Cell-free expression and characterization of membrane proteins for drug development

Sandra CORTES
Synthelis, Grenoble - France

More than 60 percent of the medicines in use today are directed against cell membrane proteins. Among these membrane proteins, G protein-coupled receptors (GPCRs) **are a major family of drug targets.** Expression and biophysical studies of GPCRs are very challenging. Indeed the production of recombinant membrane proteins by classical expression systems presents some limiting features on several aspects: difficulty to express functional membrane proteins in a reasonable amount for structural studies or functional applications. A very interesting and attractive alternative is the use of cell-free transcription/translation systems. Synthelis has developed and optimized a new membrane protein optimized cell-free expression system to provide several milligrams of functional GPCRs such as CxCR4. The tremendous advantage of this system is the production of recombinant proteoliposomes in a one-step reaction which can be easily scaled-up for industrial applications. This format allows providing the right environment for the expression of membrane proteins while preserving their structural and functional integrity.

We will present different examples including one related to natural ligands binding to the CxCR4 proteoliposomes using Horiba Scientific SPRi platform and Cisbio Bioassays Tag-lite® technology. These data provide evidence that the GPCR maintains native binding properties. We have also demonstrated that the SPRi technology was a biophysical method adapted to the study of GPCRs proteoliposome and confirm the activity of the receptors.

This approach has been successfully used to express all the main membrane target families (GPCRs, Ion Channels, Transporters, Enzymes,…) on which functionality has been assessed using various functional validations such as patch-clamp for ion channel, enzymatic assays for membrane enzymes or binding assays for GPCRs.