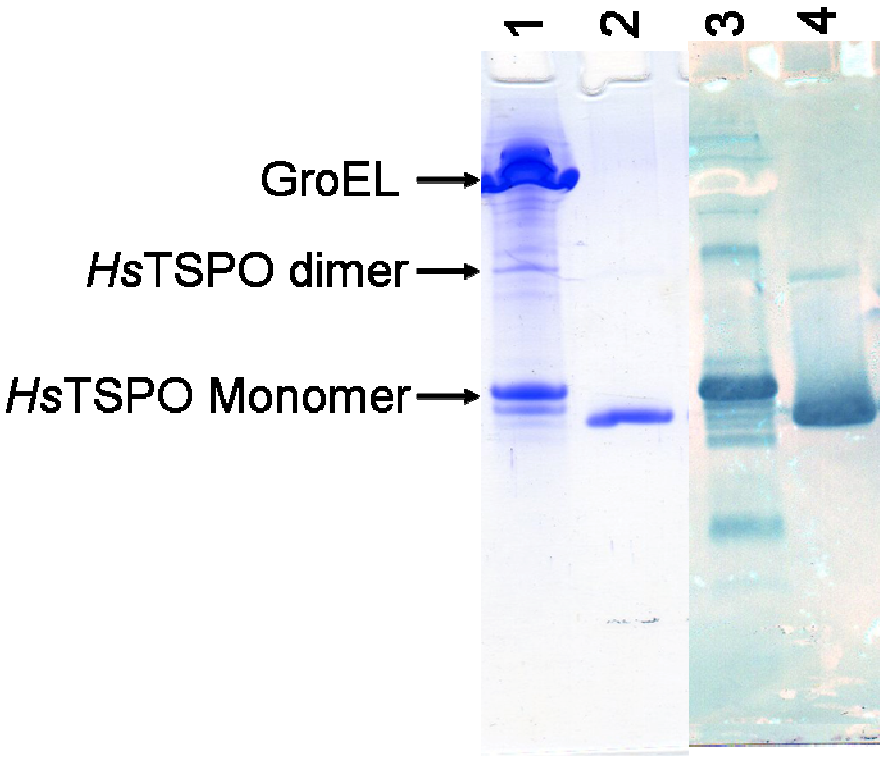
SUPPORTING INFORMATION AVAILABLE

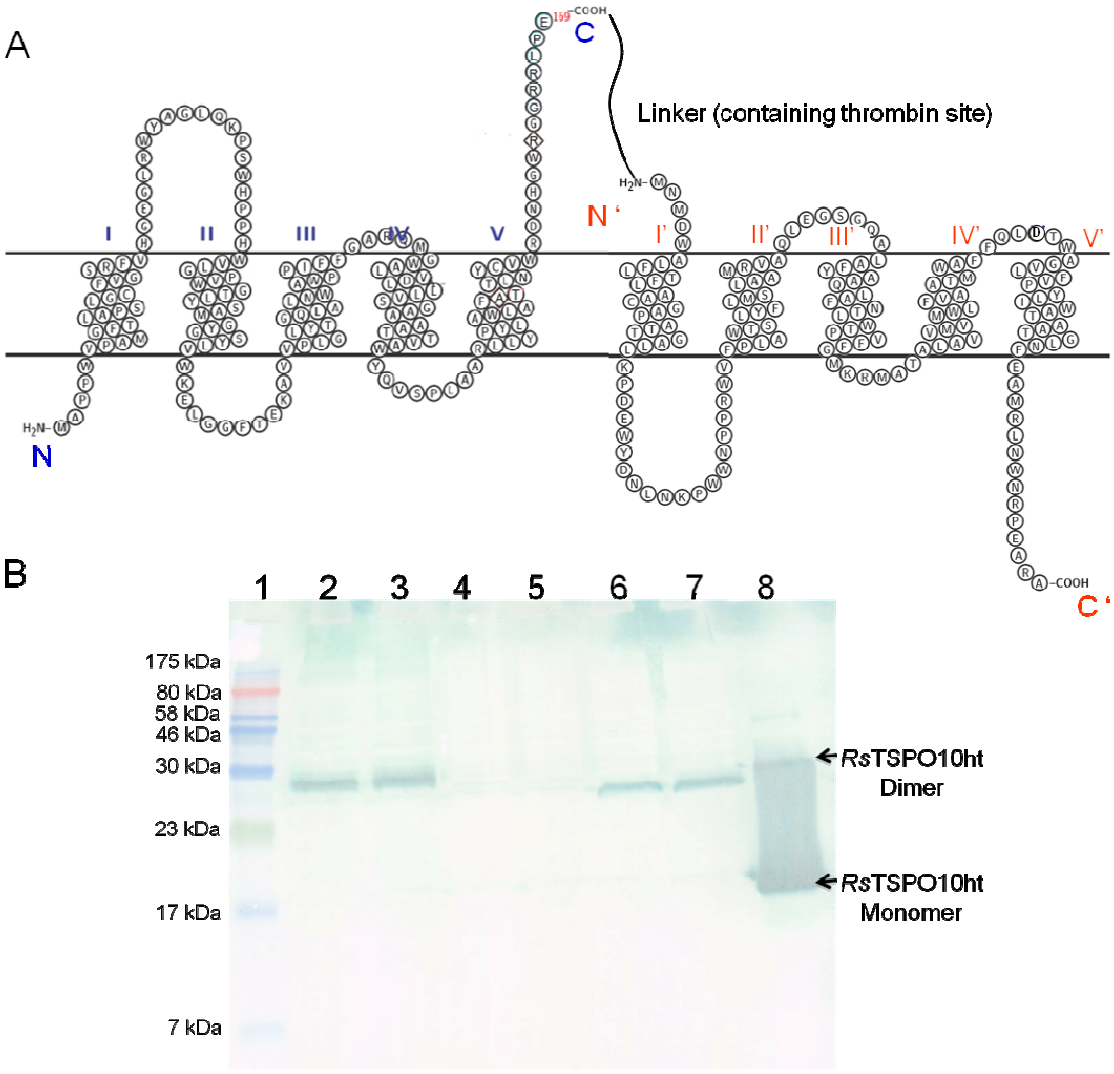
Supporting Table T1. Primer sequences used in this study. Restriction sites are underlined.

Mutant or construct	Primers used
*Rs*TSPOW38C	FW, 5'-GGCGGGTTCCAGCAGGGCTTGTTCAG -3' RV, 5'-CTGAACAAGCCCTGCTGGAACCCGCC -3'
*Hs_Rs*TSPO	Hs-FW, 5'-CGACATATGGCACCACCTTGG-3' Hs_Rs-RV, 5'-CCAGTCCATGTTGCTACCACGC-3' Hs_Rs-FW, 5'-CCGCGTGGTAGCAACATGGACTG-3' Rs-RV, 5'-GCGGAATTCGAGCTCGGTACC -3'
*Rs_Hs*TSPO	Rs-FW, 5'-ATTCATATGGACTGGGCTCTT -3' Rs_Hs-RV, 5'-CCAAGGTGGTGCGGCTTCGGG -3'; Rs_Hs-FW, 5'-GCCCGAAGCCGCACCACCTTGG-3' Hs-RV, 5'-GGGGAATTCTTAGTGGTGATGATG -3'
*Rs_Rs*TSPO	Rs-FW, 5'-ATTCATATGGACTGGGCTCTT -3' Rs_Rs-RV, 5'-CGGCCCGAAGCCAACATGGACTGG -3' Rs_Rs-FW, 5'-CCAGTCCATGTTGGCTTCGGGCCG-3' Rs-RV, 5'-GCGGAATTCGAGCTCGGTACC -3';

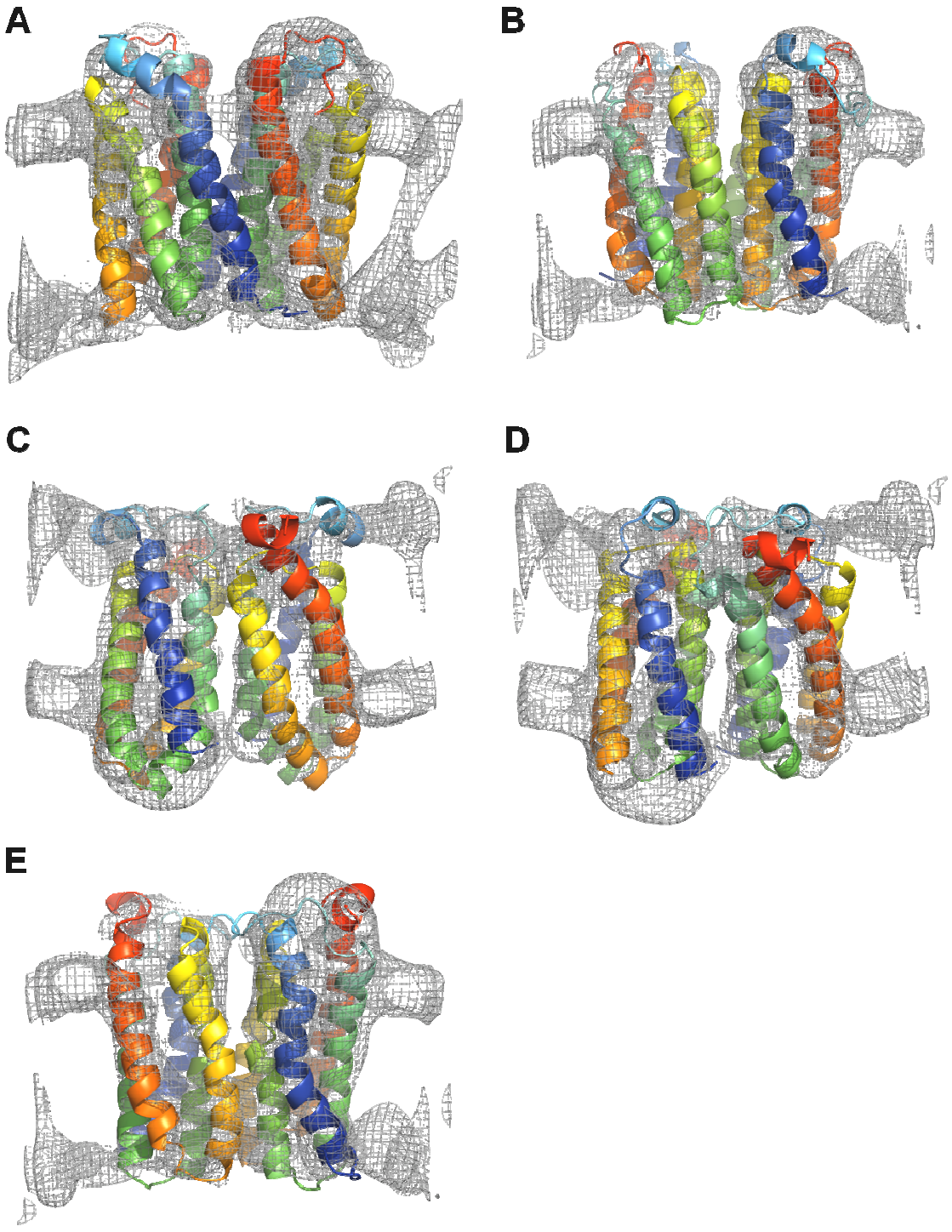
Supporting Figure S1. Purification of *Hs*TSPO. Lane 1: Coomassie stain of purified *Hs*TSPO; lane 2: Coomassie stain of purified *Rs*TSPO as control; lane 3: western blot against histag of purified *Hs*TSPO; lane 4: western blot against histag of purified *Rs*TSPO as control.



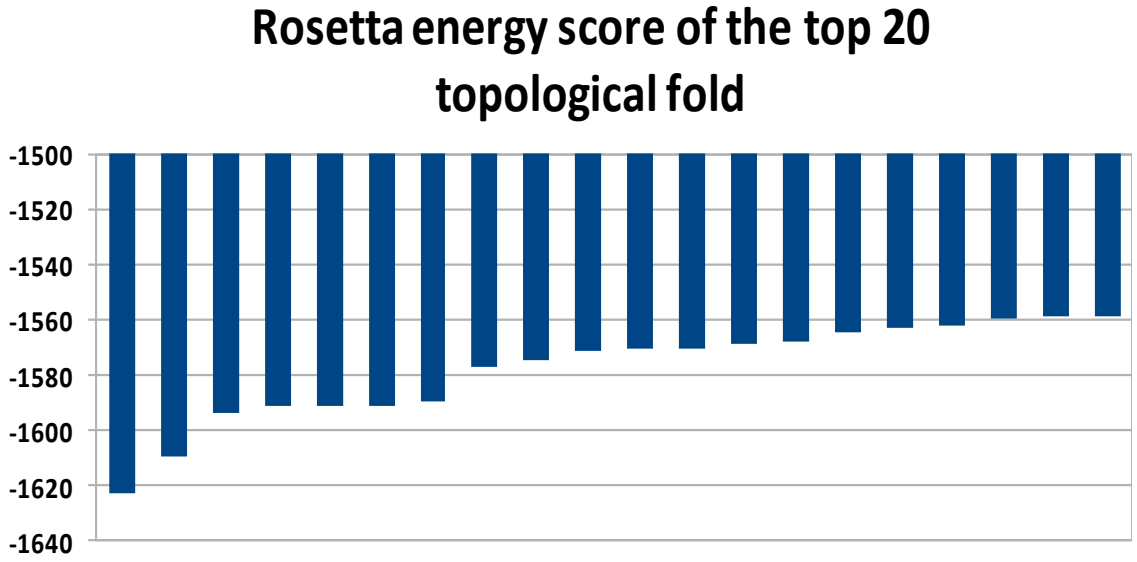
Supporting Figure S2. Expression of TSPO fusion proteins. (A) Topology illustration of fusion proteins. All three fusion proteins have the same anti-parallel configuration. Shown in the figure is the *Hs*-*Rs*TSPO fusion construct, in which a thrombin site was placed in between the C-terminus of *Hs*TSPO and the N-terminus of the *Rs*TSPO. In the cases of *Rs*-*Hs*TSPO and *Rs*-*Rs*TSPO, the fusion proteins are constructed by directly connecting the C-terminus and the N-terminus. A 10-histag was constructed at the C-terminus of all the three fusion proteins. (B) Western blot of the whole cells expressing fusion proteins with antibodies against the C-terminal 10-histag. Lane 1: size marker; lane 2: *Hs*-*Rs*TSPO10ht after 7 hrs growth; lane 3: *Hs*-*Rs*TSPO10ht after 22 hrs growth; lane 4: *Rs*-*Hs*TSPO10ht after 7 hrs growth; lane 5: *Rs*-*Hs*TSPO10ht after 22 hrs growth; lane 6: *Rs*-*Rs*TSPO10ht after 7 hrs growth; lane 7: *Rs*-*Rs*TSPO10ht after 22 hrs growth; lane 8: positive control of purified *Rs*TSPO10ht. The fusion proteins were also found to be mainly in the membrane fraction of the fractionated cells, based on estimates from the yields of total membranes.



Supporting Figure S3. Top 3-5 scoring models of *Rs*TSPO dimer. Models are displayed as rainbow diagram in order of ranking score. (A) Model 3, (B) Model 4, (C) Model 5, (D) Model 6, (E) Model 7.



Supporting Figure S4. Rosetta energy score shows that the top 7 models are significantly better than the rest.



Supporting Figure S5. Oligomerization of *Rs*TSPO. Models of *Rs*TSPO dimers were placed into the extended cryo EM electron density of the helical polymer crystal. (A) and (B): model 1; (C) and (D): model 2.