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TRIBUTE

Brighter than the sun: Rajni Govindjee at 80 and her fifty years in photobiology

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Abstract We celebrate distinguished photobiologist Rajni Govindjee for her pioneering research in photosynthesis and retinal proteins on the occasion of her 80th birthday.

Keywords Bacteriorhodopsin · Biophysics · Photochemistry · Photosynthesis

Almost everyone in the general area of the photobiology of retinal proteins, as well as many early practitioners of photosynthesis, are well acquainted with Rajni Govindjee (Fig. 1), both from her published work and from many interactions with her at scientific meetings, especially the Biophysical Society and International Retinal Protein meetings. Rajni has been a constant presence in the discipline of photobiology from her early graduate-student days, working on algal and green plant photosynthesis with pioneers of photosynthesis research Eugene Rabinowitch and Robert Emerson, to her ground-breaking work on retinal proteins.

This manuscript was edited and approved for publication by Barbara Demmig-Adams. She added "This tribute is a well-deserved celebration of Rajni's many accomplishments. Congratulations and Happy Birthday to Rajni". This tribute was also read by Sergei and Ella Balashov, Rajni's collaborators for many years; they added "This is a heart warming story of accomplished life in science, especially of someone dear to us; we were participants and witnesses of Rajni's unique qualities and her pure goodness. It is a true masterpiece on the Urbana lab of the author (Tom Ebrey)".

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Photosynthesis

Rajni Govindjee came to USA to work with Robert Emerson in 1957 and finished her PhD in 1961 under Eugene Rabinowitch at the University of Illinois at Urbana-Champaign. During her thesis work, she, using quinone Hill reaction in Chlorella cells, discovered that Emerson's two-light effect was indeed due to photosynthesis, rather than respiration as Larry Blinks had been suggesting (Govindjee et al. 1960). Further, in her work on the quinone Hill reaction, she showed that a short-wavelength form of chlorophyll a (Chl a 670) was in the same pigment system as chlorophyll b—just as shown by her husband Govindjee in photosynthesis (Govindjee and Rabinowitch 1960). She followed this line of thinking by establishing that Emerson's two-light effect was seen in NADP photoreduction by chloroplasts (Govindjee et al. 1962, 1964). Two-light effects were then studied thoroughly via whole-cell photosynthesis of red algae (Govindjee and Govindjee 1965) explaining many discrepancies in the field. Refuting claims (such as those by Otto Warburg) of low minimal quantum requirements of oxygen evolution of 2.8–4, Rajni demonstrated that photosynthetic quantum yield was consistent with a two-step model, even in synchronous young cultures of *Chlorella* and in the presence of 10 % CO_2, (Govindjee et al. 1968). After a short period of research on bacterial photosynthesis (Govindjee and Sybesma 1972; Govindjee et al. 1974), Rajni moved on to work with retinal proteins.

Rajni's active involvement in photosynthesis ended with her widely noticed overview article on primary events in

Fig. 1 A photo of Rajni Govindjee taken in 2014, in Irvine, California, by Sergei Balashov

photosynthesis published in Scientific American (Govindjee and Govindjee 1974).

Retinal proteins

Starting in 1973, Rajni and Tom Ebrey (the author, Fig. 2) started a very successful partnership that lasted for over 25 years. When Tom joined the Biophysics program at the University of Illinois at Urbana-Champaign, one of his very first acts was to sign Rajni on to work with him on

Fig. 2 Tom Ebrey and Rajni studying a map trying to figure out where they are

retinal proteins. Rajni was a master collaborator, working with dozens of people to get experiments done. Her first retinal-protein paper was a collaboration with Koji Nakanishi's group on artificial rhodopsins (Ebrey et al. 1975; Nakanishi et al. 1976) and the first report on bacteriorhodopsin (bR) was with Tom and Fumio Tokunaga collaborating with Rosalie Crouch on the first artificial bR ever created (Tokunaga et al. 1977). In this work, they substituted *retinal1* with *retinal2* (the aldehyde of vitamin A2) and went on to explore the properties of this artificial bR.

Rajni was at the center of subsequent work on many additional artificial retinal-substituted bRs with Rosalie Crouch, Don Minick, Koji Nakanishi, Valerie Balough-Nair, Laura Eisenstein, and Mudi Sheves (Mao et al. 1981; Crouch et al. 1986, and many other papers; see Fig. 3). Eventually, many retinal analogues were studied, including those that could not isomerize about their 13–14 double bond; these were important in establishing the necessity for photoisomerization in the photochemistry of bR (Chang et al. 1985a). Rajni was also one of the first to study the fluorescence of bR (Govindjee and Ebrey 1986; Govindjee et al. 1978). She also pioneered studies on the quantum efficiency of bR photochemistry (Govindjee et al. 1980, 1990; Balashov et al. 1991a, b), which became a well-established photobiological subject at Illinois in the tradition of Robert Emerson. Together with Chung-ho Chang, Rajni initiated studies ranging from the effects of cation binding to the color and photochemistry of bR (Chang et al. 1985b, 1986, 1987, 1988). Working with Bob Callender's group, Rajni provided samples and helped interpret Resonance Raman studies of bR (Schiffmiller et al. 1985) and worked with Laura Eisenstein's group on Fourier transform infrared spectroscopy (FTIR) studies of bR (Lin et al. 1987).

Another area pioneered by Rajni was the study of proton pumping by bR using flash photolysis methods. Her early studies with Qingguo Li had employed a xenon flash (Li et al. 1984), but in subsequent studies with Zsolt Dancshazy, Rajni set up a ruby laser in order to study very fast events. One of the first results was observation of an N intermediate in the bR photocycle; this had been hypothesized in the very first studies of the bR photocycle but was forgotten until Zsolt and Rajni did their careful kinetic studies (Dancshazy et al. 1986, 1988; Govindjee et al. 1989). The N intermediate turned out to be important in both proton pumping and thermal regeneration of the initial state of bR after light absorption. The effects of high pressure and temperature on the bR photocycle were studied by Rajni and Motoyuki Tsuda (Tsuda et al. 1983).

One of Rajni's most successful collaborations was with Sergei Balashov, Ella Imasheva, and Eugene Lukashev on bR mutants (see Fig. 4). Working closely with Rosalie Crouch and Don Minick and members of their groups,

Fig. 3 Participants of International Retinal Protein Meeting at the University of Illinois at Urbana-Champaign (UIUC) in 1985. Names in italics are of those scientists who have collaborated with Rajni. *First row* (*sitting*) Bob Young, Mustafa Sayed, *Colin Longstaff*, Pal Ormos, Hugh McDonnell, Toru Yoshizawa, Jimo Borjigen. *Second row Zsolt Danschazy*, Ed Dratz, Joe Cassim, *Chung-ho Chang*, *Tom Ebrey,* Mi Hong, *Rosalie Crouch*, Rajni Govindjee, Lester Packer. *Third row* Nick Gibson, Roberto Bogomolni, Brenda Flam, Judy Herzfeld*, Kim Bagley*, Walter Stoeckenius, *Suyi Liu*, *Jeff Marque*. *Fourth row Yiannis Koutalos*, Meridithe Applebury, Hans Frauenfelder, *Dieter Osterhelt*, *Valerie Balogh-Nair*, Dave Braunstein, Cora Einterz, Suolang Liu. *Fifth row* Peter DeBrunner, Camille Sandorfy, *Roy Jonas*, Janos Lanyi, *Bob Callender*, John Spudich, Bob Griffin, *Barry Honig*, Brian Kohler, David Kliger, Lionel Murray, Bob Birge. *Sixth row* Simon Rhodes, *Jianguo Chen*, Hiro Matsumoto, *Johann Lugtenberg*, *Mudi Sheves*, Burt Litman, Rich Mathies, *Bob Liu*, Warren Sherman, Lajos Keszthelyi. *Seventh row* Felix Hong, Enrico Gratton, Jim Lewis, Janos Postafi, *Koji Nakanishi*, John Termini, *Gavin Dollinger*

Fig. 4 A 2014 photo of Rajni in California. *Left* to *right* Ella Imesheva, Rajni Govindjee and Sergei Balashov

Rajni helped illuminate the role of Arg82 in controlling light-activated proton release in bR (Balashov et al. 1995; Govindjee et al. 1996). Further studies elucidated the two pKs of Asp85 (Balashov et al. 1996a, b; Misra et al. 1996). Rajni also initiated studies using site-directed mutants to elucidate the role of Tyr 57 and Lys129 in bR function (Balashov et al. 1995; Govindjee et al. 1992, 1995, 1996b, 1997). Arg82 was identified as the residue regulating pK of the group responsible for light-driven proton release in bR. Working with Saurav Misra and others, Rajni also helped elucidate the role of Glu204 of bR in the proton-pumping photocycle (Misra et al. 1997).

Rajni was also active in working on the photochemistry of visual pigments. With Jie Liang, she studied the photochemistry of cone visual pigments (Liang et al. 1993). In another study with Osamu Kuwata, Rajni used laser-induced photolysis to study the coupling of proton uptake with Metarhodopsin II formation upon photolysis of rhodopsin (Kuwata et al. 2001). Another successful collaboration was work with Bob Rando's group on the photochemical properties of methylated rhodopsins (Govindjee et al. 1988a, b).

A mentor to many, and a badminton champion

Rajni helped mentor many graduate students, including Jim Hurley, Paul Kilbride, Jianguo Chen, Boryeu Mao, Gretchen Sassenrath, Roger Calhoon, Suyi Liu, Chung-ho Chang, Yiannis Koutolos, Dipa Apte, Roy Jonas, Ching

Yuan, Masahiro Kono, Jie Liang, Saurav Misra, and Miao Lu. In addition, Rajni welcomed many visitors to the lab, such as Fumio Tokunaga, Motoyuki Tsuda, Tchiya Rosenfeld, Brian Becher, Bridgette Barry, Tadashi Nakamura, Tatsuo Iwasa, Jeff Marque, Kim Bagley, Qingguo Li, Kazuhiko Kinoshita, Koki Ohno, Zsolt Dancshazy, Mudi Sheves, Osamu Kuwata, and Akio Maeda as well as many undergraduate researchers.

While Rajni has been a life buoy to many, she has also, to mix nautical metaphors, been a wonderful home port to her two children Anita, an accomplished computer scientist, and Sanjay, an engineering professor at University of California Berkeley, as well as to her husband Govindjee (now Professor Emeritus at the University of Illinois at Urbana-Champaign).

In addition to her accomplishments in science, Rajni brought her standards of excellence and collaboration to bear on her athletics. She was a highly sought-after teammate in competitive badminton on the UIUC campus, where her athleticism, strategic execution in the game and competitive *joie de vivre* earned her the respect of an international group of enthusiasts. We know that she played badminton not only with those who were older than she, such as the late George Foster, a professor in Fine Arts who was in his late seventies, but also with students who were half her age. She also haunted several athletic facilities, where she was a regular fixture in fitness swimming. I have been told that, when she was only 10 years old, she swam across the Jamuna river in Allahabad, and her picture was published in the local newspaper.

Rajni and her collaborator in crime, the super-computnik Burr Nelson, were the glue that held things together. Rajni was the ideal collaborator, welcoming new people into the lab, and training them on all sorts of techniques. And she shared with everyone her knowledge and ideas not only in her papers but also in her presentations and discussions at many scientific meetings. Everyone in the field of photobiology will forever be in gratitude for her tangible and intangible contributions. We congratulate her on her eightieth birthday!

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References

Balashov SP, Imasheva ES, Govindjee R, Ebrey T (1991a) Quantum yield ratio of the forward and back light reactions of bacteriorhodopsin at low temperature and photo-steady state concentration of the bathoproduct K. Photochem Photobiol 54:955–961

Balashov SP, Govindjee R, Ebrey TG (1991b) Red shift of the purple membrane absorption band and the deprotonation of tyrosine residues at high pH. Biophys J 60:475–490

Balashov S, Govindjee R, Imasheva E, Misra S, Ebrey T, Feng Y, Crouch R, Menick D (1995) The two pK's of aspartate-85 and control of thermal isomerization and proton release in the argine82 to lysine mutant of bacteriorhodopsin. Biochemistry 34:8820–8834

Balashov S, Imasheva E, Govindjee R, Ebrey T (1996a) Titration of aspartate-85 in bacteriorhodopsin. Biophys J 70:473–481

Balashov S, Imasheva E, Govindjee R, Sheves M, Ebrey T (1996b) Evidence that aspartate-85 has a higher pK in all-trans than in 13-cis bacteriorhodopsin. Biophys J 71:1973–1984

Chang C, Chen J, Govindjee R, Ebrey T (1985a) Cation binding by bacteriorhodopsin. Proc Nat Acad Sci USA 82:396–400

Chang C, Govindjee R, Ebrey T, Bagley K, Dollinger G, Eisenstein L, Marque J, Roder H, Vittitow J, Fang J, Nakanishi K (1985b) Trans/13-cis isomerization is essential for both the photocycle and proton pumping of bacteriorhodopsin. Biophys J 47:509–512

Chang C, Jonas R, Melchiore S, Govindjee R, Ebrey T (1986) Mechanism and role of divalent cation binding of bacteriorhodopsin. Biophys J 49:731–739

Chang C, Liu S, Jonas R, Govindjee R (1987) The pink membrane, the stable photoproduct of deionized purple membrane. Biophys J 52:617–623

Chang C, Jonas R, Govindjee R, Ebrey T (1988) Regeneration of blue and purple membranes from deionized bleached membranes of *Halobacterium halobium*. Photochem Photobiol 47:261–265

Crouch R, Scott R, Ghent S, Govindjee R, Chang C, Ebrey T (1986) Properties of synthetic bacteriorhodopsin pigments: further probes of the chromophore binding site. Photochem Photobiol 43:297–303

Dancshazy Zs, Govindjee R, Nelson B, Ebrey T (1986) A new intermediate in the photocycle of bacteriorhodopsin. FEBS Lett 209:44–48

Dancshazy Zs, Govindjee R, Ebrey T (1988) Independent photocycles of the spectrally distinct forms of bacteriorhodopsin. Proc Natl Acad Sci USA 85:6358–6361

Ebrey T, Govindjee R, Honig B, Pollock E, Chan W, Crouch R, Yudd A, Nakanishi K (1975) The properties of several sterically modified retinal analogues and their photosensitive pigments. Biochemistry 14:3933–3941

Govindjee, Govindjee R (1965) Two different manifestations of enhancement in the photosynthesis of *Porphyridium cruentum* in flashing monochromatic light. Photochem Photobiol 4:401–415

Govindjee, Govindjee R (1974) Primary events in photosynthesis. Sci Am 231:68–82

Govindjee, Rabinowitch E (1960) Two forms of chlorophyll *a* in vivo with distinct photochemical functions. Science 132:159–160

Govindjee R, Ebrey T (1986) Light emission from bacteriorhodopsin and rhodopsin. In: Govindjee, Amesz J, Fork DC (eds) Light emission by plants and bacteria. Academic Press, New York

Govindjee R, Sybesma C (1972) The photoreduction of nicotinamide-adenine dinucleotide by chromatophore fractions from *Rhodospirillum rubrum*. Biophys J 12:897–908

Govindjee R, Thomas JB, Rabinowitch E (1960) Second Emerson effect in the Hill reaction of *Chlorella* cells with quinone as oxidant. Science 132:421

Govindjee R, Govindjee, Hoch G (1962) The Emerson enhancement effect in TPN-photoreduction by spinach chloroplasts. Biochem Biophys Res Commun 9:222–225

Govindjee R, Govindjee, Hoch G (1964) Emerson enhancement effect in chloroplast reactions. Plant Physiol 39:10–14

Govindjee R, Rabinowitch E, Govindjee (1968) Maximum quantum yield and action spectra of photosynthesis and fluorescence in *Chlorella*. Biochim Biophys Acta 162:530–544

Govindjee R, Smith WR Jr, Govindjee (1974) Interaction of viologen dyes with chromatophores and reaction-center preparations from *Rhodospirillum rubrum*. Photochem Photobiol 20:191–199

Govindjee R, Becher B, Ebrey T (1978) The fluorescence from the chromophore of the purple membrane protein. Biophys J 22:67–77

Govindjee R, Ebrey T, Crofts A (1980) The quantum efficiency of proton pumping by the purple membrane protein of *Halobacterium halobium*. Biophys J 30:231–242

Govindjee R, Dancshazy Zs, Ebrey TG, Longstaff C, Rando RR (1988a) Photochemistry of methylated rhodopsins. Photochem Photobiol 48:493–496

Govindjee R, Dancshazy Zs, Ebrey T, Longstaff C, Rando R (1988b) Photochemistry of monomethylated and permethylated bacteriorhodopsin. Biophys J 54:557–562

Govindjee R, Dancshazy Zs, Ebrey T (1989) Difference spectra of the later intermediates of the bacteriorhodopsin photocycle. SPIE Biomol Spectrosc 1–57:126–136

Govindjee R, Balashov S, Ebrey T (1990) Quantum efficiency of the photochemical cycle of bacteriorhodopsin. Biophys J 58:597–608

Govindjee R, Lukashev E, Kono M, Balashov S, Ebrey T, Soppa J, Tittor J, Oesterhelt D (1992) DTyr57Asn mutant of bacteriorhodopsin: M formation and spectral transformations at high pH. Struct Funct Retin Proteins 221:115–118

Govindjee R, Kono M, Balashov S, Imasheva E, Sheves M, Ebrey T (1995) Effect of substitution of tyrosine 57 with asparagine and phenylalanine on the properties of bacteriorhodopsin. Biochemistry 34:4828–4838

Govindjee R, Misra S, Balashov S, Ebrey T, Crouch R, Menick D (1996) Arginine-82 regulates the pK of the group responsible for the light-driven proton release in bacteriorhodopsin. Biophys J 71:1011–1023

Govindjee R, Imasheva ES, Misra S, Balashov S, Ebrey T, Chen N, Menick D, Crouch R (1997) Mutation of a surface residue, lysine129, reverses the order of proton release and uptake in bacteriorhodopsin; guanidine hydrochloride restores it. Biophys J 72:886–898

Kuwata O, Yuan C, Misra S, Govindjee R, Ebrey T (2001) Kinetics and pH dependence of light-induced deprotonation of the Schiff base of rhodopsin: possible coupling to proton uptake and formation of the active form of Meta II. Biochem (Moscow) 66:1283–1299

Li Q, Govindjee R, Ebrey T (1984) A correlation between proton pumping and the bacteriorhodopsin photocycle. Proc Natl Acad Sci USA 81:7079–7082

Liang J, Govindjee R, Ebrey T (1993) Metarhodospin intermediates of the Gecko cone pigment P521. Biochemistry 32:14187–14193

Lin S, Ormos P, Eisenstein L, Govindjee R, Konno K, Nakanishi K (1987) Deprotonation of tyrosines in bacteriorhodopsin as studied by Fourier transform infrared spectroscopy with deuterium and nitrate labeling. Biochemistry 26:8327–8331

Mao B, Govindjee R, Ebrey T, Arnaboldi M, Balogh-Nair V, Nakanishi K, Crouch R (1981) Photochemical and functional properties of bacteriorhodopsins formed from 5,6-dihydro- and 5,6-dihydrodesmethylretinals. Biochemistry 20:428–435

Misra S, Balashov S, Ebrey T, Crouch R, Menick D (1996) Arginine 82 regulates the pKa of the group responsible for the light-driven proton release in bacteriorhodopsin. Biophys J 71:1011–1023

Misra S, Govindjee R, Ebrey T, Chen N, Ma J, Crouch R (1997) Proton uptake and release are rate-limiting steps in the photocycle of the bacteriorhodopsin mutant E204Q. Biochemistry 36:4875–4883

Nakanishi K, Yudd A, Crouch R, Olson G, Cheung H, Govindjee R, Ebrey T, Patel D (1976) Allenic retinals and their visual pigment analogues. J Am Chem Soc 48:236–238

Schiffmiller R, Callender R, Waddell W, Govindjee R, Ebrey T, Kakitani H, Honig B, Nakanishi K (1985) Resonance Raman studies of bacteriorhodopsin analogues. Photochem Photobiol 41:563–567

Tokunaga F, Govindjee R, Ebrey T, Crouch R (1977) Synthetic pigment analogues of the purple membrane protein. Biophys J 19:191–198

Tsuda M, Govindjee R, Ebrey T (1983) Effects of pressure and temperature on the M412 intermediate of the bacteriorhodopsin photocycle. Biophys J 44:249–254