**Progetto e Piano formativo per un assegno di ricerca dal titolo: “Ribonuclease targeting chimeras (RIBOTACs): targeting RNA structures with small molecules” – Tutor: Prof.ssa Maria Laura Bolognesi**

This project is part of the “CN3 - National Center for Gene Therapy and Drugs based on RNA Technology”, Principal Investigator: Prof. Maria Laura Bolognesi.

This project aims to develop completely innovative therapeutic tools directed to RNA as a pharmacological target. Different classes of RNAs, including messenger RNAs (mRNAs), microRNAs (miRNAs), and long noncoding RNAs, are implicated in various diseases and are thus targets for new drugs.1 Currently, antisense oligonucleotides (ASO) and small interfering RNAs (siRNA) represent the main pharmacological approaches to modulate RNAs. Although some of these drugs have recently been approved (e.g., Patisiran for hereditary transthyretin storage amyloidosis and Nusinersen for spinal muscular atrophy), they have several limitations, such as poor cellular uptake, low tissue specificity, and, in some cases, toxicity.2 Therefore, it seems promising to develop novel small molecule-based approaches that can modulate RNAs by inducing their degradation, without the limitations of oligonucleotide-based therapies.

A new strategy for RNA degradation is represented by RIBOTACs (ribonuclease targeting chimeras).3 RIBOTACs are a novel class of bifunctional small molecules that simultaneously bind an RNA of interest (ROI) - specifically RNAs that form intricate secondary and tertiary structures - and RNase L, a latent ribonuclease. The ligand recruiting the RNase causes its dimerization and therefore its activation. Finally, the formation of a ternary complex between ROI-RIBOTAC-RNase L induces the consequent selective degradation of the ROI.

Within the "CN3 - National Center for Gene Therapy and Drugs based on RNA Technology" project, the University of Bologna Unit is responsible for the development of RIBOTACs directed to RNAs, on which the national collaborators already have experience.

The research fellow will be involved in the development of RIBOTACs. The first step will be to identify RNA-binding molecules and then convert them into RNA-degrading molecules. Starting from fragments known to bind RNA and/or for which 3D information are available, the attachment sites on the ligand suitable for binding the RNase L recruiting portion will be identified. Modifications will also be made to optimize the recognition of the ligand with the ROI. In parallel, the fragments that interact with RNase L will be synthesized. Through cycles of medicinal chemistry, a set of RIBOTACs will be synthesized by combining these two fragments through suitable linkers.

The design and synthesis of the molecules will be followed by assays to evaluate: (a) their ability to bind both to RNase L and to ROI; (b) the degradation of ROI; (c) their therapeutic potential, performed in collaboration.

It is thus necessary to activate a research grant for the specific activities described above, as already planned in the project itself.

*References*

1. M. Matsui, D.R. Corey. Non-coding RNAs as drug targets. Nat. Rev. Drug Discov., 16 (2017), pp. 167-179.
2. R.L. Setten, J.J. Rossi, S.P. Han. The current state and future directions of RNAi-based therapeutics. Nat. Rev. Drug Discov., 18 (2019), pp. 421-446.
3. M.G. Costales, B. Suresh, K. Vishnu, M.D. Disney. Targeted Degradation of a Hypoxia-Associated Non-coding RNA Enhances the Selectivity of a Small Molecule Interacting with RNA. Cell Chem. Biol., 26 (2019), pp. 1180-1186.