### Development of Molecular Markers for Oat Crown Rust Resistance

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### **Executive Summary**

Crown rust infection can reduce yields of susceptible oat cultivars resulting in regional crop losses as high as 20%. In response to this risk, resistance alleles have been purposefully deployed by oat breeders since early in the 20<sup>th</sup> century. However, in most cases the resistance genes present in specific oat lines are unknown. This project seeks to develop assays for molecular markers linked to the Crown Rust resistance genes most probably deployed in elite oat germplasm, and to evaluate markers for suitability to marker-assisted breeding and for identifying the specific forms of resistance within cultivars. To date, this project has developed assays for markers linked to 6 resistance genes (Pc38, Pc48, Pc58a, Pc68, Pc71, and Pc91) whose genomic location was already known. For all of those genes, breeders now have access to markers that can be used to track inheritance from one generation to the next during cultivar development if one of the parents carried that form of resistance. A subset of these markers can be used to identify oat lines carrying the Pc48, Pc58a, Pc71 and Pc91 genes with an error rate of less than 5%. Although most of these genes are no longer effective at most field locations, the markers developed for the Pc91 gene have been used to successfully predict oat lines with crown rust resistance. This project has also mapped the genomic location of two additional resistance genes: Pc53 andPc54. Assays are under development for markers linked to these genes.

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Crown Rust is one of the most damaging of oat diseases, causing yield losses in many growing regions. Under ideal environmental conditions, a susceptible cultivar can experience complete yield loss. Under less ideal conditions, regional yield loss of up to 20% has been documented (http://www.ars.usda.gov/Main/docs.htm?docid10123). The use of genetically disease resistant cultivars has been the primary method of controlling oat crown rust, and is still the most economical approach when possible. Unfortunately, crown rust is an extremely diverse pathogen and is able to respond rapidly when a resistance gene is widely deployed, resulting a window of effectiveness in the field of as little as five years. In order to keep ahead of pathogen diversity, breeders seek new sources of resistance to incorporate into cultivars under development. Knowing which of the lines available possess genes different from those already incorporated into a breeding program can help when it comes time to select parents for the next generation of oat cultivars. This project seeks to develop molecular markers tightly linked to known Crown Rust resistance (Pc) genes, to evaluate their ability to correctly identify oat lines carrying the linked Pc genes, and to identify oat germplasm carrying specific Pc genes.

# Markers linked to Resistance genes whose genomic location was already known

We have attempted to develop easy and reliable assays for molecular genetic markers linked to Pc genes with previously published genomic locations. Our assay method of choice has been the High-Resolution Melt Curve (HRM) method which uses commonly available real-time PCR equipment and reagents. Our successful assay results and applications are summarized below:

Resistance Gene	Marker Assays	Error Rate at Predicting	Carrier Lines Identified
	Developed	Gene Carrier Status	
Pc38	2	>20%	N/A
Pc48	3	<1%	56
Pc58a	2	2%	18
Pc68	3	>20%	N/A
Pc71	3	4%	32
Pc91	2	<1%	60

Over 500 oat lines, both resistant and susceptible, have been genotyped for the purpose of identifying which Pc genes they may carry. As we increase the number of Pc genes with tightly linked markers we anticipate that the number of oat lines characterized for the identities of the genes they carry will increase.

# Mapping Crown Rust resistance genes and finding linked markers.

Most Pc genes have no known genomic location, so developing bi-parental mapping populations has been a necessary first step towards reliable markers for these genes. This project supports primary efforts towards population development and linkage mapping of 5 genes. In addition, mapping efforts for 4 genes are being led by other scientists and this project supports collaborative efforts to develop and analyze validating populations. The progress of each mapping effort in the workflow from initial cross through marker assay development is summarized in the following table:

Gene	Role of this	Population	Genotyping	Phenotyping	Linkage Analysis	Marker
	Project	Development				Assays
Pc54	Lead	Done	Done	Done	Mrg02 at 85 cM	In Validation
Pc53	Lead	Done	Done	Done	Mrg08 at 82 cM	In Design
Pc96	Lead	Done	Done	In Progress		
Pc35	Lead	Done	DNA Ready			
Pc63	Lead	Done	DNA Ready			
Pc50	Validation	Done	Done			
Pc46	Validation	Done	DNA Ready			
Pc94	Validation	Done	DNA Ready			
Pc67	Validation	Done				