Project Proposal for Analysis of Novel *Trichoderma* Genomes

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1 Background

1.1 Trichoderma

Crop resistance to environmental stressors is a necessity for crop health and overall crop yields. Current popular methods for crop protection involve the use of pesticides and genetically modified organisms, which can be expensive and potentially politically dividing in the case of GMOs (citation needed). In addition, crops will suffer when soils are not sufficient for crop growth and health. These soil insufficiencies can include drought stress, nutrient stress and can also include solis that have been contaminated with hydrocarbons, making it difficult to grow crops in those regions and provides an opportunity for new bioremediation processes. Recently, two strains of *Trichoderma* have been identified in the prairie regions of Alberta and Saskatchewan. These two strains, named Tsth20 and DC1, have been found to have unique properties when incoluated in plants in the soils mentioned before.

Trichoderma is a type of fungi that can colonize the roots of plants in a non-toxic, non-lethal, opportunistic symbiotic relationship.[1] Many strains of *Trichoderma* have been shown to provide resistance to bacteria and other fungi in soils through the use of polyketides, non-ribosomal peptide synthetases and other antibiotic products[1][3]. In addition to these beneficial properties, the two strains mentioned above provide even further protection for plants in dry, salty soils and may also be considered as a bioremediation tool in soils contaminated with hydrocarbon content. However, little is known about how these mechanisms work in these new strains, so DC1 and Tsth20 were sequenced in an attempt to better understand the details of their genomes and secretomes.

1.2 Genome Assembly

Sequence assembly has been a long-standing issue in the field of computer science[7]. Determining the correct order and combination of smaller subsequences into an accurate sequence assembly is also computationally difficult in terms of compute resources such as memory, CPU count and storage required for input sequences[7]. In addition to these difficulties, there can be difficulties encountered during asssembly due to the nature of the data or genomes themselves. Insufficient data used in an assembly may result in short, fragmented assemblies, depending on the size of the genomes while

sequence data that is not long enough can fail to fully capture repetitive regions in an assembly. To solve this problem, a wide range of assembly tools have been developed with their own unique approaches to genome assembly problems, so it is important to use an appropriate assembler for the task at hand, and important to evaluate the assembly thoroughly. One approach to aid in the issue of genome coverage during assembly, is to use a combination of long and short reads in what is known as a hybrid assembly. Combining both highly accurate short reads with deep coverage along with less accurate but much longer reads can produce high quality genome assemblies that capture long repetitive regions. Assemblies must also be evaluated with measures such as N50, L50, coverage, average contig length and total assembled length to ensure that the genomes assemble well at least based on those metrics[7]. Following appropriate assembly protocols is essential to the further success of a project as downstream processing such as annotation depends on a high-quality assembly.

1.3 Genome Annotation

With the explosion of sequence data and genomes assemblies made available in recent years, genome annotation has become a crucial part of the sequence analyiss pipeline. Genome annotation can involve the annotation of genes as well as the annotation of structures within a genome. These annotations can be performed using either evidence oriented homology-based annotation programs or *ab-initio* statistical methods which do not consider existing evidence[6]. It may also be possible to use a combination of both in some circumstances. Furthermore, the downstream products of these genes can then be annotated for functional properties to determine what functions these genes and gene products could potentially perform.

1.4 Gene Prediction

In progress... Braker2, HMMs, evidence-based vs. *ab initio* methods

1.5 Repeat Identification and Masking

In progress...

1.6 Identification of AT-Rich Regions

In progress...

2 Research problem

Genome annotation, in the case of gene finding in this work, has been a popular computational problem for decades. The identification of possible genes provides other researchers with a valuable resource for future research avenues. Due to the importance and popularity of this topic, there have been a variety of gene finding tools that have been developed. This raises questions such as which tool is better, do different tools provide drastically different results and how much overlap exists between gene finding programs Comparative studies of gene finding tools have been performed before, however most of these studies are in reference to popular eukaryotic subjects of study such as humans, mice and in the case of plants, *Arabidopsis*. Few if any comparative studies have been performed in fungal genomes, and even fewer in the case of *Trichoderma*.

This lack of comparative analysis in fungal genomes provides a research avenue to compare results from the application of different gene finding tools to new and existing *Trichoderma* assemblies. Results from using different annotation tools will allow us to identify a 'core' genome along with genes that are not predicted by all tools in all genomes (outliers). This analysis will also allow us to compare results from both evidence-based gene finding methods as well as *ab-initio* gene finding methods. One interesting area to investigate between these methods would be in regions of low nuclotide diversity, or the AT-rich regions common to *Trichoderma* and other fungi. It is possible that gene finding programs may have difficulty with these regions of genomes, as they may contain repeat regions and as mentioned before, have low nucleotide diversity, making it unlikely for genes to be found in these regions.

3 Deliverables

- Lists of genes for each *Trichoderma* assembly considered
- Consenus or 'core' genome for each genome considered based on differ-

ent gene finding tools

- Comparison of gene finding tools in AT-rich and repetitive regions
- A potential list of true positive based on genes which have supporting RNAseq eveidence

4 Timeline

- Initial genome assemblies: September 30th
- Gene finding and annotation results: November 30th
- Analysis of gene finding results: December 31st - January 31st

5 References

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