

Abstract

Lichens are sensitive bioindicators of nitrogen oxides, common air pollutants in urbanized areas. Crustose lichens are found in polluted areas, while foliose lichens are more common in less polluted areas. Our purpose was to observe how levels of nitrogen oxides affect the biodiversity of lichen in Tenafly Nature Center and Central Park, a low pollution suburban nature preserve and a highly polluted urban center, respectively. Through the collection of lichen samples in both locations, extraction and sequencing of the lichen DNA, and a bioinformatic analysis of the results, we found that *Punctelia rudecta* was a recurring lichen species in both locations. Contrary to our expectations, the only crustose lichens were present in the Tenafly Nature Center, while all of the Central Park lichen species were foliose. These results show that although crustose lichens are known to be the most pollution-tolerant growth form, foliose lichens are also effective in tolerating air pollutants and occupying pollution-dense areas, such as Central Park.

Introduction

- Oxides of nitrogen (NOx): nitrogen dioxide, nitric oxide, nitrous oxide¹
- Released from fuel combustion
- Forms smog, acid rain, and fine particles
- Lichen: a symbiotic association between algae and fungi²
- Lack a vascular system and a waxy outer cuticle
- Absorb nutrients through cortex
- Have a high surface area to mass area
- Three main growth forms of lichens³
- Crustose: indicate highly-polluted air
- Foliose: indicate small amount of air pollution
- Fruticose: indicate clean air



Figure 1. Crustose (left), foliose (middle), fruticose (right)

Objectives

- Purpose:
 - To examine the effect of nitrogen oxides on biodiversity of lichens
 - Number and type of species
 - Growth forms
 - To find a correlation between the identified lichen species and the associated level of nitrogen oxide
- Hypothesis
 - The exposure of nitrogen oxides will reduce the biodiversity of the lichens in Central Park, while Tenafly Nature Center will harbor a more diverse set of species.

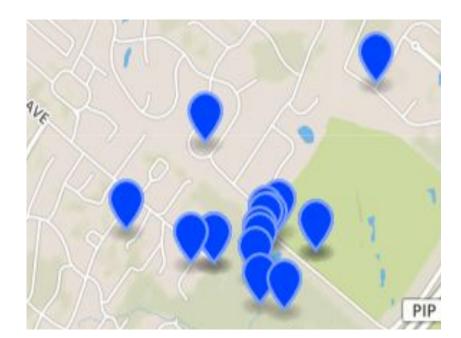
A Biodiversity Study of Lichens in Central **Park-NYC and Tenafly Nature Center**

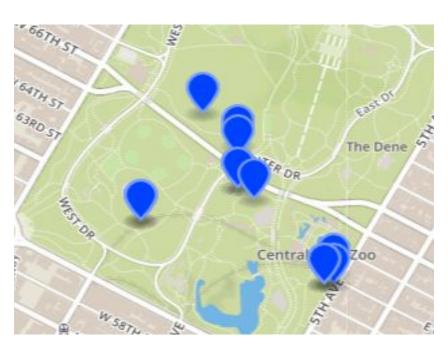
Lindsey Jung¹, Elliot Park¹ Alison Cucco², Anat Firnberg¹, Christine Marizzi² ¹Tenafly High School, ²Cold Spring Harbor Laboratory's DNALC

Methodology

Sample Collection

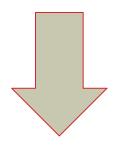
- 17 samples from Tenafly Nature Center (40°55′29″ N 73° 56′42″ W)
- 12 samples from Central Park (40°46′0″ N 73°58′17″ W)
- 1-2 cm in diameter
- Collected in late November and December
- Taken from tree branches, trunks, and rocks
- Washed with acetone and preserved in a refrigerator





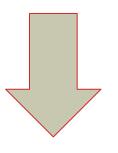
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Figure 2. Tenafly Nature Center and Central Park Sample Locations



Isolating the DNA (at Harlem DNA Lab)

- Defective samples were removed. (8 fungal species found)
- A lysis solution was used and each sample was mechanically ground.
- Each sample was centrifuged to obtain the supernatant.
- The supernatant and silica resin were mixed and incubated to isolate the DNA.
- The primer ITS mix and supernatant were placed in a thermal cycler to amplify DNA.
- Samples were analyzed through gel electrophoresis.
- Usable samples were sent to Genewiz to obtain DNA sequences.



Bioinformatics

- Samples were identified and analyzed using the DNA Subway bioinformatics platform.
- The DNA sequences were trimmed and paired to create consensus sequences.
- The BLAST and BOLD databases were used to match the DNA sequences to lichen species.
- We observed the phylogenetic trees to find similarities between samples.

Results
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GBSO9848-13 Porpidia/316-527 KJ653476.1 porpidia_albocaerul Porpidia albocaerulescens (crustose) KGY-012/340-550 Tenafly Nature Center sample (98.48 % match) HQ270129.1_Chanterelle/831-12 KGY-016/324-534 Tenafly Nature Center sample (96.14% match) KGY-017/8-115 Tenafly Nature Center sample (95.4% match) KJ653477.1 porpidia_albocaerul
KGY-020/263-471 Central Park sample (100% match) KGY-026/327-535 Central Park sample (99,45% match) RRFNL128-15 Punctelia/265-473 <i>R</i> 024456.1 punctelia_rudecta/2 <i>Punctelia rudecta</i> (foliose) KGY-015/327-535 Tenafly Nature Center sample (97,6% match GBLEC3721-13 Punctelia/254-462 GBLEC2516-13 Flavoparmelia/862 <i>Flavoparmelia caperata</i> (foliose) KGY-025/913-1118 Central Park sample (89,19 % match) LIFU007-16 Flavoparmelia/897-1 GBHYP6665-13 Cosmospora/262-48
GBTEL165-13 Phaeophyscia/684-8 Phaeophyscia melanchra (foliose) KGY-029/695-907 Central Park sample (97.78% match) AY498666.1 phaeophyscia_melanc KT695403.1 phaeophyscia_rubrop Phaeophyscia rubropulchra (foliose) KGY-022/501-713 Central Park sample (99.63% match) DQ084142.1 flavoparmelia_caper
GBTEL1342-14 Physcia/495-707 Physcia stellaris (foliose) KGY-019/557-769 Central Park sample (97.29% match) GBTEL1543-15 Physcia/251-463 AY860549.1 physcia_stellaris/2 Physcia stellaris (foliose)

^I KGY-028/656-868 Central Park sample (82.52% match)

Figure 4. Phylogenetic tree of our samples, given by DNA Subway



[2] USDA Forest Service. Lichen Habitat. Retrieved October 5, 2018, from https://www.fs.fed.us/wildflowers/beauty/lichens/ habitat.shtml

[3] APIS (2016) *Impacts of air pollution on Lichens and* Bryophytes (mosses and liverworts). Retrieved October 10, 2018, from http://www.apis.ac.uk/impacts-air-pollution-lichens-andbryophytes-mosses-and-liverworts

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Results (cont.)

Growth Forms of Lichen

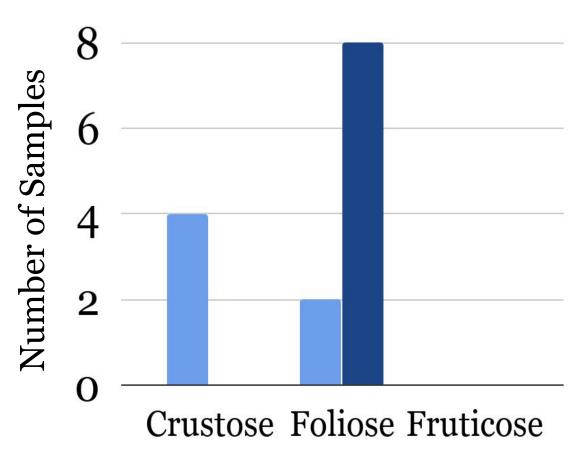


Figure 5. Growth forms of lichens identified in Tenafly Nature Center (light blue) and **Central Park** (navy)

Conclusion

Results did not support our hypothesis and predictions • No clear correlation between the identified lichen species and the associated level of nitrogen oxide • Foliose lichens were identified in Central Park. • Crustose lichens were identified in Tenafly Nature Center.

• Most tolerant of air pollution - expected in Central Park, not Tenafly Nature Center

Future Work

• If continued, we

- will take a measurement of NO, and considering other pollutants
- To find a clear distinction between levels of pollutants in Central Park and Tenafly Nature Center
- will consider soil and water quality
- wil have a larger sample pool
- will consider a more diverse set of places
- will improve accuracy in collected samples (no fungal species)

References

[1] Icopal Ltd. *Nitrogen Oxide (NOx) Pollution*. Retrieved October 5, 2018, from http://www.icopal-noxite.co.uk/nox-problem/noxpollution.aspx

Acknowledgements