PN
Physiology
News

Issue 125 / Spring 2022

Vitamin D:
One hundred years of the sunshine vitamin

The Physiological Society

Physiology News

We welcome feedback on our membership magazine, or letters and suggestions for articles for publication, including book reviews, from our members.
Please email **magazine@physoc.org**.

Physiology News is one of the benefits of membership, along with reduced registration rates for our high-profile events, free online access to our leading journals, *The Journal of Physiology*, *Experimental Physiology* and *Physiological Reports*, and travel grants to attend scientific meetings. Membership offers you access to the largest network of physiologists in Europe.

Join now to support your career in physiology:
Visit **www.physoc.org/membership** or call **0207 269 5721**.

FSC www.fsc.org MIX Paper from responsible sources FSC® C010693

Membership fees for 2022	FEES
Undergraduate or Master's Member	Free (first year of membership) £10 per year (subsequent years)
Postgraduate Member	£30 per year
Full Member	£100 per year (standard rate) £50 per year (concessionary rate)
Fellow Member	£120 per year
Retired Member	£15 per year

magazine@physoc.org

www.physoc.org

@ThePhySoc

/physoc

/company/The-Physiological-Society

/physoctv

@thephysoc

Designed and printed in the UK by The Lavenham Press Ltd.

Welcome to the Spring 2022 edition of *Physiology News*

Introduction

5 Editorial: Welcome to the first issue of *Physiology News* in 2022

6 President's View: Ireland at the heart of physiology and The Society

7 Chief Executive's View: The vital role of physiologists in tackling the long-term consequences of COVID-19

News and Views

8 Reports of The Society's recent committee meetings

10 Policy Focus: How can we develop a National Post-Pandemic Resilience Programme?

12 Celebrating physiologists and physiology at the Member Forum, Award Ceremony, and The President's Lecture

14 Book Review: The Gendered Brain: The new neuroscience that shatters the myth of the female brain by Gina Rippon

Features

16 Vitamin D

20 Phototransduction

24 Stem cell-based therapies for musculoskeletal regeneration

28 The (concise) guides to pharmacology and what they provide for physiologists

Journal Insights

32 Recent papers of interest from Society journals

34 Journal Hindsights: A look at significant papers from the past

Events

36 Long COVID: Mechanisms, Risk Factors, and Recovery

38 Europhysiology 2022

39 Processing and Modulation of Sensory Signals: From the Periphery to the Cortex

Membership

40 Congratulating our 2021 Honorary Fellows

42 Physiology Friday 2021

44 Welcome to our new Theme Leads

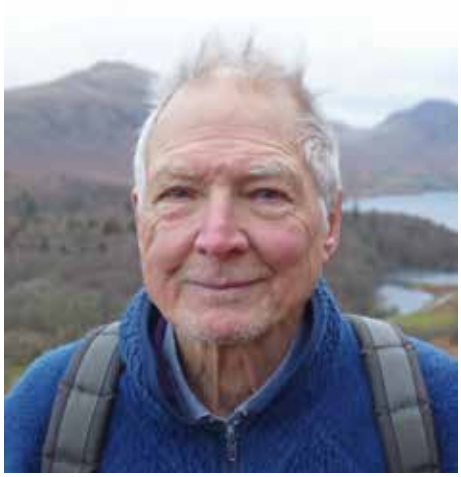
46 Obituary: Professor John Finley Benzie Morrison 1942 - 2021

Cover image: Photograph of children with rickets [© Wellcome Collection. Source: wellcomecollection.org/works/kpftgz5u. Licenced under CC BY 4.0.]

https://doi.org/10.36866/pn.125.20

Phototransduction

The decline and fall of the calcium theory



Jonathan A. Coles

Honorary Research Fellow,
University of Glasgow

The death of the calcium theory of phototransduction was announced after lunch at the Zuricher Hotel in Berlin on Friday 30 November 1984 to an international audience of some 40 researchers working on vision. The announcement instantly resolved a fierce debate that had been going on for almost 20 years and we were given the afternoon off to try to recover from the shock.

In 1965 when the pathway of sensory transduction from stimulus to electrical signal was unknown for any of the senses, vision had the attraction of the precision of the stimulus: a photon absorbed by a rhodopsin molecule. It was known that most signalling in the nervous system resulted from an increase in membrane permeability, particularly to sodium, whose influx depolarised the cell. Some invertebrate photoreceptors, such as the giant cells of the ventral eye of the horseshoe crab, *Limulus*, had been studied and they depolarised in response to light, as expected. In vertebrate retina, Svaetichin (1953) recorded from sites with a negative resting potential that gave a hyperpolarising, rather than a depolarising, response to light. Svaetichin's claim that these recordings were from photoreceptors was met with justifiable scepticism - for one thing, these recordings were too easy to make. But it took several groups, working for over a decade, to understand the situation (see Tomita, 1965 for refs). Oikawa *et al.* (1959), working on carp retinas, showed that in most recordings, the response amplitude increased when the area stimulated by light increased, but in some it did not. They suggested that the first class were horizontal cells (which extend over an area including many photoreceptors) and the second class were the photoreceptors themselves, in this case, cones. Bortoff (1964) recorded from cells in the salamander, *Necturus,* which he identified by dye labelling as rod photoreceptors and again these gave hyperpolarising responses to light. Meanwhile, Tsuneo Tomita and his students in Tokyo were working carefully and methodically to decipher the system. They introduced a range of new techniques, most notably a jolter that used an electromagnetic diaphragm to jolt the retina onto the microelectrode (Tomita *et al.*, 1967; Kaneko, 2021). They also built an elaborate light stimulator that could measure the spectral response of a cell quickly, before the electrode fell out, and used it to show that responses from cones in carp supported Young's trichromatic theory of colour vision. Toyoda *et al.* (1969) showed that the light-induced hyperpolarisation in rods was due to a fall in membrane conductance. This result defined a major problem: in rods, the rhodopsin that is isomerised by photons and initiates the receptor potential is in the membranes of the discs or sacs that are stacked inside the outer segment, and not attached to the surface membrane (Fig.1A). A tempting idea was that the photoisomerisation of rhodopsin caused some ion or molecule to diffuse through the cytoplasm from the disc and close channels on the surface membrane. Protons were quickly ruled out, but the hypothesis of Yoshikami and Hagins (1971) that the internal transmitter was calcium, received experimental support for a decade or so. On a different tack, cyclic adenosine monophosphate was known to

https://doi.org/10.36866/pn.125.20

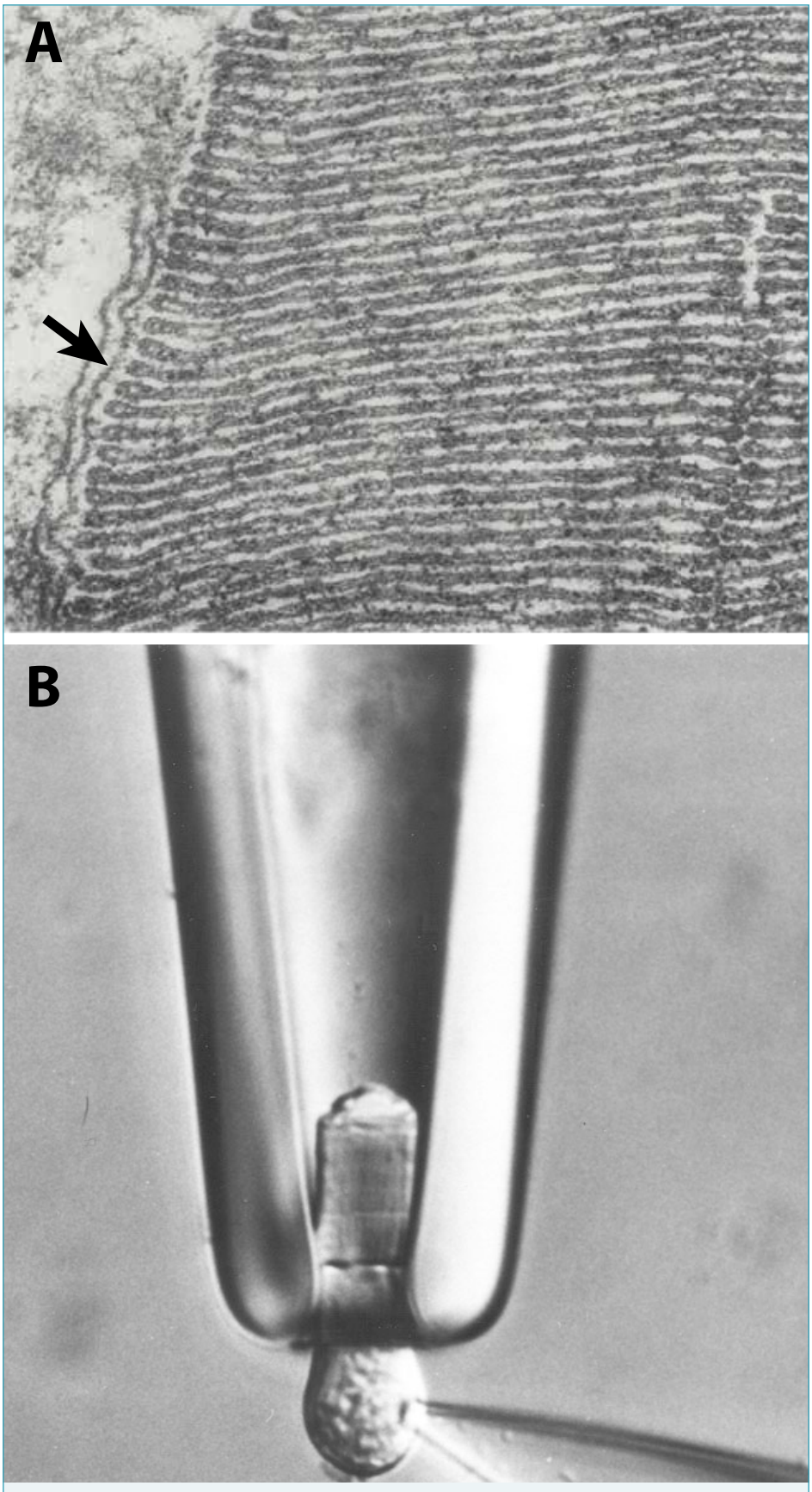


Figure 1. **A.** Electron micrograph of a frog rod outer segment showing the evenly stacked 'discs' not contacting the surface membrane (arrow). The width of the figure is about 3.0 µm. © Nilsson SE (1965) *Journal of Ultrastructure Research,* **12** 207-231. http://doi.org/10.1016/s0022-5320(65)80016-8. **B.** A rod isolated from a larval tiger salamander and held by a suction electrode, which measures current. A patch pipette was then mounted on a micromanipulator and manoeuvred in the dark until "with a little luck, the new patch pipette appeared on the screen of the TV monitor". The cell membrane was ruptured by the pipette, which was then used to introduce a calcium chelator into the cell; this increased the dark current. The diameter of the outer segment is about 10 µm. © Lamb *et al.* (1986) *Journal of Physiology,* **372**(1), 315-349. http://doi.org/10.1113/jphysiol.1986.sp016011

be a common internal messenger and the enzymes required for its production (cyclase) and hydrolysis (phosphodiesterase) were looked for, and found, in photoreceptors. But in rod outer segments, it was the guanosine system, rather than the adenosine, that was far more active, both a guanylate cyclase (Goridis *et al.*, 1973) and a guanylate phosphodiesterase. Photoisomerisation of a single rhodopsin molecule rapidly activated up to 500 molecules of phosphodiesterase converting cGMP to 5'GMP (Yee and Liebman, 1978). Manipulating cGMP levels changed the membrane potential. But how would a change in the concentration of cGMP change the membrane resistance? Its expected action was to phosphorylate proteins, or perhaps a light-activated GTPase was involved (Fleischman and Denisevich, 1979).

In the 1970s several new techniques were brought to bear. Yau and colleagues independently developed a way of measuring the current flowing through the rod outer segment membrane by sucking the outer segment into the tip of a pipette (Yau *et al.*, 1977) and, with dexterity, this could be combined with the patch electrode technique definitively described by Hamill *et al.* (1981) (see Fig.1B). In 1980, Gold and Korenbrot devised a calcium-sensitive membrane electrode on which they placed a retina and showed that light stimulation caused calcium release from rod outer segments.

It should be remembered that news was transmitted by post, by visits and sometimes by telephone, and that checking the latest literature meant going to the library and thumbing through journals. Working out a coherent description of transduction in a vertebrate sensory cell was an obvious prize and papers came tumbling out, often beautifully done and with considered but inconclusive discussions; there were, for example, six papers in 1984 on the subject in *Nature* alone.

This was the background to the Dahlem "Konferenz" (more a "workshop") that was held in 1984 on "The Molecular Mechanisms of Photoreception". Dahlem, a suburb of Berlin, is where Otto Warburg had done his biochemistry research and, in 1933, it had a fleeting connection with phototransduction when George Wald visited and found vitamin A (retinal) in the retina. Since 1974, several Dahlem Workshops a year have been held in what was the enclave of West Berlin. They are limited to 40 participants, discussion documents are prepared beforehand and, for much of the time, the meeting is split into four working groups that produce reports on different aspects. For the 1984 phototransduction meeting, the scientific organiser was Hennig Stieve, who worked on *Limulus* ventral photoreceptor, and the detailed management was by Silke Bernhard, who had 10 years of experience. The dates were 25–30 November 1984, which turned

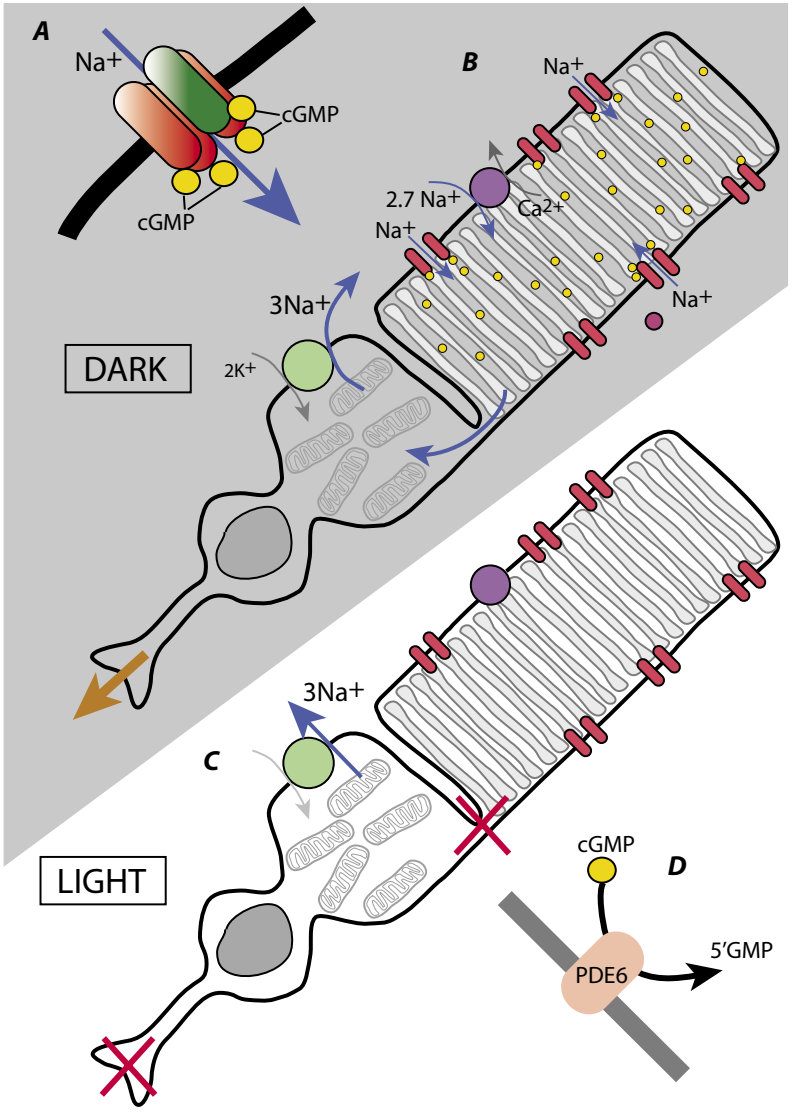


Figure 2. **Scheme of phototransduction in a rod photoreceptor with emphasis on Na^+ currents.** In the dark, cGMP is maintained high by guanylate cyclase and keeps open some of the channels in the outer segment plama membrane (A). Absorption of photons by a rhodopsin molecule leads to activation of many phosphodiesterases (D), which reduces the cGMP concentration and allows outer segment Na^+ channels to close (C). The continued extrusion of Na^+ by the Na^+,K^+,ATPase in the inner segment hyperpolarises it and reduces transmitter release at the synapse. From about 1990 onwards, interest has focused on the multiple roles of calcium in regulating the system. Recent work shows that the membrane channel is a heterotetramer with a binding site for cGMP on each subunit (e.g. Xue *et al.*, 2021) so I have shown four cGMPs on each channel.

It should be remembered that news was transmitted by post, by visits and sometimes by telephone, and that checking the latest literature meant going to the library and thumbing through journals.

out to be important. Among other things, a key paper on "Electrogenic Na-Ca exchange in retinal rod outer segment" by Yau and Nakatani (1984) had been published on 18 October.

I was something of an outsider and was shamefully relaxed about it. I enjoyed the luxury hotel overlooking the Tiergarten park, and the delightful small restaurants that Silke, a perfect host, took us to in the evenings. Of course, all this was in the Western sector. Despite the Berlin Wall, the underground railway could be used to cross the border, the main incentive being to visit the Pergamon museum with its outstanding collection of archeological treasures.

A rule at the Dahlem Workshops was that no slides were to be shown. However, early on in the meeting, Trevor Lamb, a tall, bearded New Zealander, twisted Silke's arm and was allowed to show one slide, which showed that when intracellular calcium was chelated the photoresponse increased rather than decreased (Matthews *et al.*, 1985). The meeting went on, with a curious air of uneasiness as nearly all the participants wondered what the role of cGMP was. Then, after lunch on the last day, it was revealed that two reviewers and a *Nature* Editor (all present at the meeting) had a manuscript received from the USSR Academy of Sciences at Pushchino, near Moscow, on 16 November, nine days before the meeting. Defying general wisdom, Fesenko, Kolesnikov

https://doi.org/10.36866/pn.125.20

and Lyubarsky (Fesenko *et al.*, 1985) had applied cGMP directly to the cytoplasmic face of the surface membrane of a rod outer segment and found that it increased the conductance. This result filled the missing link in vertebrate phototransduction: photoisomerisation of rhodopsin led to activation of the phosphodiesterase, which reduced cGMP levels and decreased the membrane conductance (Fig.2). As the meeting report in *Nature* (Altman, 1985) put it, the announcement "electrified the whole meeting". Disclosing results from a paper under review was not quite proper, and *Nature* had politely waited until publication of the Fesenko paper, on 24 January 1985, before publishing its report. Curiously, both this report, and the introduction to the proceedings book (Stieve, 1986) make the same mistake in the references, putting the publication of the Fesenko paper in 1984 rather than 1985, a subconscious attempt to smooth out history, perhaps. The *Nature* report also announced three further papers in press, all supporting the cGMP theory and these were published together on 14 February (Yau and Nakatani, 1985; Matthews *et al.*, 1985; Cobbs and Pugh, 1985). The "received on" dates show that all three were under review at the time of the meeting, at which four of the authors were present (and, presumably, some of the reviewers). The atmosphere in the "Internal Messengers" working group must have been interesting.

What has happened since the meeting? Most of the participants continued long and productive careers in vision: a detailed understanding of the regulation of transduction by calcium, the molecular structures of the proteins and how those structures transform, the evolution of photoreceptors, the causes of eye disease. Of the three authors of the crucial paper, Kolesnikov and Lyubarsky continued on vision, Lyubarsky having moved to Philadelphia, while Kolesnikov stayed in Pushchino. Fesenko, also in Pushchino, continues to publish frenetically on subjects that usually include magnetic fields, electromagnetic radiation or cryoprotection.

References

Altman J (1985). Sensory transduction: new visions in photoreception. *Nature* **313**(6000), 264-265. http://doi.org/10.1038/313264a0.

Bortoff A (1964). Localization of slow potential responses in the Necturus retina. *Vision Research* **4**(11), 627-635. http://doi.org/10.1016/0042-6989(64)90048-3.

Cobbs JW, Pugh EDJ (1985). Cyclic GMP can increase rod outer-segment light-sensitive current 10-fold without delay of excitation. *Nature* **313**(6003), 585-587. http://doi.org/10.1038/313585a0.

Fesenko EE *et al.* (1985). Induction by cyclic GMP of cationic conductance in plasma membrane of retinal rod outer segment. *Nature* **313**(6000), 310-313. http://doi.org/10.1038/313310a0.

Fleischman D, Denisevich M (1979). Guanylate cyclase of isolated bovine retinal rod axonemes. *Biochemistry* **18**(23), 5060-5066. http://doi.org/10.1021/bi00590a006.

Gold GH, Korenbrot JI (1980). Light-induced calcium release by intact retinal rods. *Proceedings of the National Academy of Sciences USA* **77**(9), 5557-5561. http://doi.org/10.1073/pnas.77.9.5557.

Goridis C *et al.* (1973). Guanyl cyclase in a mammalian photoreceptor. *FEBS Letters* **30**(2), 163-166. http://doi.org/10.1016/0014-5793(73)80642-8.

Hamill OP *et al.* (1981). Improved patch-clamp techniques for high-resolution current recording from cells and cell-free membrane patches. *Pflugers Archives* **391**(2), 85-100. http://doi.org/10.1007/BF00656997.

Kaneko A (2021). Recollection of my research work on the electrophyiology of the vertebrate retina. *Bioelectricity* **3**(3), 221-228. http://doi.org/10.1089/bioe.2021.0022.

Lamb TD *et al.* (1986). Incorporation of calcium buffers into salamander retinal rods: a rejection of the calcium hypothesis of phototransduction. *Journal of Physiology* **372**(1), 315-349. http://doi.org/10.1113/jphysiol.1986.sp016011.

Matthews HR *et al.* (1985). Effects on the photoresponse of calcium buffers and cyclic GMP incorporated into the cytoplasm of retinal rods. *Nature* **313**(6003), 582-585. http://doi.org/10.1038/313582a0.

Nilsson SE (1965). The ultrastructure of the receptor outer segments in the retina of the leopard frog (Rana Pipiens). *Journal of Ultrastructure Research* **12**, 207-231. http://doi.org/10.1016/s0022-5320(65)80016-8.

Oikawa T *et al.* (1959) Origin of so-called cone action potential. *Journal of Neurophysiology* **22**(1), 102-111. http://doi.org/10.1152/jn.1959.22.1.102.

Stieve S (ed.) 1986. *The Molecular Mechanism of Photoreception,* Berlin: Springer Verlag.

Svaetichin G (1953). The cone action potential. *Acta Physiologica Scandinavica* **29**(Suppl 106), 565-600.

Tomita T (1965). Electrophysiological study of the mechanisms subserving color coding in the fish retina. *Cold Spring Harbor Symposia of Quantitative Biology* **30**, 559-566. http://doi.org/10.1101/sqb.1965.030.01.054.

Tomita T *et al.* (1967). Spectral response curves of single cones in the carp. *Vision Research,* **7**(7), 519-531. http://doi.org/10.1016/0042-6989(67)90061-2.

Toyoda J *et al.* (1969). Light-induced resistance changes in single photoreceptors of Necturus and Gekko. *Vision Research* **9**(4), 453-463. http://doi.org/10.1016/0042-6989(69)90134-5.

Xue H *et al.* (2021). Structural mechanisms of gating and selectivity of human rod CNGA1 channel. *Neuron* **109**(8), 1302-1313. http://doi.org/10.1016/j.neuron.2021.02.007.

Yau KW *et al.* (1977). Light-induced fluctuations in membrane current of single toad rod outer segments. *Nature* **269**(5623), 78-80. http://doi.org/10.1038/269078a0.

Yau KW, Nakatani K (1984). Electrogenic Na-Ca exchange in retinal rod outer segment. *Nature* **311**(5987), 661-663. http://doi.org/10.1038/311661a0.

Yau KW, Nakatani K (1985). Light-induced reduction of cytoplasmic free calcium in retinal rod outer segment. *Nature* **313**(6003), 579-582. http://doi.org/10.1038/313579a0.

Yee R, Liebman PA (1978). Light-activated phosphodiesterase of the rod outer segment. Kinetics and parameters of activation and deactivation. *Journal of Biological Chemistry* **253**(24), 8902-8909. http://doi.org/10.1016/s0021-9258(17)34263-1.

Yoshikami S, Hagins WH (1971). Light, calcium and the photocurrent in vertebrate rods and cones. Biophysical Society Annual Meeting Abstracts **11**, 47a.