**Methanococcus: The New Archaea-type for Synthetic Biology**

**Establishing Methanococcus as an Alternative Platform in Synthetic Biology**

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### Abstract

When beginning research in synthetic biology, the go-to organism is the bacterium *E. coli*. Our research demonstrates the viability of using a methanogenic archaeon, *Methanococcus maripaludis*, as an alternative platform. Our lab primary focuses in this study were: 1) improving the genetic tools for synthetic biology available in *E. coli* and 2) demonstrating the viability of using *M. maripaludis* in synthetic biology. We achieved both of these goals through our research, which was aimed at manipulating *M. maripaludis* lipid biosynthesis pathway for the production of isoprenoid lipids. Our research was performed using a combination of chemical, biological, and computational approaches that included: an improved geraniol extraction procedure resulting in better recovery, a computational approach for the overproduction of isoprenoid lipids, and high-throughput genome engineering to insert any gene of interest that will be covalently linked to mCherry.

### Methanococcus maripaludis

*M. maripaludis* is a methanogenic archaeon native to salt marshes. It utilizes CO₂ and H₂, or formate through a process called methanogenesis, to form bacterial lipids and methane. This organism is the ideal organism for synthetic biology. *M. maripaludis* is an extremely versatile species, capable of being used in a wide range of applications, from environmental remediation to biorefinery processes. This organism has a relatively simple genome, which makes it easier to manipulate and engineer. Its ability to produce lipids makes it a potential candidate for the production of biofuels and other valuable products.

![Image](image1.png)

**Modeling & Flux Balance Analysis**

The supporting figure shown here are the modifications we made to the wild type *M. maripaludis* model file, which is elaborated more in the Modeling & Flux Balance Analysis section shown to the right.

**Ribosome Binding Site Library**

![Image](image2.png)

**Geraniol Production**

- **Geraniol is an acyclic monoterpene alcohol**
- Has many uses as fragrances and flavorings
- Has a higher energy density than ethanol and is a heat source
- Can be used as a potential insect repellent
- Has been shown to inhibit the growth of pancreatic, prostate, and colorectal cancers

*Geraniol is synthesized from the MEP/DOXP pathway.*

**Protein Expression/Quantification Tool**

The vector below has been optimized for expression in *M. maripaludis* and uses the red fluorescent protein, mCherry, as a fluorescent reporter. The use of... standards allow researchers to assess any gene of interest that will be covalently linked to mCherry.

**Future Work**

- Improve upon geraniol extraction methods from *M. maripaludis* cultures.
- Combine knowledge accumulated in this study to further optimize geraniol production, such as using high strength RBS sequences for better expression of the gene, and making mutations to increase geraniol yield by mapping the entire lipid biosynthesis pathway to geranyl acetate and other possible higher value chemicals.

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### References

1. Whitman, William B. "Methanococcus maripaludis strain C6, shah C6 and shah C7." *Methanococcus maripaludis C6, shah C6 and shah C7.* Department of Microbiology and Molecular Biology, University of Georgia, Athens, Georgia 30002, 2012.


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- Providing interest in the sciences to local students is an important aspect of UGA’s iGEM team. We were awarded the UGA Alumni Association Young Leaders Scholarship to engage the students in fun, hands-on experiments.

- **Human Practices**

- **Future Work**

- **Innovate: Local Outreach Projects**

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