Regular paper

The redox state of the plastoquinone pool controls the level of the light-harvesting chlorophyll a/b binding protein complex II (LHC II) during photoacclimation

Cytochrome b₆f deficient Lemma perpusilla plants are locked in a state of high-light acclimation

Dan-Hui Yang¹,⁵, Bertil Andersson¹,², Eva-Mari Aro³ & Itzhak Ohad⁴,∗
¹Department of Biochemistry and Biophysics, Arrhenius Laboratories for Natural Sciences, Stockholm University, 106 91 Stockholm, Sweden; ²Division of Cell Biology, Linköping University, 581 85 Linköping, Sweden; ³Department of Biology, University of Turku, 20014 Turku, Finland; ⁴Center for Photosynthesis Research, Department of Biological Chemistry, The Hebrew University of Jerusalem, 91904 Jerusalem, Israel; ⁵Present address: Department of Chemistry and Biochemistry, University of Guelph, Guelph, Ontario, Canada N1G 2W1; ∗Author for correspondence (e-mail: Ohad@vms.huji.ac.il; fax: +972-2-658-6448)

Received 30 June 2000; accepted in revised form 26 March 2001

Key words: cytochrome b₆f complex, Lemma perpusilla, LHC II, photoacclimation, plastoquinone pool, redox state

Abstract

A cytochrome b₆f deficient mutant of Lemma perpusilla maintains a constant and lower level of the light-harvesting chl a/b-binding protein complex II (LHC II) as compared to the wild type plants at low-light intensities. Inhibition of the plastoquinone pool reduction increases the LHC II content of the mutant at both low- and high-light intensities but only at high-light intensity in the wild type plants. Proteolytic activity against LHC II appears during high-light photoacclimation of wild type plants. However, the acclimative protease is present in the mutant at both light intensities. These and additional results suggest that the plastoquinone redox state serves as the major signal-transducing component in the photoacclimation process affecting both, synthesis and degradation of LHC II and appearance of acclimative LHC II proteolysis. The plastoquinol pool cannot be oxidized by linear electron flow in the mutant plants which are locked in a ‘high light’ acclimation state. The cytochrome b₆f complex may be involved indirectly in the regulation of photoacclimation via 1) regulation of the plastoquinone redox state; 2) regulation of the redox-controlled thylakoid protein kinase allowing exposure of the dephosphorylated LHC II to acclimative proteolysis.

Abbreviations: Chl – chlorophyll; CP43 – chlorophyll a binding protein of Photosystem II; F₉ and Fₐ – room temperature chlorophyll fluorescence yield with all reaction centers open and closed, respectively, F₅ – steady state level of fluorescence; Fᵥ – variable fluorescence = Fₐ – F₉; HL – high light; LHC II – light-harvesting chlorophyll a/b binding protein complex II; LL – low light; Mt – mutant; OG – octyl glucoside; PAGE – polyacrylamide gel electrophoresis; PFD – photon flux density; PQ/PQH₂ – plastoquinone/plastoquinol; SDS – sodium dodecyl sulfate; Sup – supernatant; Wt – wild type

Introduction

Higher plants and green algae adjust the photosynthetic apparatus activity in response to various light conditions, thereby optimizing the photosynthetic productivity (Anderson 1986; Anderson and Andersson 1988; Melis et al. 1996; Huner et al. 1998; Pfannschmidt et al. 1999b; Maxwell et al. 1999). Transient association/dissociation of the light-harvesting antennae with Photosystem II or I (PS II, PS I) occurring during short-term variation in light intensity (state transition) is regulated by reversible phosphorylation
of the light-harvesting II antennae proteins (Bennett 1991; Allen and Nilsson 1997). This process is controlled by a signal transduction chain involving reduced plastoquinone binding to the quinol oxidation site of the reduced cytochrome b6f complex leading to activation of the protein kinase (Vener et al. 1995, 1997, 1998; Gal et al. 1997; Zito et al. 1998, 1999). The possible regulation of protein phosphorylation via thiol groups, most likely through the ferredoxin-thioredoxin system, could represent a second feedback loop of redox control generated by the electron flow (Carlberg et al. 1999; Rintamäki et al. 2000).

Photoacclimation to persistent, high- or low-light levels involves changes in the composition and the structure of the thylakoid membrane (Anderson 1986; Anderson and Andersson 1988; Melis 1991; Anderson et al. 1995; Lindahl et al. 1995; Montane et al. 1998). The LHC II antenna size in plants exposed to high-light intensities decreases as compared to that of plants grown under low light (Osmond 1994; Melis 1996; Baroli and Melis 1998). The underlying mechanisms of this process are largely unknown. Recently, it was discovered that ATP-dependent proteolysis of LHC II occurs during acclimation of low-light grown spinach to high-light intensity (Lindahl et al. 1995; Yang et al. 1998). This proteolytic activity, localized in the stroma membrane domain, is regulated at both the enzyme and the substrate accessibility levels. The reversible phosphorylation of LHC II may be involved in this process (Yang et al. 1998).

The acclimation process also includes regulation of the nuclear encoded lhcb 1,2 gene expression resulting in the increase of the thylakoid LHC II content in plants acclimating to low light (Green and Salter 1996). Several hypotheses have been proposed to account for the light intensity-induced changes in nuclear gene expression (reviewed in Durnford and Falkowski 1997), including involvement of a photoreceptor, possibly phytochrome, or a blue light receptor regulating the expression of lhcb genes at both the transcriptional and translational levels (Green and Salter 1996; Melis et al. 1996; Walters et al. 1999); feedback by the chlorophyll synthesis regulatory system affecting either lhcb gene expression or LHC II protein stability (Johanningmeier and Howell 1984; Mortain-Bertrand et al. 1990; Jasper et al. 1991; Reinbothe and Reinbothe 1996) and feedback from photosynthetic electron transport carriers or a yet undefined end-product of photosynthetic metabolism (Murakami and Fujita 1993; Escoubas et al. 1995).

Photosynthetic organisms maintain a balance between energy input through photochemistry and energy utilization through metabolism. Energy balance plays a regulatory role in the acclimation to light and cold (Huner et al. 1995, 1998). The excitation pressure of PS II affects the abundance of LHC II and xanthophylls (Maxwell et al. 1995a; Montane et al. 1998; Wilson and Huner 2000). Among the thylakoid electron carriers, the plastoquinone pool is considered to be an ideal candidate to signal excess or insufficient PS II activity relative to the capacity for carbon fixation (Pfannschmidt et al. 1999a). In the green alga Dunaliella, transcription of lhcb genes is reversibly repressed by a phosphorylated factor coupled to the redox status of plastoquinone through the activation of a protein kinase (Escoubas et al. 1995; Maxwell et al. 1995b). However, this signaling system has not yet been elucidated in higher plants. It has been suggested that phosphorylation and subsequent migration of LHC II from the appressed to the nonappressed thylakoid regions may initiate a signaling cascade essential for acclimation of plants to changed environmental conditions (Pursiheimo et al. 1998a, b).

In the present study, we have used the wild type and a cytochrome b6f deficient mutant of Lemma perpusilla as a model system to test the possible role of this complex in the photoacclimation process. The results of this work demonstrate that the Lemma mutant is ‘locked’ into a state of high light acclimation when exposed even to low light intensities. This state can however at least partially be reversed to low light acclimation state by blocking the reduction of the plastoquinone pool.

Materials and methods

Growth and acclimation of plants

Lemma perpusilla strain 6746 (wild type, Wt) and mutant strain 1073 (Mt) were grown in liquid medium at 22 °C at a PFD of 5 µmol m−2 s−1 (low intensity light) or 200 µmol m−2 s−1 (high intensity light) using sucrose as a carbon source (Shahak et al. 1976). The light/dark period was 10/14 hours, respectively.