Discovery of new anti-TB compounds that target *Mycobacterial* FtsZ: high-throughput screening and molecular docking

Karpov P.A.*, Demchuyk O.M., Blume Ya.B.

*Institute of Food Biotechnology and Genomics NAS of Ukraine, 04123, Osipovskogo str., 2a, Kiev-123, Ukraine, karpov.p.a@gmail.com**

Britsun V.M., Volochnyk D.M.[#]

Institute of organic chemistry. 02660, Murmanska, 5, Kyiv, Ukraine. d.volochnyk@gmail.com[#]

Tuberculosis (TB) is one of the most common infectious diseases worldwide. According to the World Health Organization, in 2011 there were 8.7 million new TB cases and 1.4 million deaths were caused by *Mycobacterium tuberculosis*.[1, 2]. Filamentous temperature-sensitive protein Z (FtsZ), a bacterial tubulin homologue, is an essential cell-division protein that polymerizes in a GTP-dependent manner, forming a highly dynamic cytokinetic ring, designated as the Z-ring, at the septum site. Because of the requirement of FtsZ in mycobacterial (MB) cytokinesis, inhibition of FtsZ is a promising target for anti-tuberculosis drug discovery. [3]

Our present study is the joint project of IFBG NAS of Ukraine and the IOC NAS of Ukraine. First of all, we selected template Protein Data Bank (http://www.rcsb.org) structures and identify 1RQ7 structure of *M. tuberculosis* FtsZ (2.60 Å, R-Value = 0.187; R-Free = 0.242) as the most suitable for further homology modeling, high-throughput screening (HTS) and molecular docking. Also, it was built full-atomic protein model. Based on chemical library of heterocyclic compounds that were synthesized in IOC, we created virtual librarys (2286 compounds) suitable for HTS and molecular docking (*.sd, *.mol and *.mol2 formats). This database was applied to identify potential FtsZ-targeted anti-TB agents. Based on probable protein-ligand complexes predicted by I-TASSER (http://zhanglab.ccmb.med.umich.edu/I-TASSER/), as well as based on homology to known sites of different tubulin binders, we select perspective protein-ligand binding sites (besides well-characterized GTP-binding site). The reference ligand method provide us information enables to select four most perspective ligand-binding sites and built their models for HTC

and molecular docking: 1) GTP/GSP, 2) TZT, 3) HOS/T131 and 4) Taxol. The following HTS of virtual ligand library was performed by FlexX docking-algorithm, and was performed against all binding site models. Experimentally proved trisubstituted benzimidazoles design by Ojima Research Group (http://www.stonybrook.edu/commcms/ojima_group/): 1a-G4, 1a-G7, 2a-1, 2b-1, 1b-G1 and 1b-G2, were used as positive control. [3]

Based on predicted free energy of binding (ΔG), Ligand Efficiency (LE) and total "Score", 26 hits were selected among 2886 candidate compounds. New potential inhibitors of *M. tuberculosis* FtsZ were selected, and certain relation of binding site and heterocyclic core of compound we noted. In general, selected compounds have one of seven types of heterocyclic core: benzimidazole, indoline, 1,3-benzoxazole, 1,3-benzothiazole, 1,3-benzodioxole, 5,6-dihydro-[1,2,4]triazolo[5,1-b][1,3]thiazin-7-one and 4,5,6,7-tetrahydro-1-benzothiophene. To evaluate FtsZ binding *in vitro*, selected compounds were submitted to photometric testing. Final conclusions will be made based on ability of selected compounds to inhibit *FtsZ* assembly *in vivo.*

Acknowledgements: This *research was supported by* NAS of Ukraine Complex Targeted Basic Research Program grant: "Basic problems of design of new substances and materials for chemical production" for 2012-2016.

1. World Health Organization (2012) Global tuberculosis report 2012. *http://apps.who.int/iris/bitstream/10665/75938/1/9789241564502_eng.pdf*.
2. K. Kumar, et al. (2010) Discovery of anti-TB agents that target the cell-division protein FtsZ, *Future Med Chem*, **2 (8)**: 1305-1323.
3. K. Kumar, et al. (2011) Novel trisubstituted benzimidazoles, targeting Mtb FtsZ, as a new class of antitubercular agents, *J Med Chem*, **54 (1)**: 374-381.