

The Rich World of Applicable Algal Photoreceptors

Arash Kianianmomeni*

Institute of Biology, Experimental Biophysics, Humboldt- University of Berlin, Invalidenstrasse 42, D-10115 Berlin, Germany

Free-swimming algae need light not only to produce chemical energy by photosynthesis, but also to initiate and regulate many survival processes like gene expression, protein and lipid synthesis [1]. In addition, some fundamental cellular processes such as cell division and cellular differentiation during sexual and asexual development are also affected or controlled by light, especially in complex multicellular algae [2,3]. To identify suitable environments for growth and development during their fast life cycle, the photosynthetic microalgae measure the changes in light parameters, i.e. quality, quantity, direction and duration permanently. Therefore, a highly sensitive light reception system is needed for high accurate light monitoring which introduces fast changes in swimming behavior directly or indirectly or light-dependent signal transduction cascades that lead to physiological changes.

Fast reacting light-sensitive proteins, so-called photoreceptors, would be required to overcome this huge task towards adaptation of biological and physiological behavior to the permanent environmental changes. Despite the growing body of knowledge regarding light-induced movements of algae in the last century [4,5], the molecular background of light reception and signal transduction remain mostly unclear until the last couple of years, when genome and transcriptome sequencing data identified new types of photoreceptors. Algal genomes, example *Volvox* and *Chlamydomonas*, possess at least three cryptochromes and one phototropin blue light photoreceptors [6,7], similar to plants. Even though red light has been shown to be involved in controlling the expression of photosynthesis-related genes, no phytochromes or other red light absorbing protein has been identified so far in green algae. This could possibly be due to the fact that other photoreceptors may take the red light absorbing function in these species, which has been confirmed recently by investigation of animal-like cryptochrome of *Chlamydomonas* (highlighted in Figure 1 by yellow box), absorbing both blue and red light [8].

The genomes of unicellular *Chlamydomonas* and multicellular *Volvox* possess also seven rhodopsin-like photoreceptors which act as light activated ion channels or enzymes [9]. The best known examples of this group are channelrhodopsins (ChR), the light-gated ion channels, which have become widely used in neuroscience as novel revolutionary tools for fast neural control, even in freely moving mammals [10]. But to optimize the channelrhodopsins for broad application in neuroscience, some properties like photocurrent, inactivation time and absorption spectrum have to be improved toward precise control of neurons. A lot of engineered channels with improved properties have been reported in the last years [11]. In addition to engineering strategy towards perfecting these channels, the sequence data from other algae may also reveal new proteins with different ion selectivity, absorption spectra and kinetics. Moreover, they could allow the creation of new synthetic channels using domain swapping between different ChR species. Lately reported channelrhodopsins from *Volvox* and *Mesostigma* with red-shifted absorption spectra have given a leap in development of new optogenetic tools [12,13], at least through identification of responsible amino acid residues for red-shifted spectra for effective structure-based engineering [14]. Clearly, new proteins from other algae, especially other species showing different behavioral responses such as swimming speed or action spectrum of phototoxic

(like *Cryptomonas rostratiformis* [15], Figure 2) which are also far from known channelrhodopsin's revealed algae, could enrich the variety and promote faster development of optogenetic tools. In the long run,

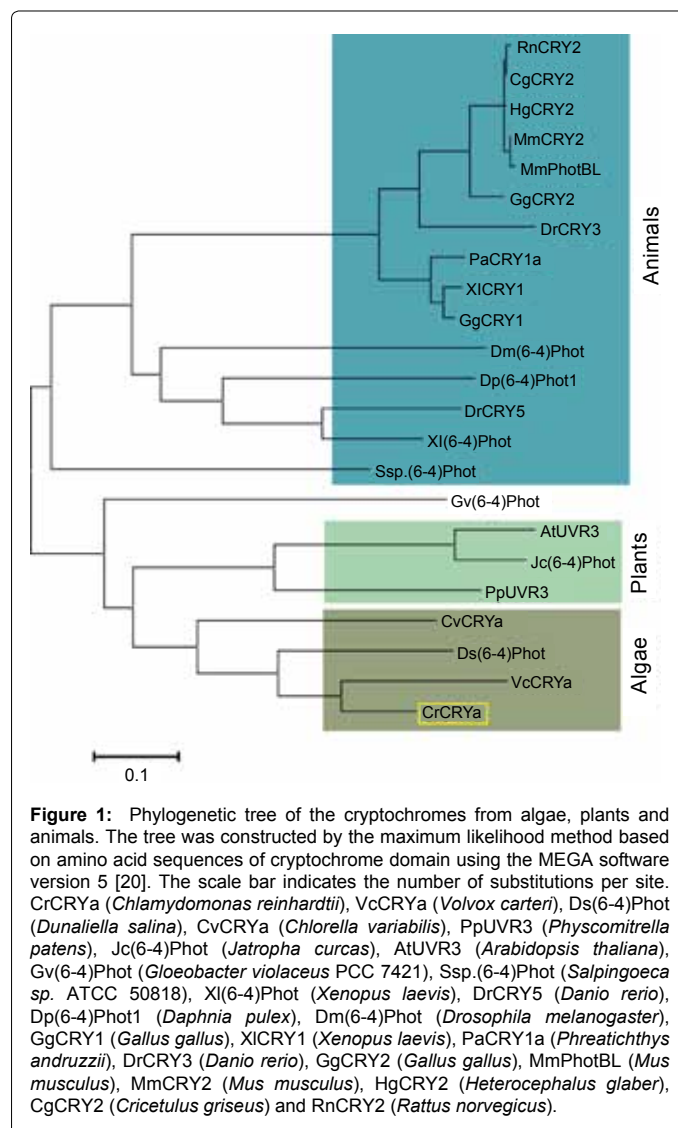


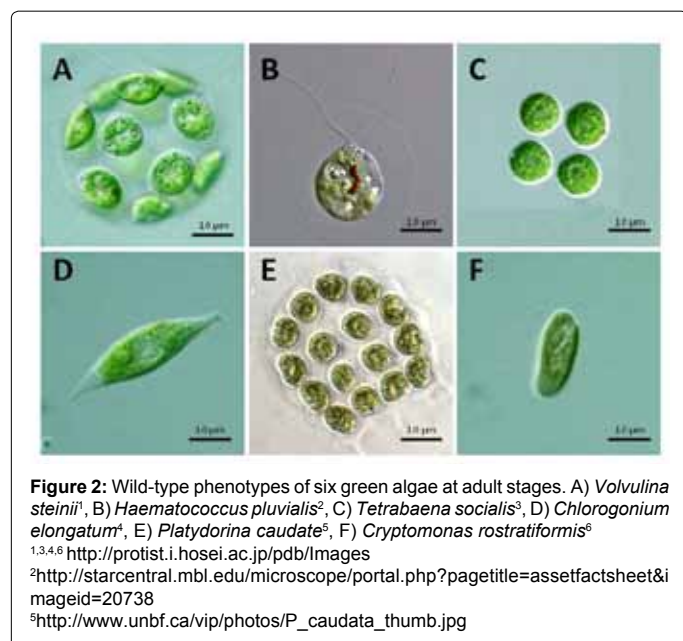
Figure 1: Phylogenetic tree of the cryptochromes from algae, plants and animals. The tree was constructed by the maximum likelihood method based on amino acid sequences of cryptochrome domain using the MEGA software version 5 [20]. The scale bar indicates the number of substitutions per site. CrCRYa (*Chlamydomonas reinhardtii*), VcCRYa (*Volvox carterii*), Ds(6-4)Phot (*Dunaliella salina*), CvCRYa (*Chlorella variabilis*), PpUVR3 (*Physcomitrella patens*), Jc(6-4)Phot (*Jatropha curcas*), AtUVR3 (*Arabidopsis thaliana*), Gv(6-4)Phot (*Gloeobacter violaceus* PCC 7421), Ssp.(6-4)Phot (*Salpingoeca sp.* ATCC 50818), XI(6-4)Phot (*Xenopus laevis*), DrCRY5 (*Danio rerio*), Dp(6-4)Phot1 (*Daphnia pulex*), Dm(6-4)Phot (*Drosophila melanogaster*), GgCRY1 (*Gallus gallus*), XICRY1 (*Xenopus laevis*), PaCRY1a (*Phreatichthys andruzzii*), DrCRY3 (*Danio rerio*), GgCRY2 (*Gallus gallus*), MmPhotBL (*Mus musculus*), MmCRY2 (*Mus musculus*), HgCRY2 (*Heterocephalus glaber*), CgCRY2 (*Cricetulus griseus*) and RnCRY2 (*Rattus norvegicus*).

*Corresponding author: Arash Kianianmomeni, Institute of Biology, Experimental Biophysics, Humboldt- University of Berlin, Invalidenstrasse 42, D-10115 Berlin, Germany, E-mail: kianiana@hu-berlin.de

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histidin kinase rhodopsins [16] which are enable to change cAMP/cGMP concentration in a light dependent manner are of high interest in designing new light-derived protein switches to control organismal physiology. Nevertheless, other light-driven ion pumps with potential application [17] could contribute to the achievement of applicable tools, especially in case of improved recovery time which allows to drive neuronal firing at higher frequencies.

The upcoming genome sequencing data from a few green algae together with the transcript sequencing of around 150 other green algae species (Onekp Project; <http://www.onekp.com>) will reveal new light-drivable molecules with modified properties. But not only the light-gated channels/pumps would then be in the focus of interest, but also other classical photoreceptors such as phototropin and cryptochromes, which have recently gained importance to be used as synthetic photo switches and fluorescent reporters in living organisms [18,19], providing additional natural tools for light control of cellular and physiological activities in living organisms.

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