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Supplemental Information

The C-Terminus and Third Cytoplasmic Loop Cooperatively Activate Mouse Melanopsin Phototransduction

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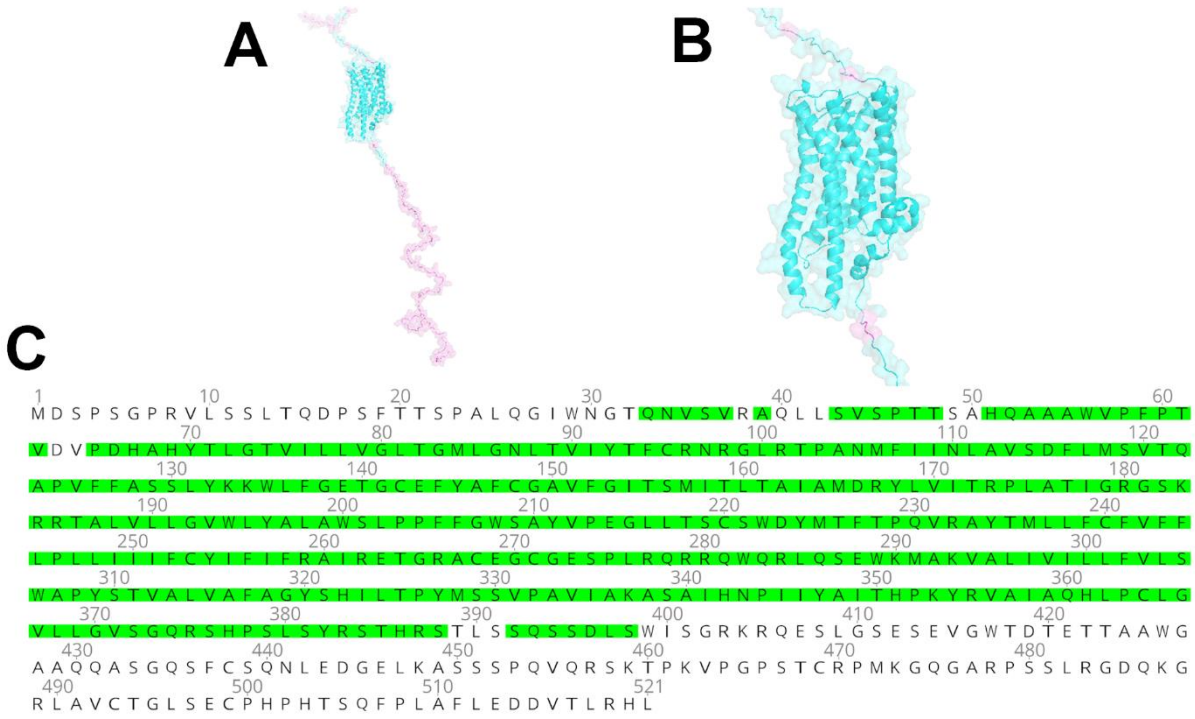
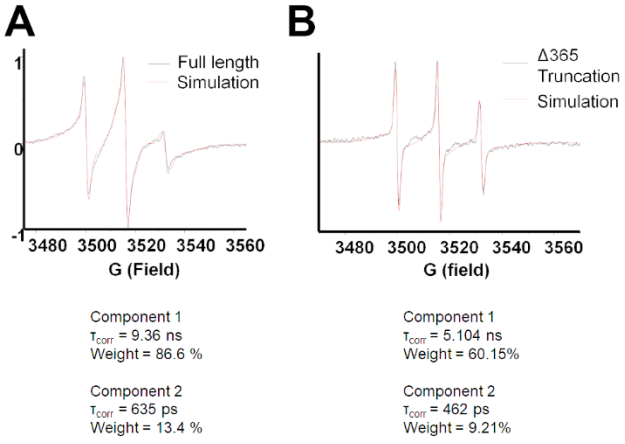
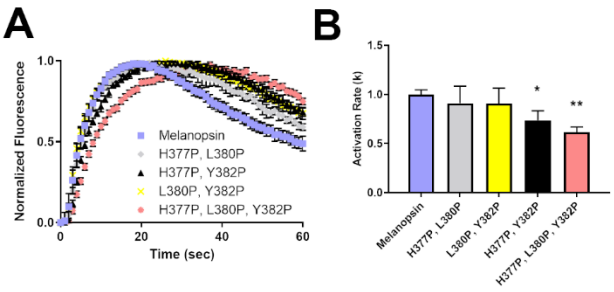


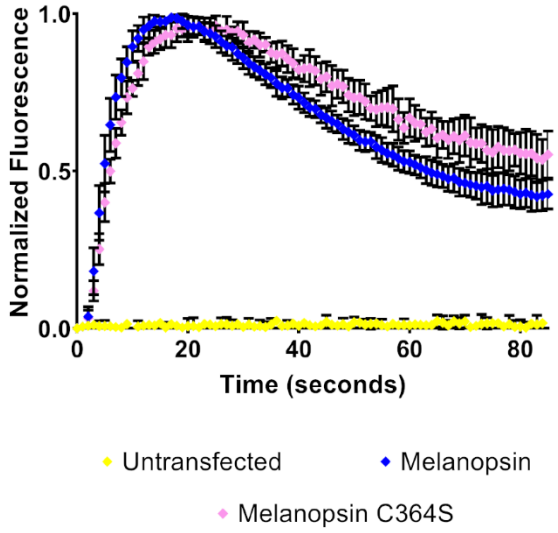
Supplemental Figure 1: *Template sequence coverage of mouse melanopsin model.* (A & B) Mouse melanopsin homology model depicting amino acids on melanopsin that were modeled using amino acids on the *T. pacificus* template. Blue residues denote residues covered by template, pink residues are non-covered residues. (C) Template sequence coverage plotted on melanopsin's amino acid sequence.



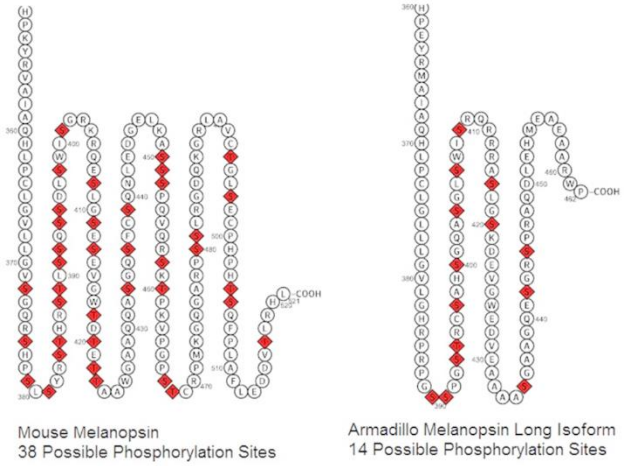
Supplemental Figure 2: *MATLAB/EasySpin simulation of experimental EPR data.* (**A**) Simulation spectrum fitting the full-length melanopsin C268 EPR spectrum or (**B**) melanopsin C268 Δ265, C-terminal truncated mutant. Rotational correlation times (τ_{corr}) calculated from the simulation, depicted below the graph. Two components calculated for each construct, the 1st and slower components represents a more immobile component, derived from spin-label attached to melanopsin, while the 2nd and faster component represents a faster component, likely from excess, free spin-label in solution.



Supplemental Figure 3: Calcium imaging of double and triple melanopsin C-terminus proline point mutants. (**A**) Calcium imaging of HEK293 cells expressing melanopsin C-terminal mutants, synthesized with combinations of point mutations at residues H377, L380, and Y382 to proline residues. (**B**) Calculated activation rates of melanopsin constructs depicted in (A). All error bars represent standard error of the mean (S.E.M.) of three independent transfections. Statistical significance tested by using Students t-test, *, **, ***, **** represent P-values <0.05, 0.01, 0.001, and 0.0001, respectively. All constructs compared to wild-type melanopsin's rate, and statistical significance is indicated over individual bars.



Supplemental Figure 4: *Calcium imaging of melanopsin palmitoylation mutant.* Representative calcium imaging of melanopsin C364S, with mutation of melanopsin's predicted C-terminus palmitoylation site, C364 to a serine residue. Error bars depict standard deviation (S.D.).



Supplemental Figure 5: *2-dimensional schematics comparing mouse and armadillo melanopsin C-termini.*