

PlasmidMaker is a Versatile, Automated, and High Throughput End-to-End Platform for Plasmid Construction

Background/Objective

Plasmids are one of the most foundational tools for recombinant DNA technologies. However, design and construction of plasmids, specifically the ones carrying complex genetic information, remains one of the most time-consuming, labor-intensive, and rate-limiting steps in performing sophisticated biological experiments.

Approach

We developed: 1) a new DNA assembly method based on *Pf*Ago-based artificial restriction enzymes that allows versatile DNA assembly; 2) a user-friendly frontend for plasmid design, as well as backend that streamlines the process; and 3) a robotic workflow for plasmid construction based on a biological foundry (iBioFAB).

Results

We were able to construct 101 plasmids from six different species ranging from 5 to 18 kb in size from up to 11 DNA fragments within few days with limited human intervention.

DESIGN BUILD TEST User-friendly WebFront Interface 2 4 Confirmation and Stock One-pot PfAao Preparation of Plates for PCR Digestion, ligation, and Reactions Transformation PlasmidMaker Quality Check Frozen Stocks List of REs Guides

PlasmidMaker: Computer-assisted plasmid design and robotic assembly.

Significance/Impacts

This platform is planned to be available for orders by CABBI and other DOE Bioenergy Research Centers and will increase research efficiency.

Enghiad, B., Xue, P., Singh, N., Boob, A., Shi, C., Petrov, V.A., Liu, R., Peri, S.S., Lane, S.T., Gaither, E.D., Zhao, H. (2022) "PlasmidMaker is a versatile, automated, and high throughput end-to-end platform for plasmid construction." *Nature Communications*. DOI: 10.1101/2021.12.31.474679.