Unveiling Molecular Interactions: Probing Trehalose-Glycolipids in Mycobacterium through Bioorthogonal Chemistry

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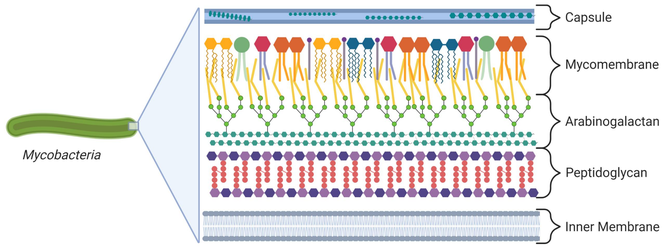
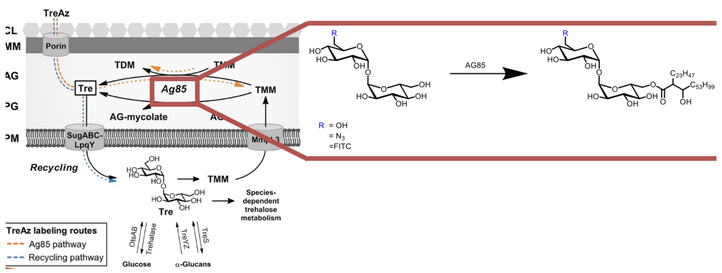
Abstract

Trehalose-modified glycolipids play a fundamental role in Mycobacterium physiology, offering avenues for metabolic engineering and chemical probing. This study proposes a bioorthogonal approach to engineer these glycolipids, integrating click chemistry techniques, with the aim of deciphering their molecular interactions within Mycobacterium systems. Initially, trehalose is engineered to possess a native-like structure for seamless incorporation into the bacterial system, while also being equipped with a versatile functional group that serves as a signature traceable moiety. This modification not only facilitates its integration within the cell but also enables further downstream chemical modifications, providing a unique and identifiable signature for tracking its fate. Leveraging retro-Cope elimination, chemical probes are seamlessly incorporated into mycobacterium, enhancing our ability to study its interactions within the cell. Upon incorporation into the bacterial system, the modified trehalose-glycolipids interact with cellular components, facilitating the formation of complex molecular networks. Utilizing bioorthogonal click reactions targeting the alkyne moiety on the probe, we pull down the interacting complexes. Subsequent cleavage using bisboron chemistry allows for the selective release of modified glycolipids for downstream analysis. Finally, through advanced analytical techniques such as mass spectrometry or mass spectrometry imaging (MASSTOF), we meticulously study the precise locations and nature of molecular interactions, unraveling the underlying mechanisms governing trehalose-glycolipid crosslinks and interactions in Mycobacterium. This integrative approach promises to advance our understanding of bacterial physiology and may yield valuable insights for the development of novel therapeutic strategies.

Introduction



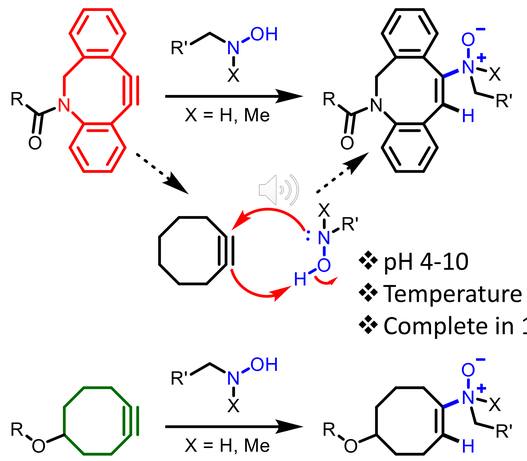
Chemically Modifying The Mycomembrane



Objectives

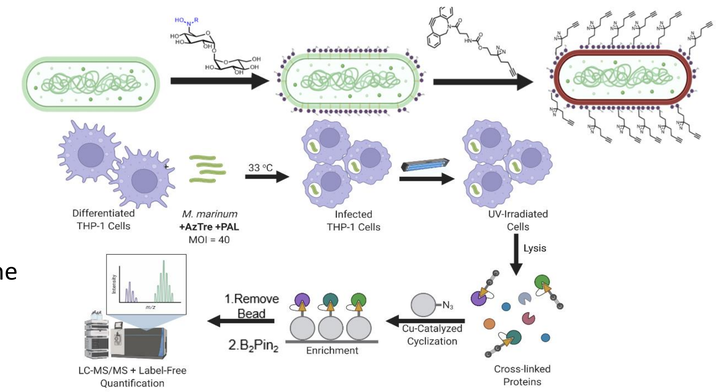
- Synthesis of Hydroxyl Methyl Amine Trehalose analog
- Develop a probe that will enable biorthogonal “Click” reactions.
- Characterization of the modified Trehalose molecule
- Utilization and optimization of the novel known retro-Cope reaction in the biological model.
- Diagnostic labeling tool of Trehalose

Retro-Cope Mechanism

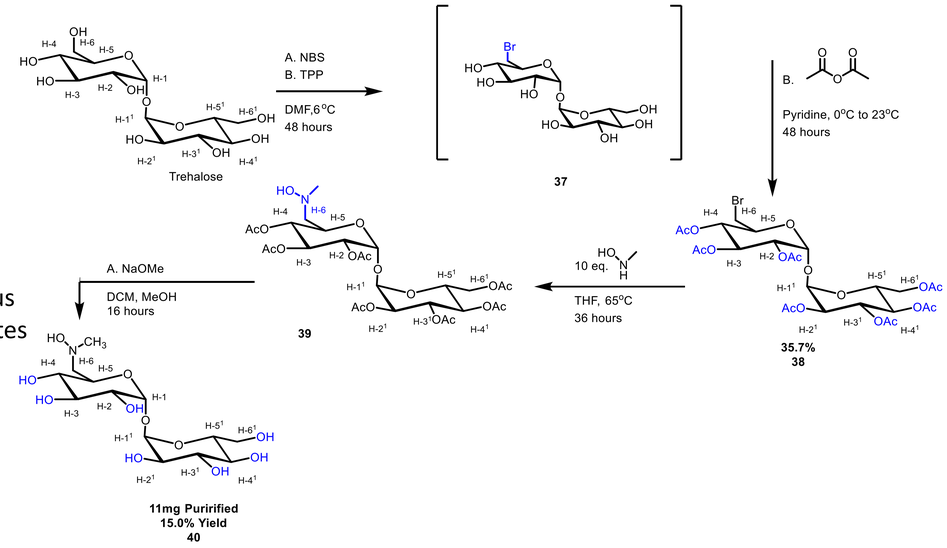


- pH 4-10
- Temperature 22°C Plus
- Complete in 10 minutes

Cross-Linking Proteomics



Current Synthesis Route



Acknowledgements

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UT System Rising STARs

1.Kang, D. ; Kim, J.Thongsomboon, JACS **2021** *143* (15), 5616-5621
2.Agard, N; Prescher,J; Bertozzi, C., *J. Am. Chem. Soc.* **2004**, 126, 46, 15046-15047

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