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ORAL ABSTRACT PRESENTATIONS

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Abstract OR-4: New Antibiotic Binding Site on the 30S Ribosomal Subunit

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Background: Antibiotic resistance becomes one of the main problems of modern medicine; therefore, the development of new antibacterial compounds is absolutely necessary. The ribosome is the target for a lot of different antibiotics; there are several main binding sites on the ribosome – decoding center, peptidyl-transferase center, and ribosome exit tunnel. Modification or mutation of nucleotides in these sites could make cells resistant to structurally different antibiotics.
Methods: pDualrep2 reporter system was used for detection of the protein synthesis inhibitors in cultural broths of new soil bacteria. By means of a cell-free translation system, the inhibitory activity and mechanism of action of Auraplanin were estimated. CryoEM data collection was performed on a Titan Krios operated at 300 kV, equipped with a Falcon II direct electron detector.
Results: In this work, we have found a new inhibitor of protein synthesis, which binds in a completely new binding site. This compound is produced by *Actinoplanes sp.* VKM Ac-2862 and by Cryo-EM study of its complex with *E.coli* ribosome, it was shown, that it binds close to 560 loop of 30S ribosomal subunit. The new compound is a derivative of tetramic acid and we called it Auraplanin, because of bright orange color of the producer strain.

Structural data are in good agreement with genetic results – resistant mutations were located close determined binding site. Substitutions C564G, G558U, and G566A significantly increase minimal inhibitory concentration, all these mutations were not detected previously. We also observed resistant mutation in ribosomal protein S4, this mutation was previously identified as error-prone. Interestingly, ribosomal ambiguity mutations, G299A and G347U, also increased resistance to Auraplanin.

Conclusion: On the basis of the genetic, structural and biochemical studies we hypothesized that Auraplanin acts prevent the transfer from an open to a closed conformation of 30S subunit, in contrast to streptomycin, which promotes the formation of a closed state.

Key Words: cryo-ET

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