Abstract

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Configuration of PKCalpha-C2 domain bound to mixed SOPC/SOPS lipid monolayers.

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Author information

Abstract

X-ray reflectivity measurements are used to determine the configuration of the C2 domain of protein kinase Calpha (PKCalpha-C2) bound to a lipid monolayer of a 7:3 mixture of 1-stearoyl-2-oleoyl-sn-glycero-3-phosphocholine and 1-stearoyl-2-oleoyl-sn-glycero-3-phosphoserine supported on a buffered aqueous solution. The reflectivity is analyzed in terms of the known crystallographic structure of PKCalpha-C2 and a slab model representation of the lipid layer. The configuration of lipid-bound PKCalpha-C2 is described by two angles that define its orientation, theta = 35 degrees +/- 10 degrees and phi =210 degrees +/- 30 degrees, and a penetration depth (=7.5 +/- 2 A) into the lipid layer. In this structure, the beta-sheets of PKCalpha-C2 are nearly perpendicular to the lipid layer and the domain penetrates into the headgroup region of the lipid layer, but not into the tailgroup region. This configuration of PKCalpha-C2 determined by our x-ray reflectivity is consistent with many previous findings, particularly mutational studies, and also provides what we believe is new molecular insight into the mechanism of PKCalpha enzyme activation. Our analysis method, which allows us to test all possible protein orientations, shows that our data cannot be explained by a protein that is orientated parallel to the membrane, as suggested by earlier work.

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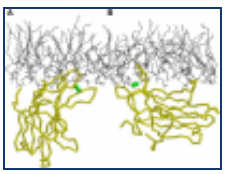
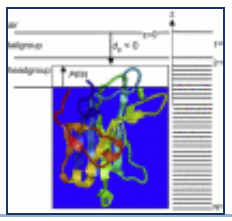
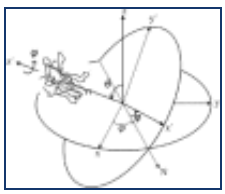
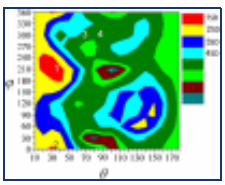
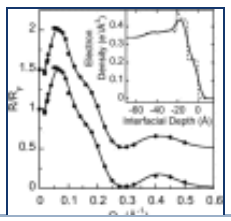
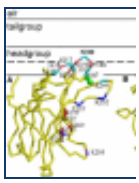

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