medium. For example, *R. viridis* is able to utilize nitrate ions, and our previous study confirmed that the expressed product of a nitrate reductase-like gene (*RvNaRL*), acquired in its nuclear genome by horizontal gene transfer, is indeed essential for the nitrate assimilation. The present study aims to investigate the function of a possible plasma membrane nitrate transporter-like gene (*RvNRTL*), also likely acquired by horizontal gene transfer, by generating an *RvNRTL* knockout (*RvNRTL* -KO) strain using CRISPR/Cas9. When cultured in a medium containing nitrate as the sole inorganic nitrogen source, the *RvNRTL* -KO cells showed lower growth rate than the wild-type cells. Microscopic observation and quantitative analysis of polysaccharides revealed an increased accumulation of cytosolic polysaccharide grains in the *RvNRTL* -KO cells, suggesting that *RvNRTL* is also essential for the nitrate assimilation and thus likely functions as a nitrate transporter. This presentation will also discuss nitrite reductase, glutamine synthetase, and glutamate synthase that are involved in the nitrate assimilation by *R. viridis*.



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