



Synechococcus as an HOOH-Consuming Helper for *Prochlorococcus*



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Introduction

- *Prochlorococcus* is a globally important marine cyanobacterium that plays a major role in primary production in the ocean.
- *Prochlorococcus*' unusually small genome (1.7-2.5 Mbp) lacks a gene encoding the enzyme catalase, which is essential for the breakdown of hydrogen peroxide (HOOH).
- HOOH is a reactive oxygen species (ROS) naturally present in the illuminated surface waters of the oceans at an average concentration of ~75-150 nM. In the absence of protective enzymes such as catalase, HOOH and other ROS can cause catastrophic cell damage and cell death.
- Previous work has demonstrated that catalase-positive heterotrophic "helper" bacteria (e.g. *Alteromonas* sp. EZ55) facilitate growth of *Prochlorococcus* by consuming HOOH present in their shared environment.
- *Synechococcus* is a cyanobacterium closely related to and commonly found alongside *Prochlorococcus*. While typically viewed as a competitor of *Prochlorococcus*, *Synechococcus* produces functional catalase and may serve as a helper for *Prochlorococcus*.

Purpose

The purpose of this investigation is to determine if *Synechococcus* can serve as a helper to *Prochlorococcus* by removing HOOH from shared culture medium.

Materials & Methods

- **Bacterial Strains:**
Prochlorococcus sp. MIT9215
Synechococcus sp. WH7803
Alteromonas sp. EZ55
- Added 0, 200, or 450 nM HOOH to *Prochlorococcus* **monocultures** (grown alone) or **co-cultures** (grown with another organism) with *Synechococcus* or *Alteromonas*. Monitored HOOH concentration ([HOOH]) daily using an **acridinium ester (AE) chemiluminescence method**. Monitored *Prochlorococcus* abundance daily using **flow cytometry**.
- Added 400 nM HOOH to *Synechococcus* and *Alteromonas* monocultures (10^6 cells/mL). Monitored [HOOH] every 30 minutes for 4.5 hours using AE chemiluminescence method.

Results

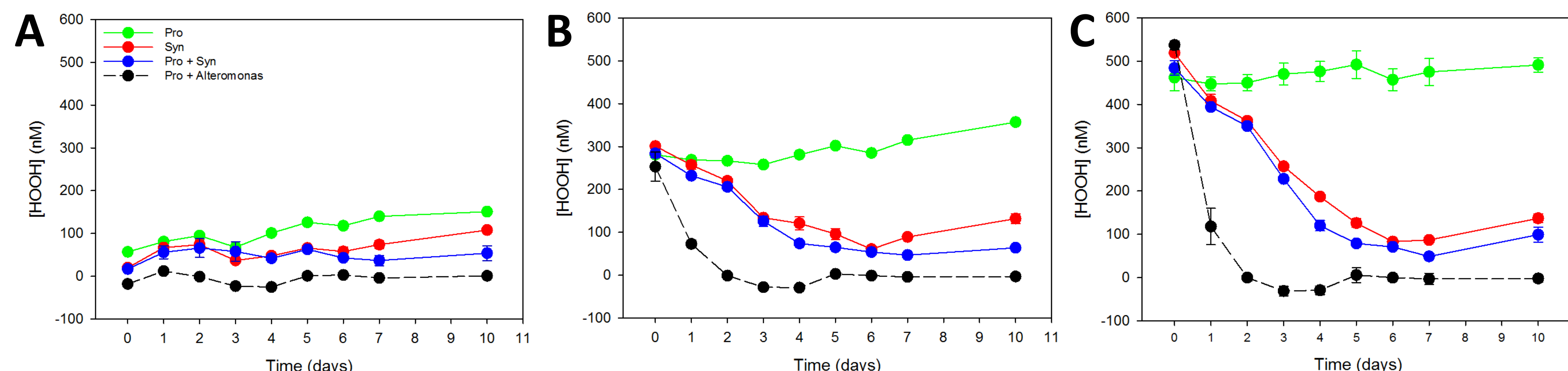


Figure 1. (above) HOOH concentrations in cultures of *Prochlorococcus* alone (Pro), *Synechococcus* alone (Syn), *Prochlorococcus* with *Synechococcus* (Pro + Syn), and *Prochlorococcus* with *Alteromonas* (Pro + *Alteromonas*). Cultures were spiked with 0 nM (A), 200 nM (B), or 450 nM (C) HOOH prior to inoculation.

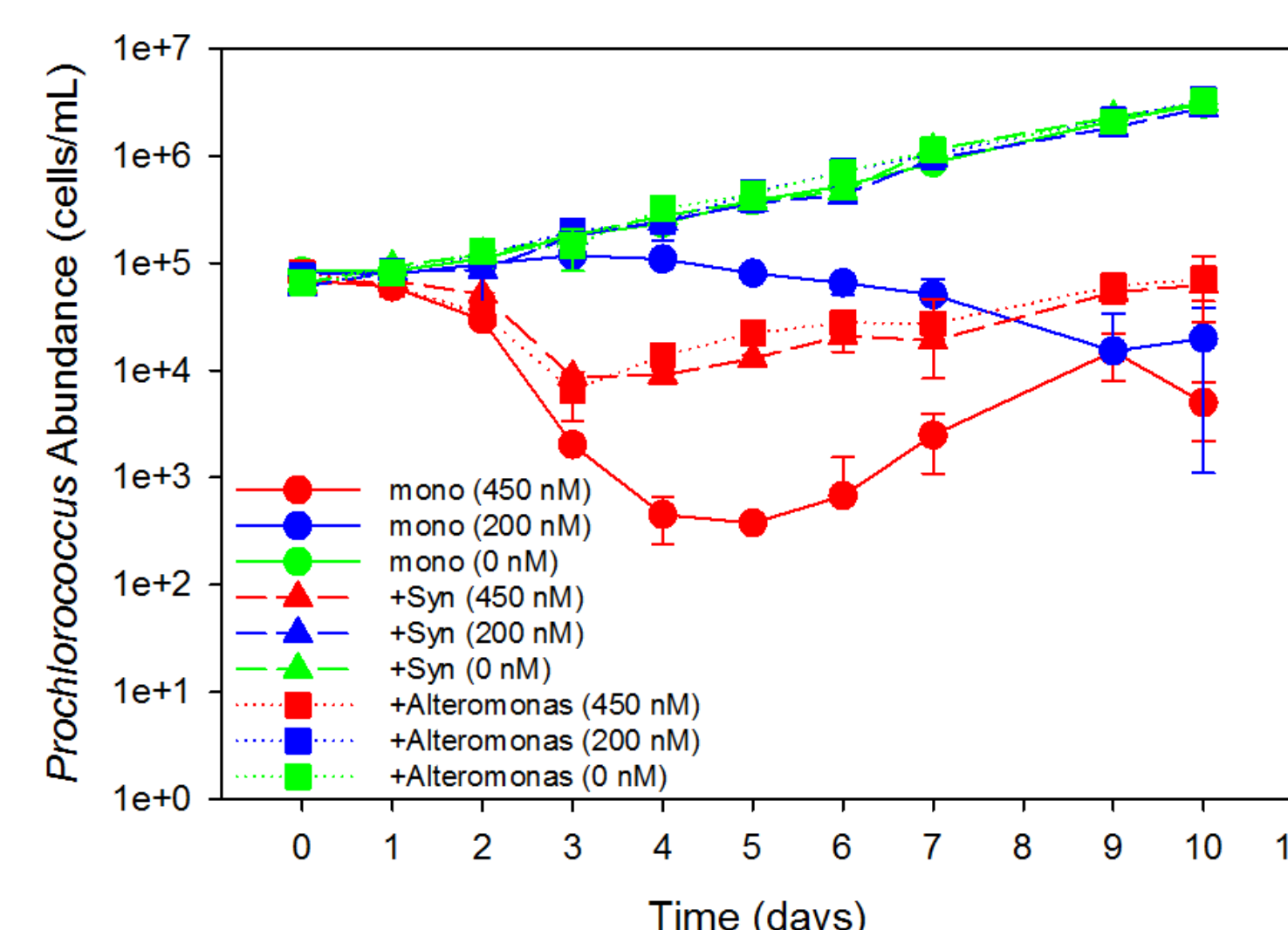
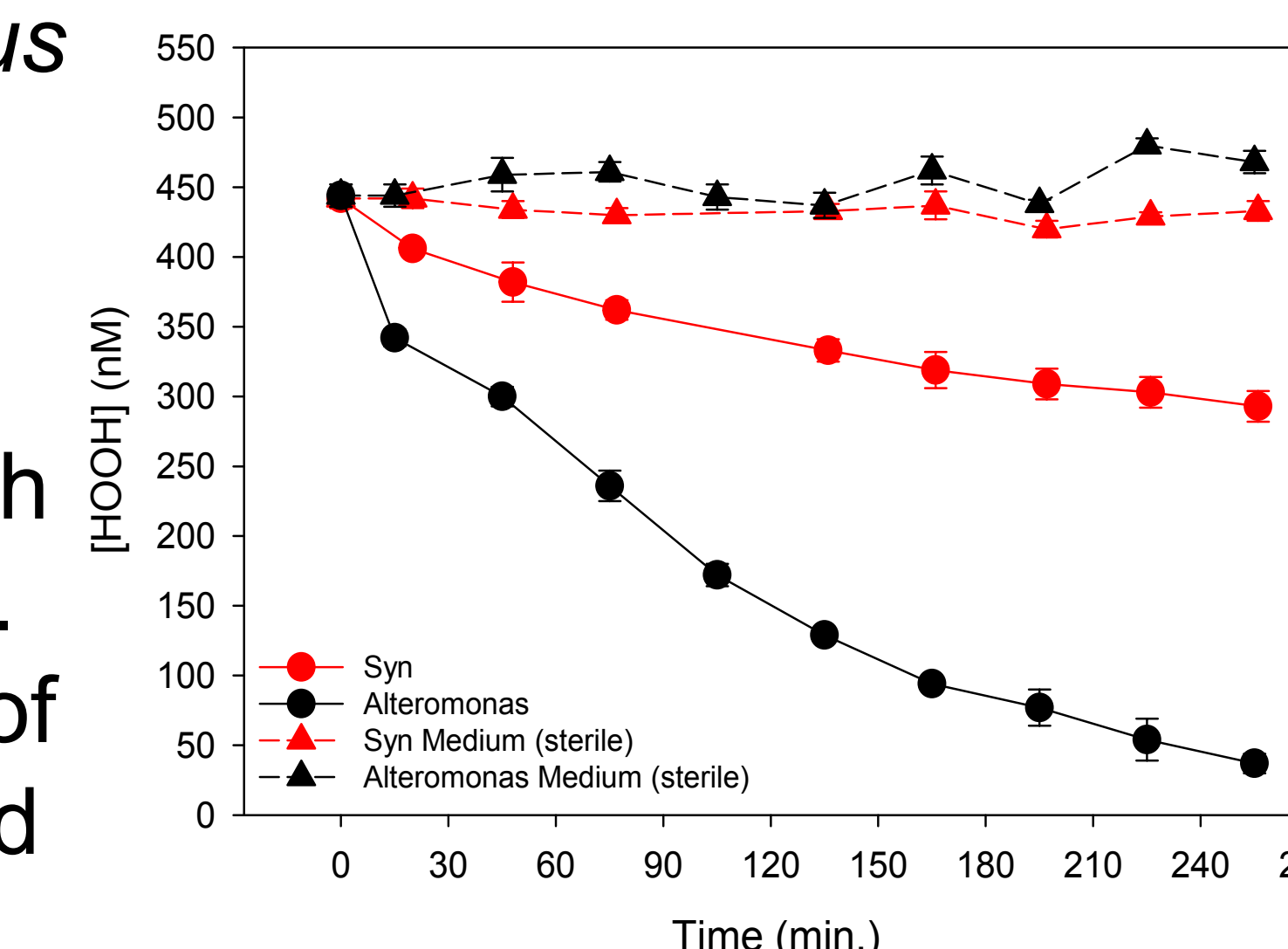


Figure 2. (left) *Prochlorococcus* abundance under various HOOH concentrations when cultured alone (mono), with *Synechococcus* (+Syn), or with *Alteromonas* (+*Alteromonas*).

Figure 3. (right) Degradation of HOOH by *Synechococcus* and *Alteromonas* over 4.5 hours.



Conclusions

1. *Prochlorococcus* alone does not remove HOOH from culture medium, leading to a decline in its population at ≥ 200 nM HOOH. Greater [HOOH] yields greater decline.
 2. *Synechococcus* steadily removes HOOH from culture medium, reducing HOOH levels from lethal to sub-lethal concentrations.
 3. Removal of HOOH by *Synechococcus* facilitates the growth of *Prochlorococcus* under HOOH stress.
 4. Positive effect of *Synechococcus* on *Prochlorococcus* growth under HOOH stress is comparable to the effect of known helper *Alteromonas* on *Prochlorococcus* growth under the same conditions.
- **Under the conditions tested, *Synechococcus* is able to serve as a helper to *Prochlorococcus* by removing HOOH from shared culture medium.**

Future Direction

- Develop a culturing system that models the natural daily flux of HOOH in the ocean environment and supports the sustained growth of *Prochlorococcus* and *Synechococcus* at ecologically relevant abundances.
- Examine the helping phenomenon in this system to gain further insight into the ecological relationship between *Prochlorococcus* and *Synechococcus*.

References/Acknowledgements

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2. Morris, J.J., Johnson, Z.I., Szul, M.J., Keller, M., & Zinser, E.R. (2011). Dependence of the Cyanobacterium *Prochlorococcus* on Hydrogen Peroxide Scavenging Microbes for Growth at the Ocean's Surface. *PLoS ONE*, 6(2):e16805.

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