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2017 Potato Symptoms in the Columbia Basin: Is there a pathogenic cause?

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Starting in early- to mid-July of the 2017 growing season, potato crops across the Columbia Basin began showing symptoms of purpling terminals, distorted leaves with warping, crinkling, and holes, and stems with bumps or blisters, often accompanied with spots of necrosis. Symptoms increased as the season went on, and by mid-August, it was difficult to find a field without symptoms. Symptomatic fields were typically 90% affected, or higher, depending upon the variety. Symptomatic foliar tissue was collected from several commercial and research fields across the Columbia Basin, and taken to the USDA-ARS research laboratory in Prosser, WA, for grafting and molecular analyses. None of the symptoms appeared graft-transmissible, as there was no correlation between field symptoms and recipient plant symptomology. In addition to this, standard molecular analyses did not detect any common bacterial or viral pathogens that correlated with these field symptoms. Results from this first year of study indicate that the causal agent of the symptoms of leaf distortion, purpling, and stem blistering in 2017 were not pathogenic in nature.

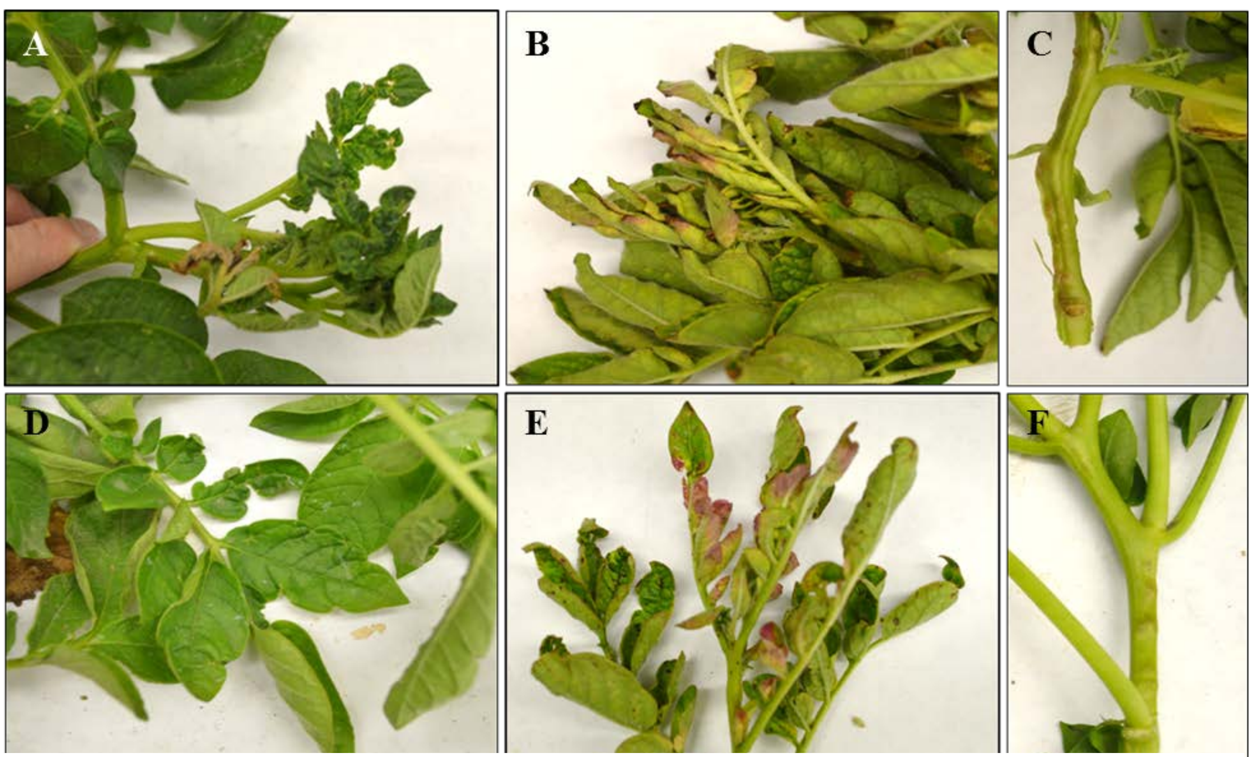

Figure 1. Symptoms observed across the Columbia Basin during the 2017 growing season. A and D) Crinkly, warped, distorted leaves. B and E) Purple terminals, negative for BLTVA phytoplasma. C and F) Stem blistering, bumps, and necrotic lesions.

Seventy five symptomatic potato plants were collected or received from eight different commercial or research fields across the Columbia Basin in July and August of 2017. Symptoms included purpling of upper leaf terminals, leaf distortion on newer growth, including leaf crinkling, warping, and holes, and bumps or blisters along the stems, often accompanied by necrotic lesions (see Figure 1). The purpling terminal leaves were similar to, or indistinguishable from, those caused by the beet leafhopper transmitted virescence agent (BLTVA) phytoplasma. Symptoms were seen across entire fields; there did not appear to be any edge effect as often seen with insect-vectored pathogens which occur when the insect migrates into a field. Standing back from several fields, it was difficult to see the symptoms, but once in the field, it was near impossible to find a plant that did not have one or more of the symptoms listed above. In fact, most fields showed symptom incidence of 90% or higher. These symptoms were observed in at least five different potato cultivars, including Ranger Russet, Alturas, Umatilla Russet, Clearwater, and Challenger. Symptoms were worse in some cultivars compared to others, specifically Umatilla Russet. An organic commercial potato field was observed to have the same symptoms, indicating that chemical damage was likely not the cause of these symptoms.

Samples were subjected to both graft analysis and molecular diagnostic analysis at the USDA-ARS research station in Prosser, WA. Grafting was used to indicate if the causal agent was capable of moving systemically within a new recipient plant, thereby indicating the presence of a pathogen as opposed to chemical, environmental, or insect damage. The ultimate goal in combining the graft analysis with molecular testing was to specifically identify if a pathogen(s) was the causal agent of these symptoms.

Materials and Methods:

Grafted plants were generated in the greenhouse at the USDA-ARS research station in Prosser, WA, each with two or three scions originating from the field-collected plants. Scions were grafted using standard technique (Secor et al. 2009) and observed on a weekly basis. Notes were taken to indicate how long the grafted scions survived, and whether any symptoms appeared on the recipient plants. Control plants were also grafted each time field-collected samples were grafted, and consisted of greenhouse-grown scions grafted to greenhouse-grown recipient plants. These controls enable the identification of any symptoms generated by the greenhouse conditions.

All samples collected or received from the field were subjected to molecular laboratory diagnostic assays to identify any pathogen(s) present in the samples. Two samples were taken from the leaf tissue of each plant for diagnostics to ensure that detection of a pathogen would not be missed. Additionally, tubers were collected or received from three different field sites and sampled for pathogens by molecular methods. Nucleic acids were extracted from each leaf and tubers sample using the standard Dellaporta protocol (Crosslin and Hamlin 2011). Extracts were then subjected to standard polymerase chain reaction (PCR) analysis or reverse transcription-PCR analysis for the detection of the following pathogens: phytoplasmas (universal target), '*Candidatus* Liberibacter solanacearum' (Lso), *tobacco rattle virus*, *potato mop top virus*, *alfalfa mosaic virus*, *tomato spotted wilt virus*, *potato leafroll virus*, *potato virus Y*, and fungal pathogens (universal target) (Crosslin et al. 2011, Crosslin and Hamlin, 2011, Cating et al. 2015).

Results: *Grafting analyses:* Forty eight field-collected samples were grafted to plants grown from greenhouse-grown minitubers at the USDA-ARS research station in Prosser, WA. Overall, no symptoms developed on the recipient plants that correlated to the field symptoms of purpling, leaf distortion and stem blistering/necrosis. Nearly all recipient plants with symptomatic scions originating from the field showed normal plant growth similar to the control plants (Figure 2).

Figure 2. All images depict recipient plants with field-collected scions grafted six and a half weeks prior. Red arrows point to the scions visible on each recipient plant.

Two recipient plants developed purpling stems and leaves, and had high levels of aerial tuber formation. The scions on these grafted plants were from field samples that tested positive for BLTVA phytoplasma, the causal agent of purple top disease in the Columbia Basin. These were the only grafted plants that tested positive for BLTVA, despite several other field-collected plants showing purpling symptoms. Purple top symptoms on BLTVA-positive and BLTVA-free plants were virtually indistinguishable (Figure 3).

Molecular analyses:

All 75 samples collected from commercial and research fields in the Columbia Basin were subjected to standard molecular diagnostic procedures in the laboratory for common pathogens of potato, as well as for all fungal species. There was no pathogen identified that correlated with the symptoms of purpling terminals, leaf distortion, or stem blistering/necrosis. Four samples with purple top symptoms tested positive for BLTVA phytoplasma. As noted above, two of these samples were grafted to healthy recipient plants in the greenhouse and showed systemic BLTVA infection, thereby confirming the molecular results.

Several of the field-collected samples were positive for various strains of PVY, specifically PVY^{NTN} and PVY$^{N:O}$/PVY$^{N\text{-}Wilga}$. However, there was no correlation in symptoms of the field-collected samples and specific PVY strains. Therefore, PVY did not appear to be associated with these symptoms.

Similarly, several foliar samples generated a PCR product when a universal fungal genetic region was targeted. Five of these samples (different cultivars and different symptoms) were subject to molecular cloning and sequencing of the unknown fungal pathogen(s). Sequencing of three clones from each sample did not identify any pathogen that was correlated with the symptoms of leaf distortion, purple top, or stem blistering/necrosis. Common fungal pathogens were however identified,

Figure 3. Comparison of purple top symptoms from a BLTVA-free (A) and a BLTVA-positive plant (B).

including *Altenaria* species and *Sclerotinia sclerotiorum*.

Tuber samples received from three fields in the Columbia Basin showed no internal defects, and appeared to be of good size during mid-August. These samples were tested for the same pathogens by molecular diagnostic methods. No pathogens were identified in these samples.

Conclusions:

The field symptoms from 2017 were not graft-transmissible, and no pathogen was correlated to the symptoms using molecular analyses in the laboratory. These results are also consistent with the findings from the plant disease diagnostic clinic at HAREC, where 0% of the samples submitted with symptoms described above tested positive for the presence of either BLTVA or Lso. Only 3% of these symptomatic plants submitted to HAREC tested positive for PVY. Based on this, it must be concluded that the cause of the 2017 field symptoms was not pathogenic in nature. Despite the above-ground symptoms, there did not appear to be symptoms in the tubers sampled, and it appeared that tuber yield was good at the time of sampling. These observations suggest that the symptoms likely did not cause quality or quantity losses for growers. Even though no causal agent of the symptoms was identified, this information is necessary for growers to know in order to continue the quest to identify a causal agent and prevent these symptoms in the future.

Despite these conclusions, repeating the graft and molecular analyses for a second year, should the symptoms arise again in 2018, will be necessary to be 100% confident that these symptoms are truly not pathogenic in nature. A second year of study will validate the results from 2017 and provide maximum confidence that the symptoms are in fact not triggered by a pathogen, and may provide further information about the true nature of these symptoms. **If you see any potato plants exhibiting symptoms of leaf distortion, stem blistering, or purpling terminals during the 2018 field season, please contact Kylie Swisher Grimm at kylie.swishergrimm@ars.usda.gov or (509) 786-9362. Carrie Wohleb (cwohleb@wsu.edu) or Tim Waters (twaters@wsu.edu) can also be contacted about symptomatic potato plants.**

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References:

Cating, R.A., C.N. Funke, N. Kaur, P.B. Hamm, and K.E. Frost. 2015. Multiplex reverse transcription (RT) high-fidelity PCR protocol for the detection of six viruses that cause potato tuber necrosis. American Journal of Potato Research 92: 536-540.

Crosslin, J.M., H. Lin, and J.E. Munyaneza. 2011. Detection of '*Candidatus* Liberibacter solanacearum' in the potato psyllid, *Bactericera cockerelli* (Sulc), by conventional and real-time PCR. Southwestern Entomologist. 36: 126-135.

Crosslin, J., and L. Hamlin. 2011. Standardized RT-PCR conditions for detection and identification of eleven viruses of potato and potato spindle tuber viroid. American Journal of Potato Research 88: 333-338.

Secor G.A., V.V. Rivera, J.A. Abad, I.-M. Lee, G.R.G. Clover, L.W. Liefting, X. Li, and S.H. De Boer. 2009. Association of '*Candidatus* Liberibacter solanacearum' with zebra chip disease of potato established by graft and psyllid transmission, electron microscopy, and PCR. Plant Disease 93: 574-583.