Combating Rose Rosette Disease: Development of rapid, efficient, user-friendly virus diagnostic tools and studying virus-vector interactions

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Summary

Garden roses, which form the cornerstone of the multi-billion dollar landscape industry, annually generate wholesale US domestic bare root and container production valued at ~ $400 million. Over the past few decades, Rose Rosette Disease (RRD) has spread from its source in the Rockies, through the Mid-West to the East coast. It now threatens to decimate the US rose industry. There is an urgent need to control RRD.

A newly-funded USDA, NIFA, Specialty Crops Research Initiative Program Project involves 17 scientists in 6 states working on a range of approaches to learn more about this disease and determine how best to manage it. The long term goal of this project is to develop roses resistant to this virus and/or the mite vector. Key to this effort will be the development of efficient diagnostic tools to enable rapid, easy-to-use and accurate detection of the viral pathogen.

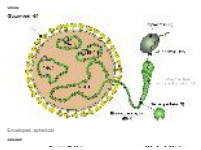
RRD is caused by the plant virus, Rose rosette virus (RRV; genus Emaravirus), which is transmitted by wind-blown eriophyid mites (Phyllocoptes fructiphilus). This virus/vector pair originated in the western part of the United States and has spread along with Rosa multiflora, a very susceptible introduced and now widespread host, throughout the eastern seaboard and the Midwest of the country. In recent years, the disease has spread onto landscape roses via the mite vector throughout this region resulting in the death of many thousands of rose bushes. Unfortunately, only a few rose species and no cultivated roses have been reported resistant to the virus.

The symptoms for RRD, although they often vary with the rose cultivar, commonly include proliferation of lateral shoots causing a witches broom symptom, unusual thorniness and reddening of these shoots and distorted flowers leading to stunting, defoliation and eventual death of the plant. Unlike other rose diseases it can kill a rose bush within two to three years of infection.

Interactions between the mite vector and rose hosts will also be studied. We hope to identify differences in leaf surface properties that can be utilized for screening breeding lines.

In the long term, this project hopes to identify additional sources of resistance and develop hybridization and genetic tools to move resistance into commercial cultivars.

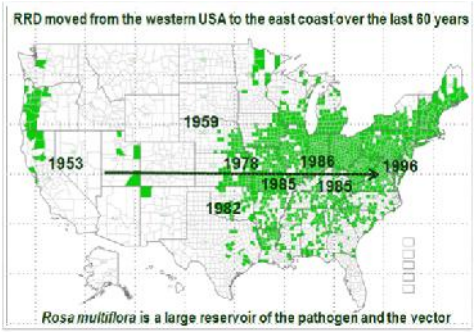
Plant Virus and Mite Vector



Rose rosette virus (RRV)

Phyllocoptes fructiphilus

Distribution of *Rosa multiflora* and Movement of RRD



Rose Rosette Disease Symptoms

RRD causes rose a cluster of shoots emerging from nearly the same point on the stem resulting in a witches' broom (rosette) appearance.

RRD causes elongated rose shoots, leaf distortion and red or yellow mottling of the leaves.

An excessive number of thorns on shoots is another symptom of RRD.

Mite Vector and Rose Host Interactions

Interactions between the mite vector and rose hosts are being evaluated. Rose genotypes that are either resistant or susceptible to mite feeding, reproduction, or RRV transmission are being examined by high resolution light, fluorescence microscopy, and low temperature scanning electron microscopy.

Paired examples of a susceptible host, Knockout (S), on the left, and a 'resistant' host, Rosa bracteata (R) on the right, showing distinct differences in numbers of hairs or trichomes on the stems and buds, visualized under light and scanning electron microscopes. Note that R. bracteata has a profusion of hairs that may impede movement of the mites.

Combating RRD: Diagnostic Objectives

Key to the effort to detect the virus and to control the disease will be the development of efficient diagnostic tools to enable rapid, easy-to-use and accurate detection of the virus.

Overarching Goal: To develop and evaluate diagnostic assays for non-skilled operators for the user-friendly detection and specific identification of RRV and to transfer these technologies nation-wide through outreach, including webinars and 'hands on' workshops.

Reagents will be designed and developed

- **RRV-specific primers and probes (for nucleic acid-based assays)**
- **Monoclonal and/or single-chain antibodies (for serological-based assays)**

Techniques and assays will be developed and refined for lab and field use

- **ELISA and immuno dipstick tests**
- **RT-LAMP (Reverse transcription-Loop mediated isothermal amplification)**
- **Self-quenched primer (SqP) technologies (for laboratory detection systems)**
- **Lateral flow devices (LFD; for both antibody and nucleotide based detection)**

The most consistent assays will be tested and validated by several diagnostic labs and transferred via outreach to other plant diagnostic labs.

Comparative analysis of potential diagnostic methods

Assay	Sensitivity	Skill required	Equipment needed	High throughput	Time required	Cost
Nucleic acid-based Assays						
LAMP	High	Medium	Yes	Medium	1-2 hrs	Med
LFD	High	Low	No	Medium	10-30 min	Low
RT-PCR	High	High	Yes	Low	6-8 hrs	High
Antibody-based Assays						
ELISA	Medium	Medium	Yes/No	High	6-18 hrs	Low
Immunostrip	Low	Low	No	Medium	10-30 min	Low

Combating RRD: Breeding Objectives

The breeding aspect of the project includes the field evaluation of 400 rose accessions for RRD resistance.

In the long term, this project hopes to identify additional sources of RRD resistant roses and develop hybridization and genetic tools to move RRD resistance efficiently into elite rose germplasm and commercial cultivars. This includes developing high throughput markers (SNPs), consensus maps and identifying marker-trait associations for RRD resistance and consistent flower productivity and quality. Diagnostic tests for RRD will contribute to this effort.

For more information on this aspect of the collaborative project – please see "Combating Rose Rosette Disease: Breeding for Resistance", Byrne et. al. at ASHS Poster #048.

Acknowledgement

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