FRET-ing over CLIC1 insertion into the membrane

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Conventionally, it has been thought that proteins adopt a well defined tertiary structure. However, it is increasingly becoming apparent that some proteins can exist in at least two or more stable conformations. A striking feature of the recently described CLIC (chloride intracellular channel) protein family is that they can exist in both soluble and integral membrane forms. The crystal structures of soluble monomeric and non-covalent dimeric CLIC1 have been solved. The monomer structure showed that the CLICs are structurally homologous to the glutathione S-transferase (GST) superfamily. However, unlike other GST proteins CLICs can traverse the membrane and display ion channel activity. Soluble CLIC1 has been demonstrated to insert into the lipid bilayer to form an active channel in the absence of other cellular protein. This change of conformation must involve large-scale structural rearrangements to confer favourable interactions with the membrane by exposing hydrophobic residues. However, the manner by which CLICs insert into the bilayer and the factors controlling this process remain unclear.

In this study, fluorescence resonance energy transfer (FRET) was used to monitor the insertion of CLIC1 into the membrane. The distance between a single native tryptophan located in the putative transmembrane membrane region of the N-domain (Trp-35) and four native cysteine residues of the C-domain individually modified with the fluorescent label 1,5-IAEDANS, were measured in the presence and absence of lipid. Upon interaction with the bilayer, the distance between Trp-35 and Cys-223 increased significantly suggesting an unfolding between the C-domain and N-domain as the N-domain is inserted into the membrane. This result demonstrates that CLIC1 indeed undergoes a large conformational change upon interaction with the membrane, reinforcing the notion of the CLIC family of proteins as dynamic entities and in turn, challenging many accepted views of protein structure and ion channel biochemistry.