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Temperature Tolerance of Phototrophs from Indiana Dunes State Park

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Abstract

Biological soil crusts (BSCs) are associations between microorganisms and the surface of the soil that occur in relatively undisturbed soils exposed to sunlight. They have been shown to stabilize soils and contribute to nutrient cycling in otherwise barren soils. Phototrophic microorganisms are important in BSCs because they initiate their development, serve as primary producers, and provide stability for the BSC microbial ecosystem. BSCs are known for their presence in open areas such as deserts, although they can be found in the more temperate environment of the Indiana Dunes State Park in the sand dunes along the Lake Michigan Lakeshore. With increasing temperatures, it is unknown as to how BSC microbial communities will respond and how a changing climate will specifically impact the phototrophs within these soils. Therefore, the objective of this research is to isolate phototrophic microorganisms from BSCs of the Indiana Dunes and observe their temperature tolerance in comparison to cyanobacteria from and deserts of the western US.

Background

BSCs are a fundamental component for arid and semiarid (dryland) areas (Fig. 1). Dryland ecosystems cover 40% of the Earth's terrestrial surface and contain 25% of global organic soil carbon (Ferrenberg et al. 2015). With temperatures increasing over the years, it has been shown that global warming has an effect on BSC distribution (Garcia-Plantefie et al. 2013). Therefore, more studies need to be done on this topic and currently there are no such studies done on the more temperate BSCs of the Indiana Dunes (Fig. 2).

The objective of this study is to analyze the temperature tolerance of phototrophs isolated from BSCs of the Indiana Dunes, Colorado Plateau (Micrurus vaginatus), and Sonoran Desert (Micrurus steenstrupii) in order to identify patterns of thermal tolerance.

Methods

The cyanobacteria in Cultures 64 and 66 from Indiana Dunes State Park were identified through PCR and sequencing of the 16S rRNA gene. Phylogenetic trees were built with the cyanobacteria taxa from this study, representative cyanobacteria from each Order, and algal plastid sequences. Phylogenies were constructed in MEGA 7 using the Neighbor-Joining algorithm with the Jukes-Cantor model and 1000 bootstrap replicates.

Temperature Tolerance Tests

• Two cultures from the Indiana Dunes, one from the Colorado Plateau (Microcoleus vaginatus), and one from the Sonoran Desert (Microcoleus steenstrupii) were grown in 12-well plates at 10, 25, 35 °C for 20 days under white light at 20-30 𝜇mol photons m⁻² s⁻¹.
• Change in growth (biomass) was measured by a change in chlorophyll a concentration.
• Comparisons were done using an ANOVA and Tukey’s HSD post-hoc analysis.

Strain Identification Results

Culture 64 had two types of phototrophs and neither culture had cyanobacteria that shared the same morphology as Microcoleus (Fig. 3). Through DNA sequencing, the cyanobacterium in Culture 64 was found to be the most closely related to Leptolyngbya (97% identity) while the identity of the cyanobacterium from Culture 66 could not be determined. The long multicellular filaments in each culture from the Indiana Dunes is morphologically similar to Klebsiellum, a eukaryotic alga. The phylogenetic analysis supports that the cyanobacterium from Culture 64 is likely a Leptolyngbya (Fig. 4), while the cyanobacterium from Culture 66 is not closely related to any of the other cyanobacteria in the tree.

Temperature Tolerance Results

While none of the cultures grew well at 10 °C or 35 °C (data not shown), at 25 °C there was growth for all of them except M. steenstrupii (Fig. 5). Culture 64, which contained a cyanobacterium and algae, grew at similar levels to the M. vaginatus strains while Culture 66, which was almost entirely the eukaryotic alga strain, did not grow as well.

Summary and Conclusions

While we were not able to compare the growth of cyanobacteria from the Indiana Dunes to those of arid deserts, we were able to evaluate the difference between cultures with and without a cyanobacterium present. Based on the growth at 25 °C, a relatively temperate condition, it appears as though the culture with an identified cyanobacterium (#64) grew better than the one without a known cyanobacterium (#66). Therefore it is possible that the cyanobacterium may have outcompeted the algae in this experiment, although no microscopy was done at the end to examine this possibility. Furthermore, it appears as though these phototrophs are sensitive to the large changes in temperature used in this study, thus changing temperatures could have an effect on the BSCs of the Indiana Dunes and other environments. Future studies will focus on comparing the growth of cyanobacteria, ideally from the same genus, and using a more narrow temperature range to capture the differences between strains from each environment.

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References

• Ferrenberg S., Reed S.C., Belnap J. (2015) Climate change and physical disturbance cause similar community shifts in biological soil crusts. PNAS 112: 39, 12116-12121.

Research Objectives

• Ferrenberg S., Reed S.C., Belnap J. (2015) Climate change and physical disturbance cause similar community shifts in biological soil crusts. PNAS 112: 39, 12116-12121.

Figure 1. Map of the US showing the geographic regions of strains used in this study.

Figure 2. Typical sampling plot in the Indiana Dunes. Note the textured sand due to stabilization by microorganisms.

Figure 3. Phototrophic strains in this study.

Figure 4. Phylogenetic tree of the cyanobacteria sequences from this study compared to other cyanobacteria and plastid 16S rRNA sequences. Bootstrap values less than 50 are not shown.