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Minireview Prologue

Membrane Protein Structural Biology Minireview Series*

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It is estimated that one-third of all proteins are integral membrane proteins, defined narrowly as proteins that completely traverse the membrane bilayer. There also exists a smaller but unknown number of *monotopic* membrane proteins, including prostaglandin endoperoxide H synthases, that interdigitate into a single leaflet of the lipid bilayer via a specific membrane binding domain (1). Additionally, there are acylated and/or prenylated proteins such as endothelial cell nitric oxide synthase (2) that depend on lipid modifications for their binding to membranes or membrane subdomains including lipid rafts (3) as well as proteins such as protein kinase C, cytosolic, Ca^{2+}-dependent phospholipase A_2 (4), and protein kinase B that undergo dissociable interactions with membranes via C1 and/or C2 or pleckstrin homology or FYVE domains in the context of cellular signaling.

Despite the fact that there now are over 14,000 independent protein structures in the Protein Data Bank only about two dozen of these are membrane proteins.[1] This reflects the difficulties of performing structural studies on proteins of this type. There appear to be two major classes of integral membrane proteins, the α-helical transmembrane proteins broadly distributed in cellular and organellar membranes and the β-barrel-containing porins found in the outer membranes of Gram-negative bacteria and mitochondrial and chloroplast membranes. This four-part minireview series focuses primarily on the α-helical and β-barrel proteins and intracellular proteins that can associate reversibly with membranes via C1 and C2 domains. Excellent minireviews on the related topics of lipids as protein chaperones (5), the structures of bacterial ion channels (6), and pleckstrin homology and FYVE domains (7) have also appeared recently.

The first minireview of this series entitled "How Membranes Shape Protein Structure" by Stephen H. White, Alexey S. Ladokhin, Sajith Jayasinghe, and Kalina Hristova focuses on the structure and folding of α-helical transmembrane domain-containing proteins. Two important concepts are developed. The first is that of the lipid bilayer being comprised of two chemically distinct regions of different hydrophobicities, a relatively polar membrane interface and a relatively nonpolar hydrocarbon core. Interestingly and importantly, these two membrane regions are of approximately equal thermal thickness per monolayer. The membrane interface is the region where much of membrane protein folding takes place. The second important concept is that of the overriding thermodynamic importance of the peptide bond and of the need to form hydrogen bonds between peptide bonds prior to insertion of α-helical proteins into the membrane. This latter concept is discussed in the context of a four-step model for α-helical membrane protein assembly that involves partitioning of the unfolded protein into the membrane interface, folding to an α-helical structure, insertion of the α-helix into the bilayer, and finally and as necessary, assembly of individual helices into bundles.

The second minireview of the series entitled "Structure and Assembly of β-Barrel Membrane Proteins" by Lukas K. Tamm, Ashish Arora, and Jörg H. Kleinschmidt focuses on the other major class of integral membrane proteins. In this minireview Tamm *et al.* describe the various known types of β-barrel proteins that contain 8, 12, 16, and 18 anti-parallel β strands assembled in different quaternary structures. Again, this review highlights and reinforces the importance of forming hydrogen-bonded secondary structures in order for any protein to be inserted into a bilayer membrane. The authors also summarize evidence from elegant experiments with OmpA, an eight-stranded β-barrel protein, for a four-step model for the folding and insertion of β-barrel proteins into membranes. This model is similar to that for assembly of α-helical proteins except that a significant part of the folding, following H-bond formation, occurs within the membrane as the β-barrel protein moves from an H-bonded but partially folded "molten disc" form to a mature "native" protein.

The third minireview entitled "Detergents as Tools in Membrane Biochemistry" by R. Michael Garavito and Shelagh Ferguson-Miller describes the physical and chemical properties of synthetic detergents used in solubilizing, purifying, and crystallizing membrane proteins. The authors emphasize the importance of understanding the phase behaviors of detergents when considering these reagents as preparative tools for obtaining purified membrane proteins for functional and structural studies. For example, they provide a current view of the dynamic structures of micelles and how these are influenced by salts, other polar solutes and proteins, as well as how detergents in various different physical states interact with and influence membrane protein structures. The authors also highlight examples of the specific binding of membrane lipids to proteins. This important concept in membrane protein structural biology has become apparent from recent high-resolution x-ray crystallographic studies of bacteriorhodopsin and cytochrome *c* oxidase. Specifically bound lipids may well be the rule rather than the exception, and in many instances it may be advantageous to retain these bound lipids in the presence of solubilizing detergents.

The final minireview of this series by Wonhwa Cho is entitled "Membrane Targeting by C1 and C2 Domains." C1 and C2 domains were first discovered in protein kinase C. The C1 domain is known to bind 1,2-diacylglycerol that is produced in the membrane as a product of the action of hormone-activated phospho-

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[1] blanco.biomol.uci.edu/Membrane_Proteins_xtal.html.

lipase C on phosphatidylinositol 4,5-bisphosphate. The C2 domain is a structural motif that has now been identified in a number of proteins including protein kinase C, cytosolic phospholipase A_2, and synaptotagmins. Different C2 domains can bind either two or three divalent calcium ions reversibly. Ca^{2+} binds to C2 domains when there are hormone-induced increases in the concentration of intracellular Ca^{2+}. Once Ca^{2+} becomes bound to a C2 domain, this domain binds to intracellular membranes. For example, protein kinase Cα binds to the plasma membrane and cytosolic phospholipase A_2 binds to the endoplasmic reticulum and nuclear envelope. Cho describes the properties of C1 and C2 domains and their modes of association with membranes.

The authors and editors hope that this minireview series on membrane protein structural biology will enable researchers in the biological and chemical sciences to appreciate and explore further this novel group of proteins.

REFERENCES

1. Spencer, A. G., Thuresson, E. A., Otto, J. C., Song, I., Smith, T., DeWitt, D. L., Garavito, R. M., and Smith, W. L. (1999) *J. Biol. Chem.* **274,** 32936–32942
2. Prabhakar, P., Cheng, V., and Michel, T. (2000) *J. Biol. Chem.* **275,** 19416–19421
3. Brown, D. A., and London, E. (2000) *J. Biol. Chem.* **275,** 17221–17224
4. Davletov, B., Perisic, O., and Williams, R. L. (1998) *J. Biol. Chem.* **273,** 19093–19096
5. Bogdanov, M., and Dowhan, W. (1999) *J. Biol. Chem.* **274,** 36827–36830
6. Rees, D. C., Chang, G., and Spencer, R. H. (2000) *J. Biol. Chem.* **275,** 713–716
7. Wurmser, A. E., Gary, J. D., and Emr, S. D. (1999) *J. Biol. Chem.* **274,** 9129–9132