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REVIEW ARTICLE

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Adaptive responses in *Chlamydomonas reinhardtii*

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Summary The photosynthetic single cellular alga *Chlamydomonas reinhardtii* has been used as a model organism to examine in detail the physiological, biochemical and molecular processes of photosynthesis, flagella synthesis and movement, mineral stress, interactions between nucleus, chloroplasts and mitochondria and other processes. In this review we summarize part of the current knowledge on adaptive responses in *C. reinhardtii* when it is exposed to oxidative stress and to changes in light intensity, concentration of minerals, herbicides and metals. The individual responses are linked in order to understand the response of the cell, which is continuously subjected to fluctuations, as a whole.

Key words Herbicides · Carotenoids · Photoprotection · Oxidative stress · Metalloregulation

Introduction

Ecosystems are continuously subjected to naturally occurring changes and additionally to disturbances originated by human activities. The impact of these changes on whole ecosystems depends to a great extent on the responses of single organisms. Generally, organisms are able to adapt to small fluctuations in their environment, or are even resistant to and can survive dramatic changes. To understand and to predict such adaptive responses of organisms, a detailed knowledge of the ecological, physiological, biochemical and molecular principles of their adaptation potential is required. On the molecular level, a detailed understanding of the adaptive responses in prokaryotic organisms has been gathered using various microorganisms, *Escherichia coli* being the major model organism [54]. In eukaryotic organisms, the yeast *Saccharomyces cerevisiae* and mammals have been for a long time the organisms of choice for studies at the molecular level [20, 27]. Only in the last two decades have other eukaryotic organisms like the plants *Arabidopsis thaliana* and *Nicotiana tabacum* gained increased attention [31]. Currently, the unicellular green alga *Chlamydomonas reinhardtii* has developed into a model organism for studying adaptive responses in photosynthetic organisms. *C. reinhardtii* has been extensively used before in studying photosynthesis, chloroplast biogenesis or flagellar function and assembly [48]. Since various molecular and genetic tools are available for this organism, and several mutant strains can relatively easily be created or are already available, *C. reinhardtii* is extremely useful to study adaptive responses at the cellular, physiological, biochemical and molecular levels [14, 28, 30, 57].

In this review, we summarize the current knowledge on different adaptive responses in *C. reinhardtii*. We limit ourselves mainly to responses to naturally occurring fluctuations in the environment and try, by linking those responses, to contribute to the understanding of the adaptive response of *C. reinhardtii* as a whole (Fig. 1).

The basic strategies that organisms in general have to maintain their homeostasis and avoid or deal with stress are the following, some of which will be discussed in more detail below:

(i) Prevention of the penetration of toxic compounds into the cell or avoidance of reactions thereof by complexation, storage or degradation.

(ii) Removal of reactive compounds before they can cause damage (see sections on light stress and oxidative stress).

(iii) Repair of damaged biomolecules (throughout the sections).

(iv) Synthesis of new, alternative biomolecules that take over functions under stress conditions, replace damaged biomolecules or protect them (exemplified in the light and the metal stress sections).

C. reinhardtii does have various complex adaptive mechanisms at its disposal with a fine-tuned regulation thereof that occurs at the transcriptional and translational level, post-transcriptional modifications of proteins and modifications of existing organic compounds. The picture becomes even more complex if the coordinated participation of different compartments is taken into account.

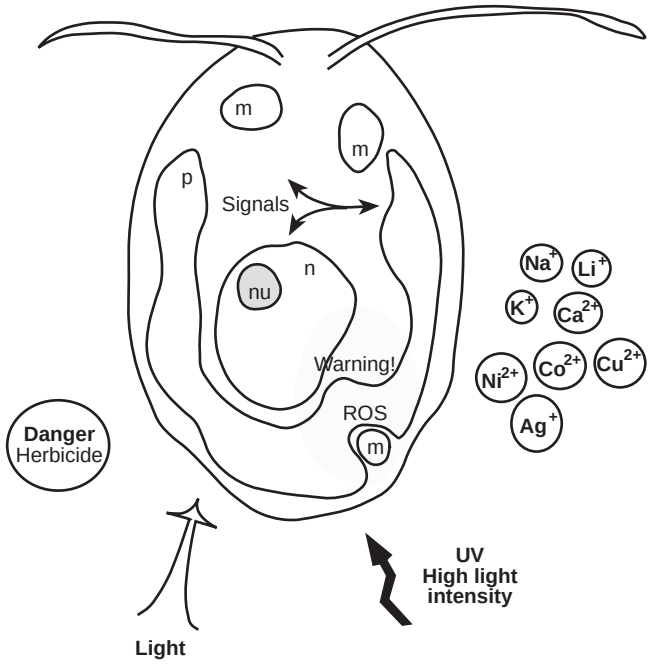

Fig. 1 Schematic representation of a *Chlamydomonas reinhardtii* cell subjected to different stresses. The figure shows that the cell receives light under normal conditions but can be stressed by high light or UV. The cell can be also exposed to other stresses like mineral stress, metal stress or the presence of herbicides. As a consequence of the different stresses, the concentration of reactive oxygen species (ROS) in the cell can increase affecting different cell compartments. The adaptive response of the cell to different fluctuations includes participation and coordination of different cell structures via different signalling pathways. Symbols: chloroplast (p), mitochondria (m), nucleus (n), nucleolus (nu)

Adaptive response to light

The optimization of photosynthesis is an enormous challenge for phototrophic organisms. Under high illumination, where light is a factor in excess, the turnover rate of the reaction centers and velocity of electron transport are rate limiting. Under these conditions, excess excitation energy from absorbed photons can not be used for the oxidation of water (photoinhibition) and the oxidized intermediates generated through light absorption may promote damage to surrounding biomolecules. Photosynthetic organisms have developed many different strategies to minimize and repair such damage. Many of these protecting mechanisms would, however, reduce the photosynthetic efficiency under low light conditions. Hence, in order to assure optimal efficiency of photosynthesis under both strong and weak illumination, protecting mechanisms must be up and down regulated. The lack of electron acceptors (CO_2), nutrient stress, drought or temperature stress, can lead to problems similar to that generated under light stress requiring a protective adaptation as well.

The photosynthetic machinery and light stress

The main damage occurring under photoinhibition is the damage of photosystem II (PSII), of which the chloroplast encoded D1 protein (*psb*A gene), the key PSII reaction center core protein, is the main target [52]. The recovery from photoinhibition requires an increased synthesis of D1 protein. During photoacclimation, the protein synthesizing machinery in the chloroplast must intially cope with a rapid need for more D1 protein, which has been demonstrated to be accompanied by a decrease in the synthesis of the large subunit (LSU) of ribulosebisphosphate carboxylase/oxygenase (rubisco) and by regulation of transcription of *cab* genes which encode the antennae proteins [13, 52]. The concomitant reduction of rubisco LSU synthesis could save cellular nitrogen or other resources that are needed to activate the translation of chloroplast D1 mRNAs.

Other chloroplast proteins are required as well for efficient replacement of the D1 protein, which turns over rapidly under strong illumination. One of these is the product of the chloroplast open reading frame 8 (*ycf*8, also named ORF31 or *psbT*). This protein (PSII-T protein) appears to be a small subunit (3.3 kDa) of PSII essential for maintaining high photosynthetic activity under high light conditions [38]. This polypeptide is highly conserved in location and predicted amino acid sequence in land plants and *C. reinhardtii*. Studies carried out with a *C. reinhardtii ycf*8 defective mutant showed that although the mutant is able to grow phototrophically, photosystem II function and cell growth are impaired under light stress conditions. Since the Ycf8 protein might span the thylakoid membrane, it could help to maintain thylakoid membrane integrity and structure around the PSII.

The relevant role of replacing damaged proteins in the photoprotection process has also been recently demonstrated in the case of photosystem I (PSI). Martin et al. [34] showed that PSII is the primary site of photoxidative damage under conditions of strong light, a combination of chilling and light, or low concentrations (1 mM) of the superoxide producing compound methyl viologen under moderate light conditions, although PSI is the primary site of damage when cells are exposed to higher concentrations of methyl viologen (5–10 mM). When photooxidative stress was caused by methyl viologen, the inhibition of the chloroplast protein synthesis accelerated the damage to PSI. This result suggests a role for newly synthesized chloroplast-encoded proteins in *C. reinhardtii* either in the antioxidative defence, in the replacement or repair of damaged proteins or in the import and processing of nuclear-encoded proteins [34]. The photooxidative inhibition of PSI might be due to oxidative damage caused by superoxide radicals to the iron-sulphur centres of PSI or ferredoxin [4]. Interestingly, oxidation of ferredoxin has been proposed as the reason for the strict anaerobiosis of green sulphur bacteria, whose photosynthetic apparatus has been demonstrated to be evolutionarily related to PSI [4, 15].

In 1996, Danon and Mayfield [7] showed that the light-regulated translation of key proteins in the chloroplast might be modulated by the binding of activator proteins to the 5´ untranslated region (UTR) of chloroplast mRNAs (*psb*A mRNA). This observation was extended by in vitro and in vivo experiments in which the binding of proteins to such mRNAs turned out to be regulated by the redox state of the proteins, suggesting that the light stimulus is transduced via a photosynthesis-generated redox potential. In another study, Nickelsen et al. [41] have shown that the chloroplast *psb*D mRNA, encoding the D2 protein of the PSII reaction center might be stabilized by the binding of a 47 kDa protein to the 5´ untranslated region of the mRNA. The studies described in this section and in previous sections suggest that both the transcription of the nuclear *cab* genes and the translation of the chloroplast D1 and D2 mRNAs are regulated by the redox status of the chloroplast. This makes sense where coordinated regulation of expression is required and where the reduction state is a good measure for photosynthetic activity and the D1/D2 state. Coregulated synthesis of D1 and D2 is further strengthened by the observation that protein D2 not only contributes to the stabilization of the PSII complex in the membrane, but is also involved in the regulation of D1 synthesis at the transcriptional or post-transcriptional level [12].

The transduction of the light stimulus by the photosynthesis-generated redox potential not only regulates the expression of genes directly involved in photosynthesis, but also seems to be a major factor in the control at the cell cycle. In *Chlamydomonas eugametos*, genes whose expression is regulated by illumination are likely to be important in controlling cell proliferation [16]. Nuclear genes that are specifically induced during the dark-to-light or the light-to-dark transitions have been already identified. In the case of one of these genes, light affects the level of expression independently from active photosynthesis. Four genes are positively or negatively regulated upon activation of photosynthesis and one is expressed transitorily at both transitions.

In the context of light-induced expression of genes and stabilization of proteins, the heat shock proteins should be dealt with as well. Although changes of temperature normally occur in nature, fast heat shock is not a usual environmental stress for aquatic organisms. Heat shock response pathways are a relevant topic in the study of *Chlamydomonas* adaptive responses since induction of heat shock proteins (HSP) has been observed not only after the exposure to a heat shock itself but also following exposure to both white light and UV light. Apparently, heat shock response is a universal cellular response that involves the increased expression of a group of genes (*hsp* genes) in organisms exposed to a variety of environmental and physiological stresses [30]. The number and nature of HSP proteins show some variations from one species to the other, but the HSP70 protein is highly conserved during evolution [19]. A chaperone function has been proposed for the HSP70 protein [22]. Under normal circumstances some unattached HSP proteins are available to interact with newly produced proteins. When cells are subjected to stress, many newly translated proteins are unstable and chaperones may need to remain attached to the proteins for longer periods. As a consequence, cells must synthesize additional HSP proteins [1]. In photosynthetic organisms, light regulated production of the HSPs has been suggested as a protection system to maintain a functional photosynthetic apparatus during stress, being involved in the synthesis of new proteins, protein import and refolding [51]. In *Chlamydomonas reinhardtii*, von Gromoff et al. [19] cloned three genes homologous to the *Drosophila hsp*70 heat shock gene and showed that the expression of these genes is regulated at the transcriptional level by both light and thermal stress. The activation of one of them (*hsp*70A gene) by heat stress and light probably occurs via two different signal pathways since the *hsp*70A promoter contains *cis*-acting sequences involved in light induction that do not participate in induction by heat stress. In addition, for the transduction of the inducing light signal the synthesis of specific components is needed [30]. These components turned out to be two chlorophyll precursors, namely Mg-protoporphyrin monomethyl ester and Mg-protoporphyrin, which may replace light in the induction of the *hsp*70A and *hsp*70B nuclear genes, encoding cytosolic and plastid-localized heat shock proteins, respectively [29].

In summary, we can assume that proteins involved in the so-called heat shock response in *C. reinhardtii* act as chaperones both in the cytosol and in the chloroplast. Different signal pathways are followed leading to their synthesis depending on the external stressor. Moreover, it seems possible that the light regulated production of the HSPs is a protection system to maintain a functional photosynthetic apparatus during stress. In order to stimulate this protective function, intermediates of chlorophyll synthesis may act as plastidic signals.

Photosynthesis and stress by herbicides or minerals

The photosynthetic machinery is not only challenged by light. It also plays a central role in response to other stress conditions, e.g. exposure to herbicides or mineral stress. In 1984, a *C. reinhardtii* mutant that was resistant to 3-(3,4-dichlorophenyl)-1,1-dimethylurea (DCMU) and 2-chloro-4-ethylamino-6-isopropylamino-s-triazine (atrazine) was isolated. These herbicides inhibit photosynthesis by preventing transfer of electrons in PSII from the primary stable electron acceptor Q_A on the D2 protein to the secondary stable electron acceptor Q_B on the D1 protein. It has been proposed that the herbicide binds the D1protein at the Q_B binding site instead of Q_A and thereby blocks the electron transfer to Q_B. Nucleotide sequence analysis identified a single T–A to G–C transversion in the *psb*A gene, encoding the D1 protein, as the reason for the observed resistance [11].

This mutation causes a substitution of a serine in the wild type protein for an alanine in the mutant protein, and may cause a structural change in the protein, altering the herbicide binding site of the D1 protein. Interestingly, the aminoacid substitution occurs in a region that is totally conserved between spinach (*Spinacia oleracea*), tobacco (*Nicotiana tabaccum*), pigweed (*Amaranthus hybridus*), soybean (*Glycine max*) and *C. reinhardtii*. Such conservations usually reflect strong functional constraints on the protein. Indeed, the herbicide resistant mutants show higher sensitivity towards excess light. In the case of the pigweed, Hirschberg and McIntosh [25] showed that a herbicide resistant mutant contains a substitution of the same serine residue for a glycine.

When *C. reinhardtii* cells are subjected to mineral stress, as caused by magnesium or sulfur deficiency, an inactivation process of PSII takes place concomitantly with a decrease in the D1 levels and in the quantity of light-harvesting complex of PSII (LHCII) [6]. After cessation of mineral stress, in a few chloroplast proteins there is a cascade of events stimulated by a changed phosphorylation leading to newly synthesized components of the photosynthetic machine. The synthesis of thylakoid proteins increases and the total amount of D1 rises 8 or 9 times. The synthesis of new proteins results in a significant increase of functional PSII and in the production of new LHCII. As a result, the ratio between PSII polypeptides and LHCII increases, which signals the start of the synthesis of new chlorophylls.

In addition to the discussed proteins, other main components of the photosynthetic apparatus show striking changes when *C. reinhardtii* is under stress. When the cells are exposed to high light, UV, oxidative stressors, etc. their carotenoid composition dramatically changes.

Carotenoids: switching from photosynthesis to photoprotection

Carotenoids are involved in at least two main functions in photosynthetic organisms, namely light collection and photoprotection. To develop such functions, carotenoids must interact with chlorophylls, either for the dissipation of energy (in photoprotection) or for passing energy to them (light harvesting in photosynthesis). In higher plants, the role of carotenoids in photoprotection has been clearly demonstrated by the finding of lethal mutations in the carotenoids biosynthetic pathways [8]. In the case of *C. reinhardtii*, in 1958 a green mutant strain was characterized that was deficient in xantophylls and contained a much lower total quantity of carotenoids than the wild type. This strain was able to carry out photosynthesis but could grow only under low light because of its very high photosensitivity. Later, several other mutants deficient in carotenoids were isolated, and many of them also showed a high light sensitivity [21]. In *C. reinhardtii*, when cells were exposed to high light for more than 2 h although no loss of the D1 protein was observed, the b-carotene content was significantly reduced. An increase of the zeaxanthin content was found, which was higher than could be accounted for by the light-induced de-epoxidation of violaxanthin in the xantophyll cycle reactions [10]. The role of the xantophyll cycle is seen in the non-photochemical quenching of excited triplet chlorophyll by zeaxanthin in the antenna and the light harvesting activity of violaxanthin as it passes excitation energy onto chlorophylls. It was postulated that a hydroxylation of b-carotene into zeaxanthin under high light conditions could be an additional source for zeaxanthin that might be involved in non-photochemical quenching (Fig. 2) [9]. Contrary to this postulation is the observation that mutants with a defect in either the a- or b-branch of carotenoid biosynthesis exhibit less non-photochemical quenching but are still able to tolerate high light [42]. A double mutant, however, affected in both a and b branches had almost no non-photochemical quenching and was extremely sensitive to high light. These results suggest that in addition to the xantophyll cycle pigments, a-carotene-derived xantophylls, such as lutein, work not only as structural components of the subunits of the light-harvesting complexes but also as photoprotective elements. Carotenoids have also been described to have a very important protective role in several heterotrophic microorganisms [Mendez-Alvarez S, Rüfenacht K, Eggen RIL, in preparation; 39]. These results suggest a conserved scavenging function of carotenoids, saving the cells from the harmful effect of reactive oxygen species generated in different processes.

Adaptive responses to oxidative stress

Molecular oxygen is used by most respiring organisms as a terminal electron acceptor. Due to partial reduction or excitation of oxygen during various processes and in various parts of the cell (e.g. at the chloroplast during photosynthesis, at the mitochondria during respiration, etc.) highly toxic reactive oxygen species (ROS) such as superoxide radicals, hydrogen peroxide, single oxygen or hydroxyl radicals may arise [20]. An increase in the generation of such ROS beyond the ability of antioxidant defenses to cope with them is called oxidative stress, also defined as a disturbance in the prooxidant-antioxidant balance in favor of the former [53], resulting in the oxidation of biomolecules and subsequently, in a decreased growth and survival of organisms [27]. As it has been discussed in other reviews, oxidative stress is generated when organisms are exposed to various environmental pollutants like transition metals, herbicides, halogenated compounds and radioactivity [2]. Moreover, the production of reactive oxygen species is also increased when organisms are exposed to higher visible- and UV-light intensities or drought [23, 40, 56]. Against these toxic oxygen species, aerobic organisms possess several enzymatic and

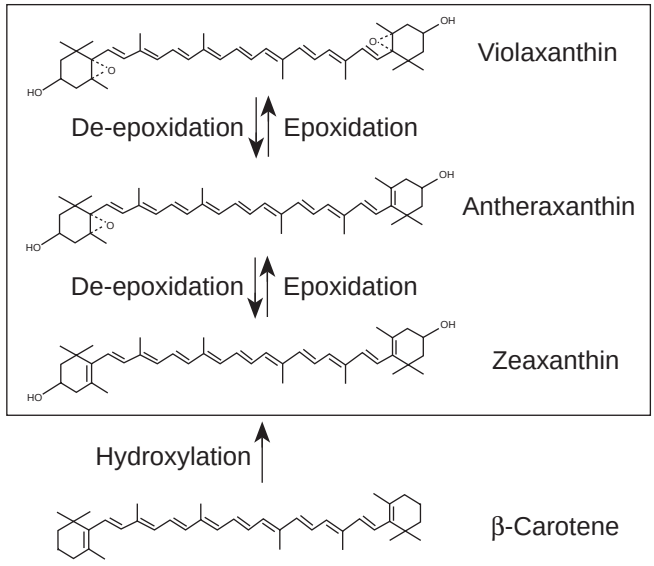

Fig. 2 In *Chlamydomonas reinhardtii* cells exposed to high light intensities, zeaxanthin is not only produced by light-induced de-epoxidation of violaxanthin in the xanthophyll cycle reactions (shadow square) but also by hydroxylation of b-carotene

non enzymatic defense mechanisms. As described above, carotenoids play an important protective role in the photo-induced oxidative stress.

The enzymatic defense known in *C. reihardtii* is mainly based on the activity of some well known enzymes like catalases, peroxidases like gluthatione peroxidase (GSH), ascorbate peroxidase (APX) and superoxide dismutases (SOD) that have already been studied in detail in higher plants [35, 43, 55]. In the non enzymatic response, glutathione, ascorbate, vitamin E and carotenoids have a special relevance as scavengers [8, 39]. Indirect antioxidant functions are carried out by enzymes that replenish the pool of scavengers and of reduced substrates for the antioxidant enzymes and by enzymes that transport and eliminate toxic compounds [32]. Several of these enzymes have been studied, cytosolic and chloroplastic activities have been compared, and attempts have been made to understand the whole cellular oxidative stress response [47]. The mentioned studies have been mainly performed at the physiological and biochemical level and only recently have the genes encoding these enzymes been isolated. This allows for a more detailed examination of the adaptive response to oxidative stress. Recently, our lab has been successful in cloning and characterizing a less known glutathione peroxidase homologous gene and an ascorbate peroxidase gene that are involved in the oxidative stress defense [Leisinger U, Rüfenacht K, Pesaro M, Zehnder A, Eggen R, Structure of a glutathion peroxidase homologous gene and its function in the oxidative stress response in *Chlamydomonas reinhardtii*. Submitted; and unpublished data]. Current elucidation of their function will give more hints towards the cellular oxidative stress response.

Adaptive responses to metals

Numerous metals are known to have specific biochemical functions and to be nutritionally essential for life. However, as a result of their chemical reactivity, many metals are also deletereous to biological systems by several different mechanisms. For this reason, organisms have developed different metal homeostasis strategies to control the availability of simultaneously essential and troublesome metals. Copper is a well known example of a metal that is essential for life but is toxic at high concentrations. For example, copper has an essential function in the catalysis of respiration (cytochrome oxidase), oxygen transport (hemocyanin) and photosynthesis

(plastocyanin). In *C. reinhardtii*, the biological significance of the copper-regulatory mechanisms is indicated by the fact that high concentrations of copper induce deflagellation of the cells and even higher concentrations may cause the cells to encyst [17]. Terrestrial photosynthetic organisms absolutely need Cu in order to preserve plastocyanin levels that are high enough for subsistence. Aquatic algae and cyanobacteria, on the contrary, can be photosynthetically active under Cu-deficient growth conditions. To maintain photosynthetic activity under copper-deficient conditions, *C. reinhardtii* substitutes a heme protein (cytochrome *c*6) for an otherwise essential copper protein, viz. plastocyanin [37]. This replacement is regulated at the transcriptional level by the availability of copper. Coordinated with the expression of the cytochrome *c*6 gene is the expression of the gene encoding coproporphyrinogen oxidase, an enzyme in the heme biosynthetic pathway. This gene is located in the nucleus and, like the cytochrome *c*6 gene, its transcription is induced by the absence of copper. This finding supposed a novel metalloregulatory response in which the synthesis of one redox cofactor (heme) is controlled by the availability of another (Cu). As another part of the copper homeostasis in *C. reinhardtii*, copper uptake mechanisms have been described [24]. The authors demonstrated the presence of a saturable and temperature-dependent copper uptake pathway which operates under both copper-containing and copper-deficient conditions. The uptake mechanism is fine-tuned and avoids that *C. reinhardtii* cells maintained in fully copper-supplemented conditions should accumulate copper in excess. Even if copper is in the environment, it might be in a non-available chemical form for the uptake mechanisms of the cells. In *C. reinhardtii*, a cupric reductase activity, which is increased under copper-deficient conditions, has been described [24]. This enzyme seems to be involved in a copper reductive assimilation pathway. This observation is supported by the fact that cupric reductase activity is regulated in coordination with copper uptake, and more importantly, that the changes in copper transport and cupric reductase activity occur in response to physiologically relevant copper ion concentrations.

In the presence of other metals *C. reinhardtii* has various adaptive mechanisms to avoid toxic effects of excess of metals. Cells that were cultured in the presence of sublethal concentrations of the toxic heavy metal cadmium synthesized and accumulated peptides consisting solely of glutamic acid, cysteine and glycine, which were able to sequester most cadmium and avoided lethality [26]. When the cells were cultured in the presence of mercury, glutathione was the principal thiol-containing compound produced at increased levels, what might indicate a heavy metals detoxification function for glutathione as described for other eukaryotic organisms [44]. Finally, cells treated with silver produced a sulfur containing peptide, although it was not accumulated at high levels [26]. The complexation of metals is also described to occur in the cell wall of *C. reinhardtii*, which possesses a high affinity for metallic cations [5]. Comparative studies of the toxicity of Cd, Co, Cu, and Ni to walled and wall-less strains of *C. reinhardtii* showed that the wall-less strain was consistently more sensitive than the walled strain to all four metals, indicating that the cell wall plays a role in conferring metal tolerance [33]. These authors described that for both strains the toxicity of all metals was pH-dependent, being less toxic at pH 5 than at pH 7. Since there were no differences in the magnitude of the pH effect between the algal strains, it was suggested that there must be considerable competition for ionic binding sites on the plasma membrane.

In the case of other cations, namely Na^+, K^+, Li^+ and Ca^{2+}, adaptive responses that are involved in regulating the internal concentrations of these cations were described. The isolation of different mutants allowed to postulate the existence of several genes involved in the maintenance of ion homeostasis, involving interlinked regulation of Na^+, K^+ and Li^+ fluxes across the plasma membrane together with the expression of proteins with osmoregulatory functions, such as the synthesis or the accumulation of glycerol in the cytosol. It has been determined that genes involved in glycerol biosynthesis and Na^+/Li^+ efflux are activated by both ionic and non ionic osmotic signals, implying that components of the regulatory pathways are shared [36, 45]. With respect to calcium, Quarmby and Hartzell [46] demonstrated that *Chlamydomonas* cells contain two distinct signal transduction pathways, which are involved in the deflagellation process and react to external changes. The deletereous effects of calcium stress on *C. reinhardtii* cells are underlined not only by deflagellation but also by the fact that phototaxis and light-induced stop responses are also known to be calcium dependent [18]. The Ca^{2+} fluxes in the cell are turned into action by changes in calmodulin (CaM) actions among others. CaM, a ubiquitous calcium-binding protein, regulates many cellular processes and stimulates the activity of several target proteins. Its action is dependent on the presence of calcium and is mediated through the binding of the Ca^{2+}-CaM complex to the respective proteins [3]. In *Chlamydomonas*, calmodulin mRNA production is possibly coupled with the synthesis of other proteins on the flagellar apparatus [58]. Moreover, CaM has a potential role in metal toxicity since, besides calcium, other metals, i.e. Cd, Zn, and Pb, are capable of binding to CaM and activating CaM-dependent enzymes [3].

Conclusion

All environments vary in space and time. Populations may respond by evolving a diverse array of responses based on phenotypic plasticity and genetic variation. Adaptive responses are regulated by coordinated cascades of events, involving different cellular compartments and structures, several kinds of biomolecules and various signaling systems. In the unicellular chlorophyte *Chlamydomonas reinhardtii*,

adaptation implies participation and coordination of different genomes, several organelles and multiple pathways to maintain cellular integrity and the cell cycle. According to its phototrophy, the *Chlamydomonas* metabolism and cell cycle are strongly dependent on light. As a consequence, the photosynthetic apparatus has a remarked relevance and nuclear and chloroplast encoded genetic informations are interlinked and co-regulated. Subsequently, adaptive responses strongly depend on different light-dark response pathways and qualitative and quantitative pigment composition. Moreover, these functions are interlinked to different enzymatic and non-enzymatic mechanisms that permit the algae to adapt under several stressful conditions such as oxidative stress, mineral stress, treatment with herbicides, presence of metals or high concentrations of other ions, etc. The coregulation of all these functions is the cornerstone that permits *Chlamydomonas* to maintain a normal cell cycle and to adapt to an always changing environment.

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