

Just Deserts: Exploring the Diversity of Melanized Fungi in Rocks and Biological Soil Crusts



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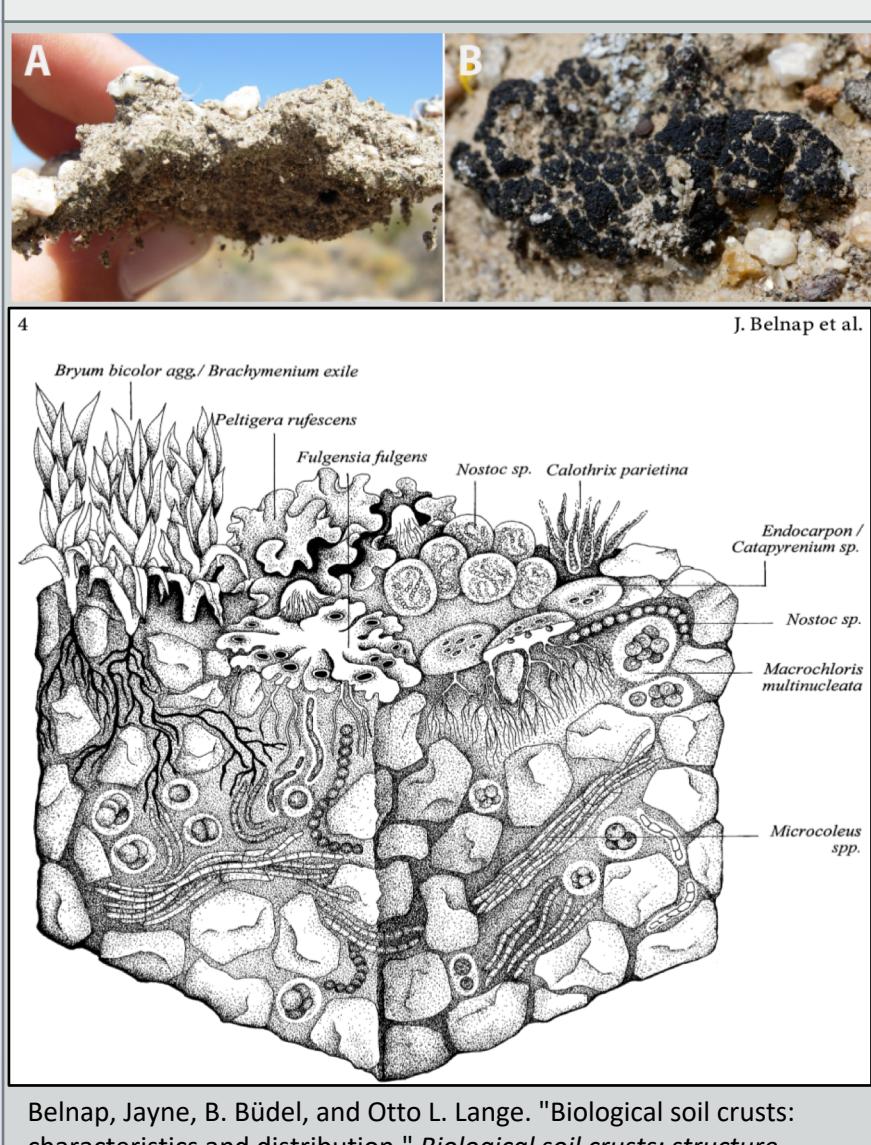


SIGNIFICANCE:

Melanized fungi are found all over the world in many different extreme environments (salters, within and on top of rocks, human patients, plant infections, dishwashers) and now desert biological soil crusts!

Culturing, isolating and understanding them from desert biocrusts could help us understand their roles in other extreme environments.

WHAT ARE BIOCRUSTS? BIOLOGICAL SOIL CRUSTS? CRYPTOGAMIC SOILS?



Biocrusts are soil with enough torsion strength to stay in shape when picked up

LAC- Light Algal Crusts- are early composed crusts

CLC- Cyano-Lichen Crusts- are late composed crusts

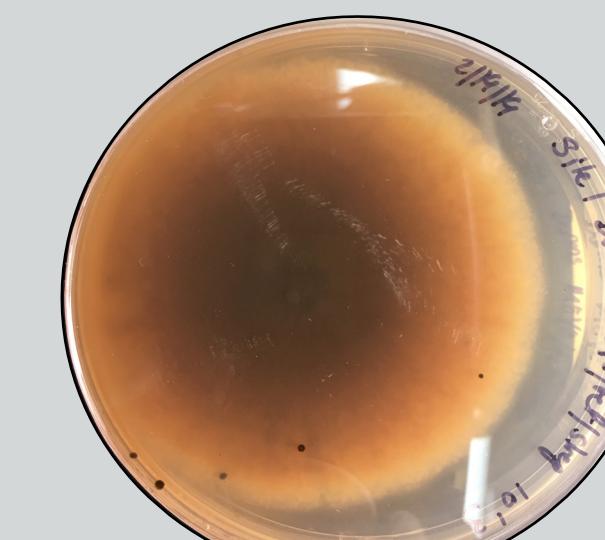
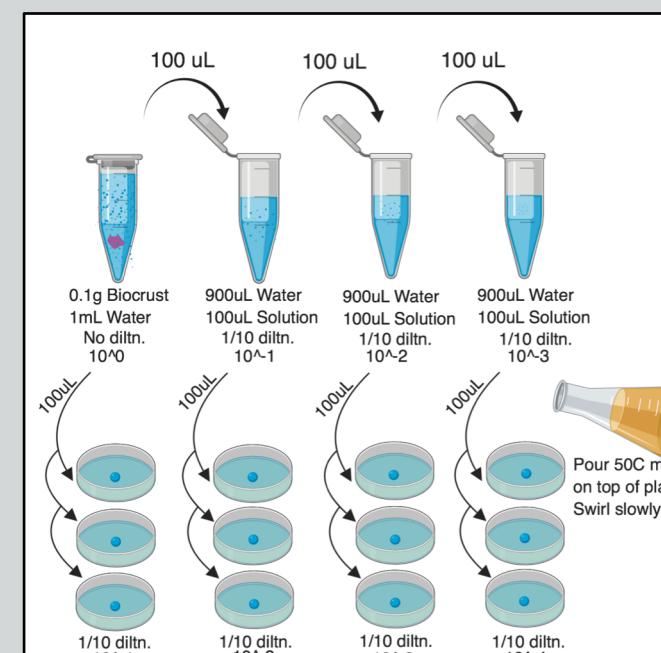
Biocrusts are an assemblage of different microorganisms working together. They help protect desert soils from weathering and are good water retainers.

OBJECTIVES:

- Role-** Observing Ecological and Metabolic Role of Melanized Fungi in Biological Soil Crusts.
- Organism-** Culturing and Utilizing Comparative Genomics Tools to Compare Different Melanized Fungi. Crusts from varying deserts will be/have been collected (Mojave, Chihuahuan, Joshua Tree, Boyd Deep Canyon, Sonoran Desert). Culture Independent analysis (NGS) established the abundant bacteria and fungi in these crust types. Culture Dependent will be done to isolate, sequence and compare to other melanized fungi.
- Gene/Metabolome-** Metabolomic Analysis of Melanized Fungi from Different Desert Regions Undergoing Different Stresses.

METHODS:

- Culturing fungi from desert biocrusts collected from Boyd Deep Canyon and Joshua Tree National Park.



- Melanized fungi were cultured and isolated from crusts- Chaetothyriales and Dothideomycetes are the two distinct clades that Melanized fungi are found in. From each culturing attempt, both filamentous fungi and melanized fungi are collected, grown on slants, small plates and frozen in -80C.
- Through culturing, extracting, PCR amplification and BLAST- identified different strains, majority of melanized fungi were Dothideomycetes. PCR was used to amplify the full ITS region (some LSU, ITS1, 5.8S, ITS2, SSU).
- High concentration DNA was isolated and 4 melanized fungi have now been deep sequenced (1 Nanopore, 3 Illumina). Comparative analysis will be completed on these soon. More strains will be sent to be Illumina sequenced. This way can use more than ITS to create trees.
- Alpha Diversity of melanized fungi was pulled from Joshua Tree Samples to generate a comparison summary

COMPARISON TREE

434 Gene Tree of Dothideomycetes vs. Chaetothyriales

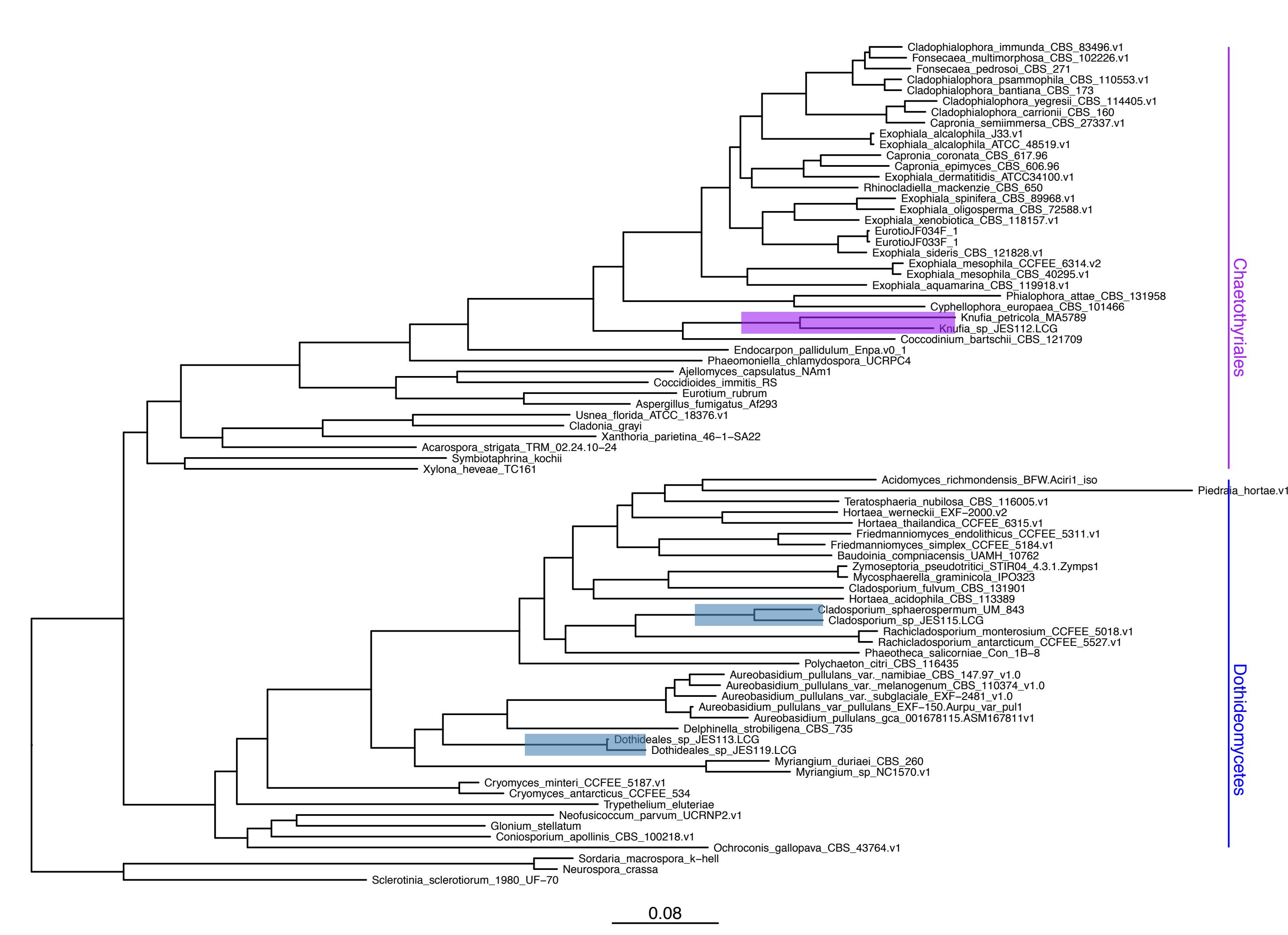


Figure 1: This tree was generated using 434 conserved genes. Highlighted strains : JES_119, JES_113, JES_112, JES_115 are melanized fungi that have been isolated from Boyd Deep Canyon and Joshua Tree National Park. These strains have been sequenced with Illumina MiSeq, while JES_119 was sequenced with Nanopore technology.

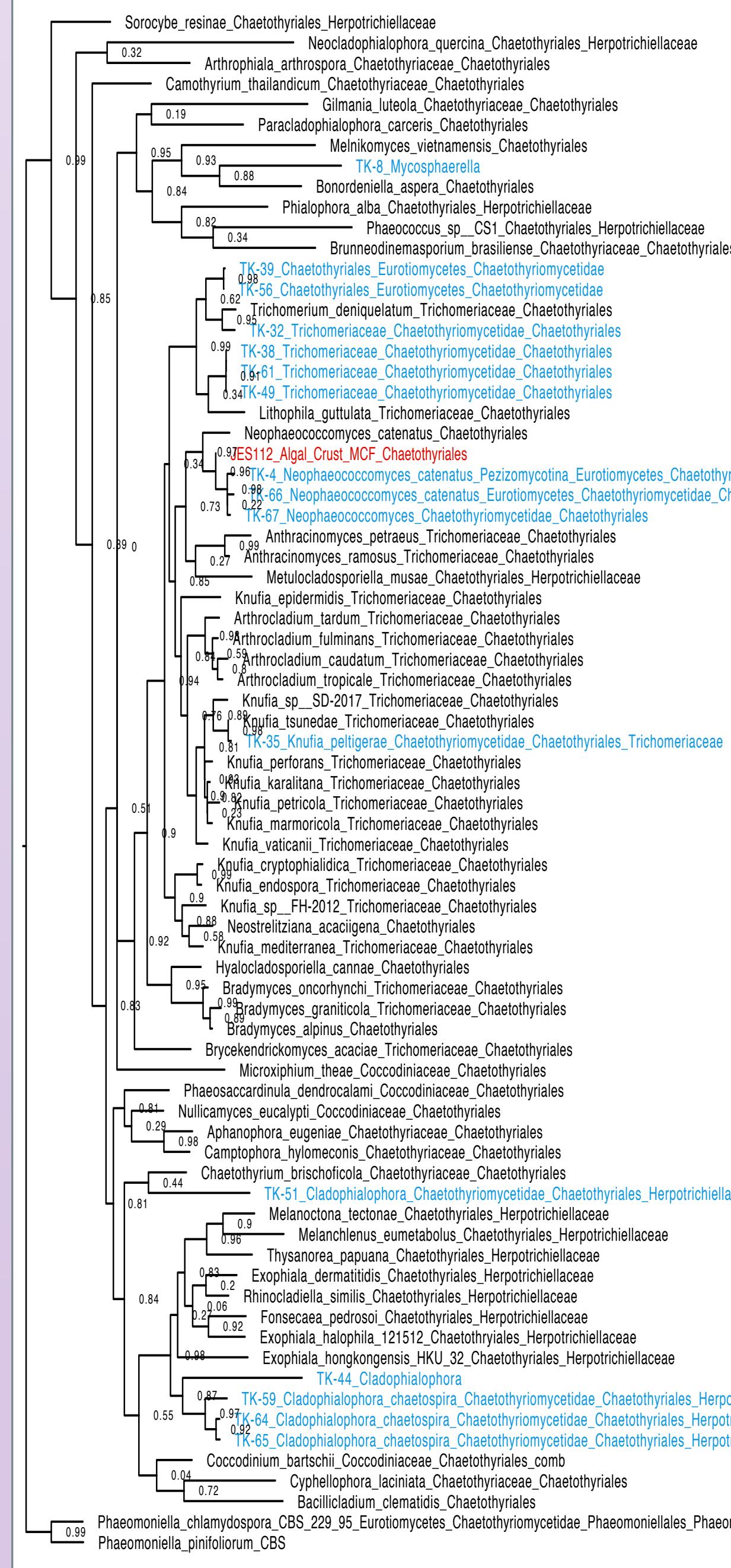
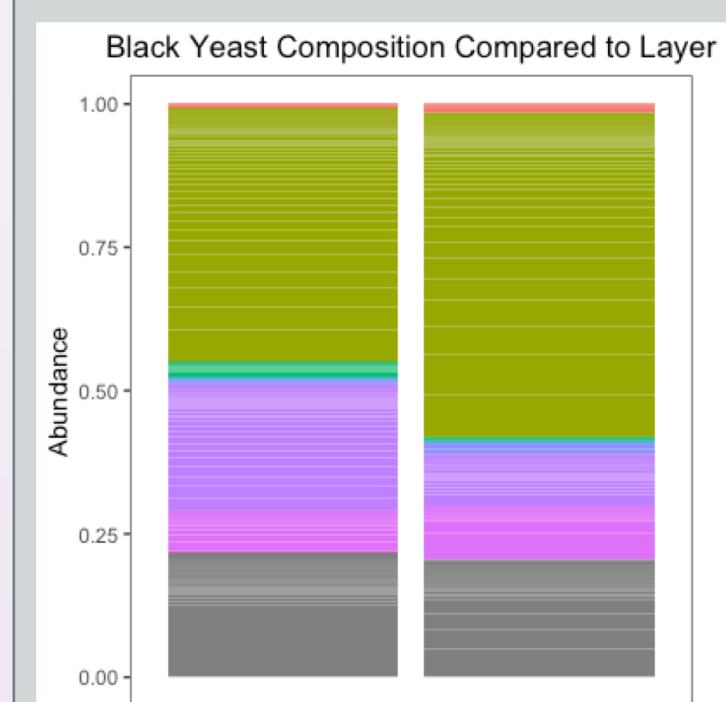


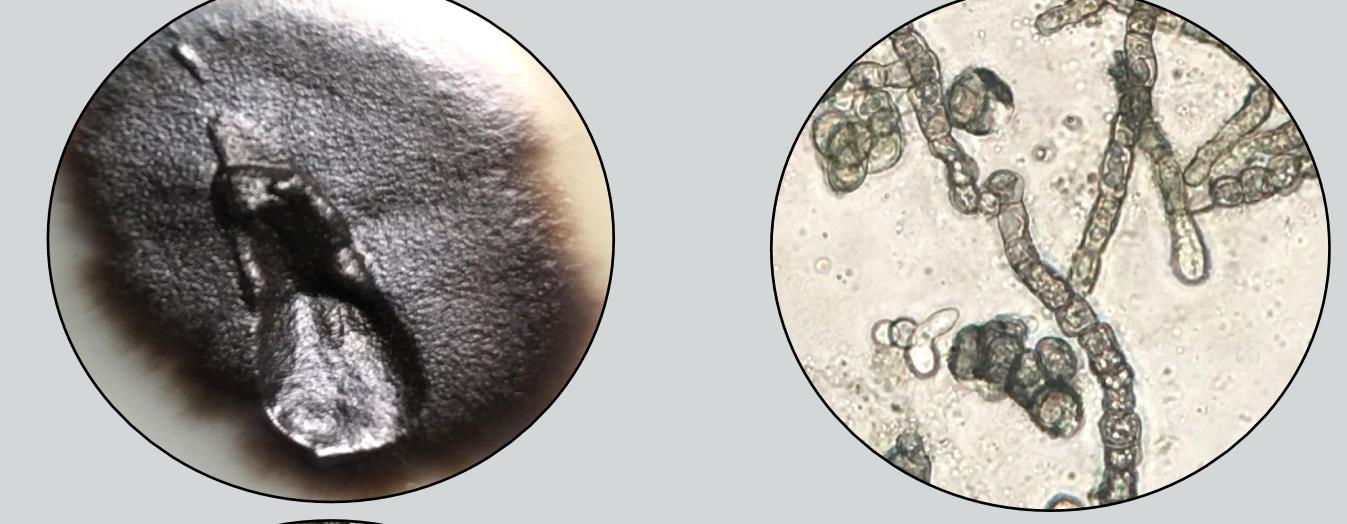
Figure 2 & 3: These two trees were generated through a collection of other fungi within the two main melanized fungi clades: Chaetothyriales and Dothideomycetes. The ITS 2 region was used to create these trees with MUSCLE as the multiple sequence aligner and FastTree as the Tree Generator. Blue are the melanized fungi that I have isolated from surface of rocks and biocrusts from Boyd Deep Canyon. Red strains are the fungi that have had full genome sequencing accomplished. Green strain is another melanized fungi that our lab has previously worked with.



RESULTS:



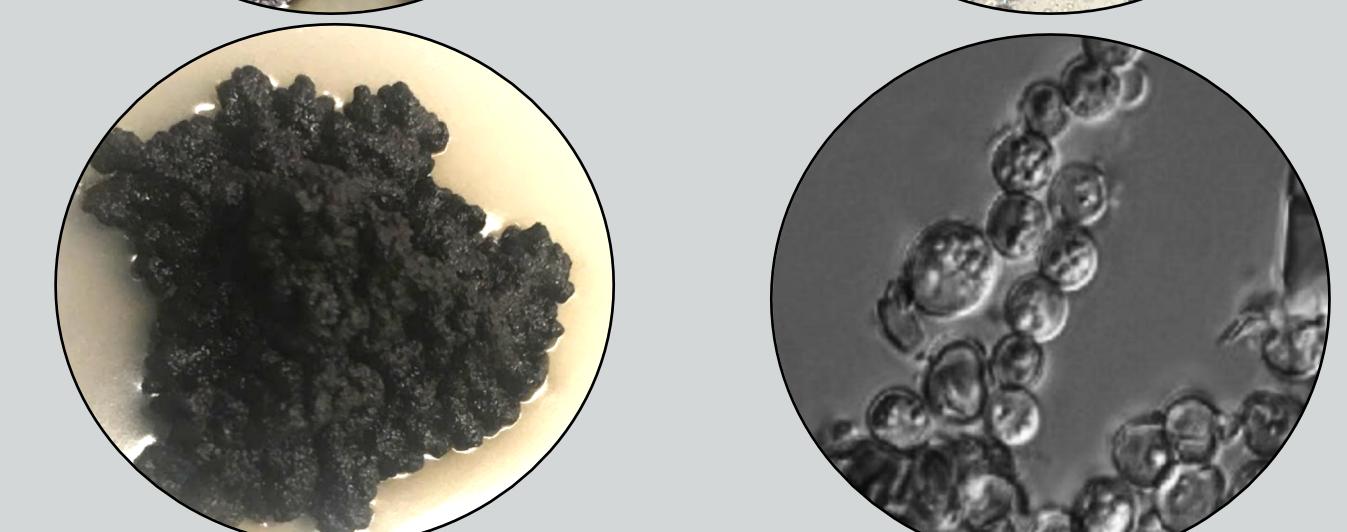
JES_112



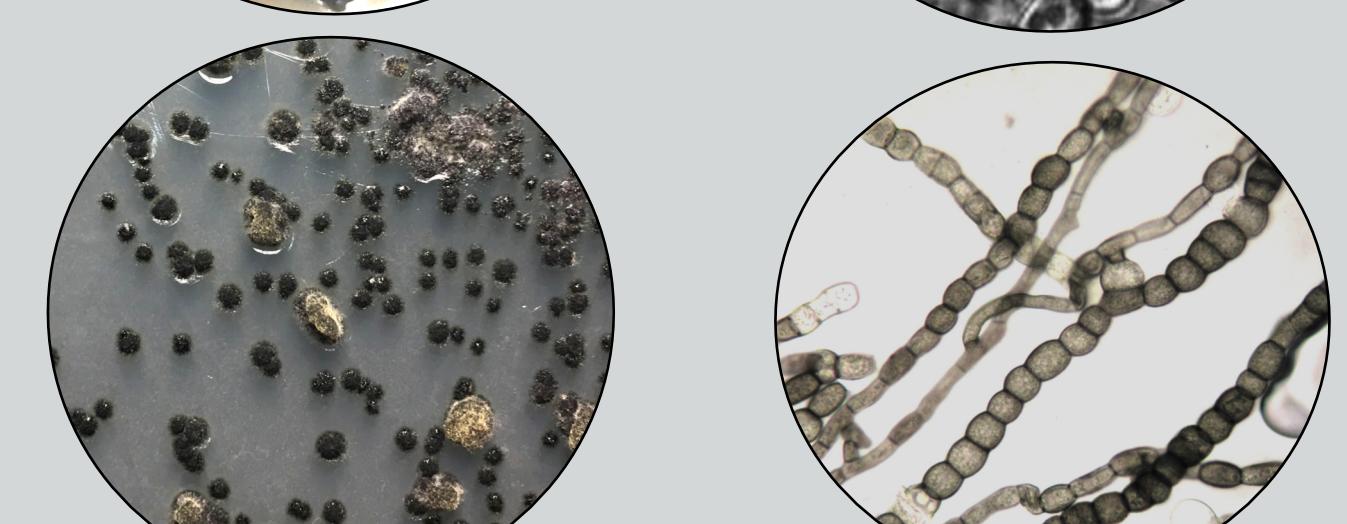
JES_113



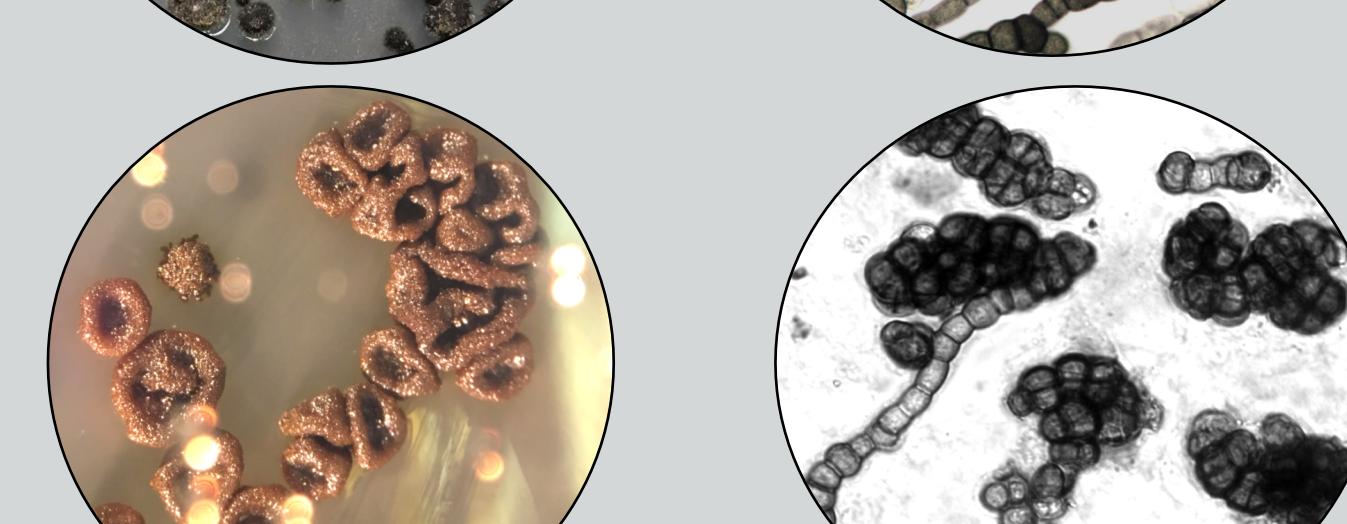
Knufia petricola A95



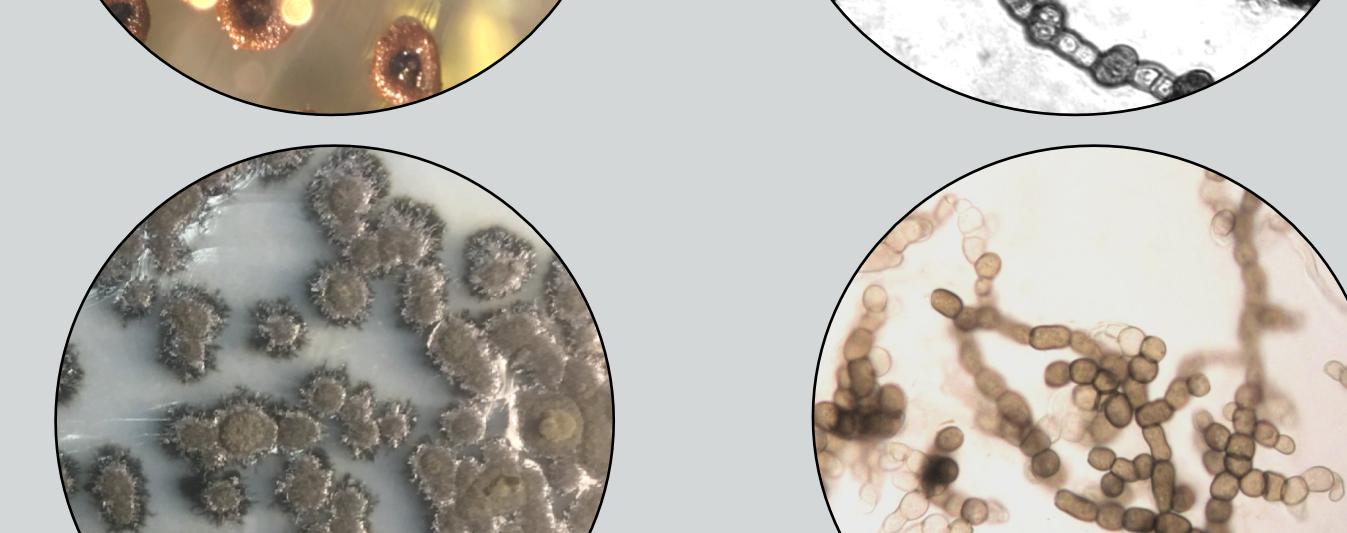
JES_115 BF2



JES_119



TK-1 C. apollonis



CONCLUSIONS & FUTURE WORK:

- We have shown the presence of melanized fungi in CLC biological soil crusts
- Sequencing MCF will help us determine the presence and absence of particular melanin synthesis genes.
- Over 30 melanized fungi were isolated from Boyd Deep Desert. 3 sequenced through Illumina, 1 through Nanopore. More will be sequenced in the future
- ITS is not a good marker to distinguish between these two orders. Creating these trees allows us to better place the newly isolated and sequenced melanized fungi in comparison to other sequenced fungi. Dothideomycetes have been better established but Chaetothyriales needs more work.
- Metabolome results from biocrust will be done soon.
- Metabolome analysis will be done on melanized fungi that has been sequenced well to observe product and genes.
- Observe melanin synthesis pathway genes through CAZY/MEROPS/PFAM/Orthologue Analysis

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