conferenceseries.com

Qiang Yan et al., Curr Synthetic Sys Biol 2017, 5:1(Suppl)
DOI: 10.4172/2332-0737-C1-008

Modularization strategies in metabolic engineering *Serratia marcescens* for N-acetylneuraminic acid production

Qiang Yan and **Stephen S Fong**
Virginia Commonwealth University, USA

Annually, large amounts of waste material are generated from seafood (crab, shrimp and lobster shells) that typically go to landfills with high expense (e.g. $150/ton in Australia). The biopolymer chitin makes up ~40% total weight of such waste and it can potentially be used as a sustainable feedstock for value-added products. In this study, we developed a consolidated bioprocess to produce N-acetylneuraminic acid (NeuNAc) directly from chitin using the chitinolytic organism *Serratia marcescens* by developing and implementing genetic engineering tools to modify *S. marcescens*. Starting from RNA sequencing data, 10 native promoters of varying strength were identified and characterized using a fluorescent reporter gene (sfGFP). Two heterologous genes (N-acetylglucosamine isomerase and NeuNAc lyase) were introduced in *S. marcescens*, and expression was transcriptionally controlled using three different strength promoters (high-high, medium-medium, and low-low). An initial 34.8 mg/L NeuNAc was produced under a high-high promoter combination. Improvements to NeuNAc were conducted by changing expression strength of the two heterologous genes to balance metabolic flux. Using a high strength promoter for the isomerase and a medium strength promoter for the lyase improved both NeuNAc production (1.42-fold) and growth (2.57-fold). Further improvements were tested by characterizing the kinetics of each heterologous enzyme and implementing alternative genes. Swapping N-acetylglucosamine isomerase gene did not show any improvement in production; however, swapping the second-step reversible NeuNAc lyase (nanA) to an irreversible NeuNAc synthase (neuB) improved NeuNAc production (3.25-fold). Overall, we identified and characterized promoter sequences that can be used for genetic engineering of *S. marcescens* and have implemented the tools to demonstrate and improve production of NeuNAc.

Biography

Qiang Yan is a PhD candidate working under the supervision of Dr. Stephen S Fong in the Department of Chemical and Life Science Engineering, Virginia Commonwealth University (VCU). His research interest focused on using molecular genetic techniques, synthetic biology and computational models to design modify and improve organism as workhorse to solve real-world problems.

yanq2@vcu.edu

Notes: