The structure of the complex of the cytoplasmic guanine nucleotide exchange factor Ric-8A with Gαi1

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The alpha subunits (Gα) of heterotrimeric G proteins are activated by guanine nucleotide exchange factors (GEFs) that catalyze the release of GDP from the Gα nucleotide binding site and subsequent loading of GTP into that site[1]. Plasma membrane-bound, agonist-stimulated G protein-coupled receptors (GPCRs) are the best characterized heterotrimeric G protein GEFs, but cytoplasmic GEFs have also been discovered. Of these, the 530-residue Ric-8A protein has been identified as both a GEF and a folding chaperone for the i, q, and 12/13 classes of Gα[2]. Here we present the X-ray crystallographic and cryo-EM structures of the complex of Ric-8A (residues 1-491) bound Gαi1 stabilized by three camelid nanobodies at 3.5-4.5Å resolution. The N-terminal 430 residues of Ric-8A adopt a mixed Armadillo/HEAT repeat fold and the disordered segment that follows includes two highly conserved serine and threonine residues that, when phosphorylated, stimulate the GEF activity of Ric-8A[3]. Ric-8A interacts with the Ras-like domain of Gαi1 at two major interfaces: with the two beta strands in the C-terminal half of the Ras domain opposite to the nucleotide binding site, and through extensive contact surface with the C-terminal alpha helix. The latter interaction is analogous to, but structurally different from, that observed in G protein complexes with with G protein-coupled receptors (GPCRs). The former has no parallel in G-protein:GPCR interactions. Together, the contacts between Ric-8A and Gα induce allosteric changes that result in the expulsion of GDP from the nucleotide binding site.

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