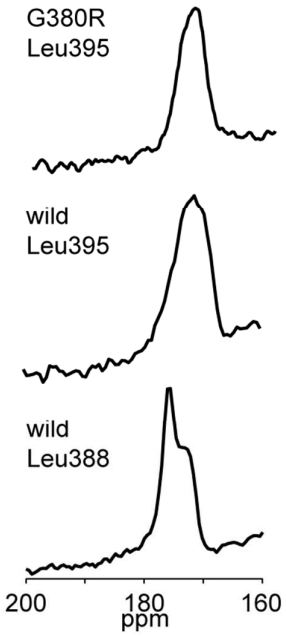
Supporting Information

Supporting Figure 1. ^{13}C CPMAS NMR of TM-JM peptide

To obtain information on the secondary structure for the TM-JM boundary region, we obtained ^{13}C spectra of TM-JM peptides in the wild type and G380R sequences containing specific backbone ^{13}C labels at the putative TM-JM boundary. TM-JM peptide contains a single ^{13}C=O labeled amino acid at Leu388 and Leu395. Leu388 is considered to be located in the middle of the TM region whereas Leu395 is at the TM-JM boundary. The spectra were obtained at a spinning frequency of 12.5 kHz and a temperature of -50 ^{o}C.

The carbonyl ^{13}C chemical shifts are sensitive to the secondary structure. Saito and Naito have correlated chemical shifts from solid-state NMR studies of fibrous and membrane proteins and have found that in helical secondary structure, carbonyl carbons exhibit chemical shifts in the range of 173-176 ppm, while in random coil and β-strand structure they are in the range of 168-172 ppm [1].

Chemical shifts for ^{13}C=O from Leu388, Leu395 on the wild type and Leu395 on G380R sequence were 175.8, 172.8 and 172.0 ppm respectively. For Leu388 in the middle of the TM region, ^{13}C on the backbone C=O showed typical chemical shift for being in α-helical structure. Contrarily, Leu395 located at the boundary region was suggested to be in extended structure. Considering the fact that two consecutive prolines exist at two residues away towards the C-terminus, the IJM region is expected to be in random extended structure.



1. Saito, H.T., Satoru; Naito, Akira. (1998) Empirical Versus N On Empirical Evaluation Of Secondary Structure Of Fibrous And Membrane Proteins By Solid-State NMR: A Practical Approach. *Annu.Rep.NMR Spectrosc.* **36**, 79-121.