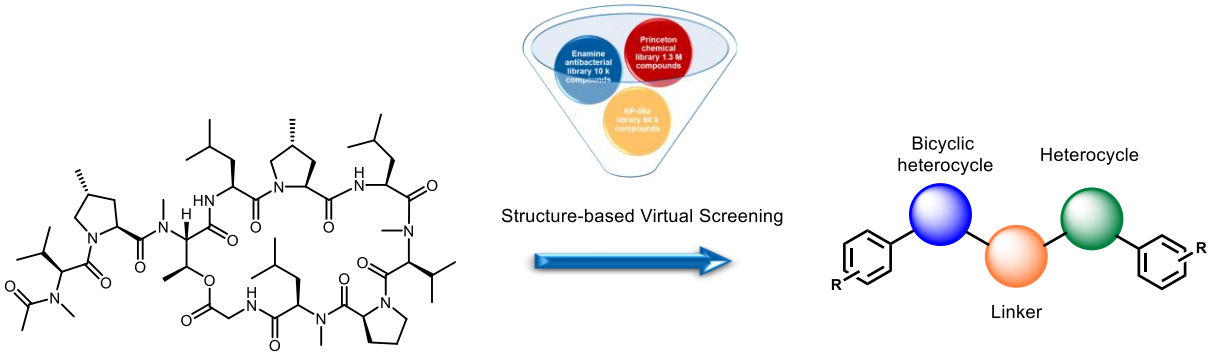
Targeting the Bacterial Sliding Clamp for the Treatment of Gram-Positive Bacterial Infections

The persistent rise of antibiotic resistance has created an urgent need for novel anti-infective agents. As current treatments increasingly fail, the search for innovative therapeutic targets becomes more critical. In this context, the bacterial β-sliding clamp (DnaN) represents a promising target for antibiotic discovery. DnaN is essential for DNA replication, highly conserved in bacteria, but structurally distinct from its eukaryotic counterpart, and its inhibition is associated with a very low frequency of resistance.[1,2]



Griselimycin
MIC (*M. tuberculosis*) = 1 µg/mL

Novel DnaN Inhibitors
MIC (*S. aureus*) = 1-4 µg/mL
MIC (*M. tuberculosis*) = 1-4 µg/mL

The natural product Griselimycin (GM) was identified as a DnaN-inhibitor with antimycobacterial activity. Through a structure-based virtual screening using a universal pharmacophore filter, we identified a small-molecule series with potent in vitro activity against priority ESKAPE pathogens and a broadened antimicrobial spectrum compared to those of GM and known inhibitors.[1,2] To confirm target engagement, we employed surface plasmon resonance (SPR), tested efficacy in an in vitro DNA replication assay, determined the crystal structure in complex with DnaN, and performed a MIC-shift assay using a DnaN-overexpressing mycobacterial strain. Moreover, the hit compound displays bactericidal activity, no-cross resistance with clinically used antibiotics, and no significant cytotoxicity.

Encouraged by these results, we proceeded with hit-to-lead optimization of this class towards an orally active antibiotic for treatment of Gram-positive bacterial infections, supported by CARB-X. Extensive structure–activity relationship (SAR) investigations, guided by structure-based design, were implemented to enhance the DnaN binding and antibacterial efficacy of the compounds, while optimizing their physicochemical and pharmacokinetic properties.

The frontrunner analogues demonstrate target engagement and potent antibacterial activities (MIC = 1-4 µg/mL) against *S. aureus* and other Gram-positive species. This work suggests that our DnaN inhibitors can be further developed as potential antibacterial agents and a valuable contribution to the fight against antimicrobial resistance.

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References

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