Biochimica et Biophysica Acta 1837 (2014) 710–716

Contents lists available at ScienceDirect

Biochimica et Biophysica Acta

journal homepage: www.elsevier.com/locate/bbabio

Review

Diversity of animal opsin-based pigments and their optogenetic potential☆

Mitsumasa Koyanagi, Akihisa Terakita *

Department of Biology and Geosciences, Graduate School of Science, Osaka City University, 3-3-138 Sugimoto, Sumiyoshi-ku, Osaka 558-8585, Japan

ARTICLE INFO

Article history:
Received 11 May 2013
Received in revised form 30 August 2013
Accepted 6 September 2013
Available online 13 September 2013

Keywords:
Non-visual pigment
Phototransduction
Opsin diversity
Optogenetics

ABSTRACT

Most animal opsin-based pigments are typical G protein-coupled receptors (GPCR) and consist of a protein moiety, opsin, and 11-*cis* retinal as a chromophore. More than several thousand opsins have been identified from a wide variety of animals, which have multiple opsin genes. Accumulated evidence reveals the molecular property of opsin-based pigments, particularly non-conventional visual pigments including non-visual pigments. Opsin-based pigments are generally a bistable pigment having two stable and photointerconvertible states and therefore are bleach-resistant and reusable, unlike vertebrate visual pigments which become bleached. The opsin family contains Gt-coupled, Gq-coupled, Go-coupled, Gs-coupled, Gi-coupled, and Gi/Go-coupled opsins, indicating the existence of a large diversity of light-driven GPCR-signaling cascades. It is suggested that these molecular properties might contribute to different physiologies. In addition, various opsin based-pigments, especially nonconventional visual pigments having different molecular characteristics would facilitate the design and development of promising optogenetic tools for modulating GPCR-signaling, which is involved in a wide variety of physiological responses. We here introduce molecular and functional properties of various kinds of opsins and discuss their physiological function and also their potentials for optogenetic applications. This article is part of a Special Issue entitled: Retinal proteins — you can teach an old dog new tricks.

© 2013 Elsevier B.V. All rights reserved.

1. Introduction

In animals, the rhodopsin-like photopigments (here after called opsin-based pigments) act as photoreceptors for vision and other non-visual functions such as circadian photoentrainment and pupil response. Most animal opsin-based pigments are typical G protein-coupled receptors (GPCR) and consist of a protein moiety, opsin, and 11-*cis* retinal as a chromophore [1–4]. Upon light absorption, the chromophore in the opsin-based pigment undergoes 11-*cis* to all-*trans* isomerization (Fig. 1), triggering a G protein-mediated signal transduction cascade, which alters the intracellular conditions within photoreceptor cells and causes a cellular response, generally an electrical response. Because the molecular properties of opsin-based pigments are largely responsible for the functional characteristics of light receptions and responses of photoreceptor cells, they have been a primary target for investigations within the fields of vision and photoreception research. In addition, opsin-based pigments are of extreme interest as "light switch molecules," which can be used for regulating physiological responses of target cells by light through exogenous introduction into the cells. Using light instead of chemicals as a stimulus allows us to modulate intracellular conditions with high temporal and spatial precision. Therefore, opsin-based pigments have the potential to be an ideal tool for the investigation of a wide variety of physiological functions. In this study, we review the diversity of opsin-based pigments, particularly non-conventional visual pigments, that have recently been identified and characterized and assess their contribution to the field of optogenetics.

2. Diversity of opsin-based pigments and the phototransduction cascade

In the early 1990s, the known members of the opsin family consisted of vertebrate visual pigments (e.g. bovine rhodopsin), which couple with transducin (Gt)-type G proteins, invertebrate visual pigments (e.g. squid rhodopsin), which couple with Gq-type G proteins, and the non-GPCR photoisomerase retinochrome, which is involved in the visual cycle. In 1994, the first non-visual pigment pinopsin, was discovered and found to be similar to Gt-coupled vertebrate visual pigments but function as a pineal photoreceptor [5,6]. Furthermore, after our discovery of Go-coupled opsin, which forms a distinct group, in scallops in 1997 [7] and the subsequent identification of other novel opsins along with the whole genome sequences of several animals and their functional analyses, the diversity of opsin-based pigments was gradually revealed. To date, several thousand opsins have been identified and phylogenetically classified into eight groups based on the members that existed early in animal evolution (Fig. 2). The classification also roughly corresponds to its molecular function, and the following are

☆ This article is part of a Special Issue entitled: Retinal proteins — you can teach an old dog new tricks.

* Corresponding author. Tel.: +81 666053144; fax: +81 666053171.
E-mail address: terakita@sci.osaka-cu.ac.jp (A. Terakita).

0005-2728/$ – see front matter © 2013 Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.bbabio.2013.09.003

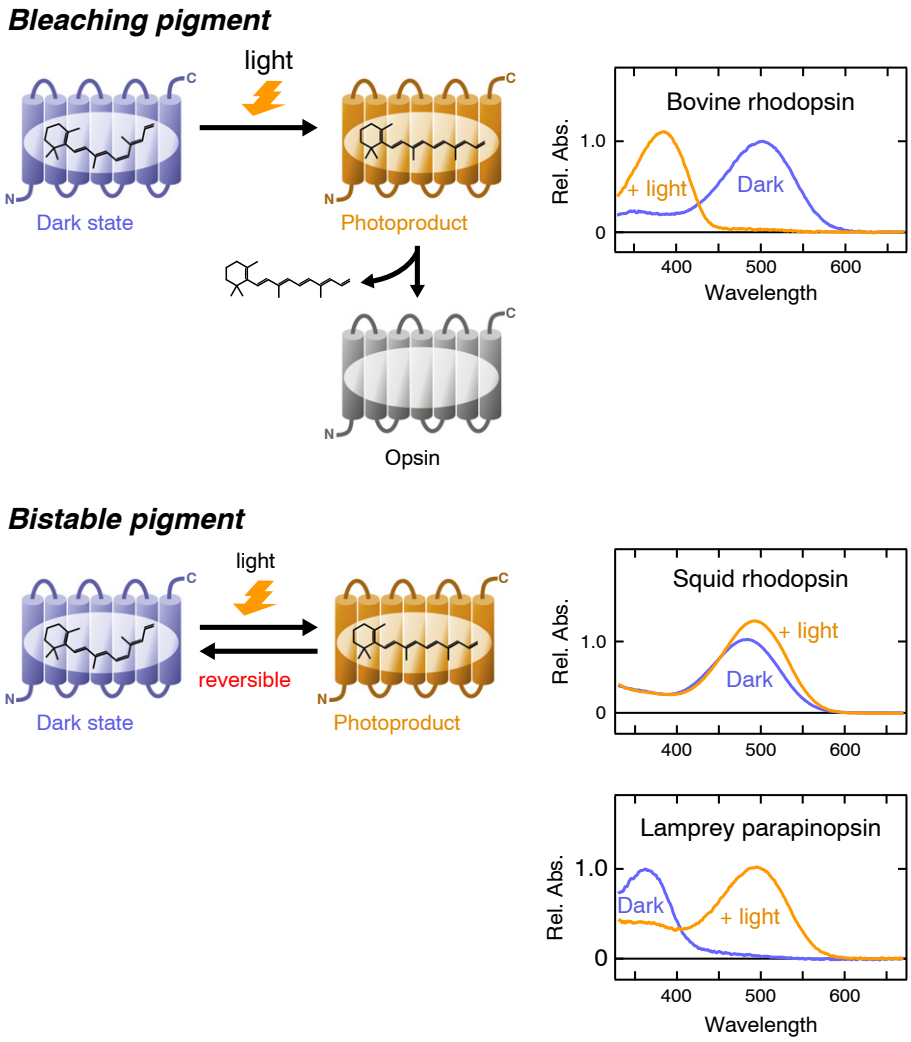

Fig. 1. Biochemical and spectroscopic properties of the bleaching and bistable pigments. The photoproduct of bleaching pigments such as vertebrate visual pigments is thermally unstable and releases its retinal chromophore. On the other hand, the photoproduct of bistable pigments is thermally stable and reverts to the original dark state by subsequent light absorption. Absorption spectra of the dark state and irradiated state of bovine rhodopsin, squid rhodopsin and lamprey parapinopsin are also shown as representatives of bleaching pigment and bistable pigment. In the case of UV-sensitive lamprey parapinopsin, the photoproduct having spectral sensitivity in visible region completely reverts to the original dark state by orange light, unlike in the squid rhodopsin, which has an absorption maximum similar to its photoproduct.

members of the six groups: Gt-coupled opsin (vertebrate visual and non-visual pigments), Gq-coupled opsin (invertebrate visual pigment and melanopsin), invertebrate Go-coupled opsin, Gi/Go-coupled Opn3 (encephalopsin and TMT opsin), Gi-coupled Opn5 (neuropsin), and cnidarian Gs-coupled opsin. These are demonstrated to function as light sensing GPCRs, and members of the other two groups, retinochrome and peropsin, are considered to be retinal photoisomerases that produce 11-*cis* retinal (Fig. 2). Because the whole genome sequences of several animals representing most phyla, including humans, have been determined, the present opsin repertoire provides an overview of the diversity of animal opsins and phototransduction cascades.

Recently, we hypothesized that animal phototransduction cascades can be evolutionary and functionally classified into two groups: phosphoinositol signaling in rhabdomeric-type photoreceptor cells and cyclic nucleotide signaling in ciliary-type photoreceptor cells [8] (Fig. 3). Rhabdomeric-type photoreceptor cells and ciliary-type photoreceptor cells are two major animal photoreceptor cell types, which are classified depending on whether the photopigment is borne on the apical microvilli or disk membrane of a modified cilium, respectively [9]. Phosphoinositol signaling is driven by Gq-coupled opsin-based pigments such as invertebrate visual pigments and melanopsin [10]. In molluscan and arthropod visual cells, which are typical rhabdomeric photoreceptor cells, Gq-coupled visual pigments activate Gq, which in turn stimulates phospholipase C to hydrolyze phosphatidylinositol 4,5-bisphosphate (PIP2) into diacylglycerol (DG) and inositol 1,4,5-triphosphate (IP3). This initiates the phosphoinositol cascade, which leads to a depolarizing response of the photoreceptor cells [2]. In *Drosophila*, proton and depletion of PIP2 underlie the opening of transient receptor potential (TRP)/transient receptor potential-like (TRPL) channels to generate a depolarizing response [11]. On the other hand, a DG-related unsaturated fatty acid can also open the channels [12]. In the arthropod lineage, Gq-coupled visual pigments have a diverse spectral sensitivity that ranges from short to long wavelengths, indicating that the arthropod color vision independently evolved from that of vertebrates during the early arthropod evolution [13]. Until recently, it was difficult to obtain a large amount of purified invertebrate visual pigment using cultured cell systems; however, honeybee UV- and blue-sensitive visual pigments, small white butterfly violet- and blue-sensitive visual pigments, and jumping spider green-sensitive visual pigments have now been successfully expressed in HEK293 cells, and such purified pigments have since been spectroscopically investigated [14–16] (Table 1). Melanopsin, also called Opn4, is an opsin gene orthologous

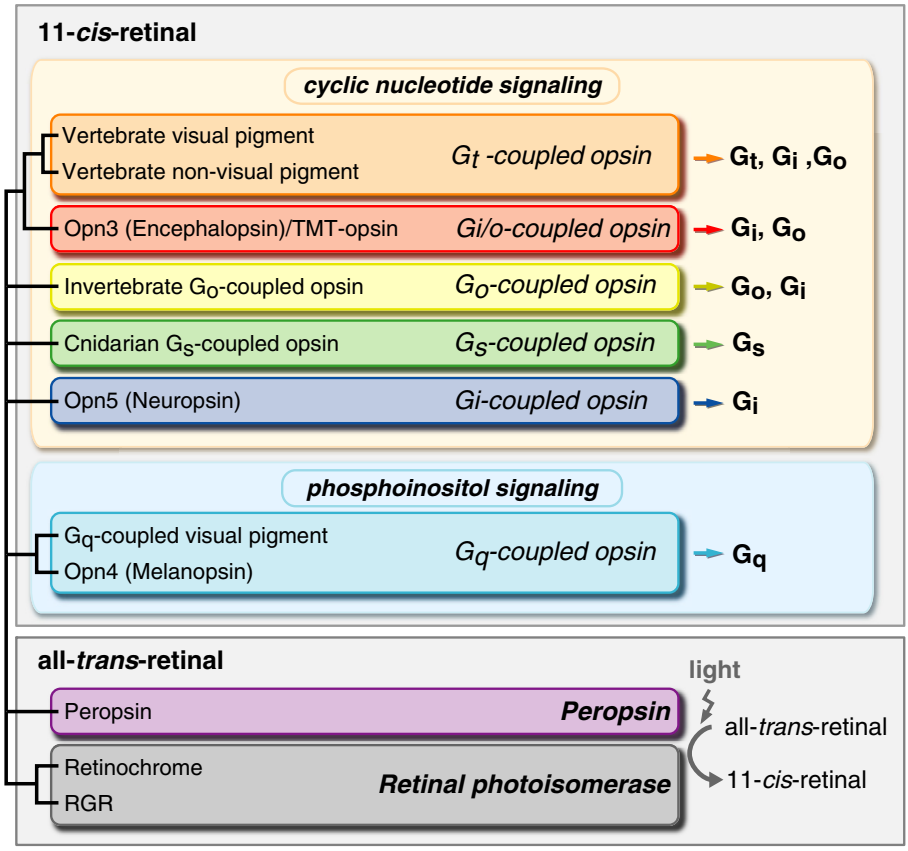

Fig. 2. Functional diversity of opsin-based pigment. Several thousands of opsins are phylogenetically and functionally classified into eight groups. Members of six groups bind 11-*cis* retinal as a chromophore and form photopigments that activate G protein-mediated signal transductions, and the signaling cascades are classified into two groups, cyclic nucleotide signaling triggered by Gt-coupled, Gi/o-coupled Go-coupled, Gs-coupled and Go-coupled opsin-based pigments and phosphoinositol signaling triggered by Gq-coupled opsin-based pigments. G protein subtypes that are activated by opsin-based pigments in each group in vitro are also shown on the right side. Members of the remaining two groups, peropsin and retinal photoisomerase groups bind all-*trans* retinal as a chromophore and light isomerizes it to the 11-*cis* form. Note that this schematic phylogeny of opsin family is based on the molecular phylogenetic analysis including opsin sequences that were revealed to function as a photopigments and their apparent homologs [8].

to invertebrate Gq-coupled visual pigments and is involved in non-image forming vision, i.e., pupil responses to light and photoentrainment of the circadian rhythm in mice [17–20]. Melanopsin has spectroscopic characteristics that are almost identical to those of invertebrate Gq-coupled visual pigments and also drives the Gq-mediated phosphoinositol signaling cascade [14,21,22]. However, overall signaling cascade of mammalian melanopsins is not yet well elucidated [23]. Melanopsin localizes in rhabdomeric photoreceptor cells in cephalochordate amphioxus and mammalian intrinsically photosensitive retinal ganglion cells, which express melanopsin [10] (Fig. 3). This raises the hypothesis that these cells and invertebrate rhabdomeric visual cells share a common origin with phosphoinositol signaling mediated by Gq-type G proteins.

Cyclic nucleotide signaling (cAMP and cGMP) involves some G protein subtypes and effector enzymes that commonly act to regulate cyclic nucleotide-gated (CNG) channels in ciliary photoreceptor cells from prebilaterian to vertebrate [8] (Fig. 2). Vertebrate visual pigments activate transducin (Gt), which in turn activates phosphodiesterase, which subsequently hydrolyzes cGMP to 5′GMP in rods and cones. A decrease in cGMP concentration in the photoreceptor cells results in closure of the cGMP-gated cation channel and leads to a hyperpolarizing cellular response [2]. Go-coupled opsins are found in scallop ciliary-type photoreceptor cells and amphioxus [7,24]. In the scallop ciliary cells, the Go-coupled opsin-based pigment activates Go, elevating cGMP levels to open the CNG channels [7,25]. In addition, parietopsin, a photopigment in photoreceptor cells of lizard parietal eyes and a member of the Gt-coupled opsin group, is reported to couple with Go [26]. Green-sensitive parietopsin and blue-sensitive pinopsin were observed to have antagonistic effects on cGMP signaling by activating Go-mediated and gustducin (a Gt-related G protein)-mediated cascades, respectively, in a single photoreceptor cell. This was identified as the mechanism underlying the detection of the ratio of blue to green light in the parietal eye [26,27]. In addition, we found that parietopsin and parapinopsin, a UV-sensitive Gt-coupled non-visual pigment [28,29], are co-localized in photoreceptor cells of the iguana parietal eye [30], where UV sensitivity has previously been reported [31]. These observations provide native examples of the regulation of cGMP signaling by different wavelengths (color) of light. Furthermore, members of the Gt-coupled opsin and Opn3 (recently revealed to be Gi/Go-coupled opsin as described later) groups can also activate Go-type G protein in vitro [32–34]. However, it should be noted that cGMP signaling can be regulated by expression of these opsins only in cells having a particular guanylyl cyclase and phosphodiesterase that are activated by G proteins directly or indirectly.

cAMP signaling can be simply regulated by Gs- and Gi-type G proteins through stimulation and inhibition of adenylyl cyclase, respectively. Vertebrate Opn5, a newly identified UV-sensitive pigment, was recently revealed to activate Gi-type G proteins [35,36]. In vitro experiments have revealed that members of the Gt- and Go-coupled opsin groups can also activate Gi-type G proteins [32,33]. In addition, we recently reported that invertebrate and vertebrate homologs of Opn3, originally called encephalopsin or panopsin, members of which are expressed in various tissues, including the brain, eye, and liver in several animals [37–40], are Gi- and Go-coupled opsins [34]. Furthermore, we

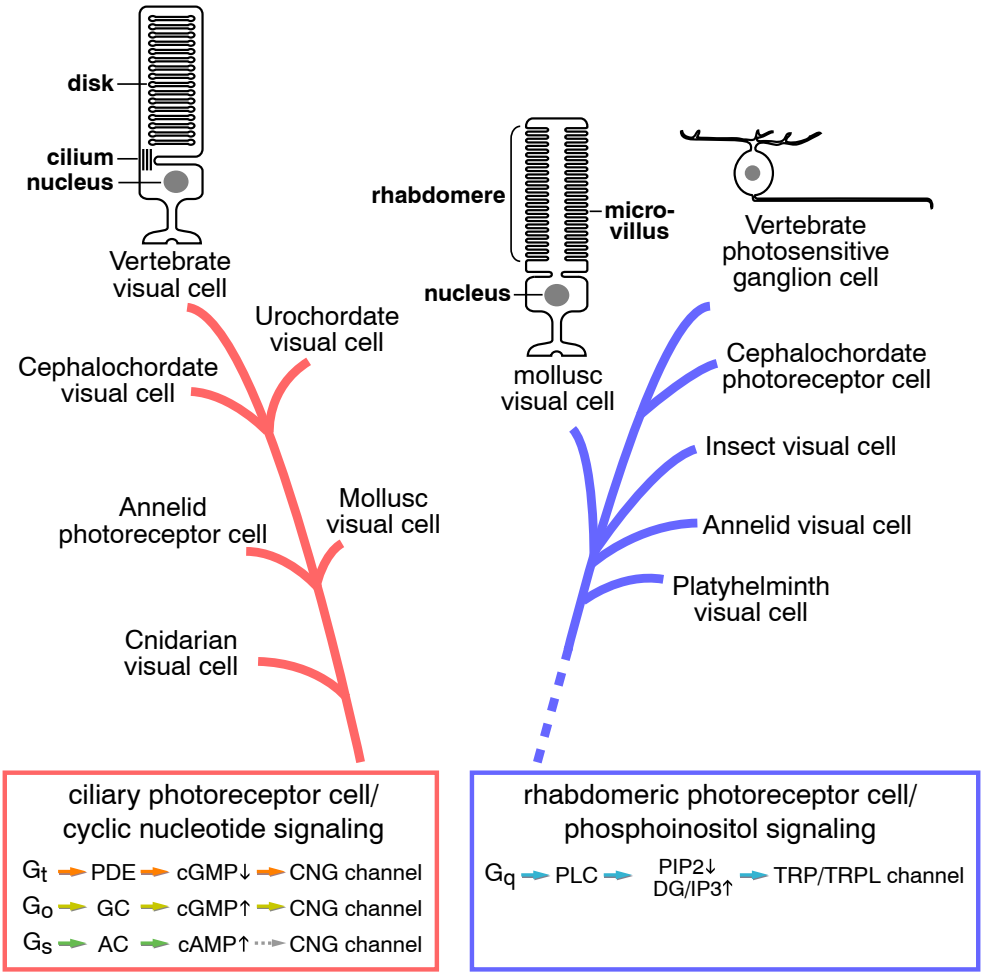

Fig. 3. Two lines of signal transductions in the animal kingdom. Animal phototransduction cascades are evolutionary and functionally classified into two groups, cyclic nucleotide signaling in ciliary-type photoreceptor cell with disk membranes of modified cilia (left) and phosphoinositol signaling in rhabdomeric-type photoreceptor cell with apical microvilli (right). Representative animal photoreceptor cells are indicated. PDE, phosphodiesterase; GC, guanylyl cyclase; AC, adenylyl cyclase; CNG channel, cyclic nucleotide gated channel; PLC, phospholipase C; PIP2, phosphatidylinositol 4,5-bisphosphate; DG, diacylglycerol; IP3, inositol 1,4,5-triphosphate; TRP channel, transient receptor potential channel; TRPL channel, transient receptor potential-like channel.

Table 1
Spectral sensitivities of non-bleaching pigments that trigger phosphoinositol signaling and cAMP signaling.

Signaling cascade	G protein	Opsin-based pigment	λmax (nm)	Ref.
Phosphoinositol-signaling	Gq	Honeybee UV	~340	[14]
		Butterfly PrV	~420	[15]
		Honeybee blue	~430	[14]
		Butterfly PrB	~450	[15]
		Mouse melanopsin	467	[22]
		Zebrafish melanopsin	470–485	[60]
		Amphioxus melanopsin	~485	[14]
		Jumping spider Rh1	~535	[16]
cAMP-signaling	Gs	Jellyfish opsin[a]	~500	[8]
	Gi	Chicken Opn5	350–360	[35,61]
		Lamprey parapinopsin	~360	[28]
		Mouse Opn5	~380	[36]
		Pufferfish TMT	~460	[34]
		Amphioxus Go-rhodopsin	~485	[24]
		Mosquito Opn3	~500	[34]

Nonconventional visual pigments that have been successfully expressed in cultured cells and analyzed by using purified sample are listed with their absorption maximum (λmax). G protein subtypes that are activated by each photopigment in vitro are also indicated.

[a] Jellyfish opsin-based pigment does not bleach and is converted to a stable photoproduct that does not revert to its original dark state by subsequent light irradiation, unlike typical bistable pigments.

demonstrated that the introduction of Opn3 homologs rendered mammalian cultured cells derived from kidney photosensitive, indicating that Opn3 can be used to downregulate cAMP signaling in non-photoreceptive tissues (Fig. 4). In contrast, jellyfish opsin discovered from prebilaterian box jellyfish visual cells is the only Gs-coupled opsin reported thus far [8]. In the jellyfish ciliary visual cells, the Gs-coupled opsin is co-localized with a large amount of Gs and adenylyl cyclase. In addition, biochemical analyses revealed that the cAMP levels are elevated in a light-dependent manner in the jellyfish visual cells, indicating that a Gs-coupled opsin-based pigment triggers a Gs-mediated signal transduction cascade that includes Gs and adenylyl cyclase in vivo. Furthermore, expression of jellyfish opsin was observed to enable a light-induced increase in cAMP levels in cultured cells, indicating that Gs-coupled opsin can be used for upregulating cAMP signaling [8,41,42] (Fig. 4).

3. Photochemical properties of opsin-based pigments

The photochemical properties of the photoproducts of opsin-based pigments can be involved in the efficiency and sustainability of light-induced signal transduction. The photoproduct of vertebrate visual pigments, including bovine rhodopsin in the Gt-coupled opsin group, releases its retinal chromophore after light activation and bleaches,

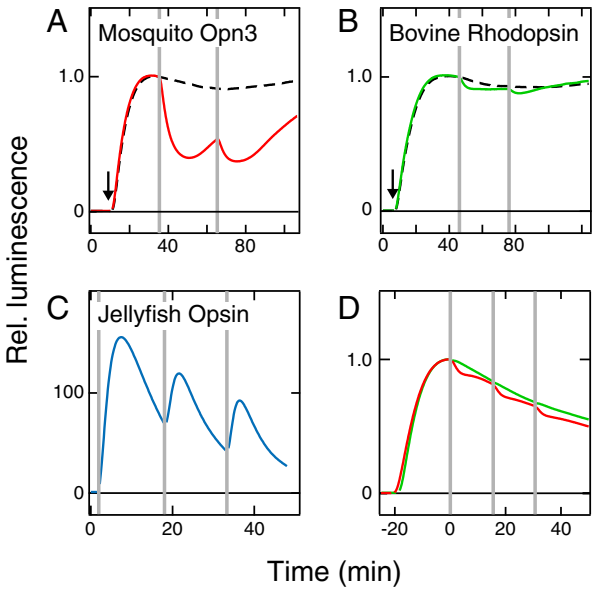

Fig. 4. Light-induced regulation of cAMP signaling in cultured cells by introducing opsin-based pigments. Changes of intracellular cAMP concentration were measured by using the GloSensor cAMP assay, which is based on a cAMP-dependent luciferase. Luminescence signals, which represent cAMP level, were first increased by forskolin treatment (arrows) and then they were decreased by light irradiations (vertical lines) via Gi activation in mosquito Opn3-expressing (A, red trace) and bovine rhodopsin-expressing (B, green trace) HEK293 cells incubated with 11-*cis* retinal. The light-induced decreases were followed by gradual recovery of luminescent signals, which is probably due to the adenylyl cyclase activity stimulated continuously by forskolin. The luminescence signals of non-irradiated cells are also shown as a control (broken traces). (C) Light-induced increases of cAMP were observed in jellyfish opsin-expressing cells (blue). (D) Changes of luminescence signals upon green light irradiation of HEK293 cells expressing the mosquito Opn3 (red trace) and bovine rhodopsin (green trace) maintained in the room light, without the addition of 11-*cis* retinal to the culture medium. It should be noted that the culture medium contains a small amount of "endogenous" retinal which is originally present in the serum. Although HEK293 cells have functional retinoid processing machinery, which could help convert the retinal in the serum to forms used by the opsins [62], the pigment formation of mosquito Opn3 can be simply explained by its capability to bind 13-*cis* retinal which was thermally generated from the all-*trans* form (see text). The luminescence values were normalized to those just prior to the irradiations.

resulting in such opsins being termed "bleaching pigments" [43] (Fig. 1). On the other hand, spectroscopic analyses of a variety of native and recombinant animal opsin-based pigments have revealed that members of the Gq-coupled opsins, including the melanopsin, Go-coupled opsin, Opn5, Opn3, and peropsin groups are "bistable pigments," i.e., they have two stable states, a dark state and a photoproduct that reverts to the original dark state by subsequent light absorption [10,34,35,44] (Fig. 1). Parapinopsin is a vertebrate non-visual pigment that is a member of the Gt-coupled opsin group, including bleaching visual pigments, and exhibits a relatively early branching in the group [45]. It is also known to be bistable pigment, suggesting that parapinopsin is an evolutionary intermediate between a bistable and bleaching pigment [28] (Fig. 1). Jellyfish Gs-coupled visual pigment does not bleach either, however it is converted to a stable photoproduct that does not revert to its original dark state by subsequent light irradiation, unlike typical bistable pigments [8]. Judging from the sustained photosensitivity of jellyfish opsin-expressing cultured cells, as described later, there is a possibility that the jellyfish opsin-based pigment would exhibit a typical bistable nature in the native condition. The wide distribution of bistable pigments in the animal opsin family suggests that animal opsin-based pigments are generally bistable pigments and that bleaching pigments evolved relatively later in the course of visual pigment evolution. The evolution of bleaching pigments can involve counterion displacement from Glu181 to Glu113 (see reviews for details). Comparative analyses of G protein activation efficiency and a site-specific fluorescence study between a bistable pigment (Glu181 counterion) and bleaching pigment (Glu113 counterion) strongly suggest that the newer counterion Glu113 could contribute to provide a larger photoactivated conformational change in opsin-based pigments and higher G protein activation efficiency [43,46]. This has led us to hypothesize that acquisition of this larger conformational change also results in the abolishment of photoreversibility and bistable nature of the pigments as a by-product.

4. Optogenetic potential of nonconventional visual pigments

Recently, another retinal-based photopigment from algae, called channelrhodopsin, which is a member of the microbial rhodopsins including bacteriorhodopsin, has been successfully applied as a tool for modulating neural responses, resulting in the establishment of the field of optogenetics [47,48]. Channelrhodopsin functions as a light sensitive cation channel and can induce inward cation currents in cells upon light absorption [49,50]. Therefore, by introducing the channelrhodopsin into neurons of living animals, neural firing can be induced by light with high temporal precision. In contrast, the use of animal opsin-based pigments as an optogenetic tool remains limited [51], which is probably because of the unsuitability of conventional visual pigments, even though they have a potential for regulating a considerable number of GPCR-based physiologies including neural responses by light. Several studies involving the use of opsins as an optogenetic tool have been reported thus far. Ectopic expression of vertebrate opsins, which can potentially activate Gi- and Go-type G proteins in addition to Gt-type G proteins [32] (Fig. 2), has enabled modulation of neural activity in hippocampal neurons [52,53] and Purkinje cells [54] through Gi/Go activation by light. Introducing engineered vertebrate visual pigments, where intracellular regions are replaced with those of Gs-coupled β2-adrenergic receptor or Gq-coupled α1-adrenergic receptor into the nucleus accumbens, enabled the triggering of cAMP signaling and phosphoinositol signaling, respectively, leading to modulation of neural activity by light [55].

However, vertebrate visual pigments are bleaching pigments and require 11-*cis* retinal, which is only produced in the photoreceptor organs, to form a photopigment. In fact, supplementation of 11-*cis* retinal is required for high sensitive and sustained photoresponsiveness of bleaching pigment-expressing cultured cells [41,42], suggesting that the same could be true for non-photoreceptive tissues, where only a small amount of 11-*cis* retinal is present. In contrast, bistable and non-bleaching pigments are bleach-resistant and reusable, which suggests that such nonconventional visual pigments can provide sustainable photosensitivity to non-photoreceptive tissues. The usefulness of nonconventional visual pigments for optogenetics was first suggested in a report showing that the addition of melanopsin renders mammalian cells photosensitive [56] and was demonstrated by a series of experiments of jellyfish opsin [41]. Jellyfish Gs-coupled opsin, which is not a typical bistable pigment, but has a stable photoproduct, has been indicated to be able to increase cAMP levels in mammalian cells by light [8,41,42], and the photosensitivity sustains after repeated light stimulations [41] (Fig. 4). Furthermore, we recently demonstrated that the introduction of Opn3 homologs rendered cultured mammalian kidney cells photosensitive without the addition of 11-*cis* retinal to the culture medium, even after continuous light exposure, whereas the introduction of bovine rhodopsin did not [34] (Fig. 4). Therefore, jellyfish Gs-coupled opsin and bistable pigments that activate Gi-type G proteins, such as Opn3, Opn5, Go-coupled opsin, and parapinopsin, can be promising optogenetic tools for the in vivo upregulation and downregulation of cAMP levels, respectively (Fig. 2). With respect to the in vivo modulation of Gq-mediated phosphoinositol signaling by light, melanopsin has been successfully employed in several cases [57–59]. While all melanopsins investigated thus far have been revealed to be blue-sensitive pigments, invertebrate Gq-coupled visual pigments have a diverse spectral sensitivity, ranging from the UV to green region. Recently, UV-, blue-, and green-sensitive Gq-coupled visual pigments have been

successfully purified and well characterized using heterologous expression in cultured cells [14–16] (Table 1). Such color-sensitive opsin-based pigments provide the highly useful possibility to choose the color of light used to trigger phosphoinositol signaling.

In addition, we have recently demonstrated that the photoresponsiveness of Opn3 homolog-expressing cells incubated in culture medium containing 11-*cis* retinal was higher than that of cells expressing the bleaching pigment bovine rhodopsin or another Gi-coupled bistable pigment, Opn5 [34,36] (Fig. 4). These results suggest that the Opn3 homologs may possess unknown molecular properties that provide an efficient decrease in cAMP in cultured cells, in addition to their bistable nature. Interestingly, we found that mosquito Opn3 acts as a light sensor when constituted with 13-*cis* retinal, a ubiquitously present retinal isomer (Fig. 5), indicating that this particular opsin would be highly advantageous for optimizing opsin-based optogenetic tools. We also propose that the UV sensitive bistable pigment parapinopsin is of great interest for optogenetic applications. The photoproduct of parapinopsin has spectral sensitivity in the visible region which completely reverts to the original dark state by orange light absorption [28] (Fig. 1) (Table 1). This spectroscopic feature would allow rapid activation and deactivation of opsin-induced signaling by UV and orange light, respectively, suggesting that the utilization of these photochemical properties for opsin-based optogenetic tools would facilitate effective modulation of cellular responses.

5. Conclusion

The discovery of novel opsin genes in several animals and the subsequent functional characterization of their molecular properties have revealed an unexpected diversity of opsin-based pigments and resulted in a number of paradigm shifts. At present, the opsin family contains Gt-coupled, Gq-coupled, Go-coupled, Gs-coupled, Gi-coupled, and Gi/Go-coupled opsins (Fig. 2), indicating the existence of a large diversity of light driven GPCR-signaling cascades. As discussed here, opsin-based pigments are generally bistable pigment, whereas bleaching is a phenomenon unique to conventional vertebrate visual pigments (Fig. 1). Finally, we note that mosquito Opn3 has the ability to form a photopigment by binding to 13-*cis* retinal as a chromophore, in addition to the more commonly utilized chromophore, 11-*cis* retinal [34]. Understanding the mechanism of 13-*cis* retinal binding ability at the amino acid level would be required to apply this optogenetic potential of Opn3 to other opsins. These recently revealed characteristics of opsin-based pigments will facilitate the design and development of highly promising optogenetic tools for modulation of the GPCR signaling, which is involved in a wide variety of physiological responses.

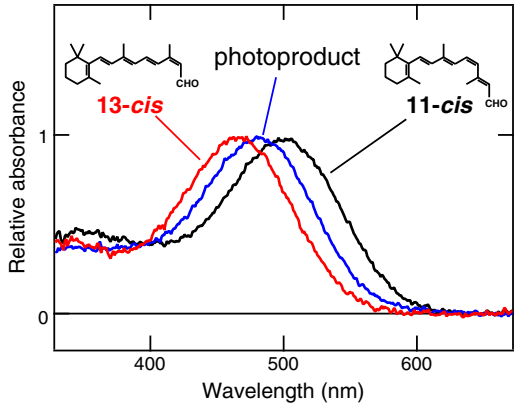

Fig. 5. 13-*Cis* retinal binding capability of mosquito Opn3. Absorption spectra of 11-*cis* retinal-bearing (black curve) and 13-*cis* bearing (red curve) mosquito Opn3-based pigments and their photoproduct (blue curve). These retinal isomers are also indicated.

References

[1] A. Terakita, The opsins, Genome Biol. 6 (2005) 213.
[2] K.W. Yau, R.C. Hardie, Phototransduction motifs and variations, Cell 139 (2009) 246–264.
[3] S.N. Peirson, S. Halford, R.G. Foster, The evolution of irradiance detection: melanopsin and the non-visual opsins, Philos. Trans. R. Soc. Lond. B Biol. Sci. 364 (2009) 2849–2865.
[4] A. Terakita, E. Kawano-Yamashita, M. Koyanagi, Evolution and diversity of opsins, WIREs Membr. Transp. Signal 1 (2012) 104–111.
[5] T. Okano, T. Yoshizawa, Y. Fukada, Pinopsin is a chicken pineal photoreceptive molecule, Nature 372 (1994) 94–97.
[6] M. Max, P.J. McKinnon, K.J. Seidenman, R.K. Barrett, M.L. Applebury, J.S. Takahashi, R.F. Margolskee, Pineal opsin: a nonvisual opsin expressed in chick pineal, Science 267 (1995) 1502–1506.
[7] D. Kojima, A. Terakita, T. Ishikawa, Y. Tsukahara, A. Maeda, Y. Shichida, A novel Go-mediated phototransduction cascade in scallop visual cells, J. Biol. Chem. 272 (1997) 22979–22982.
[8] M. Koyanagi, K. Takano, H. Tsukamoto, K. Ohtsu, F. Tokunaga, A. Terakita, Jellyfish vision starts with cAMP signaling mediated by opsin-G(s) cascade, Proc. Natl. Acad. Sci. U. S. A. 105 (2008) 15576–15580.
[9] R.M. Eakin, Evolution of photoreceptors, Cold Spring Harb. Symp. Quant. Biol. 30 (1965) 363–370.
[10] M. Koyanagi, A. Terakita, Gq-coupled rhodopsin subfamily composed of invertebrate visual pigment and melanopsin, Photochem. Photobiol. 84 (2008) 1024–1030.
[11] J. Huang, C.H. Liu, S.A. Hughes, M. Postma, C.J. Schwiening, R.C. Hardie, Activation of TRP channels by protons and phosphoinositide depletion in *Drosophila* photoreceptors, Curr. Biol. 20 (2010) 189–197.
[12] R.C. Hardie, P. Raghu, Visual transduction in *Drosophila*, Nature 413 (2001) 186–193.
[13] M. Koyanagi, T. Nagata, K. Katoh, S. Yamashita, F. Tokunaga, Molecular evolution of arthropod color vision deduced from multiple opsin genes of jumping spiders, J. Mol. Evol. 66 (2008) 130–137.
[14] A. Terakita, H. Tsukamoto, M. Koyanagi, M. Sugahara, T. Yamashita, Y. Shichida, Expression and comparative characterization of Gq-coupled invertebrate visual pigments and melanopsin, J. Neurochem. 105 (2008) 883–890.
[15] M. Wakakuwa, A. Terakita, M. Koyanagi, D.G. Stavenga, Y. Shichida, K. Arikawa, Evolution and mechanism of spectral tuning of blue-absorbing visual pigments in butterflies, PLoS One 5 (2010) e15015.
[16] T. Nagata, M. Koyanagi, H. Tsukamoto, S. Saeki, K. Isono, Y. Shichida, F. Tokunaga, M. Kinoshita, K. Arikawa, A. Terakita, Depth perception from image defocus in a jumping spider, Science 335 (2012) 469–471.
[17] I. Provencio, G. Jiang, W.J. De Grip, W.P. Hayes, M.D. Rollag, Melanopsin: an opsin in melanophores, brain, and eye, Proc. Natl. Acad. Sci. U. S. A. 95 (1998) 340–345.
[18] S. Hattar, R.J. Lucas, N. Mrosovsky, S. Thompson, R.H. Douglas, M.W. Hankins, J. Lem, M. Biel, F. Hofmann, R.G. Foster, K.W. Yau, Melanopsin and rod-cone photoreceptive systems account for all major accessory visual functions in mice, Nature 424 (2003) 76–81.
[19] R.J. Lucas, S. Hattar, M. Takao, D.M. Berson, R.G. Foster, K.W. Yau, Diminished pupillary light reflex at high irradiances in melanopsin-knockout mice, Science 299 (2003) 245–247.
[20] S. Panda, I. Provencio, D.C. Tu, S.S. Pires, M.D. Rollag, A.M. Castrucci, M.T. Pletcher, T.K. Sato, T. Wiltshire, M. Andahazy, S.A. Kay, R.N. Van Gelder, J.B. Hogenesch, Melanopsin is required for non-image-forming photic responses in blind mice, Science 301 (2003) 525–527.
[21] M. Koyanagi, K. Kubokawa, H. Tsukamoto, Y. Shichida, A. Terakita, Cephalochordate melanopsin: evolutionary linkage between invertebrate visual cells and vertebrate photosensitive retinal ganglion cells, Curr. Biol. 15 (2005) 1065–1069.
[22] T. Matsuyama, T. Yamashita, Y. Imamoto, Y. Shichida, Photochemical properties of mammalian melanopsin, Biochemistry 51 (2012) 5454–5462.
[23] S. Hughes, M.W. Hankins, R.G. Foster, S.N. Peirson, Melanopsin phototransduction: slowly emerging from the dark, Prog. Brain Res. 199 (2012) 19–40.
[24] M. Koyanagi, A. Terakita, K. Kubokawa, Y. Shichida, Amphioxus homologs of Go-coupled rhodopsin and peropsin having 11-cis- and all-trans-retinals as their chromophores, FEBS Lett. 531 (2002) 525–528.
[25] M.P. Gomez, E. Nasi, Light transduction in invertebrate hyperpolarizing photoreceptors: possible involvement of a Go-regulated guanylate cyclase, J. Neurosci. 20 (2000) 5254–5263.
[26] C.Y. Su, D.G. Luo, A. Terakita, Y. Shichida, H.W. Liao, M.A. Kazmi, T.P. Sakmar, K.W. Yau, Parietal-eye phototransduction components and their potential evolutionary implications, Science 311 (2006) 1617–1621.
[27] E. Solessio, G.A. Engbretson, Antagonistic chromatic mechanisms in photoreceptors of the parietal eye of lizards, Nature 364 (1993) 442–445.
[28] M. Koyanagi, E. Kawano, Y. Kinugawa, T. Oishi, Y. Shichida, S. Tamotsu, A. Terakita, Bistable UV pigment in the lamprey pineal, Proc. Natl. Acad. Sci. U. S. A. 101 (2004) 6687–6691.
[29] E. Kawano-Yamashita, M. Koyanagi, Y. Shichida, T. Oishi, S. Tamotsu, A. Terakita, Beta-arrestin functionally regulates the non-bleaching pigment parapinopsin in lamprey pineal, PLoS One 6 (2011) e16402.
[30] S. Wada, E. Kawano-Yamashita, M. Koyanagi, A. Terakita, Expression of UV-sensitive parapinopsin in the iguana parietal eyes and its implication in UV-sensitivity in vertebrate pineal-related organs, PLoS One 7 (2012) e39003.
[31] G. Jenison, J. Nolte, An ultraviolet-sensitive mechanism in the reptilian parietal eye, Brain Res. 194 (1980) 506–510.
[32] Y. Kanaho, S.C. Tsai, R. Adamik, E.L. Hewlett, J. Moss, M. Vaughan, Rhodopsin-enhanced GTPase activity of the inhibitory GTP-binding protein of adenylate cyclase, J. Biol. Chem. 259 (1984) 7378–7381.

[33] A. Terakita, T. Yamashita, N. Nimbari, D. Kojima, Y. Shichida, Functional interaction between bovine rhodopsin and G protein transducin, J. Biol. Chem. 277 (2002) 40–46.
[34] M. Koyanagi, E. Takada, T. Nagata, H. Tsukamoto, A. Terakita, Homologs of vertebrate Opn3 potentially serve as a light sensor in nonphotoreceptive tissue, Proc. Natl. Acad. Sci. U. S. A. 110 (2013) 4998–5003.
[35] T. Yamashita, H. Ohuchi, S. Tomonari, K. Ikeda, K. Sakai, Y. Shichida, Opn5 is a UV-sensitive bistable pigment that couples with Gi subtype of G protein, Proc. Natl. Acad. Sci. U. S. A. 107 (2010) 22084–22089.
[36] D. Kojima, S. Mori, M. Torii, A. Wada, R. Morishita, Y. Fukada, UV-sensitive photoreceptor protein OPN5 in humans and mice, PLoS One 6 (2011) e26388.
[37] S. Blackshaw, S.H. Snyder, Encephalopsin: a novel mammalian extraretinal opsin discretely localized in the brain, J. Neurosci. 19 (1999) 3681–3690.
[38] P. Moutsaki, D. Whitmore, J. Bellingham, K. Sakamoto, Z.K. David-Gray, R.G. Foster, Teleost multiple tissue (tmt) opsin: a candidate photopigment regulating the peripheral clocks of zebrafish? Brain Res. Mol. Brain Res. 112 (2003) 135–145.
[39] D. Arendt, K. Tessmar-Raible, H. Snyman, A.W. Dorresteijn, J. Wittbrodt, Ciliary photoreceptors with a vertebrate-type opsin in an invertebrate brain, Science 306 (2004) 869–871.
[40] R.A. Velarde, C.D. Sauer, K.K. Walden, S.E. Fahrbach, H.M. Robertson, Pteropsin: a vertebrate-like non-visual opsin expressed in the honey bee brain, Insect Biochem. Mol. Biol. 35 (2005) 1367–1377.
[41] H.J. Bailes, L.Y. Zhuang, R.J. Lucas, Reproducible and sustained regulation of Galphas signalling using a metazoan opsin as an optogenetic tool, PLoS One 7 (2012) e30774.
[42] W.K. Karunarathne, L. Giri, V. Kalyanaraman, N. Gautam, Optically triggering spatiotemporally confined GPCR activity in a cell and programming neurite initiation and extension, Proc. Natl. Acad. Sci. U. S. A. 110 (2013) E1565–E1574.
[43] A. Terakita, M. Koyanagi, H. Tsukamoto, T. Yamashita, T. Miyata, Y. Shichida, Counterion displacement in the molecular evolution of the rhodopsin family, Nat. Struct. Mol. Biol. 11 (2004) 284–289.
[44] H. Tsukamoto, A. Terakita, Y. Shichida, A rhodopsin exhibiting binding ability to agonist all-trans-retinal, Proc. Natl. Acad. Sci. U. S. A. 102 (2005) 6303–6308.
[45] S. Blackshaw, S.H. Snyder, Parapinopsin, a novel catfish opsin localized to the parapineal organ, defines a new gene family, J. Neurosci. 17 (1997) 8083–8092.
[46] H. Tsukamoto, D.L. Farrens, M. Koyanagi, A. Terakita, The magnitude of the light-induced conformational change in different rhodopsins correlates with their ability to activate G proteins, J. Biol. Chem. 284 (2009) 20676–20683.
[47] F. Zhang, J. Vierock, O. Yizhar, L.E. Fenno, S. Tsunoda, A. Kianianmomeni, M. Prigge, A. Berndt, J. Cushman, J. Polle, J. Magnuson, P. Hegemann, K. Deisseroth, The microbial opsin family of optogenetic tools, Cell 147 (2011) 1446–1457.
[48] J. Mattis, K.M. Tye, E.A. Ferenczi, C. Ramakrishnan, D.J. O'Shea, R. Prakash, L.A. Gunaydin, M. Hyun, L.E. Fenno, V. Gradinaru, O. Yizhar, K. Deisseroth, Principles for applying optogenetic tools derived from direct comparative analysis of microbial opsins, Nat. Methods 9 (2012) 159–172.
[49] G. Nagel, D. Ollig, M. Fuhrmann, S. Kateriya, A.M. Musti, E. Bamberg, P. Hegemann, Channelrhodopsin-1: a light-gated proton channel in green algae, Science 296 (2002) 2395–2398.
[50] G. Nagel, T. Szellas, W. Huhn, S. Kateriya, N. Adeishvili, P. Berthold, D. Ollig, P. Hegemann, E. Bamberg, Channelrhodopsin-2, a directly light-gated cation-selective membrane channel, Proc. Natl. Acad. Sci. U. S. A. 100 (2003) 13940–13945.
[51] L. Fenno, O. Yizhar, K. Deisseroth, The development and application of optogenetics, Annu. Rev. Neurosci. 34 (2011) 389–412.
[52] X. Li, D.V. Gutierrez, M.G. Hanson, J. Han, M.D. Mark, H. Chiel, P. Hegemann, L.T. Landmesser, S. Herlitze, Fast noninvasive activation and inhibition of neural and network activity by vertebrate rhodopsin and green algae channelrhodopsin, Proc. Natl. Acad. Sci. U. S. A. 102 (2005) 17816–17821.
[53] E. Oh, T. Maejima, C. Liu, E. Deneris, S. Herlitze, Substitution of 5-HT1A receptor signaling by a light-activated G protein-coupled receptor, J. Biol. Chem. 285 (2010) 30825–30836.
[54] D.V. Gutierrez, M.D. Mark, O. Masseck, T. Maejima, D. Kuckelsberg, R.A. Hyde, M. Krause, W. Kruse, S. Herlitze, Optogenetic control of motor coordination by Gi/o protein-coupled vertebrate rhodopsin in cerebellar Purkinje cells, J. Biol. Chem. 286 (2011) 25848–25858.
[55] R.D. Airan, K.R. Thompson, L.E. Fenno, H. Bernstein, K. Deisseroth, Temporally precise in vivo control of intracellular signalling, Nature 458 (2009) 1025–1029.
[56] Z. Melyan, E.E. Tarttelin, J. Bellingham, R.J. Lucas, M.W. Hankins, Addition of human melanopsin renders mammalian cells photoresponsive, Nature 433 (2005) 741–745.
[57] H. Ye, M. Daoud-El Baba, R.W. Peng, M. Fussenegger, A synthetic optogenetic transcription device enhances blood-glucose homeostasis in mice, Science 332 (2011) 1565–1568.
[58] A. Koizumi, K.F. Tanaka, A. Yamanaka, The manipulation of neural and cellular activities by ectopic expression of melanopsin, Neurosci. Res. 75 (2013) 3–5.
[59] T. Tsunematsu, K.F. Tanaka, A. Yamanaka, A. Koizumi, Ectopic expression of melanopsin in orexin/hypocretin neurons enables control of wakefulness of mice in vivo by blue light, Neurosci. Res. 75 (2013) 23–28.
[60] W.I. Davies, L. Zheng, S. Hughes, T.K. Tamai, M. Turton, S. Halford, R.G. Foster, D. Whitmore, M.W. Hankins, Functional diversity of melanopsins and their global expression in the teleost retina, Cell Mol. Life Sci. 68 (2011) 4115–4132.
[61] H. Ohuchi, T. Yamashita, S. Tomonari, S. Fujita-Yanagibayashi, K. Sakai, S. Noji, Y. Shichida, A non-mammalian type opsin 5 functions dually in the photoreceptive and non-photoreceptive organs of birds, PLoS One 7 (2012) e31534.
[62] L.I. Brueggemann, J.M. Sullivan, HEK293S cells have functional retinoid processing machinery, J. Gen. Physiol. 119 (2002) 593–612.