Beet Leafhopper and Potato Purple Top Disease: 2005 Season Recap and New Research Directions

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Introduction

During the 2002 growing season, Columbia Basin potato growers in Washington and Oregon witnessed a serious outbreak of potato purple top disease in their fields. This outbreak caused significant yield losses and a reduction in tuber quality. During ensuing years, the disease has also been observed, especially in potato fields not timely treated with insecticides. Symptoms in affected potato plants include a rolling upward of the top leaves with yellowish, reddish or purplish discoloration, moderate proliferation of buds, shortened internodes, swollen nodes, aerial tubers and early plant decline.

In response to this disease outbreak, the Washington State Potato Commission and Oregon Potato Commission funded a multi-disciplinary team to look at the situation. The team is basically composed of researchers in the Pacific Northwest, including entomologists and plant pathologists from the federal government, universities and private industry. The team's mission is to investigate various aspects of the problem, including disease causal agent(s) identification, insect(s) vectoring the disease, disease epidemiology and disease management.

Using the polymerase chain reaction (PCR) technique, a much more sensitive method than ELISA, it was determined that the beet leafhopper-transmitted virescence agent (BLTVA) phytoplasma (also known as the Columbia Basin potato purple top phytoplasma) was the causal agent of the disease. Phytoplasmas are microorganisms that have characteristics of both bacteria and viruses and have been difficult to diagnose in the past. Phytoplasmas are usually transmitted by several groups of insects, including leafhoppers, planthoppers and psyllids. Investigation of the insects potentially vectoring this potato disease in the Columbia Basin indicated that this phytoplasma was almost exclusively associated with the beet leafhopper (*Circulifer tenellus*).

The following objectives were addressed in the 2005 research season: 1) Develop information on the population dynamics of the beet leafhopper in the Columbia Basin potato production area; 2) determine the susceptibility to BLTVA of different potato cultivars important to the Columbia Basin and assess the impact of BLTVA on potato tubers; 3) develop quick and cost-effective testing protocols for phytoplasma detection in plants and insects; 4) conduct epidemiological studies on fields affected by the phytoplasma; and 5) implement management strategies for the problem.

Population Dynamics of the Beet Leafhopper in the Columbia Basin in 2005

A region-wide trapping system was conducted to monitor beet leafhoppers with yellow sticky traps in a standardized fashion from northern Oregon to north of Moses Lake in Washington. Traps were collected and replaced weekly from late spring to early November. This work was conducted to gather baseline information on leafhopper flights and populations that could be fitted into an IPM program that is the overall goal of this research project. Trapping data were regularly reported on the WSPC website: www.potatoes.com/research.cfm.Because of some limitations in 2005 leafhopper trapping capacity, data for the south WA Basin is not available. Trapping results for north and central Washington Columbia Basin and northern Oregon are presented in Figures 1, 2, 3, and 4. In Washington, beet leafhopper catch was very high early in the 2005 season (Fig. 1), but much lower than in 2004 by late season. However, early in the season, beet leafhopper populations were about 5 times higher than those recorded in 2004 in several parts of the Columbia Basin (Figs. 2 and 3); the causes of this high increase in leafhopper numbers are not well known. Similarly, beet leafhopper numbers in northern Oregon were higher in 2005 than in 2004 early in the season and the population dynamics pattern in 2005 was similar to the northern Washington (Fig. 4).

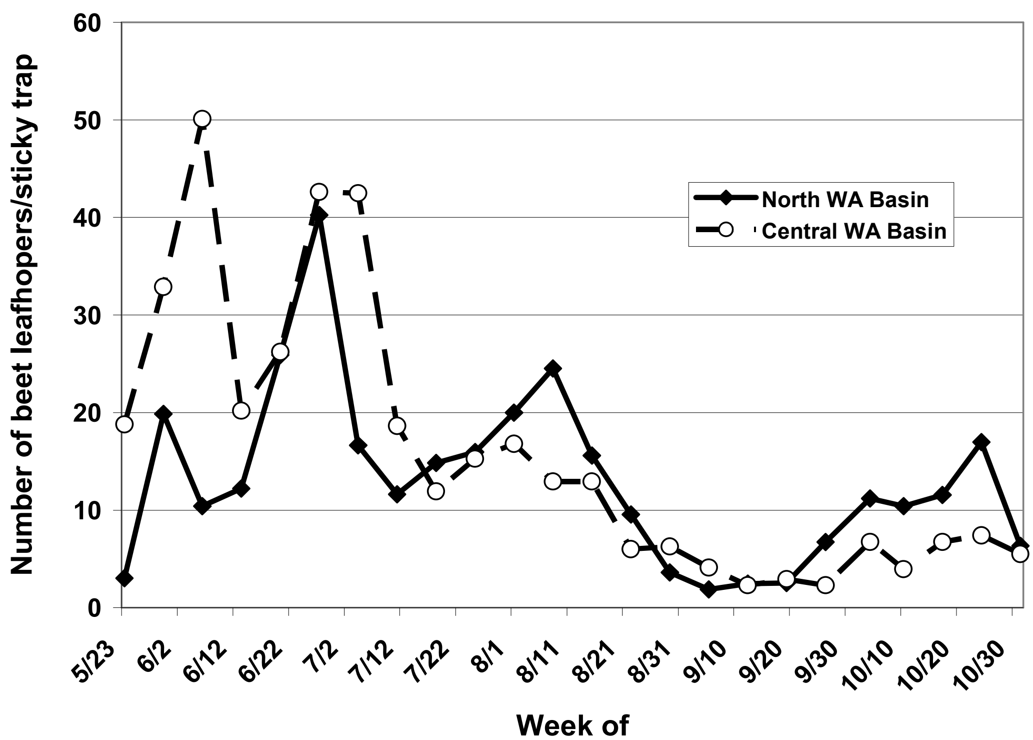

Figure 1. Average number of beet leafhoppers per sticky trap in north and central Columbia Basin of Washington in 2005.

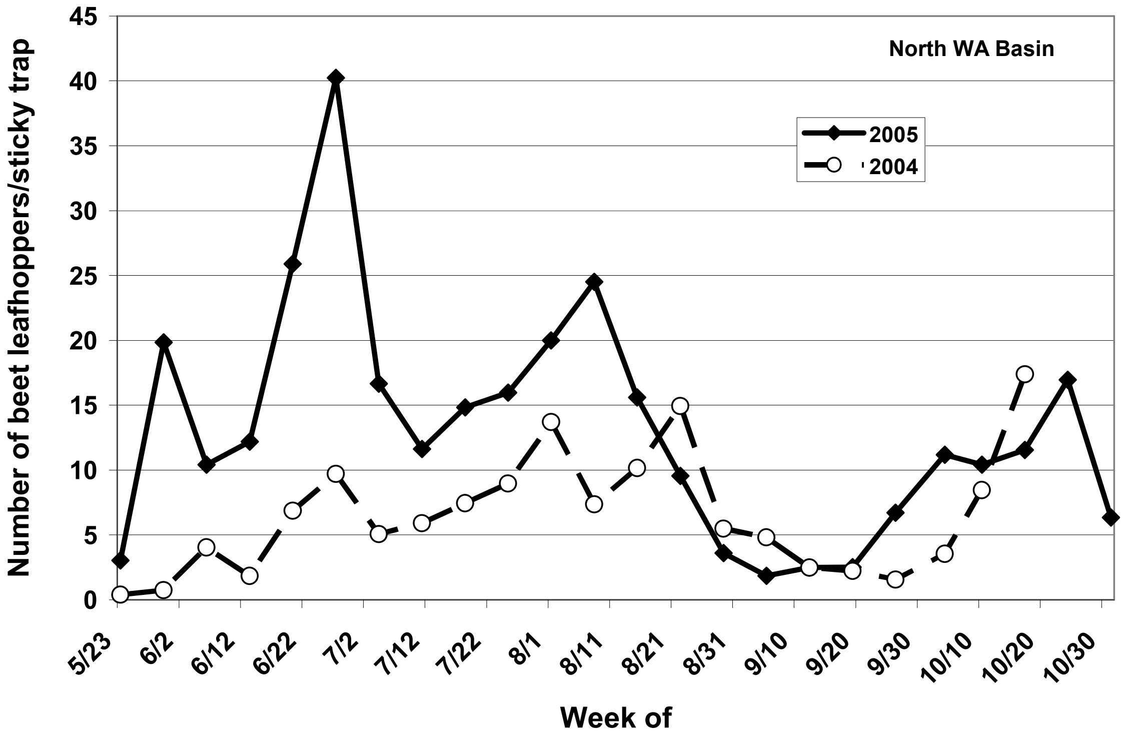

Figure 2. Average number of beet leafhoppers per sticky trap in the north Columbia Basin of Washington in 2005 compared to 2004 leafhoppers catch.

Susceptibility of Different Potato Cultivars to BLTVA Phytoplasma

The study was conducted in the laboratory at the USDA-ARS at Wapato, WA. Potato cultivars that were tested include Ranger Russet, Russet Burbank, Shepody, Umatilla Russet, Russet Norkotah, and Alturas. At the request of Frito-Lay, Inc., some chipping varieties were also assessed for BLTVA susceptibility. Different plant stages of each potato cultivar were exposed to BLTVA infective beet leafhoppers for 7 days in transmission cages in the laboratory (colonies of beet leafhoppers and various BLTVA infected plants are maintained in the Wapato Lab). More than 100 plants were exposed when plants were 1-3 inches tall. After exposure, these potato plants were removed from transmission cages in the laboratory and then maintained in large screen cages established outdoors to mimic natural conditions and to exclude aphids and leafhoppers; these plants were treated with insecticides at 2-week intervals to exclude potential vectors and other insects. Also, visual inspections of the presence of insects on the plants were conducted on a regular basis. Clean potato plants that had not been exposed to beet leafhoppers were maintained in the outdoor cages along with the exposed potatoes to verify that no unintended transmission occurred. Plants were later moved in the greenhouse to avoid freezing. Plants were observed for potato purple top symptoms throughout the experimental period and were tested for BLTVA phytoplasma using PCR at desired intervals.

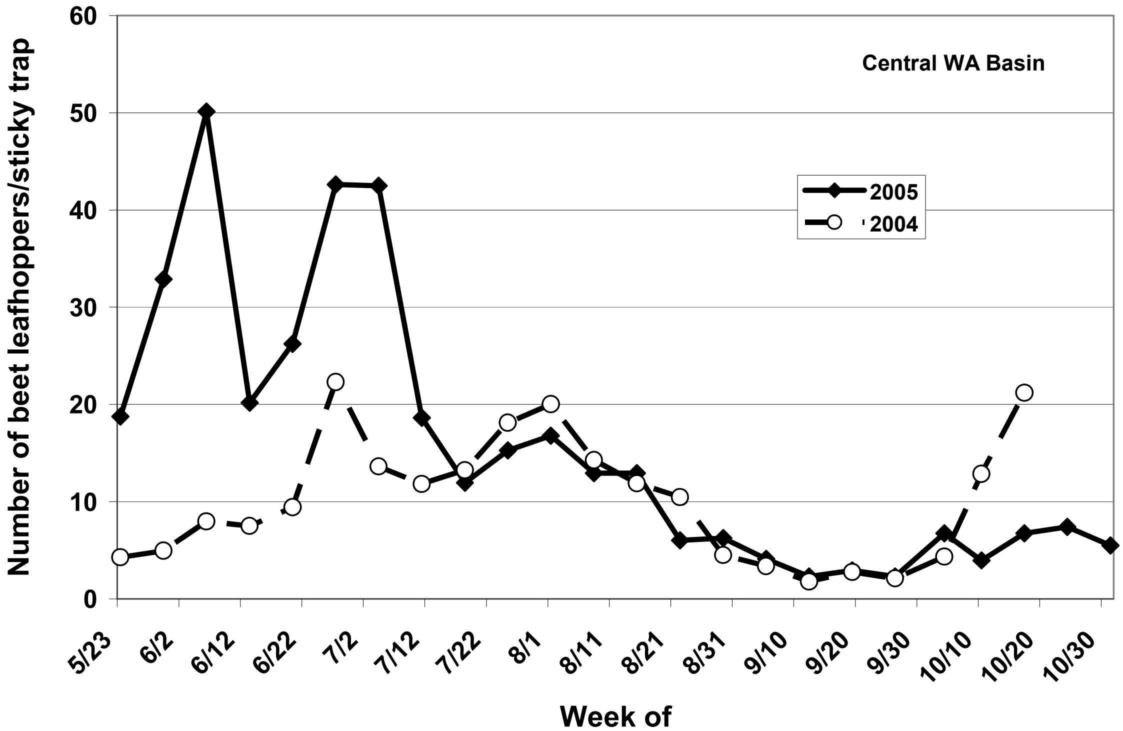

Figure 3. Average number of beet leafhoppers per sticky trap in the central Columbia Basin of Washington in 2005 compared to 2004 leafhoppers catch.

Out of 109 potato plants tested, 54 plants (49.5%) tested positive for the BLTVA phytoplasma (Table 1) and showed typical potato purple top symptoms. These symptoms included upward rolling of the top leaves with yellowish, reddish, or purplish discoloration, leaf mottling, moderate proliferation of buds, shortened internodes, swollen nodes, aerial tubers, and early plant decline. Results of the present study confirmed that the beet leafhopper effectively transmits the purple top disease phytoplasma to potatoes, and agrees with other data that this insect is the major vector of potato purple top disease in the Columbia Basin.
Current results indicate that some potato varieties are more susceptible to the phytoplasma than others (Table 1). Umatilla Russet, Ranger Russet, Russet Norkotah, and Shepody were very susceptible, whereas Alturas and Russet Burbank showed resistance to the phytoplasma. Interestingly, Russet Burbank did not test positive for BLTVA or show purple top disease symptoms until close to harvest when plants were senescing; similar observations were also made in the field. During these laboratory experiments, no infection was observed or BLTVA detected in Alturas, despite the fact that several of these plants were tested about 90 days after exposure. Chipping cultivars Atlantic and FL 1879 were very susceptible to the phytoplasma (Table 1).
Experiments using these potato cultivars are planned for the 2006 growing season to assess their susceptibility to BLTVA under field conditions.

Table 1. Susceptibility of potato cultivars to BLTVA phytoplasma; study was conducted at the USDA-ARS at Wapato.			
Potato cultivar	**Number of plants tested**	**Number of plants positive for BLTVA**	**Percent infection**
WSPC funded varieties			
Ranger Russet	13	9	69.2
Russet Norkotah	14	8	57.1
Umatilla Russet	7	5	71.4
Shepody	7	3	42.8
Russet Burbank	14	3	21.4
Alturas	7	0	0.0
Other varieties			
Atlantic	13	8	61.5
FL 1879	11	8	72.7
FL 1833	13	6	46.2
FL 1867	10	4	40.0

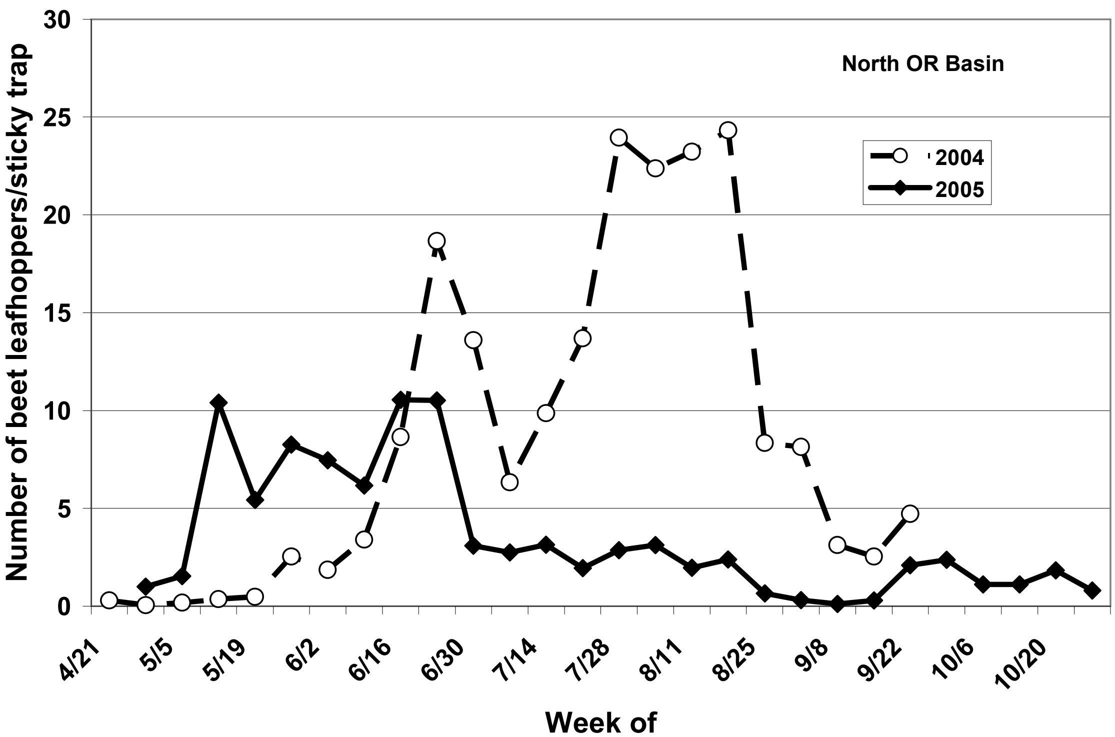

Figure 4. Average number of beet leafhoppers per sticky trap in the north Oregon Columbia Basin in 2005 compared to 2004 leafhoppers catch.

Impact of BLTVA Phytoplasma on the Quality of Potato Tubers

To look at the impact of the BLTVA phytoplasma on the potato tuber quality, two research plots were established at the UDSA-ARS research farms at Moxee (var. Russet Burbank) and Paterson (var. Umatilla Russet and Ranger Russet). Potato plants were monitored for BLTVA by collecting leaf tissues and testing them for BLTVA using PCR. These plants were marked and observed for purple top symptoms. At the end of the growing season, marked potato plants were hand harvested. Tubers from clean and BLTVA-infected plants for each cultivar were assessed for tuber quality at the potato processing plant of J. R. Simplot Company in Moses Lake, WA. To assess the tuber quality, samples of 40 tubers each were used at each processing date; all 40 tubers were measured for solids (specific gravity) and then 20 tubers were sliced and fried to test for high sugars and sugar ends whereas the other 20 tubers were checked for internal defects. Furthermore, potato tubers from chipping varieties Atlantic, FL 1833, FL 1867, and FL 1879 were collected from clean and BLTVA-infected potato plants grown in pots in the greenhouse. These potato tubers were shipped to Frito-Lay in Rhinelander, WI, for quality processing.

Results indicated that there were differences in tuber solids (specific gravity) and sugar content between BLTVA infected and non-infected potato plants (Table 2). Some of the tubers from infected potato plants showed 100% No. 4 USDA fry color, which is not commercially acceptable. It is suspected that this BLTVA phytoplasma has all long been contributing to the increasingly loss of potato tuber quality in the affected areas. Very few internal defects were observed. There was a difference in quality of potato chips between tubers from clean and infected plants, with almost all tubers from BLTVA-infected plants producing commercially unacceptable chips in all the four varieties tested. More field and laboratory experiments are planned for the 2006 growing season to look into this factor and gain more understanding of the impact of this phytoplasma on the potato tuber quality.

Weeds and Overwintering Beet Leafhoppers as Potential Sources of the BLTVA Phytoplasma

Weeds surrounding potato fields play an important role in the beet leafhopper dispersal and the epidemiology of potato purple top disease. The beet leafhopper has a very wide host range and prefers plants in the mustard family. BLTVA phytoplasma is also believed to affect a large number of plants, including several vegetable and ornamental crops. Previous studies show that, under laboratory conditions, beet leafhoppers are able to transmit phytoplasma to more than 40 host plants.

Experiments were conducted at the USDA-ARS in Prosser, WA, to determine which crop plants and weeds may be reservoirs of the BLTVA phytoplasma. Beet leafhoppers were used to transmit the BLTVA to selected crop plants and weeds, including beets. Seeds of table beets, common groundsel, shepherd's purse, kochia, prickly lettuce, clasping pepperweed, wild radish and Russian thistle were sown in commercial potting mix (Sunshine Mix Number 1; Sun Gro Horticulture, Bellevue, WA) in the greenhouse. Plants were individually enclosed in a cage and infested with 5-10 beet leafhopper adults that had been raised on BLTVA-infected daikon radish. All the plants were 4-6 inches tall when infested except the common groundsel and clasping pepperweed plants which were in rosette stage. The cages were placed in growth chambers. Some of the plants were infested twice when it appeared that most of the first group of insects did not survive for 7-10 days, at which time more insects were introduced into cages. Leafhoppers remained on the plants for 1-3 weeks before the plants were sprayed with insecticide.

The plants were observed for BLTVA symptoms and tested by PCR for BLTVA 5-7 weeks later. Results of the present study showed that the beet leafhopper transmitted BLTVA phytoplasma to 10 of 11 beets and many of the weeds tested (Table 3). The phytoplasma was transmitted to at least one plant of six of the seven weed species inoculated; none of the prickly lettuce plants were positive by PCR. The beets and most of the weeds failed to produce visible symptoms of phytoplasma infection 5-7 weeks after exposure. Infected common groundsel, however, developed flower abnormalities including virescence and phyllody, typical of phytoplasma infection. A few of the infected kochia plants senesced prematurely.

Similarly to the above report on potatoes, this is the first report of the transmission of BLTVA by the beet leafhopper to beets, common groundsel, kochia, Russian thistle, and Shepherd's purse. Before invading potatoes, it is believed that leafhoppers acquire phytoplasma from weeds. Further studies to elucidate the role of weeds surrounding potato fields in the dispersal of the beet leafhopper and the epidemiology of the potato purple top disease in the Columbia Basin are planned.

Table 3. Transmission of the BLTVA phytoplasma to table beets and selected weeds by the beet leafhopper; study conducted at the USDA-ARS in Prosser.

Common name	Scientific name	Inoculation access period (days)[1]	Number of plants infested	Number of plants positive by PCR[2]
Beets	*Beta vulgaris*	7	11	10
Kochia	*Kochia scoparia*	7-20[3]	24	8
Russian thistle	*Salsola iberica*	7	17	4
Prickly lettuce	*Lactuca serriola*	7	8	0
Shepherd's purse	*Capsella bursa-pastoris*	7-20[3]	4	4
Clasping pepperweed	*Lepidium perfolia-tum*	9-20[3]	4	1
Wild radish	*Raphanus rapha-nistrum*	10-15[3]	18	17
Groundsel	*Senecio vulgaris*	7	6	4

[1]Plants were infested with 5-10 beet leafhoppers per plant during the indicated period before being sprayed with insecticides.
[2]Plants were tested by nested PCR, 5-7 weeks after the inoculation access period.
[3]These plants were infested with leafhoppers twice.

Furthermore, a survey was conducted by researchers at the USDA-ARS at Wapato during the winter and early spring of 2005 to determine overwintering locations of the beet leafhoppers in the Columbia Basin. During this survey, beet leafhoppers were found and collected on overwintering weeds in several locations near potato fields throughout the Columbia Basin, from Hermiston/Umatilla area in Oregon to north of Moses Lake in Washington. Overwintering beet leafhopper adults collected were tested for BLTVA phytoplasma using PCR at the USDA-ARS in Prosser. The leafhopper testing revealed that about 15 percent of beet leafhoppers overwintering near potato fields in the Columbia Basin carry the phytoplasma.

Contrary to previous beliefs, this plant pathogen apparently does not necessary need overwintering weeds to survive the winter and leafhoppers are able to pass on this phytoplasma from one season to the next.

Development of Quick and Cost Effective Testing Protocols for Phytoplasma Detection in Plants and Insects

Testing individual leafhoppers and host plants has been shown useful in other phytoplasma disease monitoring programs. The team worked on developing reliable and quick methods for detecting phytoplasma in leafhoppers to aid in the determination of disease outbreak potential; most of the work was conducted at the USDA-ARS Prosser.

The nested, or two-step, PCR procedure remains the preferred method for identification of the phytoplasma in insects and plant tissue. Although the nested PCR procedure is rather lengthy and time consuming, we found that the single-step PCR procedures missed approximately 10% of the infections in potatoes and beet leafhoppers. However, for rapid screening of very large numbers of samples, the single-step PCR procedure may still be preferred.

We developed (and published) a method to use real-time PCR for detection of the phytoplasma in plants and insects. This real-time method has the advantages of a single step PCR but without the need for agarose gel electrophoresis of the reaction products. The elimination of one reaction step and electrophoresis offers the advantages of reduced materials and also reduces the chances of contamination since the PCR products do not have to be handled. The real-time PCR procedure, however, suffers from the need to quantify the DNA samples prior to analysis and also requires a real-time PCR instrument, which currently costs approximately $35,000.

Epidemiological Study on Fields Affected by the BLTVA Phytoplasma

The study was conducted by Agriculture Development Group personnel. Forty-nine commercial potato fields were monitored four times over a 4 month period during the 2005 growing season (Table 4). Fields were visited between May and August. The first visit was to locate the field and collect information regarding surrounding vegetation and crops. Subsequent visits collected information on crop health and degree of infection by BLTVA. Samples were collected from fields expressing BLTVA symptoms. Samples were provided to USDA-ARS laboratory in Prosser, WA, for BLTVA verification via PCR testing.

Results showed that fields across the Columbia Basin tested positive for BLTVA (Table 4). Although the number of fields sampled represented only 3% of the total number of fields in the Columbia Basin, there did not appear to be a difference between northern, central, or southern portions of the Columbia Basin. Another interesting finding is that the average percent of BLTVA infected plants in fields that were positive for BLTVA was relatively uniform when averaged for a county. The percentage of potato plants infected with BLTVA ranged from 3 to 25%.

A review of the vegetation associated with fields did not reveal a powerful relationship between vegetation and incidence of BLTVA. Eighty percent of fields testing positive for BLTVA and 43% of the fields testing negative for BLTVA were surrounded by kochia to some degree, indicating that kochia presence alone did not result in infection.

Table 4. Number of fields inspected for BLTVA in different counties and percent BLTVA in fields testing positive				
County	**Total number of fields inspected**	**BLTVA positive fields**	**BLTVA negative fields**	**Average % of BLTVA in fields testing positive**
Franklin	13	4	9	9.8
Walla Walla	3	2	1	15.5
Benton	4	3	1	10.0
Grant	18	5	13	10.6
Adams	11	5	6	13.8

Pesticide spray records were collected for 27 of the 49 fields. Correlating insecticide use patterns with incidence of BLTVA is difficult due the extreme variability in use patterns. One trend was extremely clear; no field with a beet leafhopper insecticide program applied to the foliage in both May and June had BLTVA. Virtually, all fields testing positive in the southern Basin (Walla Walla and Benton counties) had beet leafhopper control programs beginning in June, and a few fields had spray programs beginning the last week of May. For example, a field in Walla Walla County had 6% BLTVA and received an insecticide spray program that started on June 2. A similar nearby field that was managed virtually identically, but received an insecticide program starting in mid-May, had zero incidence of BLTVA. All fields testing positive in the southern Basin had insecticide spray programs initiated in June.

In addition, some fields with no BLTVA received no insecticide applications during May and June. Based on these results, it was concluded that BLTVA infected potatoes were somewhat uniformly distributed across a north-south gradient of the Columbia Basin, specifically in the counties of Walla Walla, Benton, Franklin, Grant and Adams. The range of infected plants in fields was relatively narrow. Fields with foliar insecticide programs starting in early May had no BLTVA. Fields with no foliar insecticide programs in May or June or with insecticide programs starting in June were at high risk of developing BLTVA. Fields in the more northern Basin that started programs after mid-May did not have BLTVA unlike the southern Basin fields which were vulnerable to BLTVA.

Insecticide Trial for the Potato Purple Top Disease Management

The beet leafhopper efficacy trial for 2005 was conducted by and located at the Agriculture Development Group Inc. research farm in Eltopia, WA. Russet Burbank seed was planted on April 6, 2005 on 34” row centers with 10” in-row spacing. Plot sizes were 4 rows by 20 ft with treatments replicated 4 times in a randomized complete block design. The soil type in the area is a sandy loam with 62% sand, 33% silt, and 5% clay with less than 1% organic matter. Rill irrigation was used at this site. Admire and Temik were applied at planting and foliar products were applied starting at emergence on May 19 and continued on a 14 day schedule until July 14 for a total of 5 applications. BLTVA samples were analyzed on July 27th.

Table 5. Beet leafhopper efficacy trial, Potatoes, 2005 (conducted in Eltopia, WA).

Treatment	Formulated Rate	BLTVA Rating
Untreated Check	---	0.25 a
Admire	16 oz/A	0.00 a
Temik	20 lb/A	0.25 a
Leverage	3.75 oz/A	0.00 a
Actara	1.5 oz/A	0.25 a
Provado	3.75 oz/A	0.50 a
Monitor	32 oz/A	0.25 a
Asana	3 oz/A	0.25 a
Messenger	2.5 oz/A	0.00 a
Guthion	1 lb/A	0.50 a

Radishes were planted around the periphery of each plot just after the potatoes were planted as a trap crop for beet leafhoppers. Insecticide control for the foliar products was considered effective through July.

At the end of July, when insecticide residues were thought to be no longer protecting the plants, 5 terminal leaves were collected within each plot and pooled together. A total of four aggregated samples for each treatment were sent to USDA-ARS in Prosser for BLTVA testing using PCR. The readings for each treatment were classified as either 0 (no plants infected), 0.25 (1 of four plots were infected), 0.50 (2 of four plots infected), 0.75 (3 of four plots infected) or 1.0 (all plots infected.)

Although beet leafhoppers were present and easily detected, rates of BLTVA infection in the trial was low. Results are summarized in Table 5. Treatments that had no BLTVA included Admire, Leverage and Messenger. Admire and Leverage both contain imidacloprid. This is the second trial in which Messenger-treated potatoes had no BLTVA.

A non WSPC funded trial was carried out looking at the ability of neonicotinoid seed and in furrow treatments to control BLTVA in potatoes. Disease pressure was even lower in this trial, making it difficult to make useful conclusions. The data suggest that thiamethoxam may have some activity against beet leafhoppers.

A pair of non WSPC funded trials was carried out on vegetable seed. The trial objective was to determine if seed treatments and foliar insecticide use could prevent transmission of BLTVA in radish and cilantro. Disease pressure in this trial was very high, with the disease infecting every single plant in areas of one trial. Two useful conclusions were drawn from this pair of trials. First, early planted (March 28) vegetable seed was more severely infected than later planted vegetable seed (April 28). It is hypothesized that at this research site in Eltopia, infected beet leafhoppers were in the plots in May and the difference in rates of infection was an interaction of growth stage of the plant and time of the year. Second, thiamethoxam was the most effective product at reducing incidence of BLTVA transmission.

Conclusions

The potato purple top disease in the Columbia Basin of WA and OR is caused by the beet leafhopper-transmitted virescence agent (BLTVA), also known as the Columbia Basin potato purple top phytoplasma. The beet leafhopper is the major vector of the purple top pathogen in this region. At least three generations of the beet leafhopper are produced in the Columbia Basin and leafhoppers overwinter locally on weeds in the vicinity of potato fields throughout this potato producing region. Leafhoppers seem to invade the Basin potato fields around mid-May to mid-June and are present in potatoes throughout the growing season. About 15% of beet leafhoppers overwintering near potato fields in the Basin carry the BLTVA phytoplasma. Observations indicate that potato infection seems to take place early in the season. Weeds immediately surrounding fields play an important role in the beet leafhopper dispersal, hosting BLTVA, and in the epidemiology of the disease. There are indications that some potato cultivars are more susceptible to BLTVA than others, at least under laboratory conditions. In addition, preliminary results indicate that the potato yield and tuber quality are affected by purple top disease. Some insecticides, when timely and appropriately applied, appear to effectively manage the beet leafhopper and the potato purple top disease.

Future Research

The following research objectives are planned for the 2006 growing season:

1. Assess the susceptibility of important potato cultivars grown in the Columbia Basin to the purple top phytoplasma under field conditions.
2. Assess the impact of the purple top phytoplasma on the potato yield and tuber quality in different potato cultivars grown under field conditions.
3. Determine the susceptibility of different potato plant growth stages to BLTVA phytoplasma under laboratory conditions.
4. Determine the relative time when BLTVA infection occurs in the field and how that may affect potato yield and tuber quality factors.
5. Determine the incidence of BLTVA phytoplasma in beet leafhoppers collected from within potatoes throughout the growing season.
6. Testing of selected weed species and crop plants for the BLTVA to determine the potential source of this potato purple top disease phytoplasma.
7. Determine if habitat manipulation around the exterior of commercial potato fields influences beet leafhopper population dynamics.
8. Continue beet leafhopper network trapping throughout the Columbia Basin.

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