Preface

Cell boundary and intracellular space are organized by biological membranes. The continuity and structural stability of the membranes are essential to their primary function as dividers between different environments. At the same time, countless normal and pathological processes involve topological rearrangement of the membranes consisting in merger of two lipid bilayers into one (fusion) or division of a single continuous membrane into two (fission). Research on the mechanisms of membrane fusion is vital for understanding synaptic transmission and viral diseases, fertilization and protein trafficking.

Ultimately, any fusion event boils down to rearrangement of membrane lipid bilayers and the pathway of these rearrangements appears to be conserved between disparate fusion processes. Due to intrinsic stability of lipid bilayers of conventional compositions, drastic changes in their structure require application of forces and input of energy, which are thought to be provided by dedicated proteins often referred to as fusion proteins.

Most studies suggest that fusion starts with a stage of hemifusion where the apposed leaflets of the fusing membranes merge whereas the inner leaflets remain separate. Subsequent and usually most energy demanding stages of the fusion pathway involve transition from this hemifusion intermediate to an expanding fusion pore that allows mixing of the contents of the fusing compartments. The specific mechanisms by which conformational energy released by restructuring of fusion proteins is used to drive the membrane rearrangements remain elusive. However, the major features of these mechanisms are likely to be shared by different membrane systems as suggested by a striking similarity of postfusion conformations of the fusion proteins of diverse enveloped viruses. Recent works emphasize the essential role of the bilayer elastic stresses, and, specifically, the curvature stresses, as mediators between refolding of the fusion proteins and restructuring of the lipid bilayers leading to merger of the latter. At the same time, along with elucidation of the shared properties of fusion processes, it is equally important to uncover their specific features in diverse biological systems characterized by dramatic variations of the regulatory mechanisms, rates, and compositions of the membranes.

This volume brings together reviews on different aspects of membrane fusion. The leading experts present both the broad picture of corresponding research fields and the in-depth discussion of the most recent developments.

A section on fusion and fission of lipid bilayers is followed by three sections discussing different families of processes involving fusion of biological membranes: fusion in viral infections, intracellular fusion, and cell-to-cell fusion in development. The last section on theoretical modeling reviews some numerical approaches used to simulate the fusion reaction and to understand the underlying physics.

The first section presents two chapters that discuss mechanisms of remodeling of lipid bilayers in fusion (J. Nikolaus et al.) and fission (T. J. Pucadyil). J. Nikolaus et al. concentrate on the pathway of lipid bilayer restructuring in fusion with emphasis on the hemifusion stage. They describe a recent direct observation of formation of an extended hemifusion diaphragm between giant lipid vesicles and discuss theoretical approaches providing insight into energetics and kinetics of hemifusion. T. J. Pucadyil focuses on a specific protein, dynamin 1, which drives bilayer fission. In addition to an overview of structural and biochemical properties of dynamin, he discusses a recently developed experimental system, which, for the first time, made possible investigation of the dynamin-driven fission of tension-free membranes under most natural conditions of constant GTP turnover.

We then proceed to three chapters covering fusion mediated by viral proteins. Most viruses including very important human pathogens deliver their genetic material into cells by fusing viral envelope to cell membranes (different enveloped viruses such as vesicular stomatitis virus and HIV discussed in A. Albertini et al. and G. Melikyan, respectively) or fusing cell membranes with each other (nonenveloped viruses, J. Boutilier and R. Duncan). The review by A. Albertini et al. focuses on the pre- and postfusion structures of viral fusion proteins and ultrastructural analysis of viral particles that suggests that fusion requires the cooperative action of a large number of fusion proteins. G. Melikyan explores the distinct stages of viral entry and fusion elucidated at the level of individual virions and describes his recent paradigm-shifting finding that HIV fuses with endosomal membrane rather than with cell's plasma membrane. J. Boutilier and R. Duncan discuss a unique family of fusion proteins of nonenveloped viruses strikingly different from other well-studied fusion proteins and thus reminding us that our current models of how proteins fuse membranes will have to be revised and extended.

The intracellular fusion section of the book focuses on exocytosis, the process of cellular secretion in which substances contained in intracellular vesicles are discharged from the cell. S. Martens and H. McMahon discuss the protein machinery that drives calcium-dependent exocytosis in chromaffin cells and neurons with the special emphasis on the mechanistic role of C2 domain-containing proteins that serve as calcium sensors. M. B. Jackson reviews the electrophysiological data on exocytosis of norepinephrine from chromaffin cells and comes to the conclusion that SNARE-mediated fusion

can proceed by a pathway that starts with an opening of a gap-junction-like proteinaceous fusion pore rather than with hemifusion. The chapter by T.-Y. Yoon et al. reviews recent studies of the molecular mechanisms of SNARE-dependent fusion explored using proteoliposomes. The chapter focuses on the small molecule inhibitors of SNARE blockers and state-of-the-art assays for dissection of fusion pathway for a single vesicle fusion event.

Cell-to-cell fusion in development is represented in the volume by two reviews. O. Avinoam and B. Podbilewicz discuss two families of eukaryotic cell–cell fusogens: fusion failure proteins identified in *Caenorhabditis elegans* and syncytins derived from proviruses in mammals. The authors analyze the similarities and distinctions between these cell fusogens and the experimental approaches used to identify and characterize them. E. H. Chen's chapter concentrates on fusion between myoblasts and discusses invasive actin-supported podosome-like structures that initiate this fusion process, essential for the development and repair of skeletal muscles. A detailed view of the fusion site at the ultrastructural level will undoubtedly stimulate significant new insights into fusion mechanism.

Overviews of numerical modeling of membrane fusion are given in two chapters. A. J. Markvoot and S. J. Marrink review the whole range of computational methods used for simulation of membrane fusion and fission. Interestingly, while some of the computations substantiate the hemifusion stalk to fusion pore pathway of fusion suggested by phenomenological theories and supported in experiments on fusion of black lipid membranes and giant lipid vesicles (see the review by J. Nikolaus et al.), other simulations predict alternative scenarios of lipid rearrangement which await their experimental verification. M. Müeller and M. Schick describe the results of a specific computational method where lipid molecules are modeled as amphiphilic block copolymers and water as a hydrophilic homopolymer. This method identifies one of the alternative fusion pathways and the related field-theoretical analysis suggests energies of the structures emerging at the intermediate stages of the fusion reaction.

In general, we believe the book both illustrates the current knowledge on membrane fusion and emphasizes that many important aspects of this exciting research field remain to be understood. For many fusion processes the proteins that mediate membrane rearrangements are yet unidentified. Even for the best characterized fusion proteins we still need to figure out the specific mechanisms by which they initiate and drive lipid bilayer restructuring. The pathway of this restructuring is also an area of active research as illustrated by conflicting data on what comes first in exocytotic fusion, hemifusion, or fusion pore (T.-Y. Yoon et al. vs. M. B. Jackson). Another example is the discrepancy between the currently available experimental data on the hemifusion structures

and phenomenological theories of the fusion pathway on one hand (reviewed in J. Nikolaus et al.) and alternative pathways of lipid rearrangements uncovered in the computer simulations on the other (M. Müeller and M. Schick and A. J. Markvoort and S. J. Marrink). Only future work will clarify the universality and biological relevance of the specific fusion motifs discussed in this book.

Leonid V. Chernomordik and Michael M. Kozlov