Implementation and application of a fragment-based screening approach to find new human cyclophilins inhibitors.

Colliandre Lionel¹, Ahmed-Belkacem Abdelhakim², Hurard Corinne¹, Roumestand Christian¹, Douguet Dominique, Bourguet William¹, Labesse Gilles¹, Pawlotsly Jean-Michel², Guichou Jean-François¹

¹) Centre de Biochimie Structurale, UM1, UM2, CNRS, Inserm, 34090 Montpellier;
²) IMRB, Inserm U955, Equipe 18, Hôpital Henri Mondor, 94010 Créteil

Cyclophilins (Cyp's) have a peptidyl-prolyl cis-trans isomerase activity. They help protein folding. It was shown that development of Cyps inhibitors could lead to new treatments against HIV, HCV, cancers or Alzheimer diseases for example.

We focused on the anti-HCV application. Because of the big genetic variability of this virus, resistances can appear rapidly during treatment. Targeting cellular factors lead to less resistances development, inhibition of cellular factors like Cyps is a complementary strategy for the actual therapy against this virus.

To design new Cyps inhibitors with low molecular mass, we applied a fragment-based screening approach on Cyclophilin D (CypD). We used X-ray crystallography and NMR that are well adapted to identify weak affinity fragments (mM).

After creation of a library of 330 fragments, we screened on CypD 19 fragments by NMR and 57 fragments by X-ray crystallography. We solved 14 crystallographic structures of CypD in complex with fragments (2,00 - 0,97 Å). All these fragments have affinity in the millimolar range on CypD.

Based on the fragments binding modes, we designed and optimized a new Cyps inhibitors family. Our lead compound have an IC₅₀ of 6,2µM on CypD in vitro and an EC₅₀ of 12,3µM for the HCV replication in cellulo. The crystallographic complex of CypD with this lead compound has been solved (1,92Å). A patent has been deposited on this compounds family.

The poster will present the combination of many techniques (chemoinformatics, X-ray crystallography, NMR, calorimetry...) to implement the fragment-based approach. It will be focused on the rational design of new human Cyps inhibitors.