

# North American Bramble Growers Research Foundation Annual Progress Report

## Interim Report

Fall 2023

**Proposal Category:** Pest management Strategies – soil borne disease and nematode management

**Title:** Genomic and phenomic technologies for evaluating and identifying root lesion nematode (*Pratylenchus penetrans*) resistance in red raspberry

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### Introduction:

Red raspberry growers in the Pacific Northwest face significant pressures from the root lesion nematode, *Pratylenchus penetrans*. Root lesion nematodes (*Pratylenchus* spp.) are one of the most important plant-parasitic nematodes to agricultural crops in the world (Jones et al., 2013). This migratory endoparasite feeds on roots, resulting in damaged vascular tissues and producing the characteristic root lesions that give *Pratylenchus* spp. their common name. The injured roots are inhibited in their uptake of water and nutrients leading to reduced plant vigor and symptoms of stunting and chlorosis.

Growers primarily manage *P. penetrans* through chemical preplant treatments and occasional postplant chemical treatments. New and revised regulations of these chemical treatments make these options increasingly limited to growers. 1,3-dichloropropene is the dominant preplant treatment (Walters et al., 2017), but this method seems to have a short-term effect with nematode population densities rebounding one to three years after application (Bélair, 1991; DeVetter et al., 2018). Other chemicals such as methyl bromide and fenamiphos were once widely used but have now been phased out (Zasada et al., 2010; Zasada & Walters, 2016). Alternative methods such as crop rotation (Forge et al., 2012), soil amendments (Gigot et al., 2013; Forge et al., 2014, 2016) and solarization (Pinkerton et al., 2000, 2002) have been investigated with little success due to limited lifetime control or costs. Development and release of resistant cultivars would aid in relieving the economic and disease pressures.

Resistant varieties have been developed in breeding programs (Bristow et al., 1980; Vrain & Daubeny, 1984) and are available in germplasm repositories (Zasada & Moore, 2014), but much remains unknown about the genetics that confers resistance to *P. penetrans*. Current knowledge suggests *P. penetrans* resistance is an additive, quantitative trait (Vrain et al., 1994) which raises challenges in studying and breeding for this trait. Recent developments in genetic analysis methodologies now allow for improved elucidation and selection for complex traits such as this one. Genomic selection is a modern breeding technique that enables plant breeders to select individuals with desirable genetics at the seedling stage using genomic estimated breeding values (GEBVs) generated from genetic marker information (Meuwissen et al., 2001). This technique has been applied in globally significant crops and demonstrated to be well-suited to assess quantitative traits, sometimes with greater efficacy compared to other genomics assisted breeding techniques such as marker assisted selection (Bernardo, 2008; Poland & Rutkoski, 2016).

Another limitation to evaluating individuals for nematode resistance has been the intensive, destructive techniques needed to examine *P. penetrans* densities in the roots. High-throughput phenotyping technologies have rapidly advanced alongside genetic technologies and present a more cost and time effective method of assessing individuals. There have been initial investigations into ground-based methods for detecting abiotic and biotic stresses (Williams et al., 2017, 2021, 2023) in red raspberry, but there have been no investigations for use in detecting nematode stress. At this time, accurate, non-destructive methods of phenotyping raspberries for *P. penetrans* resistance are not available. Such methods need to be evaluated before high-throughput phenotyping and genomic selection for this trait will be effective.

This research provides the first application of genomic prediction for quantitative disease resistance in red raspberry and will potentially identify genes associated with *P. penetrans* resistance or tolerance. It will also evaluate high-throughput phenotyping technology, which will have application not only to raspberry but to other perennial crops. Combined, this information can be used to better inform breeders and reduce development time of more resilient cultivars and encourage genomics- and phenomics-assisted breeding for other difficult or genetically complex traits in perennial crops. This project seeks to i) characterize *P. penetrans* resistance and tolerance phenotypes in a diverse population of 270 red raspberry breeding lines and cultivars

and 10 mapping populations, ii) conduct a genome-wide association study (GWAS) and perform genomic prediction of *P. penetrans* resistance using phenotypic and genotypic information, and iii) determine the effectiveness of high-throughput spectral data for assessing *P. penetrans* damage.

### **Progress to Date:**

A diverse panel of 270 red raspberry genotypes composed of released and unreleased genetic material from the USDA-ARS/OSU, Washington State University, British Columbia Berry Cultivar Development Inc. (BCBCDI) breeding programs as well as cultivars from international programs available at the USDA-ARS National Clonal Germplasm Repository were planted in a randomized complete block design with paired non-inoculated and inoculated *P. penetrans* treatments at the Washington State University Northwestern Washington Research and Extension Center (48°26'30.4"N, 122°23'38.1"W) in June 2022. The trial location is on a silt loam soil and records since 2015 have shown it has previously been planted to various grains, vegetables, herbs, and tea. There were 1,916 individual plants in this planting. Over the course of 2022 and 2023, 91 individual plants have died leaving 1,815 plants in field as of August 2023.

Aboveground biomass fresh weight for all surviving plants has been recorded for 2022 and 2023 by pruning whole plants to the crown (Figure 1) and then weighing the pruned biomass. Measurements were taken with a tabletop scale in 2022 (Figure 1) and with a hanging scale using a custom hanging arm fitted to a Gator vehicle (Figure 2).



*Figure 1: Above ground fresh weight biomass data collection from 2022. Left: Pruning of plants to the crown. Right: Recording of pruned plant biomass using tabletop scales. Photos by Savannah Phipps.*



*Figure 2: Aboveground fresh-weight biomass data collection from 2023. Plants were pruned to the ground and weighed with a hanging scale custom mounted in the back of a Gator. Photo by Shijie Zhang.*

In the 2023 season, root samples were also collected for *P. penetrans* quantification. Throughout the season high-throughput phenotyping using a AgBOT mk 1 quadcopter equipped with a MicaSense RedEdge-MX multispectral sensor and the FLIR Vue R thermal sensor for aerial imaging and an Apple iPhone 14 Pro Max with the AgerPoint Capture app as the ground-based imaging system occurred (Figure 3).



*Figure 3: Left: Constructed orthomosaic of aerial images taken May 5<sup>th</sup>, 2023. Constructed by Alexander Gregory. Middle: The AgBOT mk 1 quadcopter equipped with a MicaSense RedEdge-MX multispectral sensor and the FLIR Vue R thermal sensor. Photo by Savannah Phipps. Right: Savannah Phipps recording images of select genotypes using the AgerPoint Capture app on the iPhone 14 Pro Max. Photo by Hannah Baker.*

*Pratylenchus penetrans* extractions and quantification for the 2023 season are ongoing at the time of this report and will be finished by the end of 2023. Processing and preliminary analysis of aerial and ground-based imaging data is also in progress and expected to be finished sometime early in 2024.

### **Future Work:**

Once genotypic data has been extracted and received, we will perform a GWAS to identify potential genetic markers for resistance and develop genomic prediction models aimed at generating GEBVs for nematode resistance. One more year of phenotypic data collection will occur over the 2024 field season with data analysis concluding early in 2025. This data will subsequently be incorporated into the GWAS and genomic prediction models for further evaluation of single and multiyear data. Results from these analyses should be available in the Spring 2025 and will be publicly available in a peer-reviewed journal.

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