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Title: Exploring alternatives to the chemical control of nematodes in potato in the San Luis Valley

Most relevant funding source: CSU Cultivar Royalty Funds (from CCPGA).

Investigator name(s) and department(s): <u>Principal Investigators</u>: Drs. Jorge Vivanco (CSU-Horticulture) and Samuel Essah (CSU-Horticulture and SLV Research Center). <u>Collaborator</u>: Dr. Antoon Ploeg (University of California, Riverside – Nematology Department)

Background

The San Luis Valley is the largest potato production area in the State of Colorado. Potatoes are mostly produced for seed or for the fresh market with a few varieties targeted for the specialty markets. Apart from PVY virus which is transmitted by aphids, root knot nematodes (*Meloidogyne* spp) also cause substantial economic damage. Of the different root-knot nematode species, the Columbia root-knot nematode (*M. chitwoodi*) is of particular importance to potato as it is commonly found in potato fields and even low populations of this nematode can cause lesions and malformation of the tubers, severely impacting tuber quality. Any field in the San Luis Valley that has experienced nematode infestation cannot be planted with potatoes unless it is treated with the chemical nematicides Vydate/Telone II. These chemicals are highly toxic, and pose a serious risk to human health and the environment. In California the use of Telone is limited because it produces volatile organic compounds or VOC's which lead to air pollution problems and higher rates of respiratory issues particularly in children. Thus, there is the need to test alternatives to nematode chemical control such as biological control measures.

The use of microorganisms antagonistic to nematodes or compounds produced by these organisms may provide additional opportunities to manage the damage caused by the root-knot nematode (Radwan et al., 2012). Bacteria and fungi can produce substances that negatively affect nematodes such as antibiotics, siderophores and a variety of degrading enzymes (Tian et al., 2007). These microorganisms can also act as competitors for nematodes in the roots and rhizosphere for colonization sites and nutrients (Dong and Zhang, 2006). The fungus *Paecilomyces lilacinus* preys on *Meloidogyne* females and reduces the population density of nematodes in the soil (Khalil et al., 2012). There are currently, several biological nematicides in the market containing various formulations of *Paecilomyces lilacinus*. In addition, bacterium such as *Bacillus subtilis* have been reported to have nematicidal activity but there is limited information regarding its mode of action.

Objectives

This proposal seeks to identify ways to reduce the reliance on chemical nematicides in the San Luis Valley. The specific research objectives of our proposal are:

- 1. Test two commercial biological nematicides for effectiveness in controlling *M. chitwoodi* under greenhouse conditions.
- 2. Isolate beneficial microbes from San Luis Valley soils that could be natural antagonists of *M. chitwoodi*.

Research Team

Dr. Jorge Vivanco is a Professor of Horticulture at CSU. His laboratory works on interactions between root and the soil microbiology with particular focus on identifying beneficial microbes that could help plants deal with biological and environmental stresses. Dr. Samuel Essah is an Associate Professor of

Horticulture located at the San Luis Valley Research Center of CSU. Professor Essah's program focuses on developing the best cultural management practices for potato cultivation. Professors Vivanco and Essah have a long-standing collaboration to bring and test new technologies related to potato cultivation to the SLV. Professor Antoon Ploeg (Nematologist from the University of California, Riverside) has been invited as a close collaborator of this project.

Experimental Methodology

Objective 1. Test two commercial biological nematicides for effectiveness in controlling *M. chitwoodi* under greenhouse conditions.

Dr. Ploeg will visit the SLV as soon as this proposal is funded to train Drs. Vivanco and Essah in techniques related to nematode isolation and testing. Specific potato fields in the San Luis Valley infested by nematodes will be identified by collaborating with personnel of Agro-Engineering who have extensively mapped these areas in the valley. Soils from infected sites will be sampled and brought to the laboratory of Dr. Vivanco on the main campus of CSU. M. chitwoodi levels in the soil will be determined, using standard extraction techniques. Nematodes from these potato fields will be cultured on potato and will subsequently be used as inoculum in greenhouse experiments. All necessary permits and precautions will be followed to contain the spread of M. chitwoodi outside of the greenhouse. At least two distinct commercial biological nematicides will be tested such as MeloCon and NemaRoot on two potato cultivars that are contrasting in nematode susceptibility. The effect of these two biological products will be compared with Vydate and an untreated control. To evaluate the efficacy of the biological nematicides, their impact on nematode levels in soil and in plant roots, and on tuber yield and quality will be determined and compared to the untreated control and the Vydate standard. Data will be subjected to statistical analysis to determine the significance of treatment effects. The assistance of Dr. Ploeg at different stages of this objective will be invaluable; therefore, he will be invited to the SLV and to CSU at least 2-3 times during the one-year life of the project.

<u>Objective 2.</u> Isolate beneficial microbes from San Luis Valley soils that could be natural antagonists of *M. chitwoodi*.

This project will additionally identify and characterize soil microbial populations from the SLV that could be natural antagonists of *M. chitwoodi*. For this purpose, we will again rely on the extensive mapping of the SLV by Agro-Engineering and determine areas that are consistently nematode free yet adjacent to nematode infected locations. We will place special attention to avoiding locations that have received either chemical or biological nematicides. Soils from those areas will be collected and brought to the laboratory of Dr. Vivanco at CSU to isolate native species of *Paecilomyces lilacinus* and *Bacillus subtilis*. Soil slurries will be prepared by diluting the soils in water and plating them in specific media. There are specific medium preparations and cocktails of antibiotics available to isolate specific genera such as *Paecilomyces and Bacillus*. We propose that these native beneficial microbes from the SLV might be better prepared to control the nematode populations that affect potato cultivation in the valley than commercial biological nematicides.

Plans to Validate the Technologies

We are requesting funding for one year to test under greenhouse conditions the effectiveness of two commercial products (Obj. 1). Depending on the effectiveness of the products, we will propose in year 2 to conduct highly replicated field studies in the SLV. Additionally, in this proposal we aim to identify natural microbes from the SLV that could have activity against the nematode (Obj. 2). If successful in these isolations, we will propose in year 2 to test the effectiveness of these natural strains alone and in

combination with commercial biological products under greenhouse conditions and subsequently in the field. These combined results will constitute preliminary data to apply for external USDA funding.

Extension-Outreach Activities

A summary of the progress of the activities described in this proposal will be jointly presented by Drs. Vivanco and Essah at the 2015 Potato and Grain Conference. In addition, we will organize one forum in the SLV in which Dr. Antoon Ploeg will discuss the latest strategies of nematode control (biological, chemical and integrated) in the state of California. We will also extend an invitation to Dr. Ploeg to present research related to this project in the 2015 Potato and Grain Conference.

How this Project will Enhance Colorado Potato Grower Competitiveness

The identification of alternatives to Vydate/Telone II for nematode control in the San Luis Valley will reduce the use of these toxic chemical nematicides. Vydate/Telone II decreases microbial populations containing beneficial microbes in the soil, thus impoverishing soil health in potato fields. The use of alternative methods to control nematodes will improve the soil health of potato fields which is usually accompanied by increased tuber quality.

Budget

Personnel (\$8000). Partial salaries are requested for a technician and student-hourly aids to assist in the tasks proposed here.

Materials and Supplies (\$10,000). These expenses include chemicals, media, and disposables for the isolation of nematodes and beneficial microbes from soils. Cost of pots, soil and renting space in the greenhouse are also considered.

Travel (\$7000). Travel expenses related to visiting (4-5 visits) the SLV for soil collection purposes and to participate in the outreach/extension activities proposed here. This cost also includes the visits of Dr. Ploeg to the SLV and to CSU.

Total: \$25,000 for 1 year

References

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