Title: A novel antibiofilm compound can improve the efficacy of rifampicin against Mycobacterium biofilms in vitro

Authors:
Emilia Lintilä, Jayendrakumar Patel, Jari Yli-Kauhaluoma, Mataleena Parikka, Ashok Aspatwar

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Abstract
Tuberculosis treatment lasts a minimum of six months and comprises heavy use of antibiotics. This leads to various problems such as toxic side effects, and development of antibiotic resistance. During infection, *Mycobacterium tuberculosis* forms biofilms that contain heterogeneous bacteria population with different tolerance rates. Thus, this work was a part of DisRUPT consortium project that aims to find biofilm-disrupting compounds that could also sensitize the tolerant sub-populations to conventional antibiotics. The 25 compounds tested in this screen were designed so that they have chemical structures known to be involved in the inhibition of quorum sensing, a common bacterial communication strategy. Transgenic, bioluminescence-producing *Mycobacterium marinum* bacteria were grown into biofilms in vitro and exposed to the compounds. *M. marinum* was used because it is a BSL-2 pathogen and closely related to *M. tuberculosis*. Change in the bioluminescence was used as a measure of drug efficacy. CFU (colony forming unit) plating was additionally used to ensure the bactericidal efficacy of the most potential hits. Toxicity of the compounds was tested with an acute embryonic toxicity test using zebrafish embryos. Together with rifampicin, one compound, namely JZP-AoF-249F, inhibited the growth of 99% of the *M. marinum* population in the highest safe concentration, 40 µM, twice as fast as rifampicin alone. The combination treatment was also statistically significantly more efficient than rifampicin alone in concentrations of 10 µM-160 µM. Altogether, this screen provided important information on compounds that improve the in vitro efficacy of rifampicin without causing toxic effects on zebrafish embryos.