

Beet Leafhopper and BCTV Strain Survey

✓ SRSRSA Funded Project

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Beet curly top virus (BCTV) almost eliminated sugarbeet production in the western United States in the 1920s and early 1930s until resistant cultivars were developed. However, commercial sugarbeet cultivars only contain low to intermediate resistance. In order to maintain proper resistance to BCTV in cultivars, the strains present in southern Idaho should be monitored on a regular basis both in plants and the beet leafhopper vector. Vector population numbers will also help determine where and when stronger resistance and control will be needed.

Research Objectives

- Characterize the leafhopper species and numbers present over the growing season in the desert and sugarbeet and dry bean fields.
- Determine if biotypes are present in the beet leafhopper population
- Identify the BCTV strains present in the various leafhoppers and fields
- If phytoplasmas prove relevant, they could also be tracked in samples
- Compare captures on cards oriented horizontally and vertically

Methodology

Beet leafhopper (BLH; *Circulifer tenellus*) populations in southern Idaho were tracked in desert areas and sugarbeet and dry bean fields in four and five southern Idaho counties during 2020 and 2021, respectively. Samples were collected on yellow sticky cards on a weekly basis from mid-April through mid-September (22 weeks; the first 3 weeks were missed in 2020 because of the COVID travel restrictions) to assess all leafhopper population levels and the presence of BCTV strains. Crop plants from monitored fields were also assessed for the presence of BCTV strains. This project will establish the BCTV strains for which host plant resistance is needed and the time frame when control of BLH is necessary. This report updates our progress on the project.

Results

Vector population dynamics. The Elmore County desert site had the highest populations both years early in the season, with an average of 401 BLH per card on week 5 (May 20, 2020), and 222 BLH per card on week 7 (Jun 2, 2021). However, the BLH coming into the Elmore County sugarbeet fields did not peak until week 16 (Aug 4-5) both years with an average of 55 to 69 BLH per card. The Elmore County bean fields peaked at an average of 24 BLH per card on weeks 10 (Jun 24) and 12 (Jul 8) in 2020, while in 2021 the peak occurred on week 13 (Jul 14) with 54 BLH. The peaks in the two field sites differed from that found in the desert both in timing and numbers indicating that perhaps local weed populations may be an important source of BLH. In Owyhee County, the dry bean



field peaked at 96 BLH per card on week 15 (Jul 28), while the desert site peaked at 23 BLH in week 17 (Aug 11) and the sugarbeet field peaked at 21-22 BLH on both week 11 (Jun 30) and week 20 (Sep 1). The primary source of BLH for the bean field seemed to be the large weed population near the Snake River and not the desert. The Twin Falls County desert and bean sites had low BLH numbers both years with only an average of 0 to 7 BLH per card on any given week. The Twin Falls County sugarbeet field had a peak of 27 BLH per card on week 14 (Jul 22, 2020) and a peak of 44 BLH on week 8 (June 9, 2021), which differs from both the desert and dry bean sites both years. Local weed populations and not the desert appeared to be the source of BLH for the Twin Falls County sugarbeet fields. At all the Minidoka sites, only zero to trace levels of BLH (avg 0 to 4 BLH per card) were captured both years. In Bingham County, only trace levels of BLH were captured at the desert site both years and the 2020 sugarbeet field, while in 2021 the sugarbeet field had a peak of 16 BLH per card on week 18 (Aug 18, 2021). The Bingham County bean fields had mostly trace levels of BLH except for a peak of 23 BLH per card on week 19 (Aug 26, 2020) and 6 BLH per card on week 10 (Jun 23, 2021.). Both bean field peaks and the 2021 sugarbeet field peak differ from what occurred in the desert. These differences may have been due to local weed populations being the source of BLH and not the desert.

Card placement comparison. In Elmore County the desert sites had a sizable BLH population both years, making a good comparison of card placement possible. In 2020, there was an average of 75% fewer BLH on the horizontal cards compared to the vertical cards when significant differences were evident. In 2021, early and late in the season significant differences were not always present. However, when significant differences were present, there was an average of 51% fewer BLH with horizontal cards versus vertical cards.

Pathogen and vector molecular identification. In 2020 based on both morphology and sequencing of a 585 bp region of the cytochrome oxidase (COI) gene, 492 samples were determined to be BLH with 42% of the samples (207 samples) positive for BCTV coat protein (CP) gene. However, the percentage of CP positive samples in Elmore County in 2020 was much higher (81 to 95% positive) in weeks 8 through 16. The CP positive samples were also positive for the following BCTV strains: 93% Worland, 21% Colorado, 2% Severe, 0% CA/Logan, and 21% mixed. When the Colorado strain was found in 2020, it was

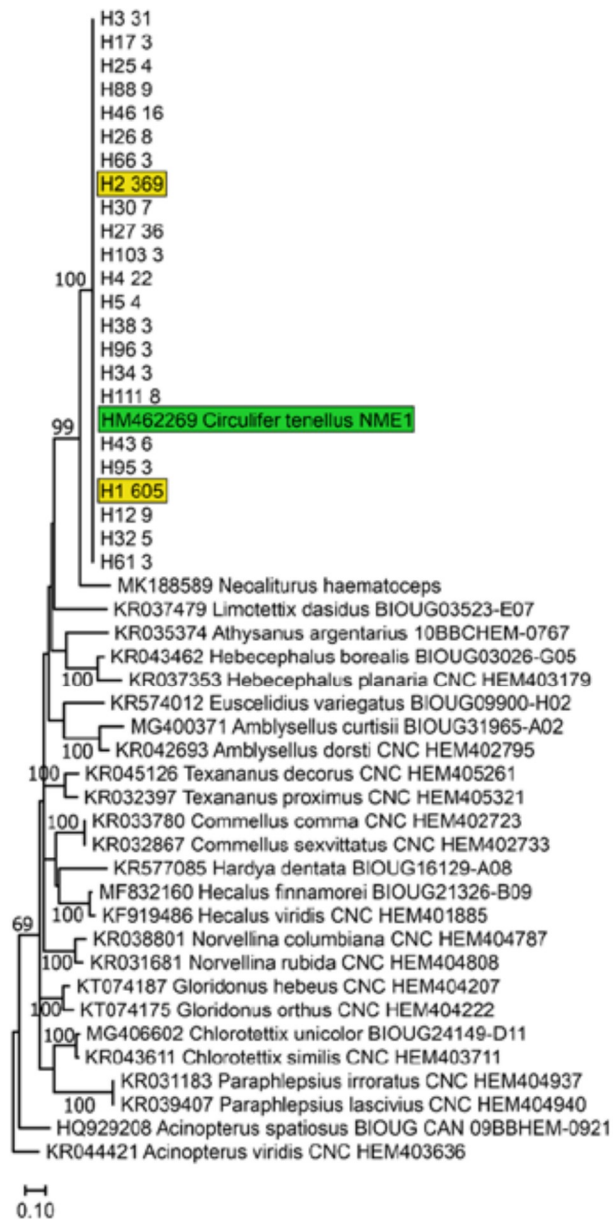


Figure 1. Phylogenetic relationships among the 23 most frequently identified *Circulifer tenellus* haplotypes from the yellow sticky card samples collected in southern Idaho were established and compared with sequences from GenBank for genera from the Deltocephalinae subfamily based on a 585 bp region of the cytochrome oxidase gene. In the *C. tenellus* samples, a total of 194 haplotypes were discovered, but the population was dominated by two haplotypes (yellow boxes) and none of the haplotypes had the exact same sequence as the *C. tenellus* GenBank accession (green box). After the haplotype number, the number of samples with that haplotype is shown.



coinfecting with Worland 86% of the time. The Severe strain was only found at Elmore and Twin Falls counties sites, while the CA/Logan strain was only found in Twin Falls County in 2020. In 2021, 854 samples were determined to be BLH with 67% of the samples (572 samples) positive for the CP gene. However, the percentage of CP positive samples in Elmore County in 2021 was much higher (82 to 100% positive) in weeks 7 through 21. The CP positive samples were also positive for the following BCTV strains: 87% Worland, 30% Colorado, 1% Severe, 5% CA/Logan, and 29% mixed. When the Colorado strain was found in 2021, it was coinfecting with Worland 85% of the time. The Severe strain was only found in Elmore and Owyhee counties sites, while the CA/Logan strain was found in all counties except Minidoka County in 2021. Sequencing



of the BLH COI region, identified 194 haplotypes over the two years. Both years the same two haplotypes were dominant with 45% of the BLH samples being haplotype 1 and 28% haplotype 2 and widely distributed throughout southern Idaho. BLASTn analysis found 96% of haplotypes (representing 99.4% of the samples) had 99.66 to 98.46% sequence identity with the *C. tenellus* NCBI accession HM462269 from New Mexico. Although closely related, the sequencing for HM462269 did not completely match any of the haplotypes found in southern Idaho. An evolutionary analysis of the haplotypes associated with more than two samples also supported that samples were *C. tenellus* and closely related to HM462269 (Fig. 1).

Curly top in plant samples. In 2020, 28% of the 40 bean samples were positive for the CP gene and the BCTV Worland strain. Only 10% of the 40 sugarbeet samples in 2020 were positive for the CP gene and currently the strain present is unknown and under investigation. In 2021, 38% of the 50 bean samples were positive for the CP gene and were found to contain the following strains: 42% CA/Logan, 37% Worland, 21% Severe, 5% Colorado, and 37% unknown. In 2021, 44% of the 50 sugarbeet samples were positive for the CP gene and were found to contain the following strains: 55% Worland, 23% CA/Logan, 9% Severe, and 27% unknown.

Parasites in samples. When amplifying a region of the COI gene in 2020 BLH samples, ten samples were found to contain parasitic fly species: *Chalarus sp.* (1 sample), *Eudorylas sp.* (4 samples), *Tomosvaryella sp.* (4 samples), and *Voria sp.* (1 sample). In 2021, five BLH samples were found to contain parasitic fly species: *Eudorylas sp.* (2 samples), *Gonatocerus sp.* (1 sample), and *Tomosvaryella sp.* (2 samples). In 2021, four BLH samples were found to contain parasitic wasp species: *Aphelopus sp.* (3 samples) and *Gonatopus sp.* (1 sample).

Conclusions

Historically, there has been a gradient in the beet curly top pressure from Treasure Valley (highest BCTV pressure) to eastern Idaho (least BCTV pressure). The BLH populations found in 2020 and 2021 matched this historical trend, with the highest BLH numbers being found in the Mountain Home area, fewer in Twin Falls, and even fewer east of Twin Falls. While a large peak occurred each year in Mountain Home around late May to early June, most of the BLH samples were not positive for BCTV until late June through July. The BLH population peaks found in desert areas did not occur at the same time as the peaks in crop fields, indicating that local weed populations, and not the desert, were the primary source of BLH migrating into fields during 2020 and 2021. The Worland strain of BCTV was dominant both years in the BLH population, but the BCTV strains found in the plant samples were more of a mix. Several BLH samples and plant samples were positive for the CP gene, but negative for strain specific primers evaluated to date. We will continue our investigation by evaluating samples for additional BCTV strains such as Pepper curly top virus, Spinach curly top virus, and Kimberly1. Preliminary testing for phytoplasma indicates that some BLH samples are positive, but so far, the plant samples evaluated have tested negative. We will wrap up the BLH portion of the study soon and begin to turn our focus to the other morphotypes found on the yellow sticky cards.

Report photos by Carl Strausbaugh

