Improvisations in phototrophy. Protein engineering and functional investigation of rhodopsin proton-pumps

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Improvisations in Phototrophy

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For all my inner and outer guides…

“Willingness to be puzzled by what seem to be obvious truths is the first step towards gaining understanding of how the world works” - Noam Chomsky

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Abbreviations

A1	all-*trans*-retinal
A2	all-*trans*-3,4-dehydroretinal
ACRs	anion channelrhodopsins
ALL-E	all-*trans*-all-E-locked-retinal
AR3	Archaerhodopsin3
ATP	adenosine triphosphate
bR	Bacteriorhodopsin
CCCP	carbonyl cyanide m-chlorophenyl hydrazine
CCRs	cation channel rhodopsins
cfu	colony forming units
ChR2	Channelrhodopsin2
DDM	aqueous solution of 1-n-dodecyl-β-D-maltopyranoside
DPC	dodecyl phosophocholine
DMAR	all-*trans*-3-dimethylamino-16-nor-1,2,3,4-didehydroretinal
DMF	dimethylformamide
DTT	1,4-dithiothreitol
GR	*Gloeobacter violaceus* rhodopsin
GR_{CTRL}	*E. coli* MG1655Δ*isc*S MUT pJBS1257- IPTG
GR_{MUT}	*E. coli* MG1655Δ*isc*S-MUT pJBS1257-IPTG, retinal, arabinose
GR_{WT}	*E. coli* MG1655Δ*isc*S-MUT pJBS1257-IPTG, retinal
GR:A1	pigment of GR with A1 as the chromophore
GR-FS	GR single mutant F260S
IPTG	Isopropyl β-D-1-thiogalactopyranoside
HR	Halorhodopsin
MMAR	all-*trans*-3-methylamino-16-nor-1,2,3,4-didehydroretinal

MOA2	all-*trans*-3-methoxy-3,4-dehydroretinal
MSP	membrane scaffold protein
MUT	mutagenesis plasmid
OG	octyl-β-D-glucopyranoside
OGNG	octyl glucose neopentyl glycol
PAR	photosynthetically active radiation
PHE	all-*trans*-1,2,3,4-didehydro-1,5-desmethyl-retinal
pmf	proton motive force
PR	Proteorhodopsin from Monterey Bay SAR86 ϒproteobacterium
PR_{CTRL}	*E. coli* MG1655Δ*isc*S MUT pJBS1255 IPTG
PR_{MUT}	*E. coli* MG1655Δ*isc*S-MUT pJBS1255 IPTG, retinal, arabinose
PR_{WT}	*E. coli* MG1655Δ*isc*S-MUT pJBS1255 IPTG, retinal
PR:A1	pigment of PR with A1 as the chromophore
PR-DNFS	PR double mutant D212N, F234S
PR-TA	PR single mutant T101A
NIR	near-infrared
Rh	bovine visual rhodopsin
RT	room temperature
SB	starvation buffer
SBAz	starvation buffer supplemented with 30 mM sodium azide
SMA	styrene maleic acid copolymer
ssNMR	solid-state nuclear magnetic resonance spectroscopy
SRI	Sensory rhodopsin I
SRII	Sensory rhodopsin II
TD-DFT	time dependent density functional theory
WT	wild type