Combining growth inhibition assays and metabolic profiling to accelerate discovery of antibiotics with unconventional modes of action

Sebastián Sosa-Carrillo, Léo Gerlin and Mattia Zampieri

Department of Biomedicine, University of Basel, Mattenstrasse 28, 4058 Basel, Switzerland

Antibiotic resistance is emerging as a major challenge for treatment of the most common and widespread infectious diseases. Moreover, classical *in vitro* susceptibility assays are limited for discovery of new antibiotics because of their restricted capabilities to reveal new modes of action. A key limitation of *in* vitro methods is that they are not able to mimic the conditions faced by the pathogen during *in vivo* infection, where nutrient availability and stresses could significantly impact bacterial physiology. Thus, such disparity in metabolic states may lead to overlook compounds targeting non-essential processes *in vitro*, which can turn out to be attractive candidates for *in vivo* treatment. Along these lines, the discovery of new compounds having *in vivo* antimicrobial activity can be accelerated by approaches that combine standard growth inhibition assays, molecular profiling and computational methods. To this end, we propose to combine growth susceptibility assays with high-throughput metabolomics to identify antimicrobial compounds with unconventional modes of action. By monitoring growth and metabolic effects of known antibiotics, as well as of other chemically-diverse compounds, in combination with common metabolic restrictions and stresses faced during infection, we aim at predicting small molecules efficacy across different conditions. Building on these notions, our approach serves as a proof-of-concept to develop strategies to more efficiently screen compound libraries and identify new and non-obvious molecules with original antimicrobial modes of action.