Metabolism-guided optimization of next-generation benzothiazinones towards highly potent antituberculosis agents.

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With over 1.5 million deaths per year, tuberculosis is still among the most fatal infectious diseases. With the dissemination of multi-drug (MDR) and extensively drug-resistant (XDR) strains, there is an increasing need for new antibiotics with novel mode of action. Recently entered clinical phase IIb**,** BTZ-043 is a highly potent benzothiazinone (BTZ) acting as a covalent inhibitor of decaprenylphosphoryl-β-D-ribose 2′-epimerase (DprE1), an essential enzyme for cell wall biosynthesis in *Mycobacterium tuberculosis*.

The discovery of an unprecedented metabolic main pathway towards hydride Meisenheimer complexes (HMC) during preclinical development of BTZ-043 prompted in-depth re-evaluation of the lead-optimization strategy and testing cascade towards next generations of this promising antibiotic. As this metabolic pathway was not represented within standard *in vitro* pharmacokinetic (PK) assays, we developed a new whole-cell assay to screen BTZ derivatives for MHC metabolite formation to efficiently guide medicinal chemistry efforts.

Our initial diversification campaign relied on the late-stage functionalization of the BTZ scaffold, i.e. 5- and 7- substitutions and expansion of the aromatic core towards benzofuran- and naphthalene-fused thiazinones. 5-methylated BTZs were the most preferred scaffolds which demonstrated a reduced HMC formation combined with potent activity, good microsomal stability and retention of the mode of DprE1 inhibition. The lead compound HKI12134085 showed a particularly favorable profile, i.e. potency against susceptible and multi-drug resistant *M. tuberculosis* strains combined with decreased HMC formation and nitro reduction. *In vivo* experiments revealed good systemic exposure upon oral administration and efficacy in a murine *M. tuberculosis* infection model.

References

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