Our perception of biological membranes and the role of lipids in membrane-related events has been changed substantially in recent years. Lipid molecules are not treated as passive structural elements anymore, but some of them appeared to be important signaling molecules. Some of the most intriguing molecules from the latter group are phosphatidic acid (PA) and ceramide-1-phosphate (C1P). Both are unusual membrane lipids mostly due to the cone-shape of their molecules, which causes deformation of membranes enriched with these lipids, and their negatively-charged headgroups, the protonation state of which may depend on pH at physiologically-relevant range. Although very similar in structure, each of the two lipids may play different or even opposite roles in particular physiological processes. Moreover the array of cellular pathways, in which each of the two lipids is involved is very broad, suggesting that regulation of their biological activity must be precise. This raises the major question on the specific recognition of each of the lipids in the context of a biological membrane by effector proteins. Remarkably, both PA and C1P exist within a cell as discrete subpopulations of molecules that differ in terms of length and amount of double bonds in their acyl chains. The key question is to what extent subtle differences in structure of backbone of C1P (sphingolipid) and PA (glycerophospholipid) and acyl chains within subpopulations of each of the lipids would affect their behavior in biological membranes and interactions with effector proteins? Such differences may reflect variable biological functions of these lipids. Going further, could each of the two lipids, together with their effector proteins, be considered as pH sensitive switches in mammalian cells? Does the recognition of PA and C1P by proteins depend on other potent signaling molecules, such as calcium ions, which would gain another level of cross-talk between different signaling pathways?

These important questions became fundamental to us while designing the current project. To address them we will employ membrane model systems, including lipid monolayers at water-air interface and lipid vesicles of different size. Such approach will enable us to strictly control key parameters that are important for understanding molecular basis of modulatory mechanisms that govern signaling lipid recognition. We will study lateral interactions of PA and C1P with cholesterol. The latter is the major modulator of physical properties of cellular membranes and plays pivotal role in the formation of membrane nanodomains (lipid rafts), which are considered as structural and functional platforms of living cells. Studying how proteins can selectively recognize PA and/or C1P in the context of lipid membrane of different composition and in different conditions will help us to elucidate whether the differences in behavior of the two lipids and their structural subspecies can be directly sensed by executory elements of cellular machinery. We should also get further information about the influence of membrane thickness and lipid packing on these cellular phenomena.

Our understanding of lipid-mediated signaling are currently limited. Execution of the proposed project will help to fill this gap by providing detailed description of mechanisms that govern molecular recognition. Phosphatidic acid and ceramide-1-phosphate both belong to a group of crucial regulators of cell functions. Due to very similar molecular structure a potential cross-talk between the two lipids may occur within a cell. Thus, decoding the mechanisms by which both lipids are selectively recognized by effector proteins are essential to understand cellular signaling pathways and consider additional levels of their regulation. Furthermore, this knowledge may help in designing innovative therapeutic strategies in the future.