Identification of novel antibiotic targets using covalent inhibitors and residue-specific proteomics

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Background
The antibiotic crisis[1]
• resistances to all marketed antibiotics
• increasing levels of multi-resistant strains
• declining effectiveness of antibiotic treatment
• antibiotics only target a limited set of pathways
• innovation gap in antibiotic development

Covalent inhibitors[2]
• prevalent as antibiotics (e.g. β-lactams)
• increased selectivity and potency
• potentially less prone to resistance development

Our approach: identifying new druggable binding sites

Competitive residue-specific proteomics[3,4]

• target identification for unmodified covalent inhibitors
• identification of the exact binding site

Compared to traditional TEV tags:
• easier synthesis
• shorter workflow
• higher coverage in bacteria
• compatible with non-trypsin digestion

isoDTB tags[4] – smaller, better, faster

• global profiling of ligandable sites with fragments
• quantification of occupancy and affinity

Cysteine profiling in bacteria[4]

• cysteine reactivity correlates with functionality
• combination of phenotypic screening and isoDTB-ABPP to identify new antibacterial targets and starting points for inhibitor development

Beyond cysteine
Aspartates and glutamates[5]

Lysines