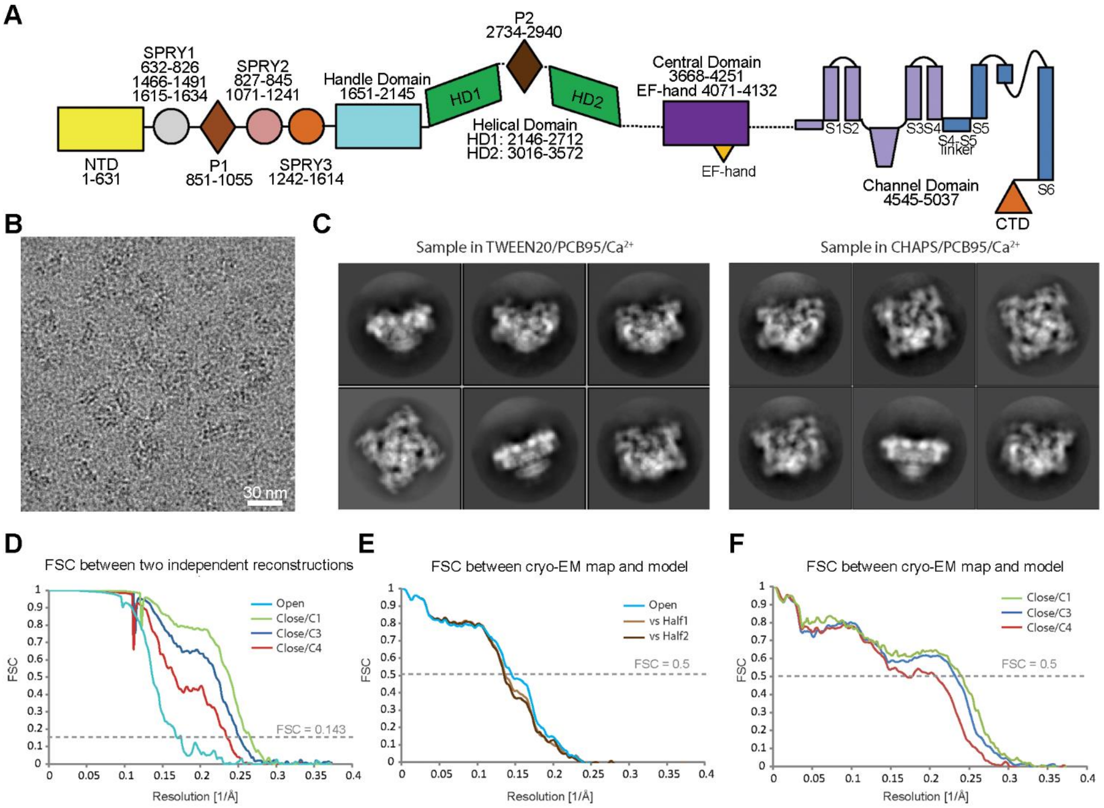

Supplementary information, Figure S1 Image processing and resolution estimation. **(A)** A schematic illustration of domain organization in one RyR1 protomer. The name and boundary of each domain are labeled. The panel is adapted from our previous publication on RyR1 [1]. **(B)** A representative micrograph. Scale bar, 30 nm. **(C)** Representative 2D class averages of the sample in Tween-20 and in CHAPS. **(D)** Gold-standard FSC curves for the maps in both open and close states. **(E)** FSC curves of model versus map for the map in the open state and cross-validation of model refinement by half maps. The light grey curves show FSC between the model and the half-map it was refined against (half1) and the dark grey curves show FSC between the model and the other half-map (half2) it was not refined against. The blue curves show FSC between the atomic model and the sum map, which the model was refined against. **(F)** FSC curves of model versus map for the maps in different close states.

1 Yan Z, Bai XC, Yan C *et al.* Structure of the rabbit ryanodine receptor RyR1 at near-atomic resolution. *Nature* 2015; **517**:50-55.