

# Analysis of Moss Biodiversity in Manhattan Parks



NEW YORK
BOTANICAL GARDEN
EST 1891

Manhattan Center



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#### Abstract

Mosses are small, spore-bearing land plants which are an important component of ecosystems. Our study examines moss biodiversity in NYC parks, based on levels of human disturbance. We sampled 43 mosses from 3 sites: 97th Street Transverse (high disturbance, 14 samples), north Central Park (medium disturbance, 13 samples), and Inwood Hill Park (low disturbance, 16 samples). DNA barcodes for the plastid gene rbcL were sequenced for 37/43 samples (86%). Samples were identified using BLAST results for barcode sequences,

suggestions from the iNaturalist computer vision model, and morphology. We identified 77% of the specimens to the genus or species level. We found mosses from 15 different genera, with the highest number of unique genera in the site with the lowest disturbance. However, more sampling is needed to examine the relationship between human disturbance and moss biodiversity. We concluded that DNA barcodes and iNaturalist are valuable tools for identifying mosses.

#### Introduction

Mosses are small, non-flowering plants that form large green mats or clumps, often in shady or damp areas.

Although the average person might think all mosses look the same, there is actually a lot of diversity within the group.

There are an estimated 461 moss species present in New York State.

Our study has two main objectives:

## To explore the effects of human disturbance on moss biodiversity.

- Mosses can be indicators of air pollution based on changes in color due to chloroplast damage and declining growth trends.
- Mosses are also sensitive to temperature increases,
   whether from climate change or urban heat island effects.

#### To compare methods for identifying mosses.

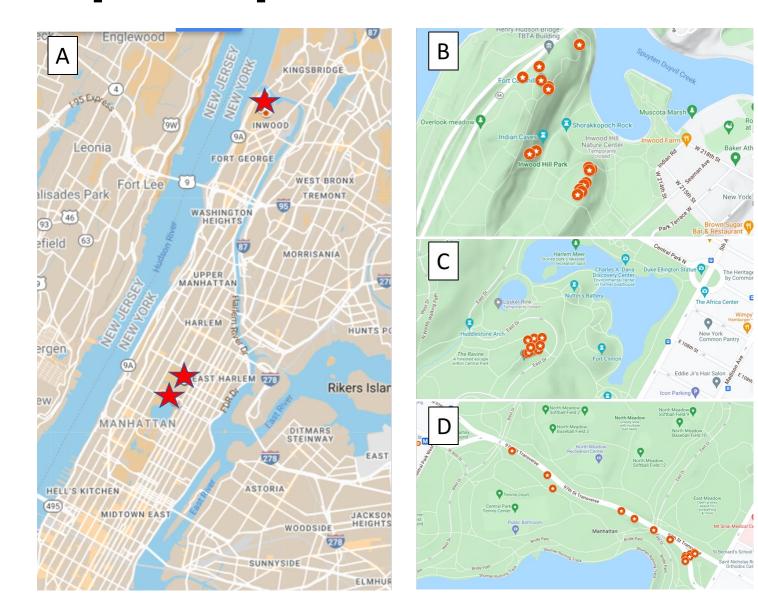
- Morphology is the most traditional method, and is based on characteristics such as the size and shape of leaves and sporophytes, some of which may only be observed using a microscope.
- DNA barcoding identifies specimens by comparing a short, standardized DNA sequence to a reference database.
- iNaturalist, a community science platform, enables users to observe and document wild flora and fauna, and includes a computer vision program to identify organisms based on photographs.





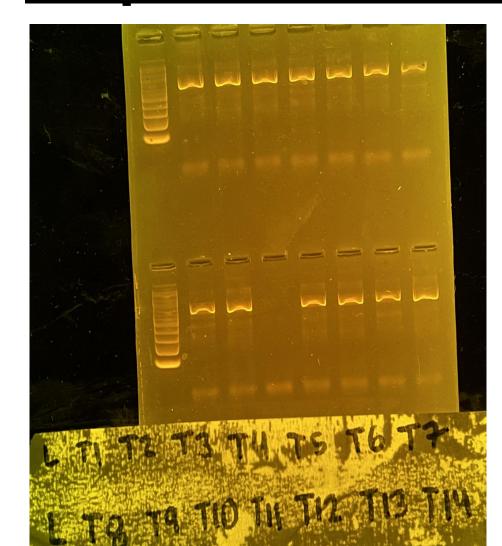
## Methodology

### **Step 1: Specimen Collection**

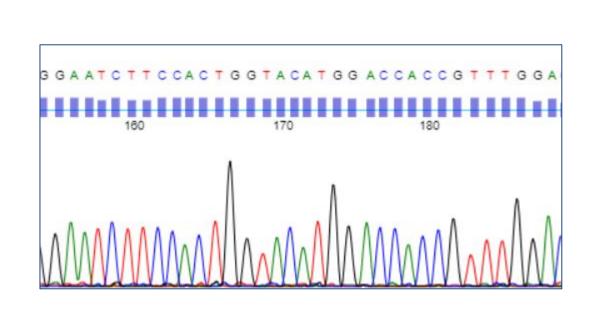


High disturbance: 97th Street Transverse; collected 14 samples Medium disturbance: northern Central Park; collected 13 samples Low disturbance: Inwood Hill Park; collected 16 samples

## Step 2: DNA Extraction & Sequencing



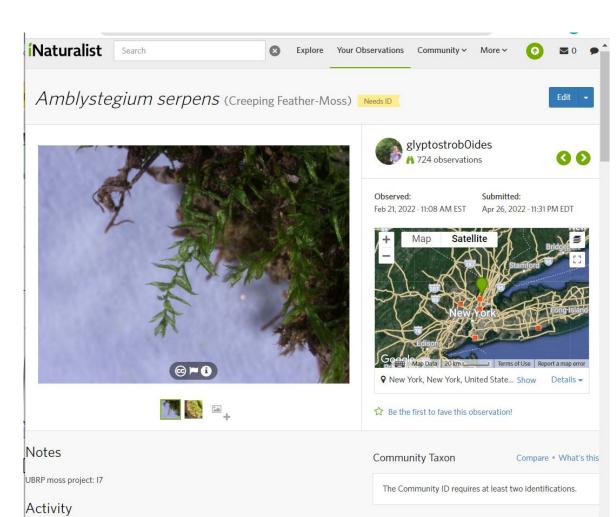
- DNA extraction
- PCR to amplify *rbcL* barcode
   Confirm success with gol
- Confirm success with gel electrophoresis
- Sent out for sequencing



## **Step 3: Data Analysis**

Sequences producing significant alignments		Download Y		Sel	ect co	lumns	∨ Show	iow 1	00 🕶
2	select all 100 sequences selected	GenBan	ı <u>k</u> G	<u>raphic</u>	s <u>Dis</u>	stance	tree of re	esults	MSA Viewe
	Description	Scientific Name	Max Score	Total Score	Query Cover	E value	Per. Ident	Acc. Len	Accession
	Atrichum undulatum voucher CUH D2 B0049 ribulose-1,5-bisphosphate carboxylase/oxygenase large subunit	Atrichum undulat	1050	1050	89%	0.0	100.00%	568	MH479287.1
/	Atrichum angustatum voucher H:Norris 83235 ribulose-1,5-bisphosphate carboxylase/oxygenase large subunit	. Atrichum angust	1026	1026	87%	0.0	99.82%	683	GU569410.1
~	Atrichum angustatum voucher Goffinet 10582 chloroplast, complete genome	Atrichum angust	1098	1098	94%	0.0	99.67%	125602	NC_058541.1
~	Atrichum flavisetum voucher H:Ignatova 06 02 ribulose-1,5-bisphosphate carboxylase/oxygenase large subunit	.Atrichum flaviset	1026	1026	88%	0.0	99.64%	686	GU569411.1
~	Atrichum angustatum ribulose-1,5-bisphosphate carboxylase/oxygenase large subunit (rbcL) gene, partial cds;	Atrichum angust	1050	1050	90%	0.0	99.48%	1337	DQ645986.1
~	Atrichum selwynii ribulose 1,5-bisphosphate carboxylase/oxygenase (rbcL) gene, partial cds; chloroplast	Atrichum selwynii	1035	1035	89%	0.0	99.47%	1335	GU295870.1
	Atrichum undulatum voucher Hyvonen 6170 ribulose 1,5-bisphosphate carboxylase/oxygenase large subunit (r	. Atrichum undulat	1035	1035	89%	0.0	99.47%	1132	AY118236.1

- DNA Subway used to process raw sequencing data and to create maximum likelihood phylogeny based on barcode sequences
- BLAST used to compare DNA barcodes to entire NCBI database
- Uploaded photos taken in the field and with a dissecting microscope to iNaturalist, and used the computer vision program to identify them.

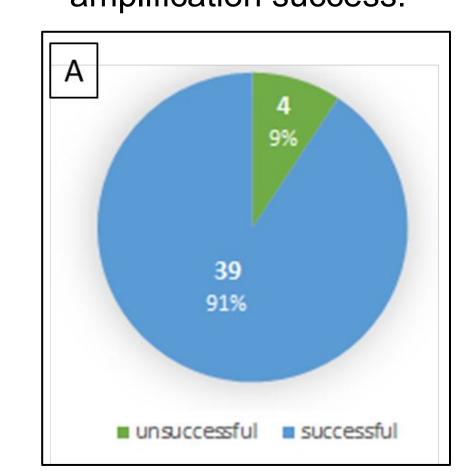


## Step 4: Comparison of ID Methods

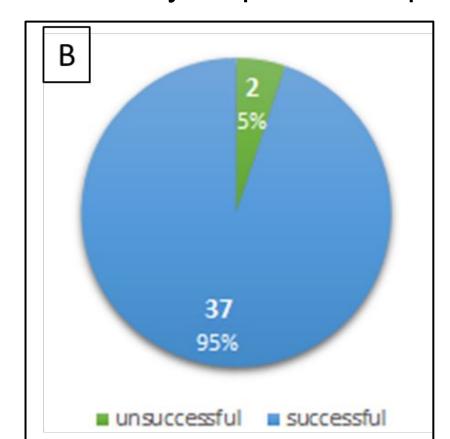
Determined identifications based on consensus of the three methods (morphological, DNA barcoding, iNaturalist computer vision)

#### Results

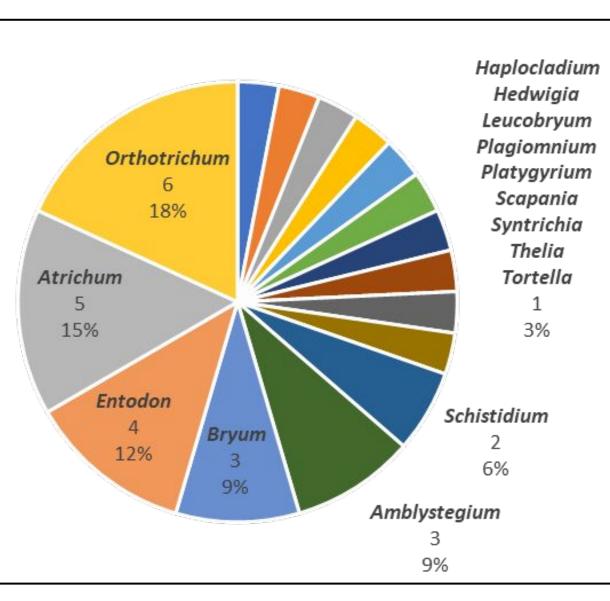
## DNA extraction and PCR amplification success.



Sequencing success of the 39 successfully amplified samples.

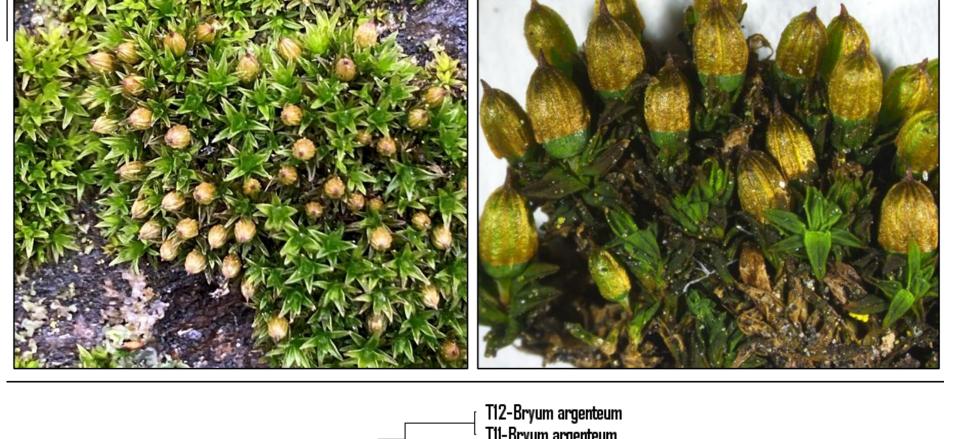


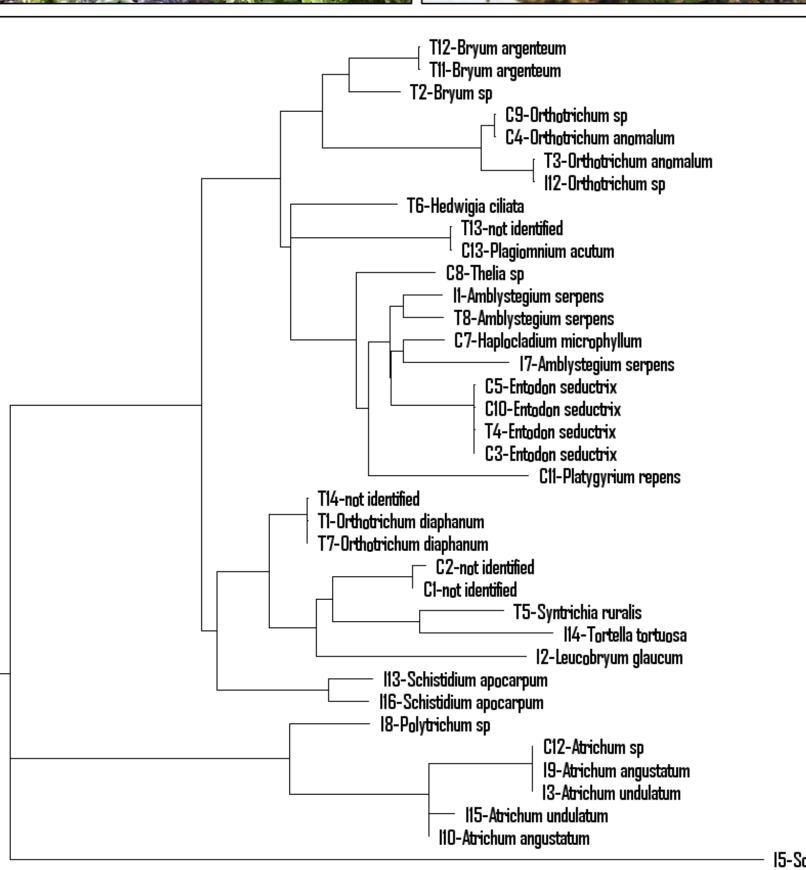
We were able to identify 27 samples to thee species level (63%), and another 6 to the genus level (14%). We were unable to identify 10 specimens (23%), 6 of which failed to sequence. Overall, factors which made samples unable to be identified included missing morphological characters (such as reproductive structures), and low-quality samples.



A breakdown of the genera of mosses collected from all 3 sites.

#### Orthotrichum anomalum (anomalous bristle-moss, sample T3)





Maximum likelihood phylogeny constructed for barcode sequences using DNA Subway. Notice that samples identified as the same species are grouped together on the tree.

#### **Discussion and Conclusions**

Morphology identifications were difficult, especially given our lack of expertise. According to the bryologist who assisted, some species are differentiated by characteristics only visible with a microscope, or at a certain stage in their life cycle.

Identifications with DNA barcoding were surprisingly challenging, due to limitations including incomplete reference database and high similarities between species. We suggest multiple DNA barcodes are necessary for more accuracy in moss identifications.

iNaturalist identification was easier than anticipated. The iNaturalist computer vision program performed better on the specimens which had high-quality photographs.

Mosses found in the highest human disturbance site were the least healthy-looking. Also, we found the highest number of unique genera in our lowest disturbance site.

A more thorough survey would need to be conducted to fully understand the moss biodiversity and the effects of human disturbance in these sites.

#### Leucobryum albidum (pincushion moss, sample I3)





#### References

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