

PREFACE

Photosynthesis is a well-regulated process utilizing solar energy for storage as chemical energy to meet the plant's metabolic needs. It is the key phenomenon that augments plant growth and development under all conditions and is considered as the ultimate source of life for all plant and human life. Although the basic processes of photosynthesis and their importance to the agricultural system for food security have been recognized about two centuries ago, there is an increasing need for intense research considering the population explosion. As the demand for food is expected to increase twofold by 2030, the efforts by plant scientists are necessary to improve the yield of food crops for sustainable development. The improvements in yield of crops can be attained by the protection of photosynthetic apparatus in the current changing environment, where degradation of soil, air and water quality has posed challenges to photosynthetic potential of plants. It has been estimated that the increasing salinization of land will result in the loss of 50% cultivated land by 2050. The situation will further aggravate with the contamination of soil and water by heavy metals. The concentration of CO₂ is expected to rise to as much as 500-1000 ppm by 2100 which can lead to increases in global mean temperature by approximately 1°C to 3°C above the present value by 2025 and 2100, respectively. Photosynthesis is one the first processes inhibited under the changing environment leading to reduction of growth and productivity of plants. The research focus on targeting mechanisms that could help in the survival of plants under the changing environment is essential for modulating photosynthetic processes to suit the climate changes. There are several processes which can be manipulated to maintain photosynthetic potential of plants. These include up-regulation of photosynthesis-related enzymes and controlling genes, enhancing source-sink potential and inhibiting the feed control. Functional genomics can also be a significant approach to understand the photosynthetic gene function under the influences of climate changes. This book provides information on the functional and structural genomics for better understanding and analysis of photosynthesis under changing climatic conditions. The central theme of the present book is to unravel the mechanisms of photosynthesis under optimal and stressful environments and gain insight into the processes which can lead to a better understanding of the photosynthetic inhibition due to various constraints and provide measures to improve photosynthetic mechanisms under these conditions.

Chapter 1 - Various growth and development phenomenon were described earlier in terms of physiology. With the advent of technology, these have been described in terms of biochemical aspects. With further advancement in technology, molecular approach is being

utilized for the study of growth and development in organisms. In the last two decades, molecular regulation, through genes controlling growth and development, has been studied extensively. Likewise, photosynthesis (a metabolic process) has also been influenced by various factors and the genes responsible has been studied. Photosynthesis is vital to all aspects of plant biology as the provider of energy and assimilator for growth and reproduction, but how it is regulated by abiotic stresses and biotic stresses and their molecular regulatory mechanisms is still not very clear. Photosynthesis is central to the performance of autotrophic plants, not in isolation but rather in combination with the processes that determine growth and development as part of the whole organism's function, reproductive performance and survival. Different environmental factors – abiotic such as temperature, water, and light and biotic such as insect, pests, or pathogens; together with the competition from other organisms – vary unpredictably. Since the mechanism of photosynthesis involves various components, including photosynthetic pigments and photosystems, the electron transport system, and CO₂ reduction pathways, any damage at any level caused by stress may reduce the overall photosynthetic capacity of a green plant. In this chapter, the molecular regulation of photosynthetic response of green plants to environmental stress factors, like extremes of light, water, and temperature together with biotic factors are discussed. Also, chloroplasts are shown as sensor of stress.

Chapter 2 - Elevated atmospheric CO₂ has been shown to influence many key physiological processes of plants. Leaf photosynthesis is typically stimulated during growth with double the present atmospheric CO₂ concentration ([CO₂]). It is not unusual for CO₂ in the rhizosphere to be up to more than 10-fold higher than the atmospheric [CO₂] due to the activities of microorganisms. The authors often question whether CO₂ level in the root-zone (RZ) could regulate plant growth. By using aeroponically-grown lettuce plants, the authors carried out a series of experiments to study the effects of elevated RZ [CO₂] on the photosynthesis of vegetable crops at different air and RZ temperatures in an environmentally controlled room and fluctuating greenhouse. Under two different temperature regimes of 28°C/22°C and 36°C/30°C (day/night) in the environmentally controlled room, it was found that elevated RZ [CO₂] stimulated photosynthetic CO₂ assimilation, with greater increase at higher temperatures. Elevated RZ [CO₂] decreased stomatal conductance at both temperatures, reducing transpirational water loss. When whole lettuce plants were grown under ambient temperatures ranging from 20 to 36°C under a maximal photosynthetic photon flux density (PPFD) of 2100 $\mu\text{mol m}^{-2} \text{s}^{-1}$ inside the greenhouse, the results indicated that elevated RZ [CO₂] protected lettuce plants from dynamic photoinhibition of photosystem II. It was supported by the fact that midday depression of chlorophyll fluorescence F_v/F_m ratios was alleviated in elevated RZ [CO₂] treated plants. Elevated RZ [CO₂] was also found to reduce the negative impacts of high air temperature on photosynthesis, N metabolism, and growth of lettuce plants, resulting in enhanced productivity. The effects of elevated RZ [CO₂] on photosynthesis, productivity, nitrate, total reduced nitrogen, total leaf soluble and Rubisco proteins under different root-zone temperature (RZTs) was also studied in a tropical greenhouse. The results demonstrated that increased photosynthesis and productivity under elevated RZ [CO₂] at different RZTs was partially due to the alleviation of midday water loss, both dynamic and chronic photoinhibition, as well as, a higher turnover of Calvin Cycle with higher ribulose-1,5-bisphosphate carboxylase/oxygenase (Rubisco) proteins. Effects of RZ [CO₂] at different air and RZT temperatures on different aspects of photosynthesis and growth were discussed.

Chapter 3 - Leaf senescence is the last step of leaf development and can be induced by many genetic and environmental factors. The chloroplast and many leaf pigments undergo a progressive degeneration. The leaf functionality is slowed down and several physiological processes are altered. The chlorophyll *a* fluorescence and its derivative indexes can be used to monitor leaf senescence. The loss of leaf functionality is tightly correlated with the photosynthetic activity. The chlorophyll *a* fluorescence and its derived parameters can be suggested as tool to monitor the leaf senescence. Natural and induced senescence such as dark incubation, detachment, salinity stress, ethylene exposure, heavy metal stress etc. will be taken in consideration. The physiological, biochemical and molecular basis connected with the photosynthesis and chlorophyll fluorescence will be described and discussed.

Chapter 4 - The phenomenon of carbon isotope heterogeneity in a photosynthetic biomass became known at the beginning of 1960s, after the pioneering works of Abelson and Hoering (1960) and Park and Epstein (1960, 1961), but the real mechanism of carbon isotope fractionation leading to uneven carbon isotope distribution in a biomass and the reasons for the isotope patterns of the biomolecules remain vague. In this chapter, it is shown that ^{13}C distribution is tightly bound to the oscillatory character of metabolic processes and to the real biosynthetic pathways of metabolites. It was found that due to oscillations, photosynthesizing cells function in regime of filling/depleting metabolic pools. In the phase of depletion the isotope effect-generating reactions are followed by a Raleigh effect, resulting in the dependence of the isotopic patterns of the molecules on the sequence of their syntheses, i.e., in the strict temporal organization of cell processes. It is shown that oscillations in photosynthesis consist of a phase of CO_2 assimilation and a phase of photorespiration. Each phase is followed by carbon isotope fractionation having the effects of opposite signs. As a result, carbon isotope heterogeneity emerges. Post-photosynthetic metabolism, mainly presented as the metabolism of the glycolytic chain, is also presented through oscillations that have two phases: glycolysis and gluconeogenesis. In glycolysis, carbon isotope fractionation occurs. This is one more reason for the isotope heterogeneity of a biomass and works in combination with biosynthetic pathways to determine the isotopic patterns of post-photosynthetic metabolites. Knowledge of the nature of carbon isotope heterogeneity provides a new delicate tool for studies on living organisms. Some possible fields for carbon-isotope data applications with the presented model are considered.

Chapter 5 - There is ample evidence in existing scientific literature that photorespiration acts towards reducing photoinhibition in plants. The internal generation of CO_2 , resulting from the oxidation of ribulose biphosphate by the enzyme RUBISCO acting as an oxygenase, is often sufficient to prevent photoinhibition partly or even completely, whereas photoinhibition is observed, when photorespiration is prevented. To prevent photorespiration in terrestrial plants it is, however, necessary to create a highly artificial composition of air that is replete in CO_2 and/or deplete in oxygen. The use of highly artificial and manipulated atmospheres makes it difficult to assess the environmental significance of photorespiratory alleviation of photoinhibition.

In contrast, aquatic plants naturally grow in an environment where carbon availability as well as ambient oxygen concentration varies from location to location as a result of varying carbonate content in the underlying soil as well as diurnally in the same location as a result of biological processes (photosynthesis and respiration). Probably as a result of the availability of free, dissolved, CO_2 , often limiting for photosynthesis due to the low diffusion rate of CO_2 in water, aquatic plants have evolved a number of different physiological carbon

accumulation strategies, including bicarbonate use, inducible C₄-type photosynthesis and CAM photosynthesis. This makes aquatic plants an obvious tool for the study of photorespiration/photoinhibition interactions.

In this chapter, the author am presenting data from a study on photoinhibition in three aquatic plants with different carbon uptake strategies: *Callitriche cophocarpa*, a plant reportedly relying entirely on dissolved CO₂ in the water for photosynthesis, *Elodea canadensis*, a bicarbonate user, and *Crassula helmsii*, an aquatic CAM-plant, taking up CO₂ during the night, storing it as malate for photosynthetic use the following day. The plants were grown under varying carbon-availability, and were compared based on their photosynthetic oxygen evolution, degree of photoinhibition (evaluated using PAM-fluorometry), and their activity of antioxidative enzymes.

Photosynthetic oxygen evolution decreased with decreasing carbon availability in *Elodea* and *Callitriche*, but not in the aquatic CAM plant *Crassula*, where the internal malate pool partly compensates for low ambient carbon availability. Photoinhibition increased with increasing carbon availability (decreasing photorespiration) in both *Callitriche* and *Elodea*, but not in *Crassula*, where the internal malate pool may prevent any significant photorespiration as well as any significant photoinhibition. Photoinhibition was higher in the bicarbonate user *Elodea*, compared to *Callitriche*, as bicarbonate uptake functions as a carbon concentration mechanism, reducing photorespiration. The antioxidative enzyme ascorbate peroxidase evinced higher activities with lower carbon availability, especially in *Callitriche*, to a lesser degree in *Elodea* and not at all in *Crassula*.

The results confirm the previously described interaction between carbon uptake physiology and photoinhibition. Interestingly, CAM-photosynthesis may prevent both photorespiration and photoinhibition, but further studies are needed.

Chapter 6 - Geological records indicate that plant life on the land has aquatic origins, started approximately 460 million years ago. However, nearly all of the land-dwelling plants are very sensitive to the inundation. Nonetheless, the most basic attributes of flooding tolerance seemingly involved an adaptive return to aquatic competence by terrestrial plants. The negative impact of flooding on terrestrial plant life is a consequence of oxygen deprivation. Flooding often disrupts plant water relations, gas exchange and stomatal conductance. Under the diffusional limitation of carbon uptake imposed by stomatal closure, the photosynthetic capacity of flood-sensitive plants would be significantly declined and can exhibit non-stomatal inhibition of photosynthesis with alterations in plant hormone concentrations induced by tissue anoxia. A tight regulation of the plant water status is thus required to maintain photosynthesis, nutrient uptake and growth in flooded environments. Here, the authors review the importance of stomatal attributes and photosynthesis for the survival of flooded land-dwelling plants.

Chapter 7 - Photosynthetic organisms sustain life by converting light energy into chemical energy. Photosynthesis is a key phenomenon contributing substantially to plant growth and development. It is a well regulated and multistep process to produce assimilatory power NADPH and ATP to synthesize carbohydrate. The changing environmental conditions have forced plant scientists to better understand photosynthetic process more importantly for agricultural sustainability. The abiotic stresses such as salt stress, heavy metal stress, water stress, heat stress, chilling stress, nutrient stress etc. adversely affect photosynthesis because of induced oxidative stress. Various mechanisms may provide protection to the photosynthetic apparatus under abiotic stress, but phytohormones have potential to modify

biochemical, physiological processes and molecular mechanisms in plants to optimize photosynthesis under optimal and stressful environments. They are a strong tool to protect and alleviate stress by modulating metabolic processes in plants. The present chapter gives an insight into the role of phytohormones in modifying biochemical, physiological and molecular responses to adapt plants in stressful environments.

Chapter 8 - Photosynthesis is undoubtedly a vital biological process which contributes substantially towards plant's growth and development. The enzyme ribulose 1, 5-bisphosphate carboxylase (Rubisco) is the most abundant enzyme on planet and plays an important role in autotrophic carbon fixation. It regulates the pool size of important photosynthetic intermediates. It is the key enzyme of the Calvin cycle which catalyzes, the carboxylation and oxygenation of ribulose 1, 5-bisphosphate (RuBP) and is the main enzyme fixing the vast majority of atmospheric carbon, acting as the natural sink for CO₂. Abiotic stress hampers the process of photosynthesis in most of the plants by modifying the ultrastructure of the organelles, concentration of various pigments and metabolites including enzymes involved in this process. It also influences stomatal regulation. Salinity is currently the most important abiotic stress that unfavorably affects the plant growth process and production potential through its impact on various physiological and biochemical process, including the imperative process of photosynthesis. One of the most prominent effects of stresses is the stomata closure. It results in lowering the concentration of intercellular CO₂, which in turn causes deactivation of Rubisco. Understanding the role of Rubisco in the biosphere is very important in today's context, with increased rate of fossil carbon liberated by human activities and flaring the problem of global warming. Further, it is important to enhance our understanding on the expression patterns of Rubisco genes under salt stress, involved in photosynthesis, in order to develop transgenic lines of different crops with enhanced photosynthetic capacity under stressful conditions. Understanding the details of the adverse effect of salinity stress on photosynthesis, focusing on Rubisco, can lead to the design of new salt-tolerant genotypes with better ability to protect photosynthetic potential under salinity stress. The present study focuses on structural and functional aspects of the Rubisco in photosynthesis under optimal and salt stress conditions.

Chapter 9 - Oxidative stress is an imbalance between reactive oxygen species (ROS) generation and scavenging of the reactive intermediates leading to damage and redox disturbances in living cells. It leads to disruption and impairment of cellular sugars, proteins, lipids and nucleic acids. Plants under stress tend to generate and accumulate ROS hindering signal cascades at organelle and cellular levels. Soluble sugars such as fructans, raffinose family oligosaccharides (RFOs), trehalose and polyols in plants have long been characterized to activate antioxidant pathways or themselves act as ROS scavengers under abiotic stress. Herein, the authors discuss their antioxidant functioning during environmental stress in cell organelles such as chloroplasts, mitochondria and peroxisomes. This physiological sugar-antioxidant interplay exerts a control over various genetic and molecular approaches facilitating favourable strategies in developing plants of agronomic and commercial importance.

Chapter 10 - Plants are constantly exposed to pathogens infection and mechanisms of defense must be triggered to avoid their deleterious effect on host physiology. Alterations on host secondary metabolism as a result of pathogens infection have been investigated by many researchers; however, studies reporting their effects on primary metabolism are yet poorly characterized. Defense induction generates energetic costs for their hosts, which require, for

instance, modifications on photosynthetic and respiratory performance besides changes on source-sink patterns, leading to reductions in plant fitness then translated in production and profit losses. It is invaluable the importance of photosynthesis in resistance against pathogens, so the development of strategies that favors plant physiology and enhance the activation of plant defense responses can provide a powerful tool for disease control, delaying or reducing the use of fungicides or pesticides since the demand for sustainable plant-protection strategies is urgent. The application of silicon as a broad plant-protection strategy has been currently evidenced; despite virtually nothing is known about the molecular basis and regulation of silicon-mediated disease resistance. The aim of this chapter is to introduce to the readers some aspects of silicon involvement on plant physiology and thus encourage the discussion and investigation on this intriguing subject.

Chapter 11 - The search of new inhibitors of the photosynthetic process through the screening of natural products is described in this chapter. Additionally, synthetic endeavors toward the development of more effective analogues are also analyzed.

Chapter 12 - According to established knowledge, only 0.1% of the total light energy (visible and invisible) that reaches the surface of our planet earth is transformed into free chemical energy through the chlorophyll molecule. If only the human knew he had the ability to dissociate the molecule of water, and do it irreversibly. With this small fraction the food chain is initiated. Explaining the existing biomass based only on photosynthesis in plants requires the acceptance of dogmas such as the source of human energy is glucose or the ability of the eukaryotic cell to amplify the ATP; concepts that although they are widely accepted cannot be explained rationally. Hence the author's discovery of the intrinsic capacity of the melanin molecule to dissociate and re-form the water molecule involves a conceptual revolution of biblical proportions. It explains and demonstrates that photosynthesis exists in humans and therefore in mammals, and in each and every living entity where melanin is present. Its primary function is the production of free chemical energy from photonic energy, a behavior unique in nature.