DiSTAP

Research Projects in the SMART DiSTAP IRG

(October 1, 2019)

1. Project Title: Engineering Bacterium to Produce VOC for Urban Farming

MIT Faculty Advisor: Gregory N STEPHANOPOULOUS and Anthony J SINSKEY
Mentor: Xiaoqiang MA
Email Address: Xiaoqiang@smart.mit.edu

Project Description

New sustainable solutions are urgently needed to support agriculture in meeting current environmental challenges and population growth. Urban farming, including vertical and rooftop farms, is rapidly becoming popular to provide a sustainable approach. One of DiSTAP's goals is to establish principles for high volume production of amino acids, lipids, vitamins and other valuable natural products in microorganisms for urban farming. In this project, our goal is to produce volatile organic compounds (biopesticide) derived from isopentanol through advanced metabolic engineering tools. We aim to develop novel and cost-effective microbial fermentative process to produce customized VOCs by linking a newly developed Isopentanol Utilization Pathway (IUP, [**1**]) with cellular central metabolites derived from renewable feedstock, such as amino acids and acetyl-CoA. Linalool (an identified biopesticide) were used as the model compounds.

In this two-month project, we will modularly optimize metabolic engineering pathway. The pathway engineering will be done by our newly developed workflow, which is based on GT DNA assembly standard [**2**].

Goals

The specific goals are listed as follows:

1) Test various linalool synthases from various sources to improve the production linalool in *Escherichia coli* (a commonly used workhouse in microbe engineering)
2) Optimize culture conditions and downstream pathway to further improve the production of linalool in *E. coli*
3) Identify the bottleneck of the involved pathway to further boost the production of linalool

Prerequisites/Skills

Chemical Engineering, Biology, or related fields/Molecular bio-techniques, cell culture, and other related skills in genetic engineering of microbes

Types of Software Applications

Matlab and other molecular and microbiology bio-techniques related ones

Individual or Team Project

Team

DiSTAP

Relevant Papers and or URLs

1) A. O. Chatzivasileiou et al. "Two-step pathway for isoprenoid synthesis", Proceedings of the National Academy of Sciences of the United States of America 116. 2 (2019) 506-511.
2) X.Q. Ma, et al. "A standard for near-scarless plasmid construction using reusable DNA parts". Nature communications 10.1 (2019): 3294.

2. Project Title: Discovery of Nano-Sensors for Plant Hormones

MIT Faculty Advisor: Michael STRANO
Mentor: Mervin ANG
Email Address: mervin.ang@smart.mit.edu

Project Description

The student will join an interdisciplinary team of chemists, engineers and plant biologists in the development of novel nano-sensors for real-time measurement of plant hormones. These novel tools enables optimal crop production and rapid development of plants suitable for high-density farming and revolutionalizes urban farming strategies. This project focuses on the chemical aspect: synthesis and characterization of polymer wrapped single-walled carbon nanotube (SWNT) suspensions which are intended as nanosensors for plant hormones such as auxins, jasmonates, salicylic acid etc. The students will be introduced to amphiphilic A-B co-polymerization techniques, as well as SWNT suspension methodologies. In particular, we will develop a series of cationic co-polymers and suspend them with SWNT, forming a corona phase. Different co-polymer wrapping results in different corona phases some of which highly selective for certain plant hormone analytes. These wrappings can be identified by *in vitro* screening of polymer library against plant hormone analyte library. Characterizing the corona phase is of particular interest as it has a direct impact on the sensor selectivity for different target analytes. The corona phase will then be characterized with state-of-the-art measurement tools and methods developed at both MIT and DiSTAP. The student will also have the opportunity to watch these nano-sensors work *in situ*. The nano-sensors developed will be infiltrated into a variety of plant species (wild-type and engineered mutants) cultivated by the Temasek Life Sciences laboratory (TLL), to demonstrate their effectiveness and generality.

Goals

At the end of this project, the student should be equipped with polymer synthetic chemistry and various optical and nanoparticle characterization skills. Throughout his/her stay at DiSTAP, the student would further expand our current synthetic polymer library and through *in vitro* screening, identify candidates that show promise as plant hormone analyte sensors.

Prerequisites/Skills

Chemical Engineering or Chemistry background

Types of Software Applications

1) Cary WinUV (UV-Vis-nIR spectroscopy)
2) Nanosight LM10 (Particle Size distribution measurement)
3) FluorEssence™ Horiba (nIR Fluorescence spectroscopy)
4) SkanIt Software (Varioskan LUX Multimode Fluorescence plate-reader)
5) Biotage® Initiator+ (Microwave polymer synthesizer)

Individual or Team Project

Individual

Relevant Papers and or URLs

1) Wong, M. H.; Giraldo, J. P.; Kwak, S.-Y.; Koman, V. B.; Sinclair, R.; Lew, T. T. S.; Bisker, G.; Liu, P.; Strano, M. S. Nature materials **2017**, 16, (2), 264.
2) Kwak, S.-Y.; Wong, M. H.; Lew, T. T. S.; Bisker, G.; Lee, M. A.; Kaplan, A.; Dong, J.; Liu, A. T.; Koman, V. B.; Sinclair, R. Annual Review of Analytical Chemistry **2017**, 10, 113-140.

3. Project Title: Development of Nanosensor Optodes for Urban Farming Applications

MIT Faculty Advisor: Michael STRANO
Mentor: Gajendra Pratap SINGH and Mervin ANG
Email Address: gajendra@smart.mit.edu; mervin.ang@smart.mit.edu

Project Description

The student will join an interdisciplinary team of chemists, engineers and plant biologists in the development of a novel portable nanosensor microneedle optode. This technology has the potential to be miniaturized and translated from laboratory to the field for commercial applications in urban farms. In the microneedle optode, a light source emits excitation light through an optical fiber into the microneedles coated with nanosensors. The nanosensors are protected by a polymer and cannot leach out into the plant. The fluorescence signal scattered back is detected and provides real-time information about plant hormone analytes. The nanosensors are based on the concept of corona phase molecular recognition, which uses polymers to wrap individualized single-walled carbon nanotubes. Different polymer wrappings result in different corona phases some of which highly selective for certain plant hormone analytes. Multiple microneedles allow multiplexing where several plant analytes can be targeted using differently functionalized nanosensors. The student will participate in the design and development of a nanosensor microneedle optode prototype based on existing nanosensors already developed at DiSTAP. He/She will proceed with testing and validation of the prototype for hormone detection in live plants.

Goals

At the end of this project, the student should build a functioning encapsulated nanosensor microneedle optode prototype for hormone detection in live plants. Throughout his/her stay at DiSTAP, the student would also be exposed to various optical and nanoparticle characterization techniques used in the development and validation of nanosensors.

Prerequisites/Skills
Electrical/Chemical/Biological Engineering background

Types of Software Applications
1) Cary WinUV (UV-Vis-nIR spectroscopy)
2) Nanosight LM10 (Particle Size distribution measurement)
3) FluorEssence™ Horiba (nIR Fluorescence spectroscopy)
4) SkanIt Software (Varioskan LUX Multimode Fluorescence plate-reader)
5) 3D printing and CAD design

Individual or Team Project
Individual

Relevant Papers and or URLs
1) Dong, J.; Salem, D. P.; Sun, J. H.; Strano, M. S. ACS nano **2018**, 12, (4), 3769-3779.
2) Kwak, S.-Y.; Wong, M. H.; Lew, T. T. S.; Bisker, G.; Lee, M. A.; Kaplan, A.; Dong, J.; Liu, A. T.; Koman, V. B.; Sinclair, R. Annual Review of Analytical Chemistry **2017**, 10, 113-140.

4. Project Title: Early Detection of Bacterial-Fungal Infection in Leafy Vegetable using Portable Raman Spectrometer

MIT Faculty Advisor: Rajeev J RAM
Mentor: Shilpi GUPTA and Gajendra P SINGH
Email Address: shilpi.gupta@smart.mit.edu

Project Description
Plant diseases are responsible for major food losses globally. Monitoring plant health and detecting diseases early are essential to reducing crop losses. Common methods for the diagnosis of plant diseases include visual inspection as well as laboratory analysis. Here, we propose to develop a portable optical instrument that can rapidly diagnose bacterial-fungal infections.

This instrument examines the spectrum of scattered laser light to study the chemical composition of the leaf. The spectrum of the scattered laser light is an optical 'fingerprint' of the various molecules within the leaf. We hope to explore several machine learning algorithms to analyze the spectrum of the scattered light and infer the disease state of the plant.

The proposed portable optical spectroscopic analyzer combined with data analysis techniques, will aid in the early detection of bacterial-fungal infection in leafy vegetables.

Goals

1) The student will learn about the applications of laser spectroscopy for real time detection of bacterial-fungal infection in plants.
2) The "spectral signatures" will be developed for early detection of plant disease levels for effective management of the diseases.

Prerequisites/Skills

Data analytics and instrumentation. An interest in analytical chemistry and plant biology

Types of Software Applications

Matlab, Python

Individual or Team Project

Team

Relevant Papers and or URLs

Not identified

5. Project Title: Early Detection of Plant Stresses using Portable Raman Spectrometer

MIT Faculty Advisor: Rajeev J RAM
Mentor: Shilpi GUPTA and Gajendra P SINGH
Email Address: shilpi.gupta@smart.mit.edu

Project Description

Any unfavorable condition or substance that affects a plant's metabolism, growth, or development is regarded as stress. The effects of stress can lead to deficiencies in growth, crop yields, permanent damage or death if the stress exceeds the plant tolerance limits. Plant stress factors are mainly categorized into two main groups; abiotic factors and biotic factors. The abiotic factors include the different environmental factors that affect plant growth (such as light, water, and temperature), while the biotic factors are the other organisms that share the environment and interact with the plants (such as pathogens and pests). To protect themselves from a stressful environment, plants have the ability to control their metabolic activity and in the process they generate a chemical response.

One of the main goals in the DiSTAP program is to detect this chemical response in real time using optical sensors. Using laser light, the leaf cells are excited and the scattered light is collected in a spectrometer. The spectrum of this scattered light contains the 'fingerprint' of the chemicals produced by the plant as part of its stress response. Here, we hope to demonstrate the first portable instruments (lasers and spectrometers) capable of detecting the chemical stress response in plants. The student will design a mechanical assembly to hold a living plant leaf in the path of the laser for analysis. The student will then acquire spectra and correlate them with the established stress response in the plants.

Goals

1) The early diagnosis of plant stress with a portable instrument offers farmers a new tool for crop management.
2) The student will learn about the applications of photonics and spectroscopy to important biological and ecological problems.

Prerequisites/Skills

Mechanical engineering design; An interest in analytical chemistry and plant biology.

Types of Software Applications

Matlab, Python

Individual or Team Project

Team

Relevant Papers and or URLs

Not identified