Supporting Information

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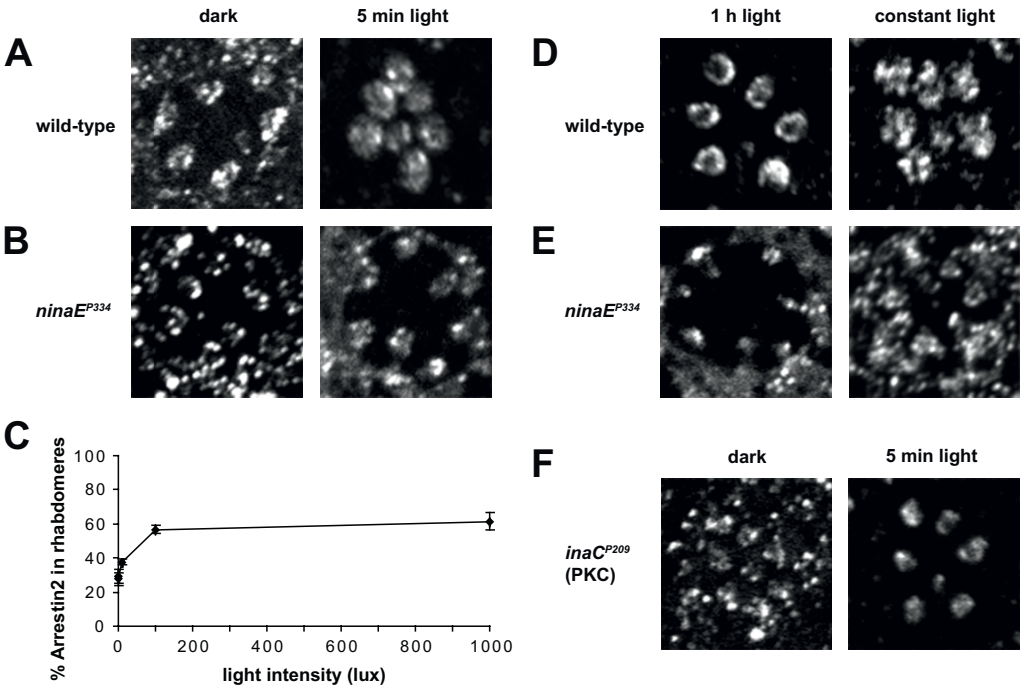

Fig. S1. Translocation of Arr2 requires rhodopsin but not protein kinase C. Tangential sections of compound eyes from (*A* and *D*) wild-type (w^{1118}), (*B* and *E*) $ninaE^{P334}$, and (*F*) PKC null mutant ($inaC^{P209}$) flies. Flies were dark-adapted for ≥10 h and subsequently remained in the dark or were exposed to white light for 5 min, 1 h, or ≥12 h (constant light). (*A*, *B*, and *D–F*) Confocal images of representative ommatidia immunostained with anti-Arr2 antibodies. (*C*) Quantification of Arr2 staining in the rhabdomeres of wild-type flies exposed to different light intensities based on examination of 11–12 ommatidia each. Error bars represent SEM.

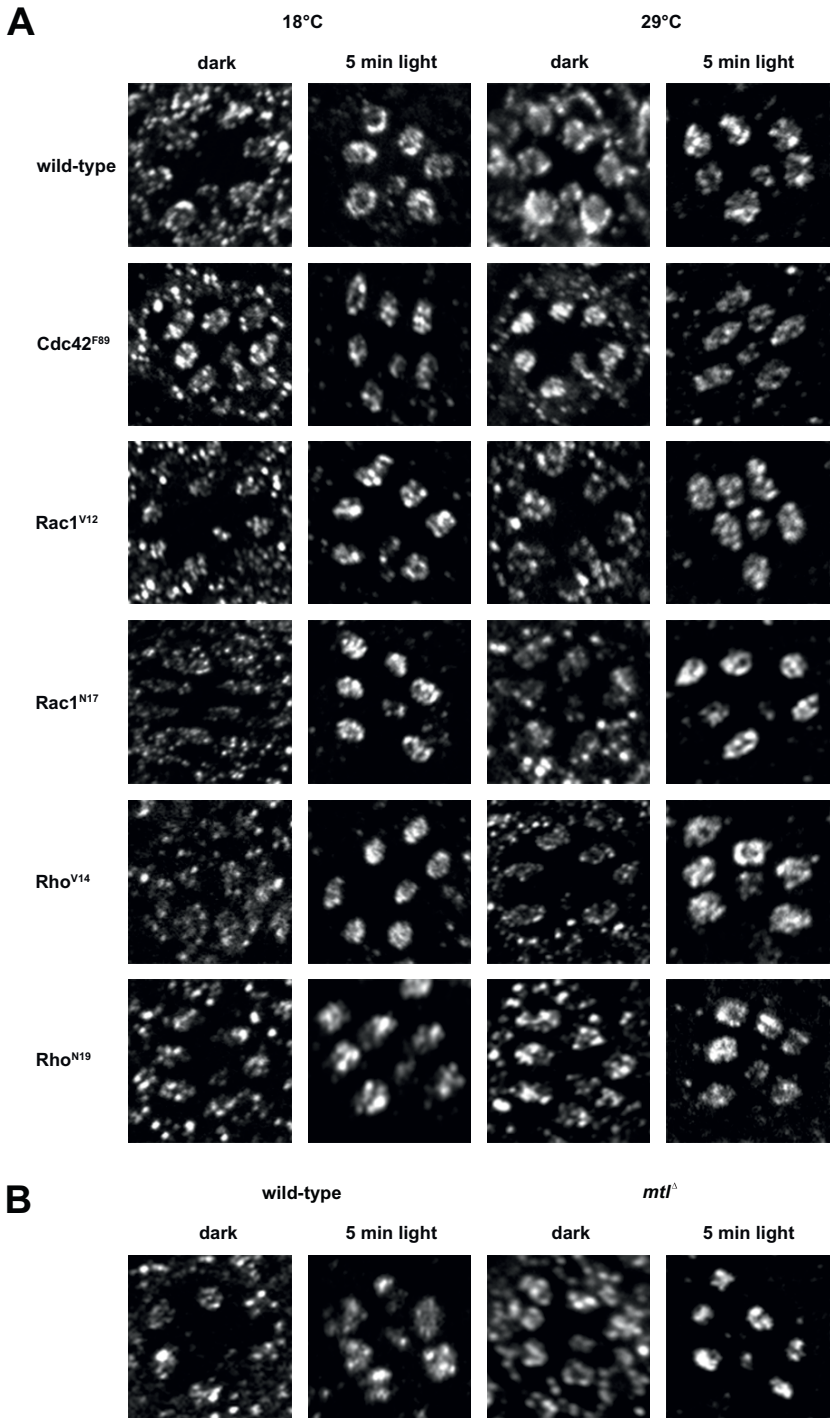

Fig. S2. Cdc42, Rac1, Rho, and Mig2-like are not involved in light-induced Arr2 translocation. Tangential sections of the distal (R7) part of the compound eye were obtained from flies of the indicated genotypes, immunostained with anti-Arr2 antibodies, and viewed by confocal microscopy. (*A*) The TARGET gene expression system (1) was used in combination with *GMR-GAL4* and a temperature-sensitive GAL80 expressed under the control of the *tubulin* promoter (*tub-GAL80^{ts}*). We efficiently blocked activation of *GAL4* transcription by maintaining the flies at the permissive temperature (18 °C) for GAL80^{ts}. When the flies are kept at the nonpermissive temperature (29 °C) for GAL80^{ts}, GAL4 initiated transcription of *UAS* transgenes. The flies (≤3 days old) were reared under a 12-h light/12-h dark cycle at 18 °C. Flies were shifted to 29 °C for 72 h to induce expression of the constitutively active or dominant-negative small GTPases. Control flies were kept at 18 °C. Before the experiment, the flies were dark-adapted for ≥10 h and subsequently either exposed to white light for 5 min or maintained in the dark. (*B*) Localization of Arr2 in sections obtained from wild-type and genetically mosaic flies in which the eyes were composed exclusively of *mtl*$^{\Delta}$ cells.

1. McGuire SE, Mao Z, Davis RL (2004) Spatiotemporal gene expression targeting with the TARGET and gene-switch systems in *Drosophila*. *Sci STKE* 2004:pl6.

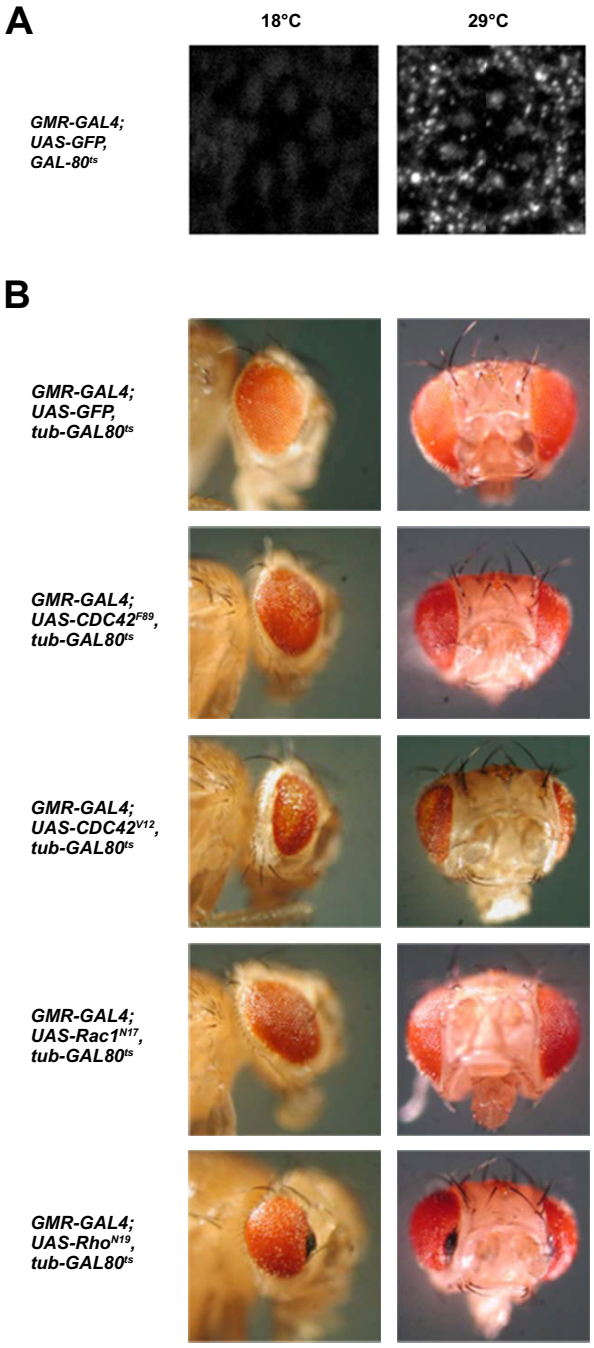

Fig. S3. The TARGET expression system efficiently controls expression of *UAS* constructs in fly eyes. (*A*) Tangential sections of eyes obtained from flies expressing GFP (*UAS-GFP*) under control of the TARGET system (1) (using *GMR-GAL4* and *tub-GAL80^{ts}*). Flies (≤3 days old) were reared under a 12-h light/12-h dark cycle at 18 °C and shifted to 29 °C for 72 h to induce GFP expression. Control flies were kept at 18 °C. Sections of single ommatidia were immunostained with anti-GFP antibodies and viewed by confocal microscopy. (*B*) To determine whether the expression of the small GTPases using the TARGET system was induced at 29 °C, the flies were reared at 29 °C and the effects on eye morphology were analyzed. Flies carrying the *UAS-GFP* transgene, and therefore expressing GFP, were used as a control.

1. McGuire SE, Mao Z, Davis RL (2004) Spatiotemporal gene expression targeting with the TARGET and gene-switch systems in *Drosophila*. *Sci STKE* 2004:pl6.

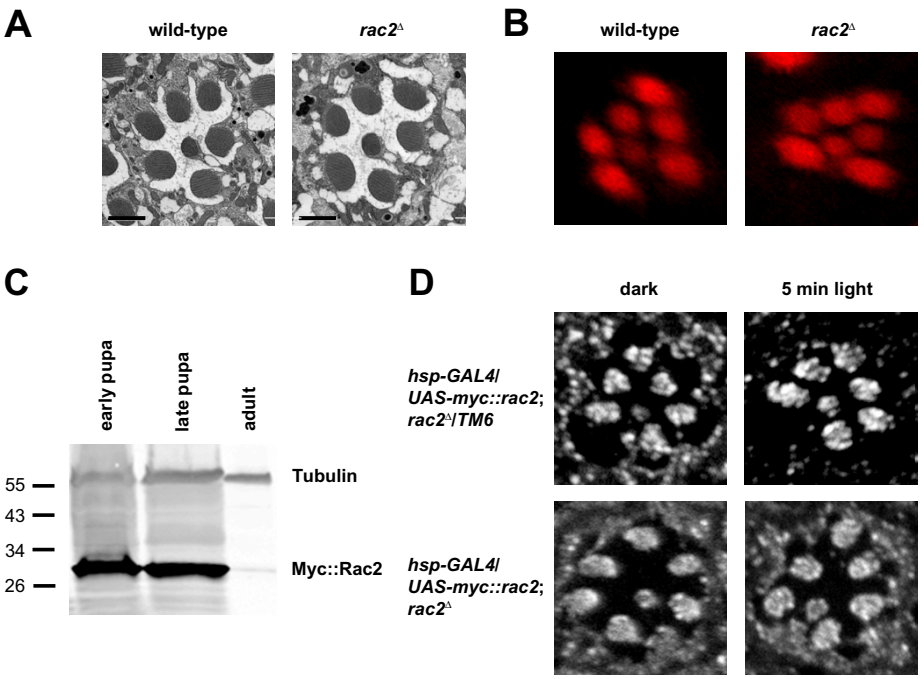

Fig. S4. Deletion of *rac2* does not alter photoreceptor cell morphology. (*A*) Eye morphology of wild-type and $rac2^{\Delta}$ ommatidia examined by transmission electron microscopy. The cross-sections were obtained from young flies (≤3 days old) kept under a 12-h light/12-h dark cycle. (Scale bars, 2 μm.) (*B*) Phalloidin staining of whole-mount retina. Shown are confocal images of single ommatidia from wild-type (ry^{506}) and $rac2^{\Delta}$ flies. (*C*) Western blot containing extracts prepared from either early pupae, late pupae, or adult flies overexpressing Myc-tagged Rac2 under the control of *hsp-GAL4* during development. The blot was probed with anti-Myc and anti-Tubulin antibodies. The positions of protein size markers are indicated. (*D*) Tangential sections of the compound eye were obtained from flies expressing Myc::Rac2 during development, immunostained with anti-Arr2 antibodies and viewed by confocal microscopy. The flies were either heterozygous (upper panel) or homozygous for $rac2^{\Delta}$ (lower panel).

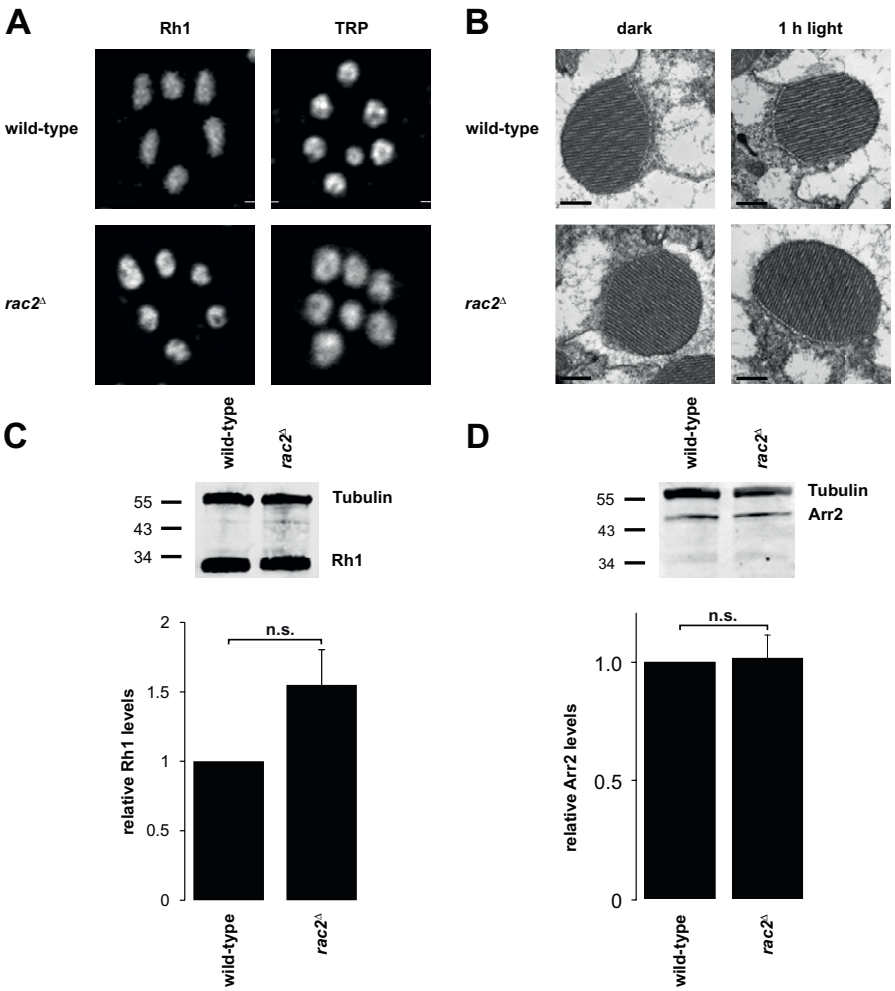

Fig. S5. Spatial distribution of Rh1 and TRP in *rac2*$^{\Delta}$ flies and morphology of *rac2*$^{\Delta}$ rhabdomeres. (*A*) The spatial distributions of Rh1 and TRP were analyzed in tangential sections of compound eyes from wild-type and *rac2*$^{\Delta}$ flies. Ommatidia were immunostained with anti-Rh1 and anti-TRP antibodies and viewed by confocal microscopy. (*B*) Transmission electron microscopy of single rhabdomeres. Tangential sections of the compound eye were obtained from wild-type and *rac2*$^{\Delta}$ flies. The flies were dark-adapted for ≥10 h and subsequently maintained in the dark (left) or exposed to light for 1 h (right). (Scale bars, 0.5 μm.) (*C* and *D*) Expression levels of Rh1 and Arr2 were not decreased in *rac2*$^{\Delta}$ flies. Western blots containing head extracts prepared from wild-type and *rac2*$^{\Delta}$ flies were probed with anti-Tubulin antibodies and either anti-Rh1 or anti-Arr2 antibodies, respectively. The bar graphs show quantification of the relative expression levels of Rh1 and Arr2. The error bars represent SEM, and n.s. indicates no significant differences using the Student's *t* test.