Cave algae pigments

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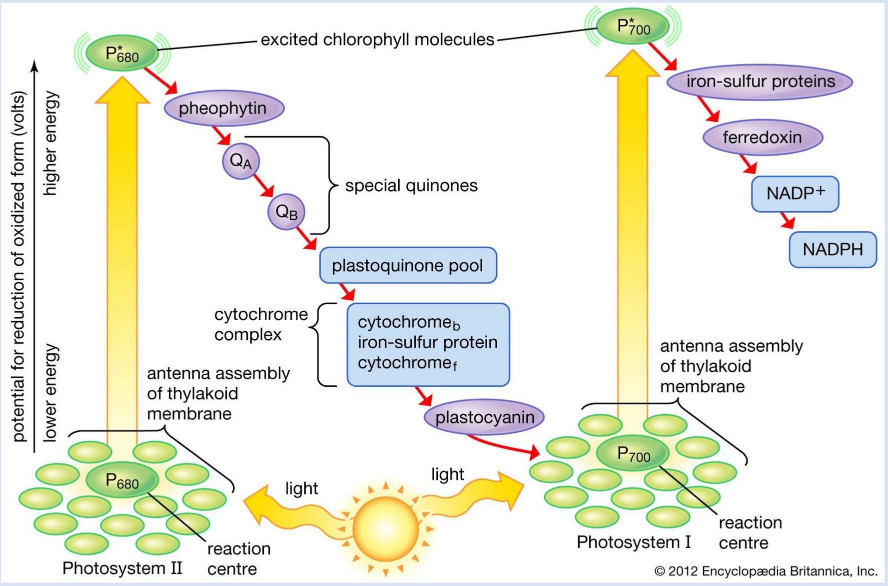
Abstract

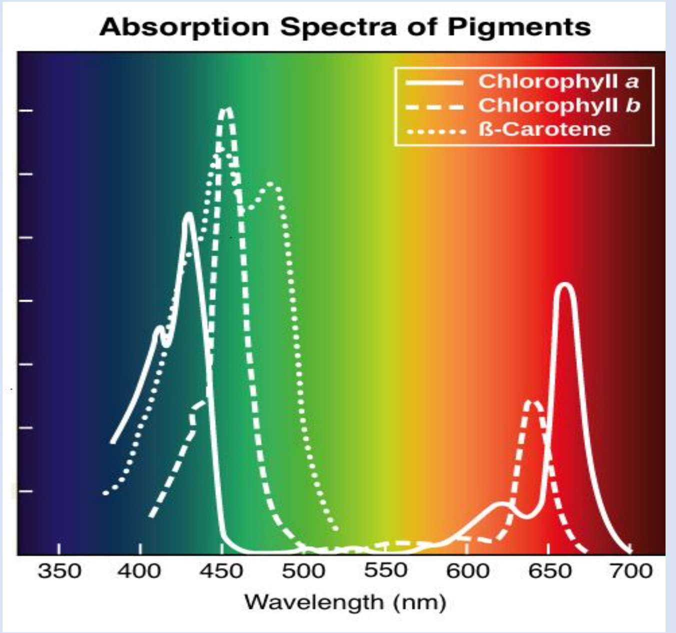
Mammoth Cave National Park in Kentucky has approximately 7 miles of illuminated cave trails. These illuminated trails, along with high humidity and a constant temperature of 54oF, provide an environment that allows small algae (lampenflora) to grow on the cave walls. The objective of this project was to study the lampenflora adaptation to low light, and use UV- visible spectrophotometry to see what wavelengths are being absorbed.

Background

Chlorophyll and other pigments harvest photons and send energy to PS II & PS I reaction centers.



The pigments absorb light energy at specific wavelengths.



Algae grows in Mammoth Cave near tour lights

Objective and Methods

Objective: To characterize the photosynthesis pigments and how the cave algae have adapted to low light (less than 1/100th of surface light)

Measure chlorophyll a and phycocyanin fluorescence (measure of pigment) in the cave

Collect and run chromatography and UV-Vis spectrum to see what wavelengths are absorbed.

Methods and Materials

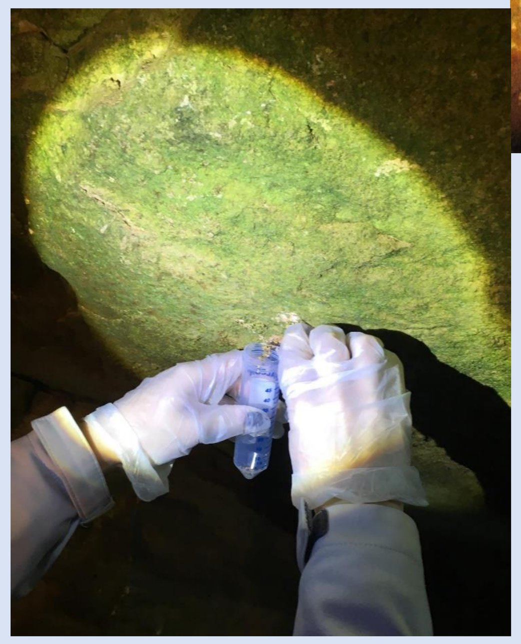
Selected 4 different types of algae growing at 3 lights.

In the image I was using a photometer to measure the amount of light the algae growing on the wall was absorbing.

In the images below there are examples of each type of algae sampled.

Light # color of algae

- D150-GW (Green-&-white)
- D150-T (Turquoise)
- D152-B (Brown)
- D-178-G (Green)



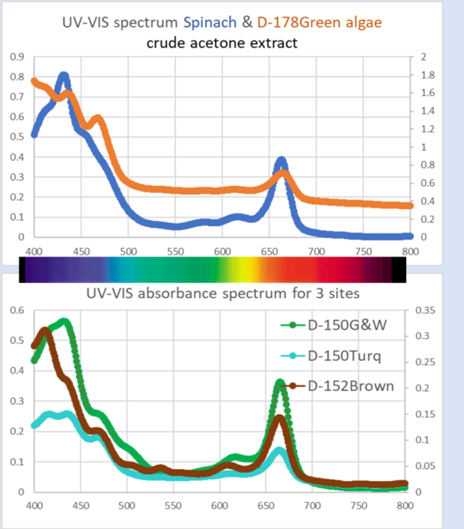
Higher PC values indicates blue-green algae
Higher Chl-a indicates green algae

Site	Avg Chl_a fluorescence (n=3)	Avg Phycocyanin fluorescence (n=3)
D150-GW	0.65	0.73
D150-T	0.32	0.96
D152-B	0.38	0.50
D178-G	0.74	0.20

Higher PC values indicates blue-green algae
Higher Chl-a indicates green algae

Preliminary UV-Vis results

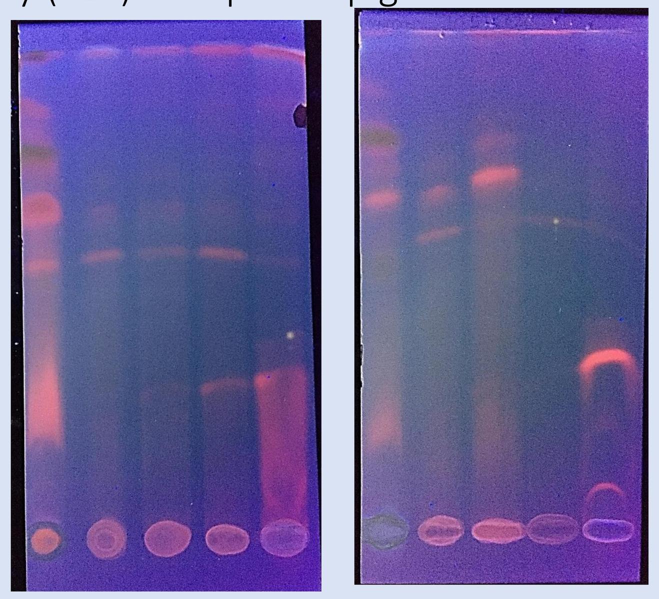
Ran scan on crude acetone extracts from the 4 cave algae colonies and spinach. Note different absorbance peaks. (different pigments for photosynthesis)
Some of the pigment-absorbance peaks may be hidden under the big peaks.



Used Thin Layer Chromatography (TLC) to separate pigments & run UV-VIS spectroscopy

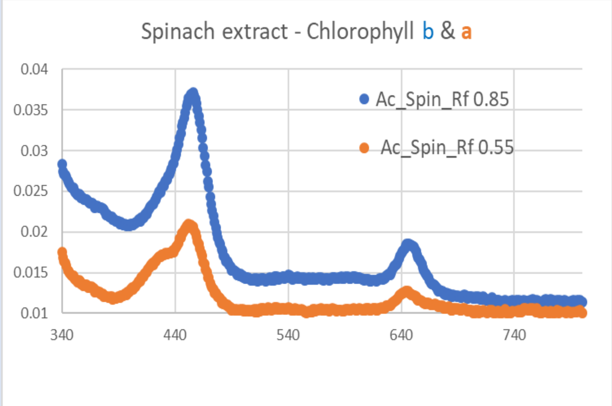
Extract pigments with acetone (hydrophobic) & with MeOH (hydrophilic)
Used spinach as standard (well studied)
Used Toluene : Acetone (60:40) as mobile solvent
Identified fluorescent spots

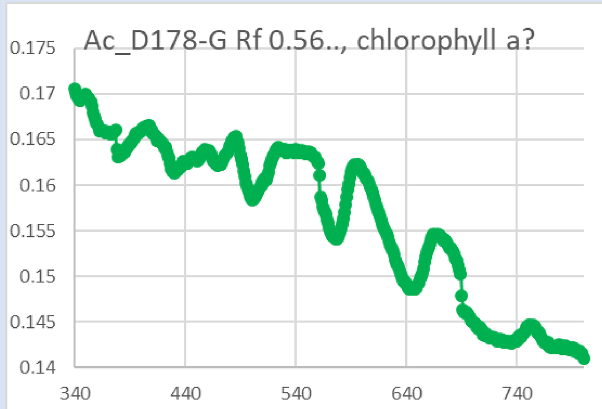
Calc. R_f = spot distance from baseline / solvent distance from baseline

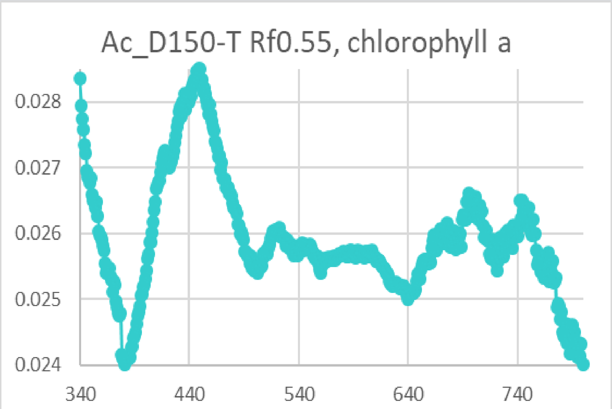


Scrape spots off and redissolve in Acetone / MeOH & run UV-Vis spectrophotometry

Comparing spots with similar Rf

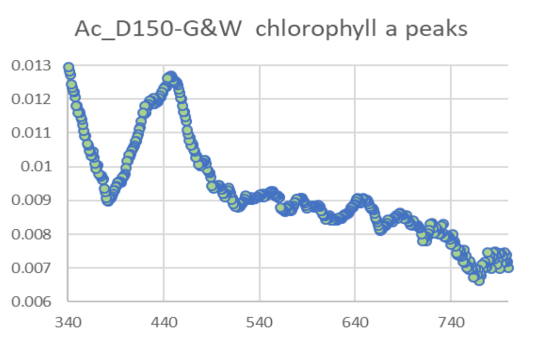


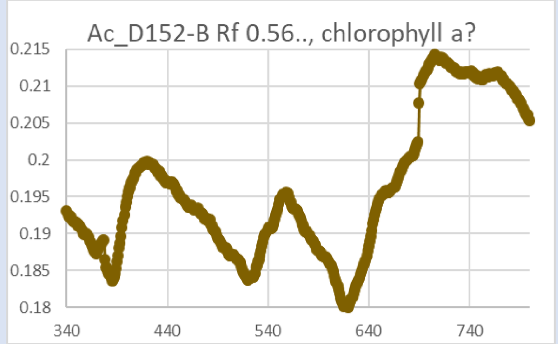


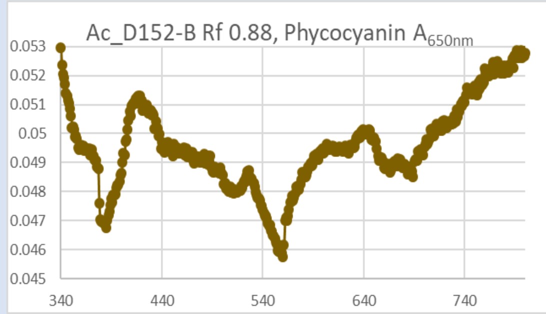


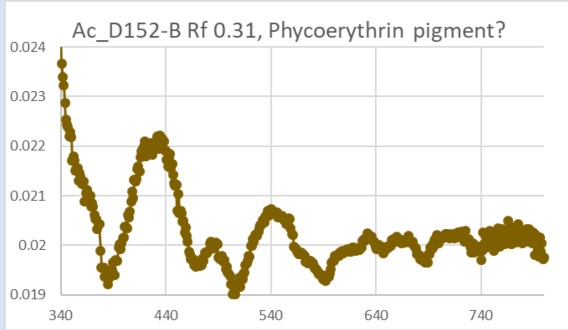
Significance of results:

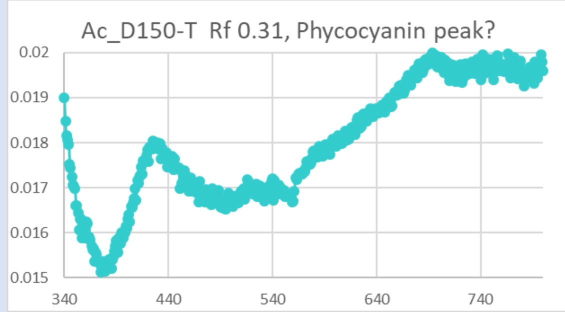
1. Chlorophyll b is absent in algae
2. Normal chlorophyll a does not absorb wavelengths 450-600 nm
3. Algae absorbed energy in 450-600 nm range











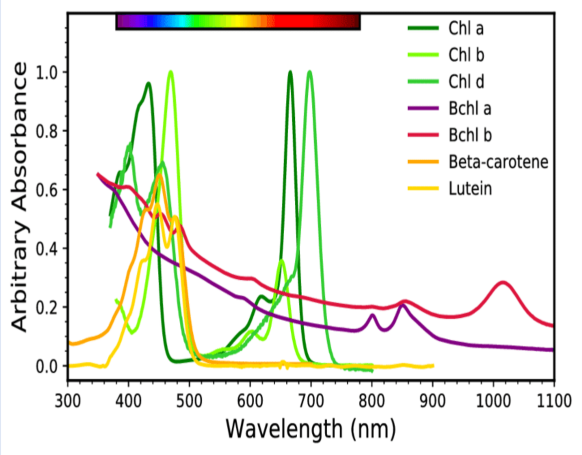
In these examples we can see that Algae appear to have adapted pigments that absorb wavelengths not normally absorbed.

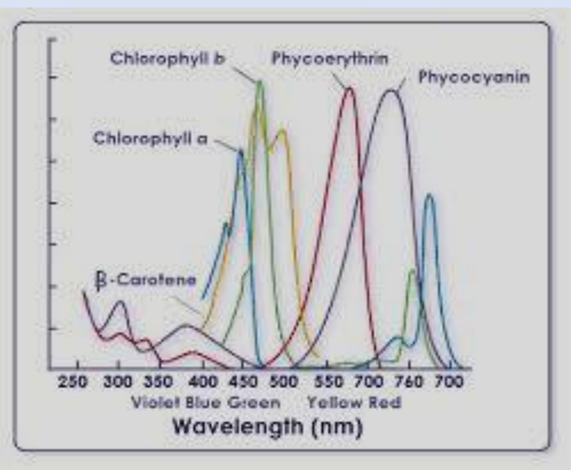
Summary

1. The algae have adapted to extremely low cave lighting
2. The algae pigments have shifted from absorbing standard chlorophyll a and b wavelengths to include additional wavelengths.
3. Attempts by the National Park Service to use lighting that shines at 500-600 nm wavelength will not stop the algae from growing since they can use that spectrum for photosynthesis.
4.

Future studies

- Do additional studies to characterize the pigments
- This includes more UV-VIS spectroscopy, fluorescence, and better standards for the pigments to help identify them.
- Compare the cave algae to surface algae to see how the pigment concentrations changed in response to living in a cave
- Try to raise the cave algae in the lab under low light and full light to see if they can re-configure their pigments to be more like surface algae.





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