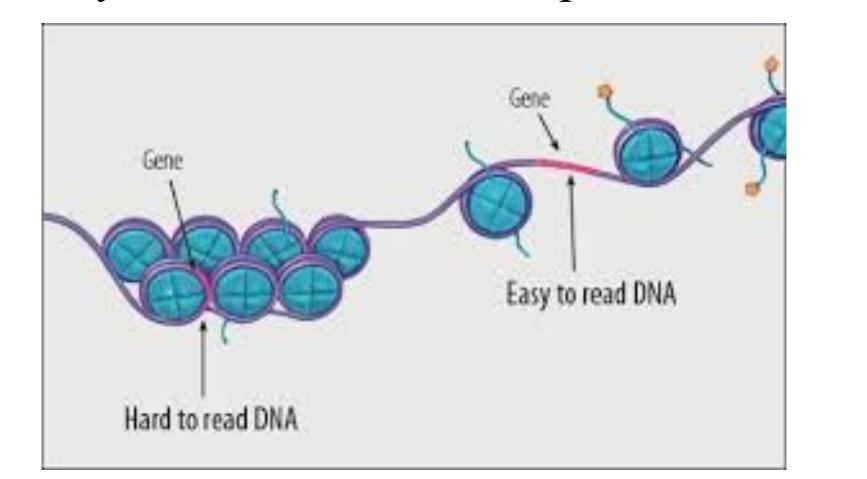
The Walking Forest: Epigenetic Response to Environmental Stress in Picea Glauca Vanderbush, K., Katz, J., Obioha, C., Nicoloff, M., DeCarlo, L., Gentile, D., Cox, R.L. Lisman Laboratory for Molecular Ecology, Riverdale Country School, New York, NY USA

• Epigenetics is the study of differentiated gene expression without differences in the DNA sequence. Methylation is an epigenetic process that inhibits gene expression. Global DNA methylation refers to the percent of methylated DNA in the sample.



- Genomic DNA was isolated from the needles.
- Global DNA methylation percentages were measured for each sample using Zymo's Global Methylation Assay.
- Global DNA methylation was analyzed for trends both across each site individually and after grouping sites by similar environmental

Figure 3: Proteins interact with DNA strands changing how they are read and expressed.

 Spruce needles are exposed to harsh arctic winters. Methylation in these needles may fluctuate in response to the stresses of living at or close to treeline.

> Embryonic (First year)



- conditions.
- Methods for the study were analyzed using additional data from the Black Rock Forest.

Results

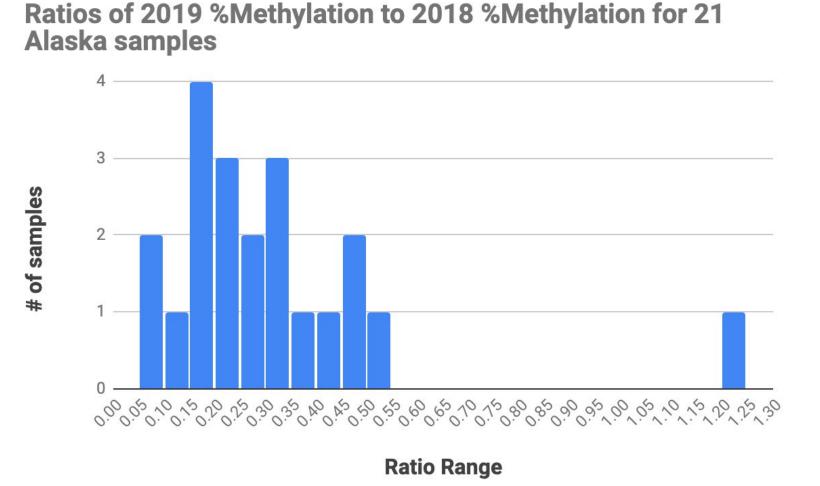
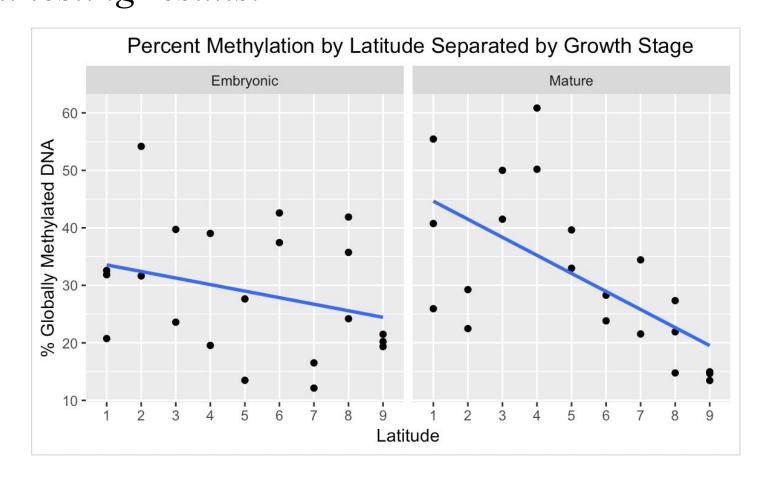


Figure 5: Ratios of experimentally derived methylation levels in samples stored in freezer over 1 compared with original testing results.



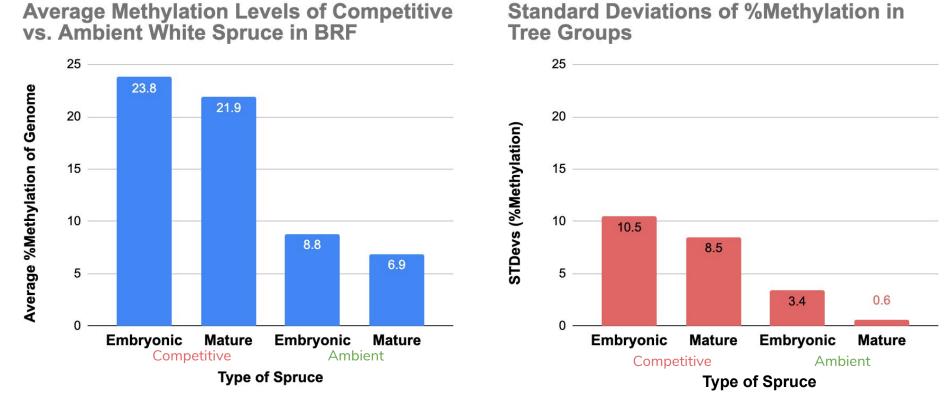


Mature (3rd year)

Figure 4: Embryonic and mature growth on a spruce branch

Methods

Needle samples from spruce tree individuals at eight latitudes in the Alaskan Boreal Forest and two sites in Black Rock Forest were collected.
Individuals at most sites are monitored by Kevin Griffin of the Lamont-Doherty Earth Observatory at Columbia University. Figure 6: 2018 latitudinal methylation trends compared between embryonic and mature growth. Latitude 1 is North Treeline (AK), 2-8 represent the Boreal Forest (AK), and 9 is Black Rock Forest (NY).



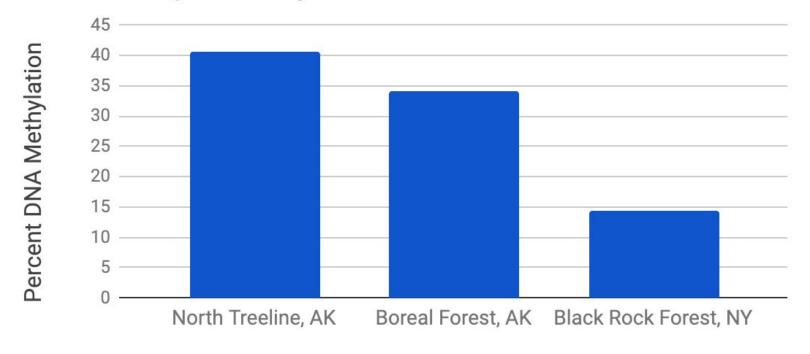
Figures 7 & 8: Mean and standard deviation of methylation levels in two Black Rock Forest Picea groups



Research Goals

- As treeline moves northward with rising temperatures, we strive to determine the nature of spruce trees' adaptations to climatic stress.
- We hypothesized that trees in more stressful environments, such as the northern edge of

Percent of Global DNA Methylation of Old Arctic Spruce **Growths Organized by Location**



Latitude Group

treeline in Alaska, would exhibit higher global DNA methylation than trees in less stressful environments (i.e: Black Rock Forest) and that these trends would appear only in mature growth needles.

• This year we also aimed to analyze the efficacy of our methodologies and sampling protocols through several studies such as re-analyzing DNA from arctic samples stored in a -20C freezer.

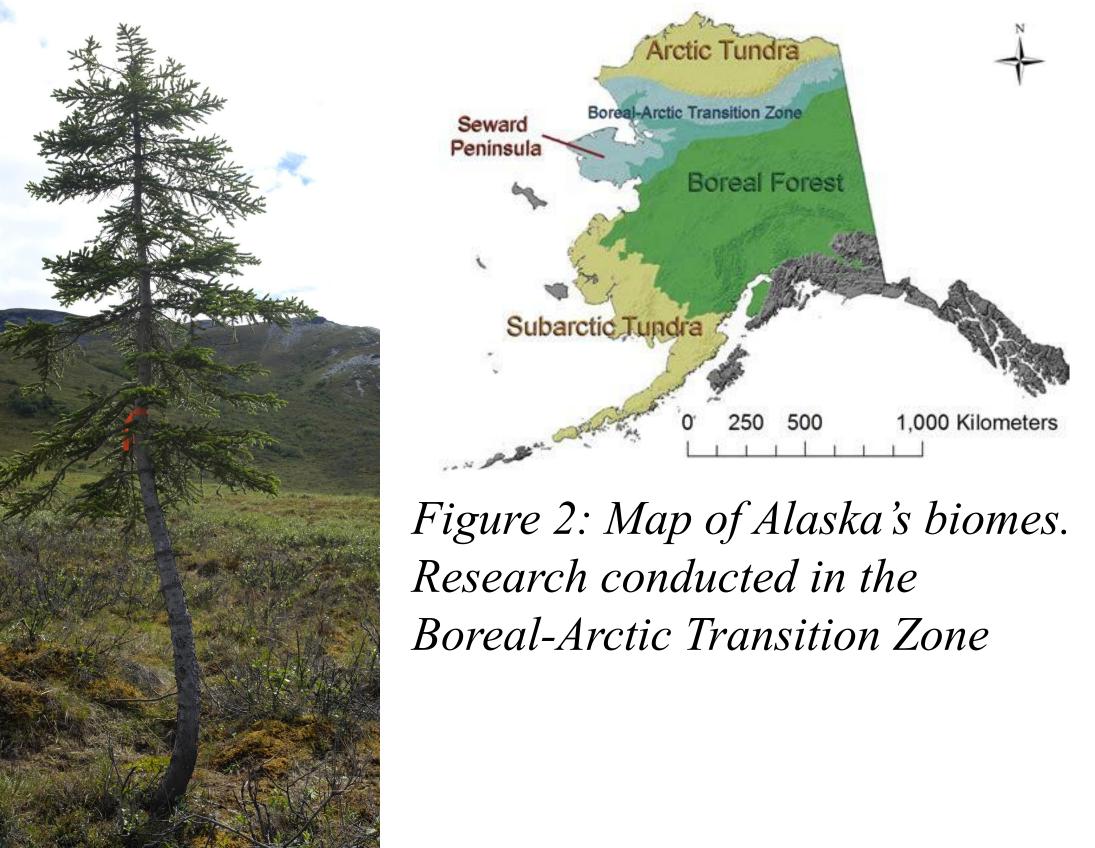
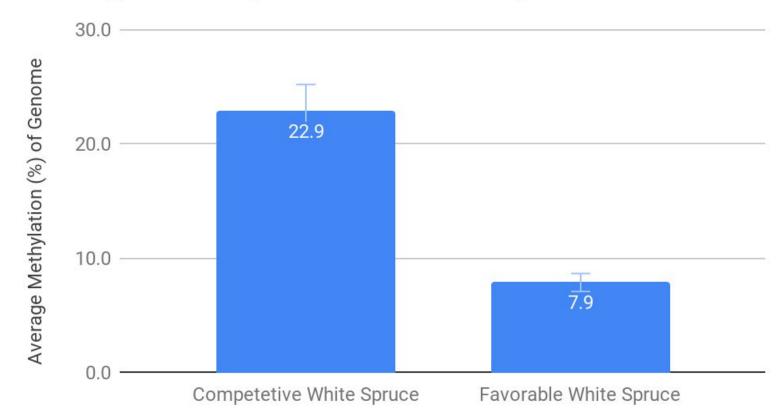


Figure 9: Average percent of global DNA methylation across a latitudinal transect from Northern Alaska to New York.



Higher methylation in the "Competitve" Picea

Figure 10: Average percent of global DNA methylation in high-competition white spruce vs. ambient "favorable" white spruce

Conclusions

• Stored samples, embryonic growth, and samples from more stressed populations display more erratic behavior in methylation, making it harder to find precise trends between samples. • Various environmental stressors may cause organisms to shut down "excess" gene transcription and protein production through methylation, a mechanism essential to their survival.

Figure 1: The northernmost spruce tree in Alaska. Northern edge trees are highly exposed, resulting in more harmful environmental stressors.

Epigenetics and Arctic Spruce

• Arctic spruce, specifically *Picea glauca*, is a foundation species of the Boreal Forest ecosystem.

Future Research

- We will use our methods findings to enhance our strategy for collecting and analyzing data.
- We will search for specific genes being shut off or modified.

Acknowledgements

- Marjot Foundation
- Black Rock Forest Consortium
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