MS06-1-10 Structural exploration of FemX interaction with tXNA conjugates : identification of one potential antibiotic
#MS06-1-10

I. Li De La Sierra Gallay [1], L. Rietmeyer [2], G. Schepers [3], D. Dorchêne [2], L. Iannazzo [4], D. Patin [1], T. Touzé [1], H. Van Tilbeurgh [1], P. Herdewijn [3], M. Ethève-Quelquejeu [4], M. Fonvielle [2]
[1]I2BC, UMR9198 - Gif-sur-Yvette (France), [2]CRC, UMRS1138 - Paris (France), [3]LMC - Leuven (Belgium), [4]LCBPT, UMR8601 - Paris (France)

Abstract
FemX is a non-ribosomal aminoacyl transferase. This enzyme transfers an alanyl residue from Ala-tRNAAla to the L-Lys side chain in the peptidoglycan precursor UDP-N-acetyl-muramyl-pentapeptide, a crucial reaction for the production of bacterial cell walls. This reaction is the first step of synthesis of the L-Ala-L-Ser-L-Ala side chain, which is completed by other Fem transferases. Because of their key role in the peptidoglycan metabolism, Fem transferases are considered as attractive targets for the development of novel antibiotics.
We have previously analyzed the interaction of several in vitro synthetized peptidyl-RNA conjugates and solved the X-ray structure of one peptidyl-RNA conjugate in complex with FemXWv of Weissella viridescens, a model enzyme of Fem family (1). We are now interesting on the study of Xenobiotic Nucleic Acids (XNA) analogs as substrates or inhibitors of FemX. XNAs, are characterized by replacement of the ribose (or deoxy-ribose) by a non-natural sugar, to create a nucleic acid that usually possesses unaltered Watson-Crick (or other) base-pairing properties and a similar 3D structure when compared to its natural DNA or RNA analogue.
In the present work we explored the impact of the XNA modification in aminoacyl-tRNA (aa-tRNA) analogs. We synthetized and characterized a subset L-Ala-tXNA microhelices and solved the Xray-structure of four complexes of FemXWv / tXNA conjugates. Where the tRNA in the tXNA conjugates is replaced with an 8nt XNA microhelix, the lysine-side chain branch is replaced with an artificial 1,4-triazole linkage and the sugar is replaced with : six-carbon sugars (HNA), 2'-F ribose (2'F-RNA), 2'-F arabinose (2'-FANA) and deoxyribonucleic acids (DNA). The combination of structural and enzymatic information allowed us to shed some light on the chemical groups implicated in the FemX inhibition at the molecular level. HNA and 2'F-RNA aminoacylated substrates have essentially unaltered catalytic and structural properties relative to L-Ala-tRNA microhelix controls. DNA and 2'F-ANA, conversely, abolish catalysis and substrate binding. These are all shown to be FemX inhibitors, with the 2'F-RNA being the most potent. A stabilizing interaction with one of the F atoms is thought to account for the high affinity in the case 2'F-RNA compound.

References
(1) Fonvielle M e*t al.* Angew. Chem. Int. Ed 2013 Jun 6. doi: 10.1002/anie.201301411