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Supplemental Information

Oncogenic K-Ras Binds to an Anionic Membrane in Two Distinct Orientations: A Molecular Dynamics Analysis

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SUPPLEMENTARY DATA

Oncogenic K-Ras binds to an anionic membrane in two distinct orientations: A molecular dynamics analysis

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Running Title: Oncogenic K-Ras membrane interaction

Movie S1: A movie showing the transition from one orientation state (OS1) to another (OS2). The movie was generated from one of the Nocmap-K-Ras simulations (panel 3 of Figure 2B in the main text), which is the only trajectory that underwent such a transition. The "toehold" region comprising the C-terminus of helix-3 and loop7 is shown in purple, helices 3 and 4 are green, and strands β1-3 and helix 2 are in orange. The residues in licorice are those within 4 Å of the bilayer (gray lines). The elements in licorice are colored as follows: nitrogen (blue), oxygen (red), carbon (cyan), sulphur (yellow).

Movie S2: A movie showing the "rocking" motion via the "toehold" in a NormK-Ras simulation (panel 4 of Figure 2A in the main text). Color scheme is the same as in S1.

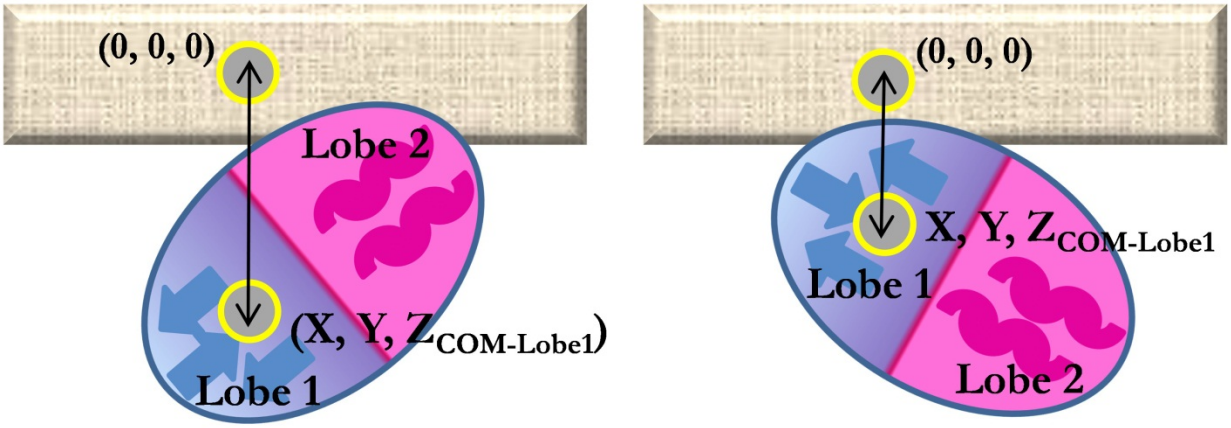

Figure S1: A schematic illustration of one of the two reaction coordinates used in this paper, $Z_{COM\text{-}lobe1}$, defined as the z-coordinate of the center of mass of lobe1 (residues 1-86) of K-Ras after aligning the bilayer center at the origin. The bilayer (rectangle) is centered at the origin so that $Z_{COM\text{-}Lobe1}$ measures the displacement of lobe 1 (in blue) to and away from the bilayer. $Z_{COM\text{-}Lobe2}$ was defined similarly to monitor the displacement of lobe 2 (pink) along the membrane normal.

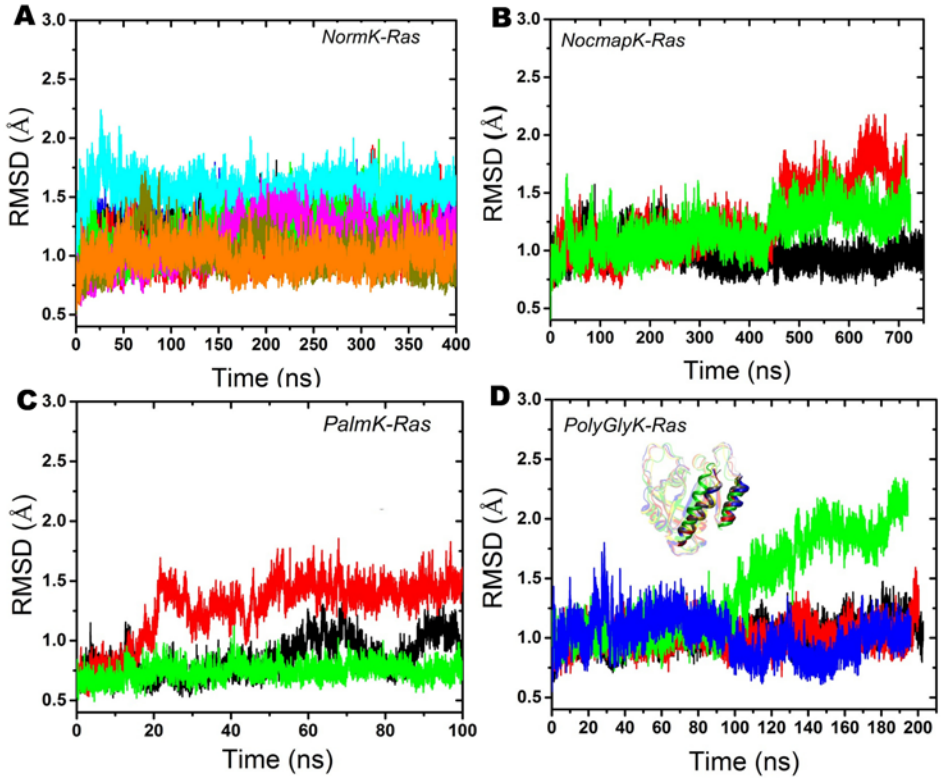

Figure S2: Backbone root mean square deviation (RMSD) of the catalytic domain during **(A)** eight NormK-Ras simulations where the CD makes direct contact with the bilayer, **(B)** three NocmapK-Ras simulations, **(C)** three PalmK-Ras simulations, and (D) four PolyGlyK-Ras simulations. The inset in **(D)** shows an overlay of the last snapshot from the four runs, highlighting the significant conformational change of helix 3 in one of them. The flexible residues of switch 1 (25- 40), switch 2 (57-75) and the HVR (residues 169-185) were excluded. The equilibrated structure was used as a reference in each case except the following. As mentioned in Methods of the main text, the NormK-Ras simulations consisted of two sub-groups, where in one group PG to PS replacement was performed without a short no-CMAP MD relaxation whereas in the other PG to PS replacement was done after a 20 ns relation without CMAP. In the latter, the backbone RMSD using the initial structure as a reference was relatively large (1.8-2.5 Å) due to conformational changes prior to the application of CMAP; this was confirmed by the finding that using the crystal structure as a reference reduced the RMSD to about 1.2 Å within the first 20 ns. Therefore, we regarded the 25 ns snapshot as a better-equilibrated reference structure for this sub-group of simulations.

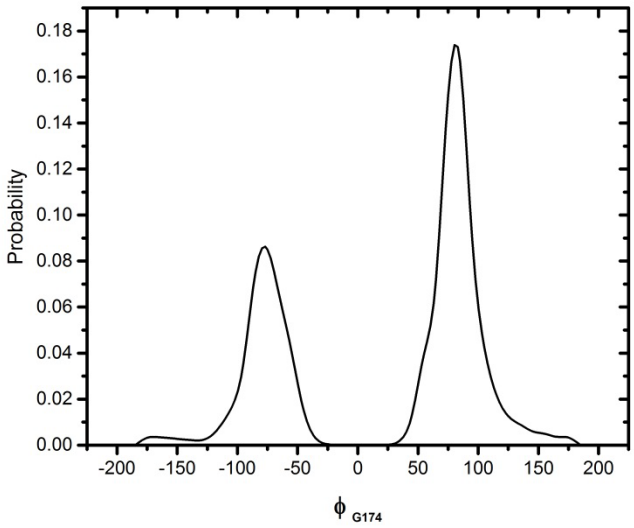

Figure S3: Distribution of the dihedral angle phi of Gly74 among conformers in which the catalytic domain makes direct contact with the bilayer, derived from only the NormK-Ras simulations.

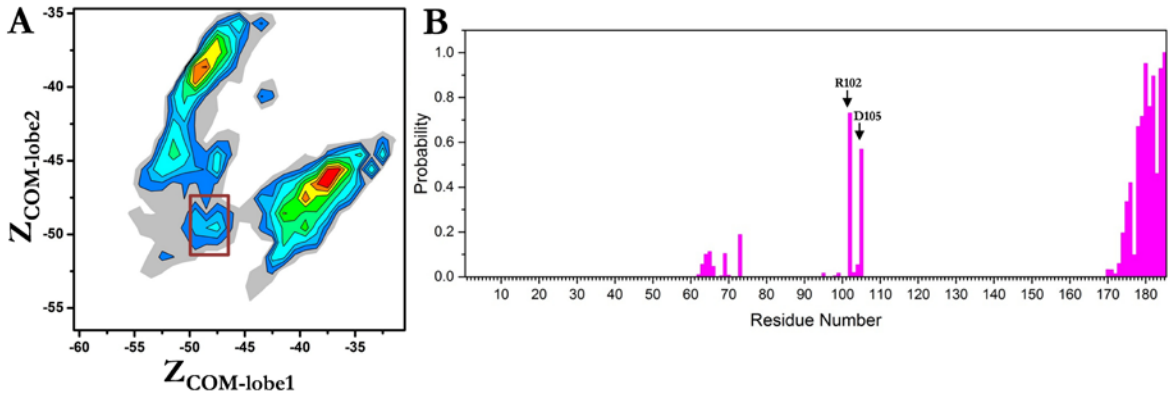

Figure S4: A putative intermediate orientation state. (A) Population density plot highlighting a putative intermediate orientation (purple box) identified by the $P(Z_{COM\text{-}lobe1}, Z_{COM\text{-}lobe2})$ analysis described in the main text. **(B)** Normalized probability distribution of bilayer contact of K-Ras residues in the intermediate state. The data was derived from the NormK-Ras simulations.

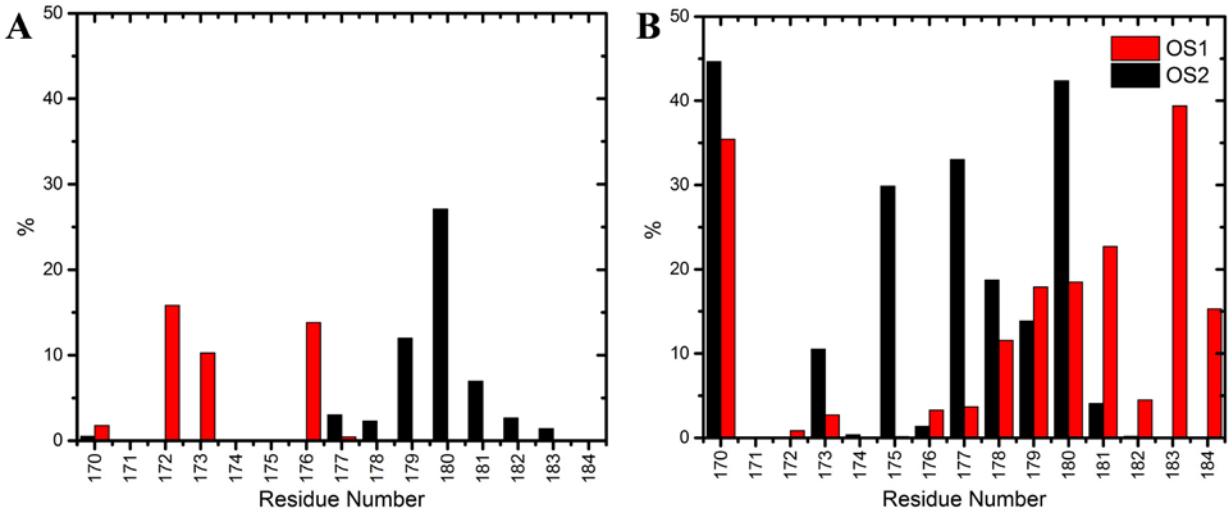

Figure S5: HVR-core interactions. Shown are percentages (y-axis) of cumulative heavy atom contacts made by individual HVR residues (x-axis) with residues on the surface of the β1-β3 region **(A)** and loop 7 **(B)** in OS1 (red) and OS2 (black).