

Figure S1 All-*trans*-retinal (ATR) is required for light-evoked behavior in Chrimson and CoChR expressing animals

(A, B) Backing behavior as in Figure 1B and C, in the presence (left panel) or the absence (right panel) of all-*trans*-retinal(ATR) in the growth medium; +ATR: $n \ge 33$ animals; -ATR: $n \ge 20$.



B TRN::Chrimson::GFP





Figure S2 Expression of Chrimson::GFP and CoChR::GFP

(A, B) Representative fluorescence photomicrographs and corresponding schematic interpretations of transgenic animals expressing CoChR::GFP and Chrimson::GFP. Scale bars: $80 \ \mu m$



Figure S3 Excitation spectra of blue and red light stimuli

Comparison of the excitation spectra of blue and red light stimuli used in this study.

File S1

Blue light-evoked backing behavior in a FLP::CoChR animal

The video shows the behavioral response of a *FLP::CoChR* animal crawling at the surface of a food-containing NGM plate and repeatedly stimulated with blue light.

Available for download as a .wmv file at www.genetics.org/lookup/suppl/doi:10.1534/genetics.115.177956/-/DC1

Table S1 Plasmids used in this study

Plasmid	Description		
GH50	GFP::unc-54UTR in slot3 Entry vector, gift from Erik Jorgensen		
pMH473	unc-54UTR in slot3 Entry vector, gift from Mark Hammarlund		
mg207	pDEST_R4R3, gift from Miriam Goodman		
mg277	SL2::mCherry::unc-54UTR in slot3 Entry vector, gift from Miriam Goodman		
dg9	unc-122p::RFP		
dg68	mec-3 promoter in slot1 Entry vector		
dg229	QUAS promoter in slot1 Entry vector		
dg239	mec-4 promoter (2kb) in slot1 Entry vector		
dg240	QF in slot2 Entry vector		
dg241	QS in slot2 Entry vector		
dg243	[mec-3p::QF], generated by LR recombination between mg207, pMH473, dg68 and dg240		
dg245	[mec-4p::QS::SL2mCherry], generated by LR recombination between mg207, mg277, dg241, and mg239		
dg260	[mec-4p::Chrimson::GFP], generated by LR recombination between mg207, GH50, mg239, and dg263		
dg262	[mec-4p::CoChR::GFP], generated by LR recombination between mg207, GH50, mg239, and dg265		
dg263	Chrimson, codon optimized and containing artificial introns in slot2 Entry vector, generated through gene synthesis in the pUC57-Kan backbone		
dg264	Chronos, codon optimized and containing artificial introns in slot2 Entry vector, generated through gene synthesis in the pUC57-Kan backbone		
dg265	CoChR, codon optimized and containing artificial introns in slot2 Entry vector, generated through gene synthesis in the pUC57-Kan backbone		
dg276	[QUAS::CoChR::GFP], generated by LR recombination between mg207, GH50, mg229, and dg265		

Table S2 Strains used in this study

Strain name	Genotype	Comment
N2	wild type	
KG1180	lite-1(ce314) X	Mutant displaying no behavioral response to UV and intense blue light.
AQ2313	ljIs123[mec-4p::ChR2 codon optimized; unc-122p::rfp]	TRN::ChR2
DAG252-255	lite-1(ce314) X; domEx252-255[mec- 4p::Chrimson::GFP, unc-122p::rfp]	TRN::Chrimson Injected plasmids: dg260 (20 ng/μl), dg9 (20 ng/μl). We observed similar behavioral responses with all four independent lines and Figure 1 reports aggregated data.
DAG260-263	lite-1(ce314) X; domEx260-263[mec- 4p::CoChR::GFP, unc-122p::rfp]	<i>TRN::CoChR</i> Injected plasmids: dg262 (20 ng/μl), dg9 (20 ng/μl). We observed similar behavioral responses with all four independent lines and Figure 1 reports aggregated data.
DAG272	lite-1(ce314) X; domEx272[mec-3p::QF, mec-4p::QS, QUAS::CoChR::GFP, unc122p::RFP]	<i>FLP::CoChR</i> Injected plasmids: dg243 (5 ng/μl), dg245 (40 ng/μl), dg276 (20 ng/μl), dg9 (20 ng/μl).
DAG342-344	lite-1(ce314) X; domEx342-344[mec- 4p::Chrimson::GFP, mec-3p::QF, mec- 4p::QS, QUAS::CoChR::GFP, unc122p::RFP]	FLP::CoChR;TRN::Chrimson Injected plasmids: dg243 (5 ng/μl), dg245 (40 ng/μl), dg276 (20 ng/μl), dg260 (20 ng/μl), dg9 (20 ng/μl). We observed similar behavioral responses with all three independent lines and Figure 2 reports aggregated data.
DAG355	lite-1(ce314) X; domIs355[mec-3p::QF, mec-4p::QS, QUAS::CoChR::GFP, unc122p::RFP]	<i>FLP::CoChR</i> DAG272 was integrated and outcrossed twice with KG1180 <i>lite-1(ce314)</i> to produce DAG355.
DAG356	domIs355[mec-3p::QF, mec-4p::QS, QUAS::CoChR::GFP, unc122p::RFP]	<i>FLP::CoChR</i> DAG355 was crossed with N2 to remove the <i>lite-1</i> mutation and create DAG356.