An Analysis on the Growth Effects of *Pseudomonas fluorescens* on Hexadecane Under Various Aquatic Conditions

Alison Kirby, Jacob Gooch, Yolanda Wilson
North Carolina State University, Department of Microbiology, 4510 Thomas Hall, Campus Box 7615 Raleigh, NC 27695
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Introduction

As stated in Meghan Shea's paper, The Effect of Nitrogen, Sulfur, and Phosphorus Compounds on Bioremediation of Oil Spills by Pseudomonas fluorescens and Bacillus subtilis, marine oil spills are devastating and being able to prepare for them would limit damage. Our experiment was designed using Shea's fundamental idea for *Pseudomonas fluorescens* as an option for bioremediation, a technique in which organisms are used to remove pollutants from a place that is contaminated (1). The goal was to find which type of water created the best environment for the growth of *Pseudomonas fluorescens* (*P. fluorescens*) on hexadecane. Hexadecane, an aliphatic hydrocarbon, is a model representative of diesel fuel and our pollutant. By studying the growth of *P.fluorescens* on hexadecane, we were able to see how the organism's growth was affected based off the water used. It is probable that a water sample most like ocean water will provide the best environment for the growth of *P. fluorescens* on hexadecane.

Materials and Methods

Initially we needed to see how and if growth of P.fluorescens was inhibited by hexadecane. The 10x Luria Broth (LB) with tryptone and yeast extract provided enough nutrients where growth was not inhibited with the addition of hexadecane. A 250 µl sample was made and 200 µl was loaded into the Bioscreen C instrument, which was used to measure the optical density allowing for the construction of a growth curve. We depleted the nutrients, so *P. fluorescens* had to adapt or find other sources for nutrients. Once we lowered the nutrients to .5 µl of both tryptone and yeast extract with 50 µl of hexadecane varying water types was possible. Our water samples included filtered water from Lake Johnson in Raleigh, filtered instant ocean from Dr. James Brown's saltwater tank that will be referred to as Marine water, 1 M NaCl, tap water from the lab in Thomas Hall at NC State University, and distilled water used as a control.

Results

Based off maximum growth, Marine water provided the best conditions for the growth of *P. fluorescens* on hexadecane.

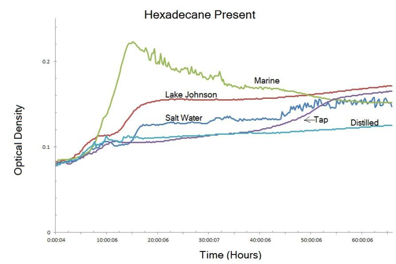

Fig. 1. Growth of P. fluorescens with hexadecane present of .5µl of tryptone and .5µl of yeast extract in various water to simulate different aquatic environments.

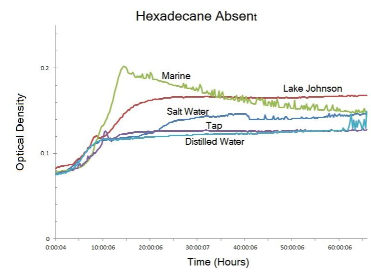

Fig. 2. Growth of P. fluorescens with no hexadecane present in .5µl of tryptone and .5µl of yeast extract in various water to simulate different aquatic environments.

Maximum Growth

Type of Water	Hexadecane Present	Hexadecane Absent
Salt Water (NaCl)	0.158	0.147
Lake Johnson	0.171	0.168
Marine Water	0.222	0.201
Tap Water	0.165	0.129
Distilled Water	0.125	0.149

Table 1. This shows the maximum growth of P. fluorescens in each of the different types of water.

Discussion

Although the water samples from Lake Johnson and the saltwater tank have unknown nutrients which allow us to have realistic data with our simulated aquatic environments, a large scale model of the reaction between *P. fluorescens* and hexadecane in different types of water would be more representative of a real world oil spill.

References

1) Shea, M. (2013). The effect of nitrogen, sulfur, and phosphorus compounds on bioremediation of oil spills by pseudomonas fluorescens and bacillus subtilis. The Journal of Experimental Secondary Science, 30-37.

Acknowledgements:

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