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PERFORMING INSTITUTION:

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DEVELOPMENT OF COMPREHENSIVE STRATEGIES TO MANAGE POTATO VIRUS Y IN POTATO AND ERADICATE THE TUBER NECROTIC VARIANTS.

NON-TECHNICAL SUMMARY: Potato Virus Y (PVY) has re-emerged as a serious disease problem in many seed potato production areas in the United States. In the past, PVY has been managed effectively by potato seed certification programs, but the widespread acceptance of cultivars that are tolerant of infection (plants are diseased, but do not show symptoms) and the emergence of necrotic strains of PVY bring new challenges to seed certification, seed trade and potato production. Many of these necrotic viruses induce mild foliar symptoms in potato and are more difficult to identify during field inspections, yet they are more damaging to tubers. The current practices in potato seed certification which are dependent upon visual assessment of virus infection levels in the growing crop cannot effectively manage the new strains of PVY in the current array of cultivars, and they will not prevent the necrotic strains from becoming prevalent. Inaccurate field inspections and improper diagnosis have contributed to increased incidence of some necrotic strains leading to some unnecessary trade restrictions. Necrotic strains of PVY pose a significant threat to the potato industry due to reductions in yield and lower tuber quality. Additionally, because they are regulated pathogens, the necrotic strains will continue to impact interstate and international seed trade. An evolving national management plan sanctioned by the industry provides guidelines that will be refined by our research to reduce overall PVY incidence and limit the introduction of necrotic strains. Several of the principal investigators of this project have served as the scientific advisors for the plan development and revision. The US-Canada Management Plan for Viruses that Cause Tuber Necrosis in Potato Tubers, signed into effect in April 2006, is designed to facilitate the trade of potatoes within and between the US and Canada while protecting the potato industry from several emerging viruses, including PVY, that cause yield and quality losses in the potato crop. Management strategies and research needs of the industry are listed in the plan and by design are reviewed and refined by the industry on an annual basis. The overall objective of this project is to refine the current management strategies based on industry needs to reduce overall levels of PVY and prevent the spread of necrotic strains. Specifically, we will 1) Work with seed certification agencies to develop new tools to better identify and eliminate PVY from seed stocks. These will include training aids to better identify infected plants in the field, and improve diagnostics and post harvest testing procedures to better evaluate the tuber crop, 2) Work with breeders to develop PVY resistant cultivars or cultivars with recognizable symptoms. Current cultivars and breeding lines will be evaluated in regional trials to look for

new sources of resistance and to identify the level of susceptibility to a range of PVY strains, 3) Work with growers to develop cost effective, on-farm virus control strategies that are aimed at reducing the amount of PVY introduced to the crop and controlling the spread of the virus in the crop.

OBJECTIVES: Potato virus Y (PVY), once managed effectively by strict seed certification practices, has reemerged as a serious disease problem in the seed potato crop. The seed potato industry has identified PVY as its most serious disease problem and has set goals to reduce PVY incidence below tolerance limits set by seed certification programs, and to eliminate the new tuber necrotic isolates. Our overall objective in this proposal is to refine the current PVY management strategies used by growers and seed certification agencies to reduce virus levels in seed stocks and spread of virus within the crop. Specifically, we will a) Assist seed certification agencies to amend current practices and improve their ability to detect, monitor and eliminate PVY from seed stocks. b) Assist breeding programs to develop improved methods to screen for PVY resistance and symptom expression. c) Assist growers to develop cost effective, on-farm virus control strategies. Reducing the overall incidence of PVY to manageable levels and eliminating the tuber necrotic strains will require an adjustment of seed certification practices and a more aggressive use of on-farm management strategies by both seed and commercial potato growers. Similar strategies have been successful in eliminating other potato pathogens from seed production by learning important control points for each of the pathogens, implementing control measures on farms, and making appropriate changes in seed certification and regulatory practices. The goal of this proposal is to achieve a similar level of control for PVY. The expected outcomes and deliverables of our proposed research include: 1. Improved diagnostic tests capable of differentiating all the PVY strains affecting U.S. potatoes. 2. Educational training aids for seed inspectors and growers to recognize all the various foliar and tuber symptoms caused by the new PVY strains. 3. Information back to each participating seed programs on the PVY strains collected from their state. 4. Alternatives to current post harvest test methods, including a comprehensive economic analyses of the different methods currently in use and for those developed in this proposal. 5. Knowledge of the susceptibility of the major U.S. potato varieties to potato tuber necrotic ringspot disease cause by the recently introduced necrotic and recombinant strains of PVY 6. Establishment of regional trials to evaluate susceptibility, symptom expression, and resistance in varieties and advanced breeding lines to the range of PVY isolates now present in the U.S. 7. Improved minituber production methods that would allow varieties to reach the market earlier and reduce the potential for PVY infection of minituber crops planted in the field. 8. Forecast and economic models that relate the levels of PVY in seed lots of major cultivars to the levels of PVY incidence determined in the field and harvested tubers. 9. New information on the aphid transmission efficiency of the new PVY strains to help predict the likelihood of spread of these strains within and between crops. 10. New information on the efficacy of new antifeedant compounds that could prevent aphid inoculation of PVY to potato.

APPROACH: Efforts to improve the effectiveness of seed certification will include the development of new training materials to help inspectors better identify virus infections. This will also include evaluations of diagnostic methods that are being developed in this and other projects. Improvements to post harvest test procedures will focus on new lighting systems that may allow operations to move out of the field and into controlled environments that would allow more uniform growth and faster testing. An economic analysis of various post harvest test methods will provide certification agencies with short and long term cost savings projections if they choose to implement new testing procedures. Potato tuber necrosis is the most serious aspect of the emerging PVY strains. Field and greenhouse evaluations of existing cultivars and breeding lines for their susceptibility to tuber necrosis and their reactions to all PVY strains will provide valuable risk assessment information.

Regional trials of material should also identify new sources of resistance to PVY that can be introduced into breeding populations. Finally, studies to improve on-farm management options will focus on reducing initial inoculum and reducing the spread of PVY into and within a crop. Both the mechanics and economics of identifying maximum threshold levels of PVY that will still allow satisfactory disease management and high economic return are featured aspects of these studies. Epidemiological studies on reducing virus spread in the field will focus on aphid vector phenologies and control, as well as weed reservoir management. A cost-benefit analysis will be conducted for each of the scientific recommendations when appropriate to assist the industry and regulatory agencies in deciding acceptance of any proposed changes to the national plans. Cost-benefit analyses also can assist state certification and regulatory agencies and individual growers in determining whether to adopt individual practices that are not mandated at the federal level. Each of the outlined objectives of this proposal meets current needs as defined by the industry. To ensure the research continues to address relevant issues, growers, industry representatives, seed certification officials, and federal regulatory officials meet annually with the Co-Investigators on the proposal to discuss research findings and potential recommendations to modify the current Management Plan, as well as changes in direction and scope of proposed research needs. In addition, Seed Certification officials from each of the 16 seed producing states meet in December with scientists and grower representatives to focus on seed certification issues. A WERA-089 meeting held each March brings together scientists working on virus and phytoplasma diseases of potato. These are often attended by seed certification, regulatory and industry representatives. Technology and information is also transferred to stakeholders at regional and national grower meetings, farm and field-day events, statewide vegetable newsletters, and through popular press and scientific articles.

PROGRESS: 2012/09 TO 2013/08

Target Audience: Potato growers, seed certification officials and inspectors, seed potato specialists, extension agents, scientists, graduate students Changes/Problems: Nothing Reported What opportunities for training and professional development has the project provided? Eight graduate students, three postdoctoral associates and numerous undergraduate students have been partially supported by this project and have participated in many different aspects of the research and extension activities. Students and postdocs are expected to attend meetings and present findings to stakeholders. They are also involved in developing educational and training materials that is used primarily by growers and seed certification programs, but some of the materials are also useful for the research community. Students and postdocs are also involved in developing reports, budgeting and grant administration. They are also involved in visiting collaborating laboratories to learn new techniques and to share information and technologies. How have the results been disseminated to communities of interest? Information is disseminated through scientific publications and presentations at scientific meetings. Members of our project attend most of the state and national potato grower meetings and give presentations and provide written materials that describe our project and its findings including methods to improve on-farm management of PVY. Our group also interacts with seed certification agencies in all seed potato producing states to convey up to date information on the incidence and distribution of the endemic and emerging strains of PVY affecting the potato crop. All PIs on the project, including members of their laboratories, attended the Multistate Research Project, WERA89, "Potato Virus and Virus-Like Disease Management" meeting in March 2013. This meeting brings together most of the federal and state researchers working on insect transmitted diseases of potato. Additionally, many extension and seed certification professionals, as well as researchers and managers from agrobusiness attend. This is an opportunity for all members of our project to present findings and get feedback from many of our research and extension related stakeholders. Members of our project also attend international meetings

on potato production and potato diseases to provide information on PVY as it affects US potato production. In June 2013 our group organized and hosted the 6th International meeting on Potato virus Y in Idaho Falls, ID. Thirty six participants from nine countries met for three days to present and discuss research. It was an opportunity to share findings from our project with researchers in Europe and South America. Europe experienced a PVY resurgence in the 1990s and we learned much from comparing and contrasting multiyear epidemics on both continents. What do you plan to do during the next reporting period to accomplish the goals? Continue with lab to lab communications. Our project meeting will be held in conjunction with the 2014 Potato Expo in San Antonio so that we may involve many of our stakeholders in updates and discussion, and in the planning of new projects based on industry needs.

IMPACT: 2012/09 TO 2013/08

What was accomplished under these goals? PVY continues to be the number one disease issue affecting seed potato production. Results from the post harvest test of the 2012 crop indicated very high virus levels in many areas of the country due in part to high aphid numbers and relatively high PVY incidence in seed planted in 2012. These high levels of virus are expected to carry over into the 2013 crop, although the aphid populations were much lower than 2012 in many production areas. In many of the seed production states, PVY is detected in 40-60% of the seed potato lots and virus levels exceed 2% (the cutoff for interstate and international trade) in nearly 20% of the seed lots. The ordinary strain of PVY, PVYO, continues to become less prominent, while the recombinant strains continue to increase in prevalence and distribution. The tuber necrotic strain is now found in all seed production areas, but is still contained in relatively few seed lots. These tuber necrotic strain-infected seed lots are being identified by our testing of seed lots during the post harvest survey and states are encouraging growers not to plant these seed lots back into seed production areas. By doing this we are keeping the tuber necrotic strain in check. The Wilga strain of PVY is now predominant in most seed production areas due to several factors researched in this project. The recombinant strains are transmitted more efficiently plant to plant by aphids and a greater percentage of tubers are infected with the recombinant strains. The reliance on visual symptoms to evaluate virus levels in the crop continues to select for the mild symptom recombinant strains, although many of the state seed certification programs are adopting serological testing of some or all seed lots as an alternative to visual inspection. A national harmonization standard that requires seed potatoes moving across state and international borders be certified and post harvest tested was adopted by the national potato industry and most of the seed production states. These standards were developed and championed in large part due to the research provided by this project. The standards are providing regulations that can reduce the overall incidence of PVY and the movement of the virus between production areas. Two new diagnostic methods were developed, one that uses a multiplex PCR-based strategy to differentiate nine different PVY strains and strain variants found in the US; another provides a rapid dot-blot serological format that couples sampling of a large number of individual plants in field with a quick, high throughput, serological detection of virus in the lab. Both of these techniques will be useful for seed certification programs to better assess virus levels in the crop and to identify those lots infected with the tuber necrotic strain of PVY. We continue to work with an increasing number of potato breeding programs to identify new sources of PVY resistance and to determine the effectiveness of the various resistance genes against the wide genetic diversity of PVY strains that we have identified and researched during this project.

Resistance to the virus will ultimately be the best method to control PVY and eliminate the tuber necrotic strains. On-farm PVY management options continue to be refined and are based mainly on minimizing the planting of infected tubers, removing infected plants that do emerge, and minimizing the spread of virus within the crop by controlling aphid vectors and preventing aphids from feeding on plants. The success of virus management on the farm

depends on knowledge of high risk periods of disease spread so that appropriate control methods can be applied at those times. Studies continue to identify the most important aphid vectors, the sources of these aphids and the timing of aphids moving into and through the crop. Predictive models are being developed and tested to identify high risk periods. Economic analyses are providing evidence of PVY impact to growers, especially producers of potatoes sold to grocery stores or food processors, so they can better understand the need to abide by virus tolerance limits in seed potatoes being planted. There is an average loss of \$20 per acre for each 1% of PVY in the crop. Considering that the typical incidence in production potato fields is above 20%, the economic and yield loss is very significant. The loss for seed producers is much higher. Further analyses of crop data are allowing us to develop a risk index for the various cultivars of potato. Some cultivars are far more inclined to have high incidence of virus and therefore pose a greater risk for growers. If growers can select low risk cultivars, especially when virus incidence is predicted to be high, they can minimize the impact of the virus on their crop. A better understanding of how the virus is changing its makeup and geographic distribution, how it affects the various potato cultivars, and how it can best be detected and controlled, has allowed us to provide recommendations to growers, seed certification programs and regulatory agencies to better manage the economic and trade impacts and to adopt practices that will minimize the incidence of virus in seed stocks.

PUBLICATIONS (not previously reported): 2012/09 TO 2013/08

1. Type: Journal Articles Status: Published Year Published: 2013 Citation: Chikh-Ali, M., Gray, S. M., and Karasev, A. V. 2013. An improved multiplex IC-RT-PCR assay distinguishes nine strains of Potato virus Y. *Plant Dis.* 97:1370-1374.
2. Type: Journal Articles Status: Published Year Published: 2013 Citation: Karasev, A. and Gray, S.M. 2013. Continuous and Emerging Challenges of Potato virus Y in Potato. *Annu. Rev. Phytopathol.* 51:571?86
3. Type: Journal Articles Status: Published Year Published: 2013 Citation: Frost, K. E., R. L. Groves, and A. O. Charkowski. 2013. Integrated control of potato pathogens through seed potato certification and provision of clean seed potatoes. *Plant Dis.* 97-1268-1280
4. Type: Journal Articles Status: Accepted Year Published: 2013 Citation: Fulladolsa, A. C., R. Kota, and A. O. Charkowski. 2013. Optimization of a chemiluminescent dot-blot immunoassay for detection of potato viruses. *Amer. J. Potato Res.* 90:306-312.
5. Type: Conference Papers and Presentations Status: Published Year Published: 2013 Citation: Fulladolsa-Palma, A. C. K. E. LaPlant, and A. O. Charkowski. Effects of seed type and variety on the agronomic performance of potato minitubers and the incidence of Potato virus Y. American Phytopathological Society, Austin TX Aug 2013.
6. Type: Conference Papers and Presentations Status: Published Year Published: 2013 Citation: Zhang, C. and R. L. Groves. Resistance to infection by Potato virus Y among selected varieties for improved seed potato production. American Phytopathological Society, Austin TX Aug 2013.
7. Type: Conference Papers and Presentations Status: Other Year Published: 2013 Citation: Charkowski, A. O. Integrated control of potato pathogens through seed potato certification and provision of clean seed potatoes. International Congress Plant Pathology, Beijing China, Aug 25-30, 2013.
8. Type: Other Status: Published Year Published: 2013 Citation: Nolte, P. More good news on PVY in Idaho seed potatoes. *Potato Grower Magazine.* 42(5):35. May, 2013.
9. Type: Journal Articles Status: Published Year Published: 2013 Citation: Quintero-Ferrer, A. and Karasev, A.V. (2013) First report of Potato virus Y in potato in Jalisco, Mexico. *Plant Disease* 97: 430-430.
10. Type: Journal Articles Status: Published Year Published: 2013 Citation: Karasev, A.V. and Gray, S.M. (2013) Genetic diversity of Potato virus Y complex. *American Journal of Potato Research* 90: 7-13.

PROGRESS: 2011/09/01 TO 2012/08/31

OUTPUTS: Continuing surveys of PVY strains in seed lots and follow-up characterization of the viruses define the distribution and relative incidence of existing and emerging PVY strains. Emerging, recombinant PVY strains that induce mild symptoms in many potato cultivars are problematic for Seed Certification agencies dependent on observable symptoms to inspect and grade seed lots. Short term efforts to improve PVY management focus on assisting Certification programs to better evaluate levels of PVY in seed lots. We continue to assess foliar and tuber symptoms induced by all PVY strains on the major potato cultivars. Tabular and photographic information is available on www.potatovirus.com. and discussed at industry meetings. Evaluation of current and historical data from Certification programs on virus levels observed during the summer growing season and the winter post harvest test were analyzed to determine if the summer data accurately predict final virus disease incidence in the crop and if accuracy differed among potato cultivars. Furthermore, comparisons were made between visual and serological assessments of virus incidence in seed lots. These data are made available to seed certification agencies so they can determine the extent of post harvest testing needed to accurately determine virus incidence in seed lots. Due to changes in the genetic structure of the virus populations determined by the surveys, the currently employed diagnostic procedures were reevaluated for their ability to correctly identify all of the virus strains infecting the potato crop. Suggested changes in diagnostic protocols are reported on the project website and distributed to certification and regulatory agencies and to diagnostic testing labs. Efforts to improve on-farm best management practices for PVY continue on several fronts. In partnership with U.S. based public and private potato breeding programs, advanced potato lines are evaluated to identify new sources of resistance to the various PVY strains. Data from regional trials in NY, WI and ID are distributed back to the breeding programs. Oils and new antifeedant chemistries offer some degree of control of aphids transmitting PVY, but optimal application schedules are needed to minimize economic and environmental impact. Seasonal phenologies of aphid vector populations are correlated with virus disease incidence data and the efficacy of oil and chemical treatments are evaluated. To complement field data, laboratory studies determine the transmission efficiency of the PVY strains by various aphid species to better predict which need to be targeted for control. Since most of the initial virus inoculum in the field comes from planting infected tubers, the effect of initial levels of PVY in a field, determined by virus levels present in the seed, on final disease incidence is being determined to help growers and the certification programs evaluate threshold levels of virus to avoid significant yield and quality impacts. Preliminary data on all these longer term studies is presented at grower and industry meetings as well as scientific conferences. Popular press articles also outline some of the findings.

PARTICIPANTS: Stewart Gray is the overall project manager and developed the project website. Research includes PVY surveys, the evaluation and long-term storage of PVY isolates, evaluations of PTNRD symptom expression in greenhouse and field experiments, evaluation of quantitative resistance in breeding lines, and transmission efficiency of PVY by aphids. Eric Wenninger evaluated aphid transmission of PVY strains and cultivar reactions to PVY, and assisted with evaluations of aphid populations and virus incidence in hairy nightshade plots. Amy Charkowski investigated minituber versus conventional seed potato production and evaluation of potato germplasm for symptoms and resistance to PVY in the regional trial network. Russ Groves investigated PVY infection rates in minituber versus conventional seed production, established the relationship between initial PVY infection level in seed and disease progress plus final incidence at the end of the growing season, documented the seasonal phenology and dispersal of the soybean aphid and determined their relationship to PVY disease progress, and evaluated the effects of foliar protectants to limit or minimize PVY incidence in established field plots. Pamela J.S. Hutchinson investigated the role of hairy nightshade in aphid and PVY incidence. Alexander Karasev is responsible for large-scale genetic (RT-PCR) and serological (ELISA and Western blot) typing of all the field PVY

isolates collected by Seed Certification agencies and individual researchers working on this project. Karasev's lab will provide pure cultures of various PVY recombinant strains from Idaho, Wisconsin, or New York, for potato genetic screening experiments. Christopher McIntosh is responsible for the economic evaluation of and risk assessment of in-season PVY field inspections versus post-season tests, the economics of minituber versus conventional seed production, and the economic impact of PVY on commercial potato production, which has direct impacts on the value of seed and seed certification programs. Phillip Nolte is responsible for receiving and coordinating information from the seed certification agencies, the production of PVY training manuals, and will perform field trials to measure the impact of seed borne PVY on yield and the influence of seed borne PVY on the amount of PVY present in the harvested crop. Jonathan Whitworth's role is to plan, coordinate and administer national PVY field trials and to evaluate cultivars for foliar and tuber symptoms to PVY strains. Seed certification agencies from ME, NY, MI, WI, MN, ND, NE, WY, MT, CO, ID, CA, WA, OR, AK participate in this project by supplying plant and tuber samples, and by providing data on summer and post harvest test evaluations of seed lots. TARGET AUDIENCES: The outputs and outcomes of this project are targeted mainly at the various segments of the potato production industry, including seed producers, fresh-pack and process growers. We also provide information to seed certification agencies, regulatory agencies and to the general scientific community. PROJECT MODIFICATIONS: Nothing significant to report during this reporting period.

IMPACT: 2011/09/01 TO 2012/08/31

Continuing surveys of PVY strains present in seed lots and commercial fields from many seed production states indicated that the ordinary strain of PVY is no longer the predominant strain in most seed production areas. The recombinant tobacco necrotic strain predominates in most of the states surveyed. The tuber necrotic strain is present in nearly all production areas but remains associated with a limited number of seed lots. This has led Certification agencies to conduct more laboratory testing, especially for the tuber necrotic strain, to better evaluate PVY incidence in seed lots. Growers are urged not to reenter into certification those seed lots infected with the tuber necrotic strains and not to plant that seed in seed production areas. Our research has driven in part the adoption of new national standards for the movement of seed potatoes between states and Canada. Many state Certification agencies ask growers to abide by the same rules for seed moving within a state and many states have adopted strict seed laws that require certified seed for planting. Negotiations are ongoing to amend the bi-national PVY management plan adopted by Canada and the US in 2004 to include the updated standards. The identification of PVY strain variants that can be misidentified by current diagnostic protocols has led to the development of revised diagnostic testing protocols that have been adopted by Certification testing labs. These changes have also been submitted to NAPPO and are undergoing review prior to adoption by Canada, U.S. and Mexico. Four advanced breeding lines from public programs were identified as resistant to all strains of PVY, several lines from the private program also show promise. The evaluation of symptom expression on multiple cultivars induced by multiple isolates representing each of the PVY strains indicates that in general the emerging recombinant strains induce milder and often transient foliar symptoms whereas the nonrecombinant ordinary strain induces more severe foliar symptoms. However, many cultivars are asymptomatic when infected with most PVY isolates, and isolates within a strain can cause a range of symptom types and severity within a cultivar. This necessitates testing using multiple isolates within a strain to adequately evaluate cultivar reactions. Furthermore, tuber necrosis is not restricted to the tuber necrotic strain of PVY, but can be induced by isolates of the virus classified in other strain groups. Analyses of seed certification data identified Russet Burbank, Russet Norkotah and Atlantic as cultivars that cannot be evaluated for PVY incidence by summer field inspections and that require visual and laboratory post harvest testing. Economic modeling of yield effects of PVY

indicate that on average the fresh market income per acre will fall by \$8.32 for each percent of PVY in the Russet Burbank crop.

PUBLICATIONS: 2011/09/01 TO 2012/08/31

Whitworth JL, Hamm PB, Nolte PL, 2011. Distribution of Potato virus Y strains in tubers during post-harvest period. Amer. J. Potato Res. 89:136-141 McCue KF, Ponciano GP, Rockhold DR, Whitworth JL, Gray SM, Fofanov Y, Belknap WR, 2011. Generation of PVY coat protein siRNAs in transgenic potatoes resistant to PVY. Amer. J. Potato Res. 89:136-141 P. Nolte. 2012. Combatting PVY. Spudman Magazine, 50(1):42-44. P. Nolte. 2012. PVY Update. Potato Grower Magazine, 41(3):59. P. Nolte. 2012. California Grow Outs. Potato Grower Magazine, 41(4):42-43. Galvino-Costa, S.B., Figueira, A., Camargos, V.V., Geraldino, P.S., Hu, X., Nikolaeva, O.V., Kerlan, C., and Karasev, A.V. (2012) A novel type of Potato virus Y recombinant genome, determined for the genetic strain PVYE. Plant Pathology 61: 388-398. Nikolaeva, O.V., Roop, D., Galvino-Costa, S.F.B., Figueira, A.R., Gray, S.M., and Karasev, A.V. (2012) Epitope mapping for monoclonal antibodies recognizing tuber necrotic strains of Potato virus Y. American Journal

PROGRESS: 2010/09/01 TO 2011/08/31

OUTPUTS: Continuing surveys of PVY strains occurring in seed lots and commercial fields from many seed production states indicated that PVYO remains the predominant strain in the East and West, but not the Midwest. Also, incidence and distribution of tuber necrotic strains is increasing, but is still at manageable levels. Molecular and biological analysis of PVY isolates from the surveys have redefined the phylogenetic relationships among PVY strains and identified new sequence relationships that have potential to develop improved diagnostics. Major U.S. potato cultivars are being evaluated for foliar and tuber reactions to various PVY strains and strain variants. A photo gallery depicting these reactions has been posted on www.potatovirus.com along with information on PVY disease development and management. A national field trial to evaluate the susceptibility of advanced breeding material to the predominant PVY strains was established in NY, WI and ID. Three clones were tentatively identified as resistant to all strains of PVY. None of the material was observed to be susceptible to tuber necrosis disease. Preliminary cost-based risk assessment analysis of summer field inspection versus post harvest testing to determine PVY levels in seed lots suggests that correlations between values vary widely among cultivars. Seed Certification Agencies can use this information to better allocate post harvest test resources to cultivars whose PVY index levels are not accurately determined by summer inspections. To better advise the industry on levels of PVY that should be acceptable in seed stocks, field experiments were conducted in ID, WI and NY to measure the spread of virus in plots with different initial inoculum densities. This will provide information on yield and quality loss associated with levels of PVY in seed. We are also comparing minituber against conventional seed production in ID and WI to determine economics of production and vulnerability of minitubers to PVY disease. First year data are being evaluated. Hairy nightshade density and plant age appear to be a factor in PVY incidence in potato fields. Continuing field experiments will investigate the importance of weed management in maintaining low incidence of PVY and aphid vectors. Aphid vector management can reduce overall virus incidence, but improved economic outcomes and vector control can be achieved by lowering the number of sprays and timing them during high risk of virus transmission. The seasonal phenology of major PVY vectors is being determined and used to time applications of mineral oils and plant derived compounds that may have antifeedant activities. All of the field experiments will provide contributing data to develop Best Management Practices that growers can use to limit current season spread of PVY on the farm. A total of 16 presentations were made at various grower meetings, additional factsheets and popular press articles were developed (see publication list) and two new cultivars with PVY resistance were released. **PARTICIPANTS:** Stewart Gray is the overall

project manager and developed the project website. Research includes PVY surveys, the evaluation and long-term storage of PVY isolates, evaluations of PTNRD symptom expression in greenhouse and field experiments, evaluation of quantitative resistance in breeding lines, and transmission efficiency of PVY by aphids. Eric Wenninger replaces Juan Alvarez and evaluated aphid transmission of PVY strains and cultivar reactions to PVY, and assisted with evaluations of aphid populations and virus incidence in hairy nightshade plots. Amy Charkowski investigated minituber versus conventional seed potato production and evaluation of potato germplasm for symptoms and resistance to PVY in the regional trial network. Russ Groves investigated PVY infection rates in minituber versus conventional seed production, established the relationship between initial PVY infection level in seed and disease progress plus final incidence at the end of the growing season, documented the seasonal phenology and dispersal of the soybean aphid and determined their relationship to PVY disease progress, and evaluated the effects of foliar protectants to limit or minimize PVY incidence in established field plots. Pamela J.S. Hutchinson investigated the role of hairy nightshade in aphid and PVY incidence. Alexander Karasev is responsible for large-scale genetic (RT-PCR) and serological (ELISA and Western blot) typing of all the field PVY isolates collected by Seed Certification agencies and individual researchers working on this project. Karasev's lab will provide pure cultures of various PVY recombinant strains from Idaho, Wisconsin, or New York, for potato genetic screening experiments. Christopher McIntosh is responsible for the economic evaluation of and risk assessment of in-season PVY field inspections versus post-season tests, the economics of minituber versus conventional seed production, and the economic impact of PVY on commercial potato production, which has direct impacts on the value of seed and seed certification programs. Phillip Nolte is responsible for receiving and coordinating information from the seed certification agencies, the production of PVY training manuals, and will perform field trials to measure the impact of seed borne PVY on yield and the influence of seed borne PVY on the amount of PVY present in the harvested crop. Jonathan Whitworth's role is to plan, coordinate and administer national PVY field trials and to evaluate cultivars for foliar and tuber symptoms to PVY strains. Seed certification agencies from ME, NY, MI, WI, MN, ND, NE, WY, MT, CO, ID, CA, WA, OR, AK participate in this project by supplying plant and tuber samples, and by providing data on summer and post harvest test evaluations of seed lots.

TARGET AUDIENCES: The outputs and outcomes of this project are targeted mainly at the various segments of the potato production industry, including seed producers, fresh-pack and process growers. We also provide information to seed certification agencies, regulatory agencies and to the general scientific community.

PROJECT MODIFICATIONS: Nothing significant to report during this reporting period.

IMPACT: 2010/09/01 TO 2011/08/31

Our surveys of PVY strains affecting the U.S. potato crop and follow-up characterization of the viruses has defined the existing and potential problems that the emerging strains impose on the potato industry. Emerging, recombinant PVY strains that induce mild symptoms in many potato cultivars are problematic for seed certification agencies that depend on observable symptoms to inspect and grade seed lots. To assist with this, we are documenting foliar and tuber symptoms on the major potato cultivars grown in the U.S. caused by these PVY strains. This photographic information is made available on www.potatovirus.com. Several of the state seed certification agencies are conducting more laboratory testing than in the past to better evaluate PVY incidence during the summer growing season and during the winter post harvest tests. Laboratory testing is focused primarily on those cultivars that show mild or transient symptoms. The emergence of PVY strains causing tuber necrosis disease in the U.S. has also prompted some seed certification agencies to begin testing PVY infected material specifically for the tuber necrotic strains. Growers are urged not to reenter into certification those seed lots infected with the tuber necrotic strains and not to plant that seed in seed production areas. As a result of our

recent research, the U.S. potato industry, working through the National Potato Council, has adopted decisive changes in state and international requirements for moving seed potatoes, namely that all seed moved across domestic and international borders be certified and subjected to a post harvest test. The maximum tolerance level determined in the post harvest test is 2% for seed lots re-entering the certification process. Maximum tolerance level determined in the summer inspection is 3% for total virus, 2% for mosaic. Many state certification agencies are asking growers to abide by the same rules for seed moving within a state and many states have adopted strict seed laws that require certified seed for planting. To minimize the release of asymptomatic cultivars from breeding programs, we have established national trials with most of the U.S. -based potato breeding programs to evaluate their advanced clones for susceptibility and symptom expression when infected with the three predominant PVY strains found in the U.S. Understanding the seasonal phenology of aphid vector populations enhances grower options for PVY management, ultimately minimizing environmental impact and promoting IPM. Our work has identified when elevated risk for PVY transmission occurs and which aphid vector species should be targeted for control. Precisely timed applications of foliar, behavioral modifying plant protectants (mineral oils and natural plant extracts) can reduce PVY spread in the field and thus protect high value early generation seed stocks.

PUBLICATIONS: 2010/09/01 TO 2011/08/31

Karasev, A., Hu, X., Kerlan, C., Nikolaeva, O. Crosslin, J., Brown. C., and Gray, S. 2011. Genetic diversity of the ordinary strain of Potato virus Y (PVY) and origin of recombinant PVY strains. *Phytopathology* 101:778-785 Kerlan, C., Nikolaeva, O., Hu, X., Meacham, T., Gray, S., and Karasev, A. 2011. Identification of the molecular make-up of the Potato virus Y strain PVYZ: genetic typing of PVYZ-NTN. *Phytopathology* 101:1052-1060 Cavatorta, J., Perez, K., Gray, S., Van Ek, J., Yeam, I., and Jahn, M. 2011. Engineering virus resistance using a modified potato gene. *Plant Biotech. J.* 9:1014-1021 Mello, A.F.S., Olarte, R.A., Gray, S.M., and Perry, K.L. 2011. Transmission efficiency of Potato virus Y strains PVYO and PVYN-Wi by five aphid species. *Plant Dis.* 95:1279-1283. Nolte, P. Potato Pointers: The New Face of Potato Virus Y. *American Vegetable Grower.* 58(112):57 December, 2010. Karasev, A. and P. Nolte, P. Taking Precautions: Keeping the Industry Clean from PVY. *Potato Grower.* 40(1):54-55 January,

PROGRESS: 2009/09/01 TO 2010/08/31

OUTPUTS: Surveys of PVY strains occurring in seed lots from 11 of 15 seed production states indicated that PVYO remains the predominant strain affecting potato, but that incidence and distribution of tuber necrotic strains has increased since a 3 year survey was completed in 2006. Eighteen potato cultivars were evaluated in the greenhouse for susceptibility to tuber necrotic ringspot disease caused primarily by the PVYNTN strain. Four were highly susceptible, 4 moderately susceptible and 2 had mild symptoms. Isolates within the PVYNWi and PVYO strains were also found to induce PTNRD. Field experiments were conducted to: 1. evaluate foliar and tuber symptoms on 42 potato cultivars 2. determine the role of nightshade populations on aphid and virus incidence in potato 3. determine the relationship of initial levels of PVY in the seed to final disease incidence 4. compare PVY epidemiology in plots grown from minitubers verses conventional seed 5. investigate the seasonal phenology of potential vectors and the relationship to disease incidence 6. evaluate the efficacy of several compounds with antifeedant activity to control PVY Data from all field trials are currently being analyzed. A total of 14 presentations describing research and extension efforts related to this grant were made at various state and national potato grower meetings. A website (<http://www.potatovirus.com/>) was developed to describe the activities of the grant and to provide information and outreach to various sectors of the potato industry. The site features a Discussion Forum where anyone can sign in and ask questions, provide information or comment on postings. The latest information on PVY and

other potato viruses is posted as it is developed by the researchers on the project or others that want to contribute information. The website also functions as a principal mechanism for researchers and collaborators on the project to share data and information with each other on secure pages not accessible by the general public. A training manual for seed certification officials and inspectors, as well as, growers is being developed that provides information on and pictures of foliar and tuber symptoms on major North American cultivars that are caused by the various PVY strains. This is available on the above mentioned website. A video was developed to help growers, seed certification and regulatory officials, consultants, and other practitioners in the United States understand how and why PVY is affecting the seed potato crop and if left unchecked why it will become a major disease issue for the food sector of the potato industry. We address the reemergence of PVY as a disease problem in seed potatoes, how regulatory, business and science-based activities have addressed the problem, and finally what steps can and are being taken to manage the disease and prevent the newly introduced tuber necrotic strains of the virus from becoming a major quality issue for potato growers.

[http://www.plantmanagementnetwork.org/edcenter/seminars/potato/Potato VirusY/](http://www.plantmanagementnetwork.org/edcenter/seminars/potato/Potato%20VirusY/)
Additional factsheets and popular press articles were developed (see publication list) and two new cultivars with PVY resistance were released. PARTICIPANTS: Stewart Gray is the overall project manager and developed the project website. Research includes the evaluation and long-term storage of PVY isolates, evaluations of PTNRD symptom expression in greenhouse and field experiments, and evaluation quantitative resistance in breeding lines. Juan Manuel Alvarez evaluated quantitative resistance in selected breeding lines using aphid inoculations, and assisted with evaluations of aphid populations and virus incidence in hairy nightshade plots. Amy Charkowski investigated minituber versus conventional seed potato production and evaluation of potato germplasm for symptoms and resistance to PVY in the regional trial network. Russ Groves investigated PVY infection rates in minituber versus conventional seed production, established the relationship between initial PVY infection level in seed and disease progress plus final incidence at the end of the growing season, documented the seasonal phenology and dispersal of the soybean aphid and determined their relationship to PVY disease progress, and evaluated the effects of foliar protectants to limit or minimize PVY incidence in established field plots. Pamela J.S. Hutchinson investigated the role of hairy nightshade in aphid and PVY incidence. A.V. Karasev's lab will be responsible for large-scale genetic (RT-PCR) and serological (ELISA and Western blotting) typing of all the field PVY isolates collected by Seed Certification agencies and individual researchers working on this project. Karasev's lab will provide pure cultures of various PVY recombinant strains from Idaho, Wisconsin, or New York, for potato genetic screening experiments. Christopher McIntosh will be responsible for the economic evaluation of and risk assessment of in-season PVY field inspections versus post-season tests, the economics of minituber versus conventional seed production, and the economic impact of PVY on commercial potato production, which has direct impacts on the value of seed and seed certification program. Phillip Nolte is responsible for receiving and coordinating information from the seed certification agencies, the production of PVY training manuals, and will perform field trials to measure the impact of seed borne PVY on yield and the influence of seed borne PVY on the amount of PVY present in the harvested crop. Dr. Whitworth's role is to plan, coordinate and administer national PVY field trials and to evaluate cultivars for foliar and tuber symptoms to PVY strains. Seed certification agencies from ME, NY, MI, WI, MN, ND, NE, WY, MT, CO, ID, CA, WA, OR, AK participate in this project by supplying plant and tuber samples, and by providing data on summer and post harvest test evaluations of seed lots TARGET AUDIENCES: The outputs and outcomes of this project are targeted mainly at the various segments of the potato production industry including seed producers and fresh-pack and process growers. We also provide information to seed certification agencies, regulatory agencies and to the general scientific community. PROJECT MODIFICATIONS: Nothing significant to report during this reporting period.

IMPACT: 2009/09/01 TO 2010/08/31

In recent years, new strains of PVY have been discovered in the U.S. Some cause severe necrosis in potato and tobacco and are a threat to commercial production. U.S. regulatory agencies, and their international equivalents, test for these necrotic strains using antibody based lab tests. We discovered a PVY variant that is identified as a necrotic strain by these antibody tests, but this variant does not cause necrosis in the host plant. Detection of this variant has resulted in shipments of potatoes being refused at border checkpoints and/or being destroyed rather than being sold. We characterized the new virus variant and determined that a minor change in one virus protein allows the virus to react with the antibody currently used to detect necrotic strains of PVY. This simple change in one protein does not affect any other property of the virus. We tested other commercially available antibodies and identified one that does not identify this variant as a necrotic virus, but it does recognize the real necrotic viruses as such. A simple change in testing procedures has been adopted by U.S. seed certification agencies and changes are being implemented in the North American Plant Protection Organization standards that will avert these erroneous results and facilitate trade of potatoes across state and international boundaries. (Karasev et al. 2010) PVY has become the number one disease problem for the seed potato industry and the most important virus disease affecting potato. PVY was successfully managed for many decades by seed certification practices, but the introduction of new strains of the virus, the release of several popular asymptomatic cultivars, and an increase in late season disease spread have all reduced the effectiveness of visual inspections of the potato crop during the growing season as a means of weeding out those fields that have high virus infection. New knowledge of the strains composition now found in the U.S. crop has led to a revision of seed certification and disease management practices (Gray et al 2010).

Successful management of PVY in the future will require an integrated approach of seed certification, on-farm disease control and the integration of virus resistance into new cultivars. To this end, our work has led to state seed laws that now require seed moving interstate or internationally to be certified using information from a post harvest test and strict tolerances on the level of PVY allowed. Similar provisions are being proposed for inclusion into a U.S. - Canada Tuber Necrotic Virus Management Plan that allows for unrestricted movement of seed potatoes across the international border. The release of potato cultivars that are susceptible to PVY, but do not express symptoms contribute to the increased levels of PVY in seed and commercial crops. We have initiated a collaborative project with most of the major U.S. potato breeding programs to develop national potato trials that will evaluate breeding material against a set of PVY strains in multiple locations to determine the level of susceptibility and symptom expression, foliar and tuber, so that this information is available prior to release of a cultivar.

PUBLICATIONS: 2009/09/01 TO 2010/08/31

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