Submitting Researcher

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Institution

USDA-ARS

Research Proposal

I study the molecular interactions between wheat leaf rust and wheat by understanding how the plant recognizes virulence effectors leading to the activation of defense mechanisms. I am faced with many roadblocks with this system. Puccinia triticina is an obligate biotroph, dikaryotic, cannot be cultured in vitro and creating sexual populations can take several years. Thus, a genomics approach was taken to try to understand the gene content, identify pathogen effectors, and study how the pathogen can quickly overcome disease resistance in the host.

My collaboration with the Broad Institute released a Roche 454 draft genome of P. tritiicna, estimated to be 135 Mb in length in 14,818 supercontigs, is 54%repetitive, and has 14,880 genes. The assemblers were not able to separately assemble the two karyotypes. P. triticina contains 1,358 predicted secreted proteins, believed to contain the core set of effectors used by the fungus to modify host cells and are also key factors recognized by the plant resistance proteins to induce cell death. Because of the difficulties in culturing and producing a mapping population, I have resorted to using fast neutrons to cause deletions to physically map these effectors. I have isolated 32 P. triticina mutants with effector knockouts and have sequenced them using Illumina.

Now the issue. As I have tried to align the sequencing reads to the genome, it appears that the genome assembly is not complete enough for me to identify the deletions. I am in need of a more complete assembly via BioNano and PacBio with pseudo chromosomes from each karyotype so that I can accurately identify where the deletions are. A PacBio and BioNano assembly will help me sort this out.