

Development and Characterization of Gold Nanoparticles for Plant Genetic Engineering



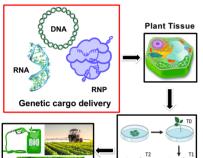
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Salwan Butrus¹, Natalie S. Goh², Gozde S. Demirer², Huan Zhang², Francis J. Cunningham², Markita P. Landry²

¹University of Michigan Department of Chemical Engineering, ²UC Berkeley Department of Chemical Engineering

Motivation

Plant genetic engineering is poised to fortify several key industries



Existing plant transformation

Applications

methods lack practical applicability. have a narrow host range, are toxic. and inefficient

Agrobacterium Gene Gun

Research Objectives

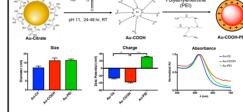
- **Design** biocompatible vehicles capable of efficient transport into plant cells of mature leaves
- Tune physicochemical properties of carriers to screen for optimal formulations
- Integrate nanoplatforms into model plant systems to gauge gene silencing and expression efficiency

Methods and Results

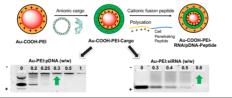
Nanoparticle Synthesis and Characterization

Approach 1: Electrostatic laver-by-laver siRNA and pDNA nanocarrier assembly

Carboxvlation of AuNPs to obtain a stable negative surface for cationic polymer deposition

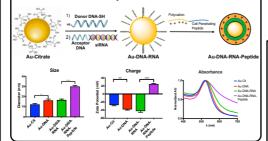


Successful loading of siRNA and pDNA onto engineered cationic nanoparticles

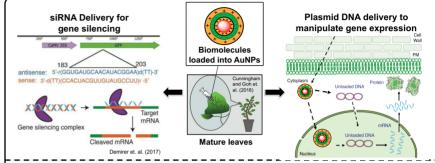


Approach 2: siRNA conjugation to AuNPs via complementary base pair hybridization

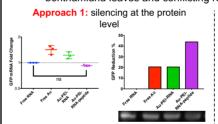
Thiolated Donor DNA attachment to AuNPs and hybridization to GFP siRNA

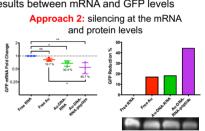


Administration of Nanoparticles to Mature Leaves



Preliminary results show GFP silencing in transgenic Nicotiana benthamiana leaves and conflicting results between mRNA and GFP levels





Approach 1: preliminary confocal imaging results show modest levels of GFP expression in wild-type Nicotiana benthamiana leaves









Au-pDNA

Au-pDNA-Peptide

Conclusions and Future Work

We developed two nanoplatforms for the delivery of siRNA and pDNA to intact leaves. The following work must be conducted in the future:

- Repetition of all experiments with biological replicates to conclusively confirm or deny
- Determination of critical AuNP toxicity concentrations to evaluate system scalability
- Evaluation of AuNP internalization into plant cells to gauge the platform's passive cell
- Delivery of sgRNA to Cas9-expressing plants for precise plant genetic engineering

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References