



(A) Schematic view of the domain structure of VanS proteins. These comprise two transmembrane helices (TM1 & TM2), a periplasmic sensor domain, a membrane-proximal HAMP domain, and two catalytic domains: a dimerization & histidine-phosphorylation domain (DHp) and a catalytic & ATP-binding domain (CA). MWs for different VanS proteins are ca. 42-45 kDa, and the protein is an obligate dimer. (B) Model for nanodisc formation. The structure shown is the AlphaFold prediction for the A-type VanS protein (VanS_A). (C) Coomassie-stained SDS PAGE gel showing nanodisc preparations for the A- and B-type VanS proteins. MSP1D1 is used as the scaffolding protein for both preparations. (D) Size-exclusion chromatogram demonstrating homogeneity for purified protein-nanodisc preps of the A- and B-type VanS proteins. (E) Representative images collected on the Thomas Jefferson Univ. Glacios instrument showing VanS-containing nanodiscs (in red boxes). (F) Enlarged views of individual nanodiscs in two different orientations.