Interactions and growth dynamics of *Prochlorococcus*, *Rhodospirilaceae*, and *Alteromonas* in a coculture environment

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Introduction

Interactions between different species of microorganisms have a significant role in the growth dynamics within the environment. (Morris 08) The specifics mechanisms of these interactions, however, are overall poorly understood.
The purpose of this study is to study the interactions between the heterotrophs *Rhodospirilaceae* (EZ54), and *Alteromonas* (EZ55) and the cyanobacterium *Prochlorococcus* (Vol 1).
Here we establish the effects the cyanobacterium has on the growth dynamics of each of the heterotrophs as well as the effect the two heterotrophs have on one another.

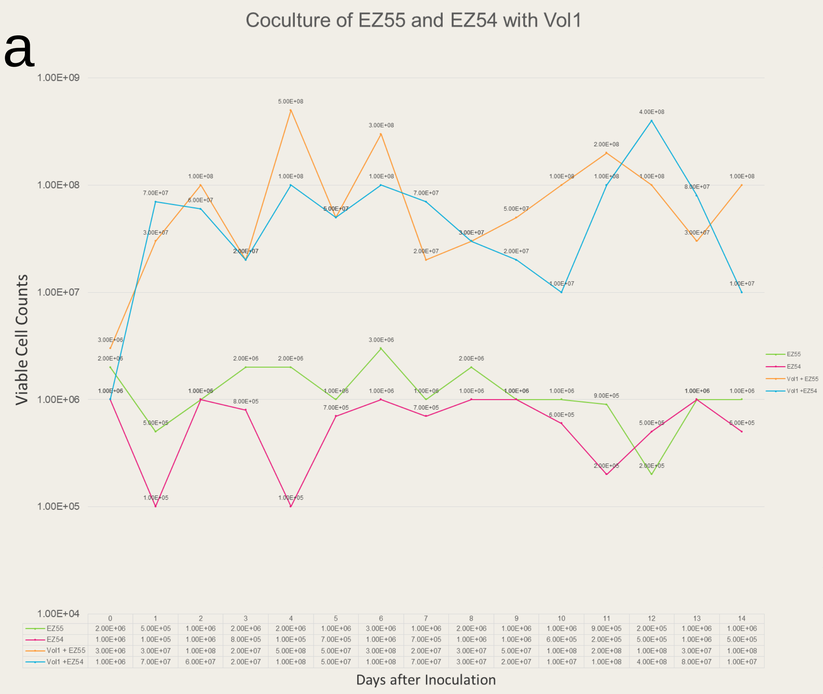
Methods

EZ54, EZ55 and VOL1 grown in tubes of minimal Amp-A media,
designed to simulate the natural environment. Four separate combinations of organisms were prepared in duplicate. EZ55 and EZ54 were grown in monoculture to establish the base growth rate of each. Two cultures were prepared contained each of the heterotrophs in coculture with VOL1. The growth rate of each culture was measured by taking viable counts every 24 hours. The results from both replicates were averaged together to generate the results graph.

A separate experiment performed over the course of four days determined the relative abundances of EZ54 and EZ55 when the two were grown in coculture with one another. Two tubes of Amp-A media were prepared and inoculated with both EZ54 and EZ55. Growth was the measured via daily viable cell counts utilizing the distinct colony morphology of each strain to distinguish between the organisms after plating. Their growth rate was then compared to that of the monocultures measured in the first experiment.

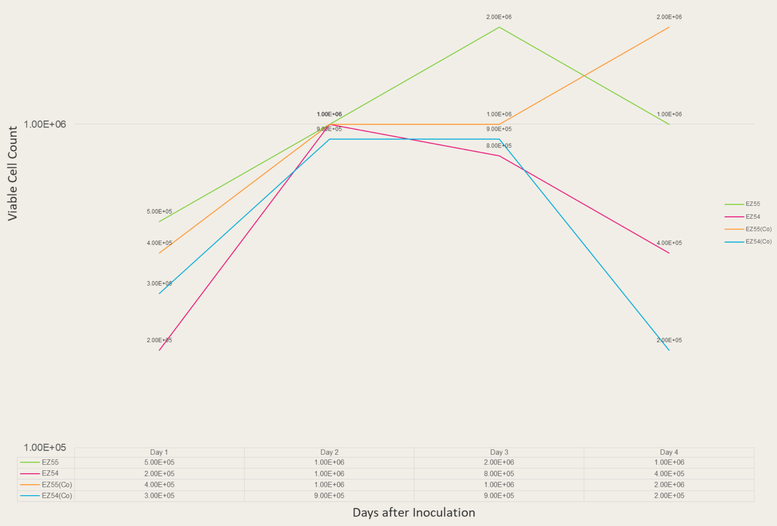
Results

a



	0	1	2	3	4	5	6	7	8	9	10	11	12	13	14
EZ55	2.00E+06	5.00E+05	1.00E+06	2.00E+06	2.00E+06	1.00E+06	3.00E+06	1.00E+06	2.00E+06	1.00E+06	1.00E+06	9.00E+05	2.00E+05	1.00E+06	1.00E+06
EZ54	1.00E+06	1.00E+05	1.00E+06	8.00E+05	1.00E+05	7.00E+05	1.00E+06	7.00E+05	1.00E+06	1.00E+06	6.00E+05	2.00E+05	5.00E+05	1.00E+06	5.00E+05
Volt + EZ55	3.00E+06	3.00E+07	1.00E+08	2.00E+07	5.00E+08	5.00E+07	3.00E+08	2.00E+07	3.00E+07	5.00E+07	1.00E+08	2.00E+08	1.00E+08	3.00E+07	1.00E+08
Volt +EZ54	1.00E+06	7.00E+07	6.00E+07	2.00E+07	1.00E+08	5.00E+07	1.00E+08	7.00E+07	3.00E+07	2.00E+07	1.00E+07	1.00E+08	4.00E+08	8.00E+07	1.00E+07

b



	Day 1	Day 2	Day 3	Day 4
EZ55	5.00E+05	1.00E+06	2.00E+06	1.00E+06
EZ54	2.00E+05	1.00E+06	8.00E+05	4.00E+05
EZ55(Co)	4.00E+05	1.00E+06	1.00E+06	2.00E+06
EZ54(Co)	3.00E+05	9.00E+05	9.00E+05	2.00E+05

Heterotroph cultures grown in monoculture were shown to have a significantly lower cell density than those cocultured with the cyanobacterium *Prochlorococcus*. (Fig a)

Heterotrophs grown in coculture with one another did not show any significant difference in their cell density in comparison to those grown in monoculture. (Fig b)

Conclusions

The results of the Heterotroph and VOL1 coculture indicate that the cyanobacterium has a positive effect on the heterotrophs' ability to grow in minimal media. This indicates that VOL1 produces some molecules beneficial for heterotrophic growth.

The coculture of the two heterotrophs, meanwhile, shows that there is not significant overlap in the nutrient requirements of *Rhodospirilaceae*, and *Alteromonas*, as being placed in a coculture together does not appear to harm the growth rate of either organism.

Since both heterotrophs benefit from the presence of *Prochlorococcus*, and their nutrient requirements do not overlap, the Cyanobacterium may be producing multiple molecules beneficial to the surrounding heterotrophs.

Future Research

The next step in this experiment would be generation of auxotrophic mutants of the heterotrophs in order to identify the exact molecules that each organism receives from the Cyanobacterium. Measurements also need to be taken of the growth rate of *Prochlorococcus* in the presence and absence of the heterotrophs in order to establish how heterotrophs affects its ability to grow. Finally all three should be placed in a single coculture and the growth of each observed. This would show whether *Prochlorococcus* provides distinct nutrients to each organism, or if the heterotrophs would compete for the limited supply of excess nutrients produced by the cyanobacterium.

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Citations
Morris, J. Jeffrey et al. "Facilitation of Robust Growth of *Prochlorococcus*Colonies and Dilute Liquid Cultures by 'Helper' Heterotrophic Bacteria ." *Applied and Environmental Microbiology* 74.14 (2008): 4530–4534. *PMC*. Web. 17 Apr. 2018.