

Dottorando: Dott.ssa Federica Campana

Titolo del progetto: “Molecular dynamics investigations of drug-cell membrane interactions”.

The cell membrane functions as a platform for the assembly of many signal transduction pathways and provides an additional level of regulation in cell signaling networks.

The complex dynamic structure of the plasma membrane allows lipid–lipid and lipid–protein interactions, as well as the interaction of lipid–protein complexes with the submembrane cytoskeleton. The existence of membrane microdomains adds further complexity to such interactions, as well as the messages propagated in cells through G proteins and other non-permanent (extrinsic) membrane proteins.

Each G protein can simultaneously bear a myristoyl, palmitoyl and isoprenyl moiety and therefore, many lipid molecules associated with G proteins may arise in G protein coupled receptor (GPCR)-rich membrane microdomains. These lipids can regulate the biophysical properties of membranes, which in turn modulate the interaction and activity of G proteins. The aim of the first part of my work was to understand the effect of these moieties on membrane structure and G protein-membrane interactions. Although recent studies found that the G $\beta\gamma$ dimer drives the interaction of G-proteins with nonlamellar-prone membranes, little is known about the molecular basis of this interaction. For this reason, I also investigated the interaction of the C-terminus of the G γ protein with model membranes with or without the isoprenyl moieties.

From the very beginning of my work, my studies have focused on the interaction of saturated and unsaturated fatty acids with cell membranes. Many of these molecules, in fact, have proven to be active against certain types of cancers and other diseases. Among all these molecules it is important to remember the Minerval, an analogue of oleic acid developed in the laboratory of Prof. Escribà, which has led to a reduction of up to 80% in the development of lung cancer and glioma cells.

A promising molecule with high anti-inflammatory activity and synthesized by the group of Prof. Escribà like the previous one, is a modification of arachidonic acid: the 2-hydroxy arachidonic acid (AAOH). Due to the similarity with arachidonic acid (AA), that is the natural substrate of cyclooxygenase (COX), the second part of my work consisted in the investigation of the interaction of AAOH with COX-1 and COX-2. The results, in terms of free energy of binding and the Fukui function, demonstrated the potential of AAOH as non-toxic non-steroidal anti-inflammatory drug (NSAID).

Recent findings pointed unambiguously to membranes as additional cellular sensors in activating a stress protein response, from prokaryotes to mammalian cells, at the beginning of temperature rise or other stresses. Aging or pathophysiological conditions can also be linked to the development of subtle membrane changes or “membrane-defects”, responsible for a dysregulated expression of heat shock proteins (HSPs).

Chaperone co-inducers, among which can be mentioned the hydroxylamines such as BGP-15 and NG-094, are substances that cannot induce HSPs by itself, but can enhance HSP induction in combination with other mild stresses. A chaperone co-inducer also has the ability to lower the temperature threshold of the heat shock response and may provide suitable therapeutic candidates for many disease states since they are capable of affecting stressed rather than unstressed cells.

The third part of my work consisted in molecular dynamics investigations of BGP-15 and NG-094 on membranes made of sphingomyelin and cholesterol at different composition to understand some aspects of membrane functioning. BGP-15 and NG-094 can induce an alteration in membrane’s fluidity similar to those induced by heat and a strong reorganization of sphingomyelin headgroups with an increased penetration of water. Taken together, all the results suggested that hydroxylamines have a strong effect on microdomains reorganization showing a great potential to become a new class of pharmaceuticals to combat most various protein-misfolding diseases and aging.

All these investigations are relevant in the context of the physiology of cells, whose alterations may lead to pathologies whose treatment could be addressed by modifying membrane lipid composition and structure through so-called “Membrane-lipid therapy”.