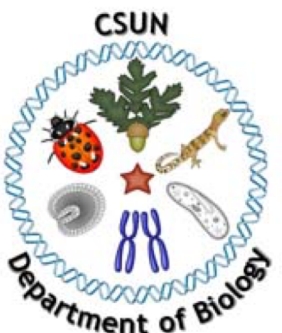




The Search for Fungi in Ancient Permafrost

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INTRODUCTION

Permafrost (permanently frozen soil), is widespread across the Arctic and sub-Arctic. Despite harsh conditions, such as extreme cold and low nutrient content, microbial communities are able to persist and reproduce within permafrost^{1,2,3}. These communities are of interest for two main reasons. First, climate change is triggering widespread permafrost thaw. This releases undecomposed carbon to the action of microbial communities, who degrade the carbon and “breath” it into the atmosphere as carbon dioxide and methane². Second, it is relevant to the field of exobiology. If life exists outside of our planet, it likely is in frozen conditions since the most of the planets, moon, comets, and asteroids persist at sub-zero temperatures³. Understanding how microbes are able to survive in permafrost for millennia may give us clues as to what could enable survival in other parts of our solar system¹.

Since most microbes living in permafrost cannot be cultured in a laboratory setting, metagenomic analysis is often used to study these populations⁴. Metagenomics is a process by which all DNA is extracted from environmental samples and sequenced. DNA sequences can then be compared to databases to determine taxonomic origin and function. Current research investigating microbial communities in permafrost has mainly focused on bacterial and archaeal communities, leaving eukaryotic populations largely understudied. To contribute to our understanding these microbial communities I aim to explore the eukaryotic microbes that persist in Siberian permafrost and environmental conditions that shape these populations.

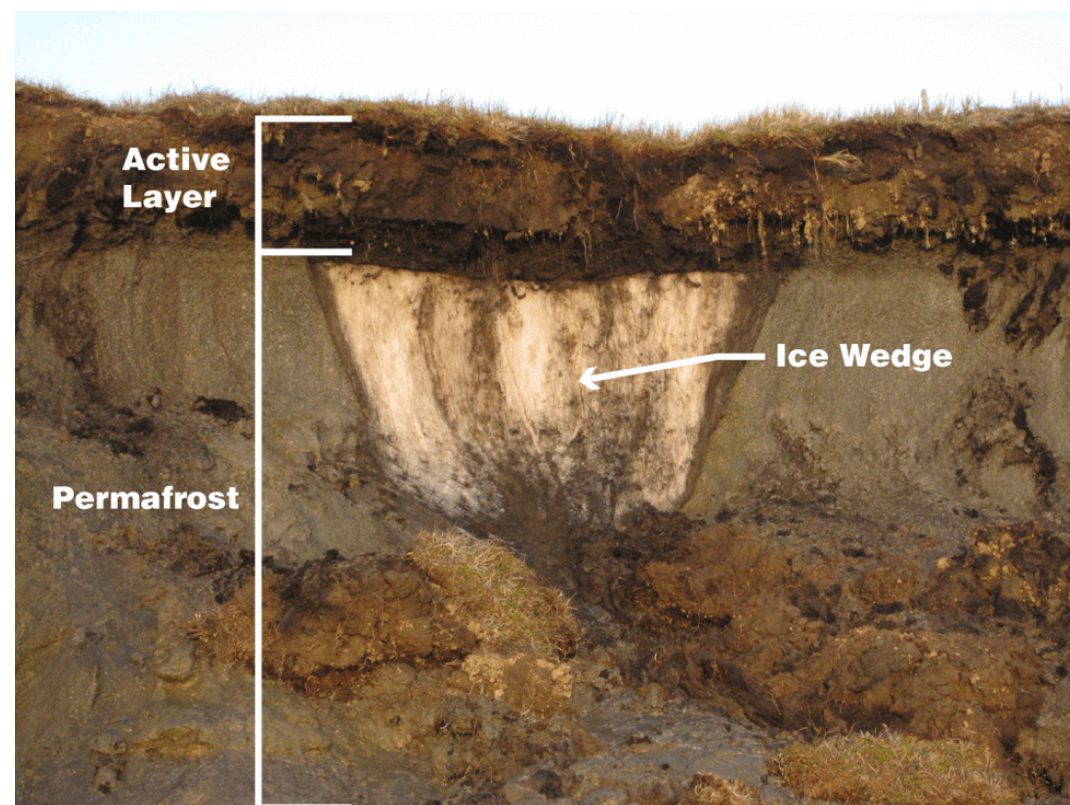


Figure 1. The layers of permafrost. Photo credit: Benjamin Jones, USGS. Public domain

QUESTIONS

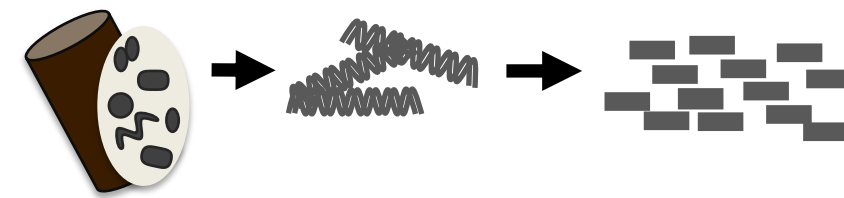
What eukaryotes are able to survive in permafrost?

What traits enable survival under frozen conditions throughout geological time?

Do factors such as permafrost location, depth and age have a significant effect on eukaryotic diversity and abundance?

METHODS

- Isolate and sequence community DNA from permafrost



- Identify and classify eukaryotic sequences by comparing them to the NCBI refseq databases with Kaiju



- Sorted classified reads by taxonomic group and compared the identity and abundances of eukaryotic taxa between samples



RESULTS

Relative Abundances of Eukaryotic Phylum

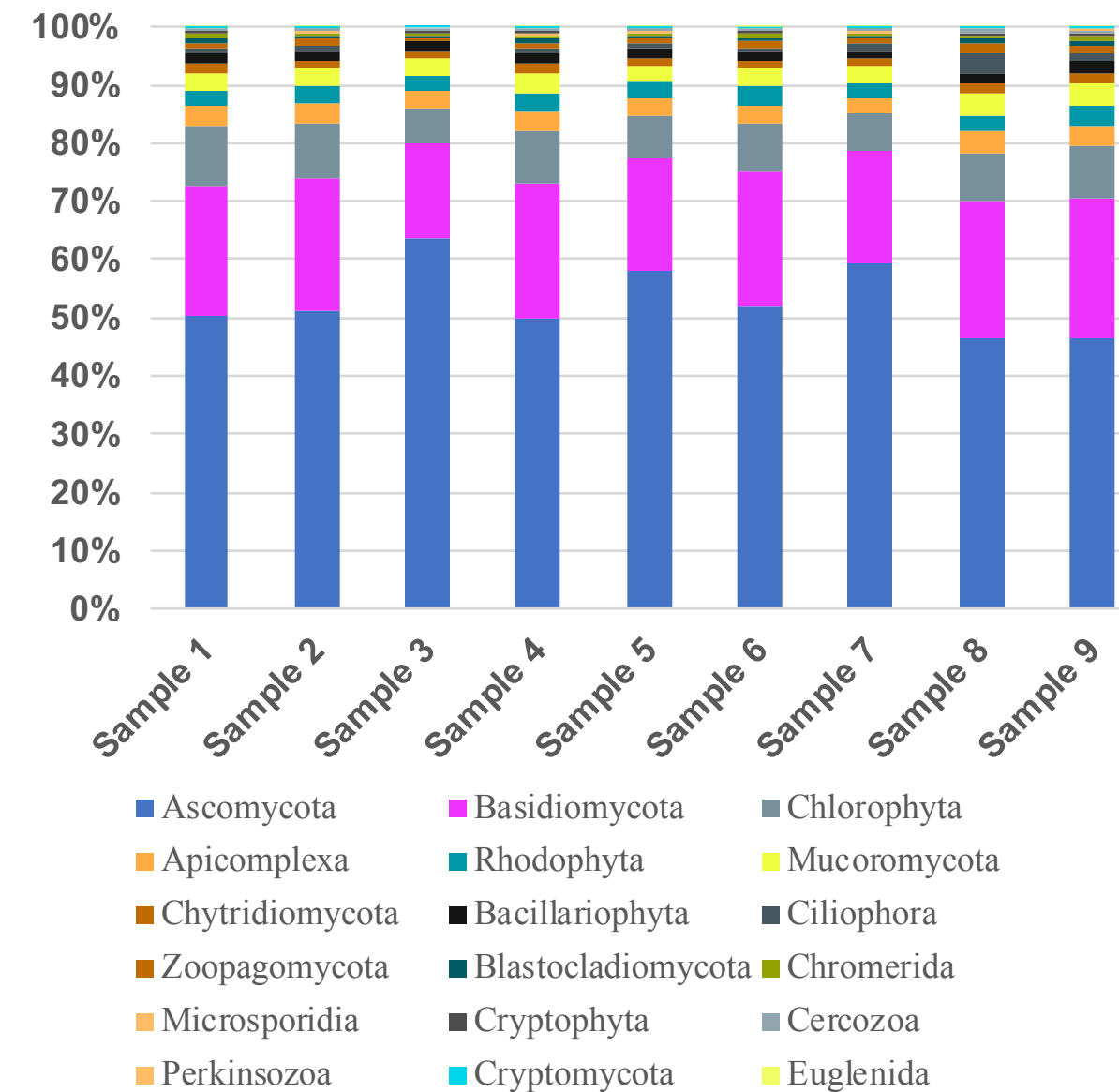


Figure 3. Graph showing the relative abundances of eukaryotic phyla present in Siberian permafrost samples

Relative Abundances of Eukaryotic Phylum

Phylum	Sample 1	Sample 2	Sample 3	Sample 4	Sample 5	Sample 6	Sample 7	Sample 8	Sample 9	Sample 10	Sample 11	Sample 12	Sample 13	Sample 14	Sample 15	Sample 16	Sample 17	Sample 18	Average
Ascomycota	50.3	51.1	63.6	49.8	57.9	51.7	59.4	46.3	46.4	47.6	49.8	54.5	56.4	59.3	49.1	55.1	48.8	48.6	52.6
Basidiomycota	22.5	23	16.0	23.5	19.5	23.2	19.5	23.8	24.0	25.0	23.8	22.4	20.8	22.6	23.2	21.7	20.0	20.5	21.9
Chlorophyta	10.0	9.5	6.4	8.8	7.3	8.2	6.1	8.2	9.2	7.9	7.9	6.7	6.9	4.0	6.0	9.2	12.4	11.9	8.1
Apicomplexa	3.5	3.3	2.8	3.5	2.9	3.3	2.7	3.8	3.4	3.7	3.1	2.9	2.7	2.8	4.0	3.1	3.5	4.2	3.3
Rhodophyta	2.9	3.1	2.6	3.1	2.9	3.1	2.5	2.8	3.5	3.4	3.6	3.2	2.9	5.2	4.1	2.4	2.3	3.6	3.2
Mucoromycota	2.7	2.8	30	3.3	2.7	3.0	2.9	3.8	3.5	3.1	3.0	2.6	2.9	1.2	4.6	3.3	3.6	3.3	3.1
Chytridiomycota	1.7	1.5	1.4	1.6	1.4	1.5	1.5	1.7	2.1	1.9	1.6	1.5	1.3	1.5	2.1	1.3	1.7	1.5	1.6
Bacillariophyta	1.8	1.4	1.4	1.7	1.5	1.8	1.3	1.5	1.9	1.8	1.6	1.4	1.7	1.4	1.4	0.4	1.7	1.6	1.5
Ciliophora	0.8	0.9	0.3	0.9	0.7	0.7	1.1	3.8	1.4	1.6	2.3	1.4	0.7	0.4	0.5	0.6	2.2	1.5	1.2
Zoopagomycota	1.2	1.2	0.5	1.1	0.1	1.1	0.9	1.5	1.4	1.2	1.1	1.2	0.9	0.3	0.9	0.3	1.1	0.7	1.0
Blastocladiomycota	0.8	0.4	0.1	0.7	0.5	0.7	0.5	0.8	0.6	0.8	0.7	0.9	1.5	0.4	0.7	1.6	0.9	1.2	0.8
Chromerida	0.7	0.6	0.7	0.6	0.5	0.5	0.5	0.5	0.8	0.6	0.6	0.4	0.6		1.3	0.5	0.4	0.2	0.6
Microsporidia	0.2	0.3	0.1	0.3	0.2	0.2	0.2	0.2	0.3	0.4	0.2	0.1	0.2		0.4	0.2	0.3	0.5	0.2
Cryptophyta	0.4	0.2	0.4	0.4	0.2	0.5	0.3	0.3	0.4	0.4	0.3	0.4	0.2		0.8		0.3	0.3	0.3
Cercozoa	0.3	0.2	0.4	0.4	0.3	0.3	0.3	0.9	0.5	0.3	0.2	0.2	0.1	0.4	0.4	0.1	0.6	0.3	0.3
Perkinsozoa	0.2	0.1		0.1	0.1	0.2	0.1	0.2	0.2	0.3	0.2	0.1	0.1	0.3		0.2		0.1	0.1
Cryptomycota	0.1	0.2	0.1	0.1	0.2	0.1	0.1	0.2	0.3	0.1	0.1		0.1			0.1	0.2		0.1
Euglenida														0.1	0.4		0.1		0.1

Figure 2. Table showing the relative abundance of eukaryotic phyla present in Siberian permafrost samples

CONCLUSIONS

The two most abundant phyla were Ascomycota (comprising just over 50% of the eukaryotic population) and Basidiomycota at approximately 21%. Members of these fungal phyla contain many spore formers, which suggests that dormancy may contribute to their ability to survive in harsh conditions.

Chlorophyta, commonly known as green algae, represented 8% of the population. This was not expected since Chlorophyta are typically photosynthetic organisms. Further research is required to determine whether they are relics from prior to permafrost formation or if they have adaptations allowing them to survive in a completely dark underground environment such as permafrost.

Population size and diversity of Eukaryotes did not vary significantly by permafrost location, depth, or age. This was surprising since bacterial and archaeal vary significantly by location. Further analysis at the class and order level will likely give us better insight into the variations between these communities.

FUTURE DIRECTION

Do eukaryotic populations vary by location with respect to class or order?

What adaptations help these populations to persist through such harsh conditions?

Further research into the viability of these organisms and their ability to adapt to changing environments is also required. For example, when permafrost thaws what role might they play in nutrient cycling? Since many microbial fungi form symbiotic relationships with plants and root systems, what effect will they have on local plant populations?

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