



**Supplementary Figure S1.** Location of RHO mutation and conservation of amino acid residues at position 104 among species. (A) Two-dimensional sequence of rhodopsin and localization of the identified mutation (V104I). H1–H7 indicates rhodopsin transmembrane helices 1–7; H8 is intracellular helix 8; N, amino terminus; C, carboxyl terminus; EL, extracellular loop; CL, cytoplasmic loop. (B) Sequence alignment of selected regions of RHO. The position of identified p.V104I mutation was comparatively conserved across species. (C) Structural implications of RHO amino acid substitution (V104I). The wild-type residue at position 104 is colored purple and shown in a red dashed box. The mutated residue is colored brown, and was located at the interface between the N-terminal cap (blue) and EL1, and is displayed on EL1. The N-terminal cap strengthens the stability of the EL2  $\beta$ -sheet that serves as a ‘lid’ to block rapid exit of the retinal from the pocket.<sup>1</sup> The V104I mutation does not increase the contact area between the N-terminal cap and EL1. The amino acid transition at position 104 is shown as a stick model in the orange dashed box. Structures were prepared with pymol; corresponding PDB code is 4ZWJ.

## References

1. Zhou XE, Melcher K, Xu HE. Structure and activation of rhodopsin. *Acta Pharmacol Sin.* 2012;33:291–299.