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Research

Curly Top Viruses and Phytoplasmas in Sugar Beets, Common Beans, and Beet Leafhoppers Along with Vector Population Dynamics in Southern Idaho

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Abstract

Beet curly top in sugar beet and common bean is a major yield-limiting factor that is caused by beet curly top virus (BCTV) and is vectored by the beet leafhopper (BLH; *Circulifer tenellus*). BLH populations in southern Idaho were tracked during the 2020 and 2021 growing seasons in desert areas and sugar beet and common bean fields with yellow sticky cards to assess BLH population levels and identify the curly top virus species/strains and phytoplasmas present. Plants from monitored crop fields were also assessed for the same pathogens. In one desert area (Elmore Co.), BLH populations began increasing in May and were present in double-digit numbers per card through the summer at all sites in both years. However, the BLH numbers at other desert sites were at or near zero. Local weed populations and not desert areas appeared to be the primary source of BLH in crop fields. Based on cytochrome oxidase gene sequence, two haplotypes dominated the BLH population. In both years, BCTV strains Worland and Colorado were the primary strains in BLH and plant samples. The California/Logan, pepper curly top (PeCT), and severe strains of BCTV were also detected in BLH, along with spinach curly top Arizona virus (SpCTAV). Phytoplasmas were detected in 1% of BLH samples in both years. Phytoplasmas, SpCTAV, and PeCT were not detected in plant samples. This project established the curly top species/strains for which host plant resistance is needed, as well as the time and areas when crops are at the highest risk for infection.

Keywords: beet curly top, beet curly top virus, beet leafhopper, *Beta vulgaris*, *Circulifer tenellus*, common bean, *Phaseolus vulgaris*, phytoplasmas, spinach curly top Arizona virus, sugar beet

Beet curly top in sugar beet (*Beta vulgaris* L.) and common bean (snap and dry beans; *Phaseolus vulgaris* L.) caused by beet curly top virus (BCTV) is a serious yield-limiting disease in semiarid production areas worldwide (Bennett 1971; Gharouni Kardani et al. 2013; Strausbaugh et al. 2017; Yazdi et al. 2008). Beet curly top infestations start in the

spring when weeds in desert areas begin to dry out and viruliferous beet leafhoppers (BLHs; *Circulifer tenellus* Baker; syn. *Neoaliturus tenellus*; Hemiptera:Cicadellidae) disperse into local commercial crop fields (Bennett 1971). Besides sugar beet, numerous dicotyledonous plants are impacted by beet curly top, including basil, cantaloupe, common bean, coriander, cucumber, hemp, muskmelon, pepper, pumpkin, tomato, spinach, squash, watermelon, and numerous weed species (Bennett 1971; Blickenstaff and Traveller 1979; Chen and Gilbertson 2009; Chen et al. 2014; Creamer 2020; Creamer et al. 1996; Melgarejo et al. 2022; Swisher Grimm et al. 2021). Early surveys in southern Idaho recorded 40 host species (Haegele 1927); later studies documented hosts from 13 families, including 17 introduced species (Douglass and Hallock 1957). After the initial dispersal from the desert, additional generations can occur in production fields. In southern Idaho, northern Utah, and the Columbia River basin in Washington and Oregon, there are generally three broods of BLH, whereas in Arizona, California, and New Mexico, five broods can occur (Cook 1967; Harries and Douglass 1948). The western United States is considered to have six major BLH breeding areas: the San Joaquin Valley, CA; the lower Colorado River Valley; the Rio Grande area of New Mexico and Texas; scattered breeding grounds in Colorado, Utah, and Nevada; the lower Snake River plains of Idaho and Oregon; and the Columbia River area of Washington and Oregon (Douglass and Cook 1954). However, genetic testing of these breeding populations should be conducted to determine if they intermix.

Beet curly top can be caused by three *Curtovirus* spp. (BCTV, *Spinach severe curly top virus* [SpSCTV], and *Horseradish curly top virus* [HCTV]) and two *Becurtovirus* spp. (*Beet curly top Iran virus* [BCTIV] and *Spinach curly top Arizona virus* [SpCTAV]) (Strausbaugh et al. 2017; Varsani et al. 2014a, b). Within BCTV, 11 strains have been described in the literature, but in recent surveys of symptomatic plants in Idaho sugar beet fields, only five of the strains have been detected: California/Logan (CA/Logan), Colorado (CO), Kimberly 1 (Kim1), Severe (Svr), and Worland (Wor) (Strausbaugh et al. 2017). The BCTV strains affecting common bean have not been thoroughly investigated, but a recent publication evaluated common bean cultivars versus CA/Logan and Wor-like strains (Soler-Garzón et al. 2023). Also, the incidence of BCTV strains in sugar beet was shown to vary between the 2006 to 2007 and 2012 to 2015 samples in southern Idaho (Strausbaugh et al. 2008, 2017). In 2006 to 2007 samples, the BCTV strain incidence was 87% Svr, 60% Wor-like, 7% CA/Logan, and 59% mixed infections, whereas in 2012 to 2015 samples, the incidence was 2% Svr, 87% Wor-like, 30% CA/Logan, and 16% mixed (Strausbaugh et al. 2008, 2017). Sequencing the genomes of 39 Wor-like primer-positive samples established that 56% of these samples were CO, whereas 39 and 5% were Wor and Kim1, respectively (Strausbaugh et al. 2017). Thus, there was a transition from Svr and Wor-like strains and mixed infections in sugar beet plants in 2006 to 2007 to primarily Wor-like strains (mostly CO and Wor) and single infections in the 2012 to 2015 samples (Strausbaugh et al. 2008, 2017). Most of the 2012 to 2015 sugar beet leaf samples had only relatively mild symptoms (more than 50% of the upper surface of the leaf visible) despite commercial cultivars containing only low to moderate resistance to BCTV.

However, a major change in management occurred in 2008 when neonicotinoid seed treatments became fully labeled for use on sugar beet (Strausbaugh et al. 2006, 2010, 2012, 2014, 2016). The seed treatments proved to be a highly effective supplement to host resistance because yields could be increased by 15% or more in commercial sugar beet fields with high curly top pressure (Panella et al. 2014; Rojas et al. 2018; Strausbaugh et al. 2006, 2012, 2014). Although the neonicotinoid seed treatments have a small environmental footprint compared with using numerous less effective foliar sprays, they are not without concern, especially with regard to nontarget effects and insecticide resistance (Bonmatin et al. 2015; Dewar 2022; Thompson et al. 2022; Warren 2021; Woodcock et al. 2021). Systemic insecticide seed treatments and host resistance are the most effective controls in common bean as well (Singh and Schwartz 2010; Soler-Garzón et al. 2023).

With shifting weather patterns and drought conditions occurring during 2019 to 2021 in southern Idaho, some desert areas had few annual weeds during the winter months in recent years (C. A. Strausbaugh, *personal observation*). Because winter annuals in desert areas were historically thought to be the primary source of BLH, industry stakeholders requested that BLH numbers be tracked in both desert and crop lands. The strains of BCTV present in the BLH is also of concern because some sources of host resistance to BCTV are strain specific (Montazeri et al. 2016). Knowledge of which strains are most prevalent can help guide resistance-breeding programs. Thus, in five southern Idaho counties, the BLH populations were monitored in desert areas and sugar beet and common bean fields for both vector population numbers and the curly top virus species/strains they carried. Because BLH can also carry plant pathogens important to other crops, such as phytoplasmas and spiroplasmas (Crosslin et al. 2006; Golino et al. 1987; Greenway 2022; Rivedal et al. 2022; Swisher et al. 2016, 2018), assays for these pathogens were also conducted.

Materials and Methods

Leafhopper collections

To monitor BLH populations during the 2020 growing season, 10 × 15 cm yellow sticky cards (Alpha Scents, Canby, OR) were placed in the Bingham, Elmore, Minidoka, and Twin Falls southern Idaho counties where desert areas and sugar beet and common bean fields were in close proximity (Fig. 1). The yellow sticky cards were collected weekly for 22 weeks from mid-April through mid-September (Supplementary Table S1). With four counties, three sample sites (desert, sugar beet, and common bean) within a county, three cards per site, and 22 weeks, there were a total of 792 cards collected throughout the growing season. Cards for sugar beet and common bean fields were placed near the edge of the fields but far enough away that they were not impacted by sprinkler irrigation. The cards were placed on three different sides of each field, if practicable, or at least 30 m apart. In the desert locations, the cards were placed at least 30 m in from the access road (unless the access road was surrounded by desert on both sides) and at least 30 m apart. The global positioning system (GPS) coordinates for card locations can be found in Supplementary Table S2. The elevation range of the card locations in each county over the two years was 928 to 944 m in Elmore, 1,049 to 1,177 m in Twin Falls, 1,294 to 1,308 m Minidoka, and 1,359 to 1,412 m in Bingham. Each card was affixed with a metal clip on a wooden lath stake with the bottom edge approximately 8 cm off the ground based on research by Meyerdirk and Oldfield (1985). The vegetation in a 1-m-diameter area around the card was maintained at less than 5 cm in height. When retrieved, the cards were placed in a cooler for transportation back to the laboratory. Once back at the laboratory, the cards were placed in a −20°C freezer. From each card, BLHs were counted and identified based on macro- and micromorphology (Jensen 2008). The BLHs were about 3 to 4 mm in length, relatively pale, and their heads were rounded in front with no dark spots. To supplement visual identification, from each card, up to a maximum of 10

BLHs were sampled and pooled into groups of up to five BLHs in a 2-ml microcentrifuge tube and stored at −80°C for molecular analysis. The study was repeated during the 2021 growing season using sites in the same general areas assessed during the previous season and the same methods. In 2021, one additional county, Owyhee Co., was added at the request of the funding organization. The elevation of the Owyhee Co. collection sites ranged from 746 to 758 m.

Plant samples

To compare the BCTV strains present in BLHs collected on the yellow cards with the strains present in crop plants, three-hole punches (9 mm in diameter; each from a different leaf) were collected using the lid of a 2-ml microcentrifuge tube from plants in commercial fields with curly top symptoms (stunted plants with yellow curling thickened leaves). Ten plants were collected from each of the monitored sugar beet and common bean fields ($n = 10$ per field). With plants in four counties being tested, there was a total of 40 samples per crop in 2020. With plants in five counties being evaluated in 2021, 50 samples per crop were collected. In 2020, the plant samples were collected from the common bean fields from August 19 to 24 and from the sugar beet fields from September 2 to 8. In 2021, all the plant samples were collected from July 23 to 24. The plant samples were placed in a cooler for transportation back to the laboratory and then stored at −80°C.

Pathogen and vector identification

Molecular methods were used to confirm morphological identifications of BLH and to evaluate for the presence of curly top viruses, phytoplasmas, and spiroplasmas in both BLH samples and plant material. If BLH parasitoids are found in the BLH samples, they will also be documented. The samples were centrifuged at 19,000 × *g* for 30 s to be sure the samples were at the bottom of the tube and then they were lyophilized. The samples were then pulverized using a Retch MM301 mixer mill (Retch, Newton, PA) with 5-mm-diameter stainless steel balls. Plant DNeasy kits (Qiagen., Valencia, CA) were then utilized to extract DNA via spin columns. The DNA was assessed via gel electrophoresis and quantified using a NanoDrop 2000 spectrophotometer (Thermo Fisher Scientific, Waltham, MA).

Initially, the BLH samples were analyzed using PCR assays that amplified a region of the cytochrome oxidase gene using the LCO1490/HCO2198 primers (Folmer et al. 1994). BLH and plant samples were then tested individually with BCTV coat protein (CP) primers and species and/or strain-specific primers (Supplementary Table S3). Polymerase chain reaction (PCR) assays were performed using 20-µl volumes using 2 µl (approximately 20 ng of DNA) of nucleic acid extract, 0.2 µl of GoTaq DNA polymerase (Promega, Madison, WI), 2 µl of 3 µM each primer (Integrated DNA Technologies, Coralville, IA), 0.4 µl of 10 mM dNTP (Promega), 0.6 µl of 25 mM $MgCl_2$ (Promega), 4 µl of 5× Green GoTaq buffer (pH 8.5 with 7.5 mM $MgCl_2$; Promega), and 8.8 µl of UltraPure DNase/RNase-free distilled water (Invitrogen, Waltham, MA). The amplification cycle consisted of 3 min at 95°C, followed by 35 cycles of 95°C for 30 s, 50 to 58°C (depending on primers as indicated in Supplementary Table S3) for 30 s, and 72°C for 1 min. Following the final cycle, the reaction was held at 72°C for 5 min and then 12°C. Reactions without template DNA served as negative controls, whereas template DNA from samples previously identified as positive for a specific pathogen served as positive controls. Amplification products were visualized on a 1.6% agarose gel after GelRed (Biotium, Freemont, CA) staining.

For universal phytoplasma detection, a nested PCR assay was conducted using outer primers P1/P7 (Deng and Hiruki 1991; Schneider et al. 1995; Swisher et al. 2018) and FU5/RU3 (Lorenz et al. 1995; Swisher et al. 2018) as inner primers. The P1/P7 PCR product was diluted 1:20, and then 2 µl served as template for the second round FU5/RU3 reactions. Additional Mg was not added, as was just mentioned for the strain-specific PCR reactions, and the extension time was increased to 2 min. To confirm the identification of the phytoplasmas found via sequencing, the RFLP pattern was also investigated via iPhyClassifier (Zhao et al. 2009). For spiroplasma detection, PCR was conducted with primer pair P89-F/P89-R (Swisher et al. 2016; Yokomi et al. 2008). To confirm if the amplification product for the BCTV strains and other pathogens were working properly, sequencing was conducted for eight or more samples containing each strain or species found, if possible. Amplicons generated for sequencing were sent to TACGen (Richmond, CA) for bidirectional sequencing. The Sanger sequencing was repeated to achieve 4× coverage. The sequences were evaluated using BioEdit version 7.2.6.1 (Hall 1999), and consensus sequences were generated and deposited in GenBank (COI: OR433969-OR434003; Virus: OR451585-OR451659; 16S: OR428568-OR428572). Consensus sequences were initially compared with those on GenBank via BLASTn. For the COI sequences, the haplotype sequences that occurred more than twice were compared with the only GenBank accession for *C. tenellus* (HM462269) and sequences for closely related species from the Deltocephalinae subfamily to infer their evolutionary history and support their identification. The sequences were aligned using ClustralX Ver. 2.1 (Larkin et al. 2007). The Bayesian information criterion was used via MEGA 11.0.10 (Kumar et al. 2016) to determine that the best substi-

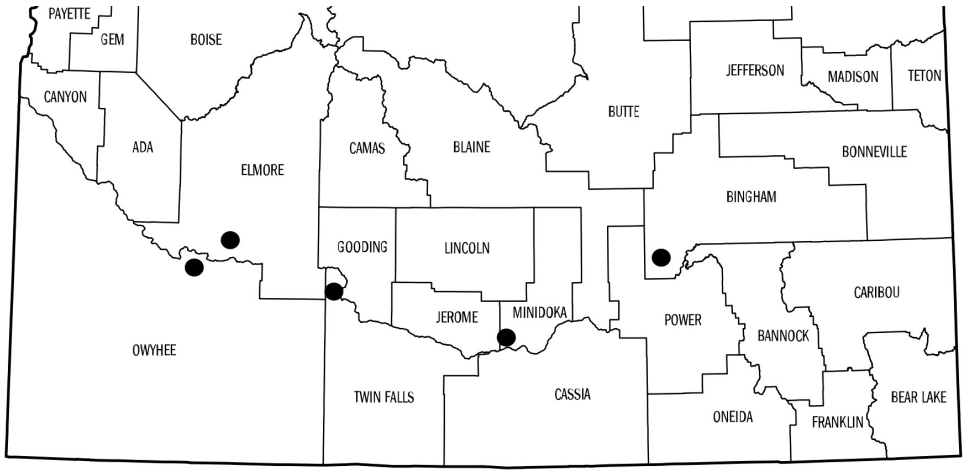

FIGURE 1
Beet leafhoppers, *Circulifer tenellus*, were collected using yellow sticky cards from five counties (Bingham, Elmore, Minidoka, Owyhee, and Twin Falls) in southern Idaho. The sample locations are designated with black dots.

tution model was GTR+G+I. Using this substitution model, an evolutionary analysis was conducted by the maximum likelihood method in MEGA 11.0.10. An initial search (two replicates) was conducted to estimate the model parameters, which were then fixed for a bootstrap analysis of 1,000 replicates.

Data analysis

When mean values are followed by ± x, x refers to the standard error. Comparison of strain frequencies between studies was conducted using a contingency test in SAS (version 9.4; SAS Institute, Cary, NC) via the Proc Freq procedure with the χ^2 statistic.

Results

Vector population dynamics

In Bingham Co., only trace levels of BLH (average of 0 to 2 per card) were captured at the desert site in both years and in the 2020 sugar beet field, whereas in the 2021 sugar beet field, there was a peak of 16 BLHs per card on week 18 (18 August) (Fig. 2; Supplementary Table S1). The Bingham Co. bean fields had mostly trace levels of BLH, except for a peak of 22 to 23 BLHs per card on weeks 18 and 19 (Aug 18 to 25) in 2020 and six BLHs per card on week 10 (23 June) in 2021. These peaks in Bingham Co. fields differed from what occurred in the desert. In both years, the highest populations of BLH in the study were at the Elmore Co. desert site with an average peak of 401 BLHs per card on week 5 (19 May) in 2020 and a peak of 222 BLHs per card on week 7 (2 June) in 2021 (Fig. 3). Two smaller Elmore Co. desert peaks occurred in 2020 later in the summer at 69 and 63 BLHs per card on weeks 16 (4 August) and 19 (25 August), respectively. In 2021, several small Elmore Co. desert peaks later in the summer were found at 58, 60, and 58 BLHs per card on weeks 16 (4 August), 18 (18 August), and 20 (1 September), respectively. On week 16 (4 August), the BLH coming into the sugar beet fields in Elmore Co. peaked at an average of 69 and 55 BLHs per card in 2020 and 2021, respectively. In Elmore Co. bean fields, the BLH population peaked at 24 BLHs per card on both week 10 (23 June) and week 12 (7 July) in 2020, whereas in 2021, a peak at 54 BLHs occurred on week 13 (14 July), which was offset from the BLH peaks in the surrounding desert sites and sugar beet fields. At the Minidoka desert and crop sites, zero to trace levels of BLH (average 0 to 4 BLHs per card) were captured in both years (Fig. 4). The Owyhee desert site peaked on week 17 (11 August) with an average of 23 BLHs per card, whereas the sugar beet field peaked at 21, 18, and 22 BLHs on weeks 11, 18, and 20, respectively (Fig. 5). The Owyhee bean field peaked at 59 and 96 BLHs on weeks 9 and 15, respectively. The Owyhee bean field peaks differ from the other sites in size and timing. The Twin Falls desert sites only averaged five and seven BLHs per card at their peaks in 2020 and 2021, respectively (Fig. 6). The Twin Falls 2020 sugar beet field had a peak of 27 BLHs per card found on week 14 (21 July) and a peak of 44 BLHs on week 8 (9 June) in 2021, which differs from what was seen in the desert and bean sites in both years. In Twin Falls Co., the bean sites had only trace levels of BLH (average 0 to 4 BLHs per card) during both years.

Molecular evaluation of BLH samples

In 2020 based on both morphology and sequencing of a 585-bp region of the COI gene, 492 samples were determined to be BLH, with 51% of the samples (251 samples) positive based on results for the PCR tests with the BCTV CP primers (Tables 1 and 2). However, the percentage of BCTV CP-positive BLH samples in Elmore Co. in 2020 was much higher (81 to 100% positive) in weeks 8 through 16. The BCTV CP-positive samples were also positive for the following BCTV strains: 82% Wor-like, 18% CO, 5% pepper curly top (PeCT), 2% Svr, and 1% CA/Logan (Table 2). In addition, 16% of the samples were positive for SpCTAV. The 2020 BLH samples contained virus mixtures of BCTV

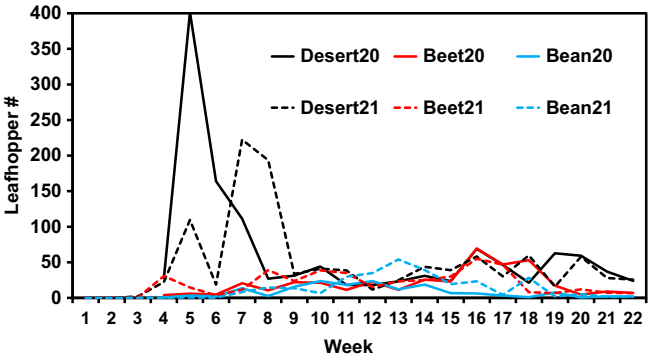

FIGURE 3
The mean number of beet leafhoppers, *Circulifer tenellus*, in Elmore County on 258 cm^2 of yellow sticky cards after 7 days in desert areas or next to sugar beet or common bean fields collected for 22 weeks in 2020 and 2021 beginning April 21 in southern Idaho.

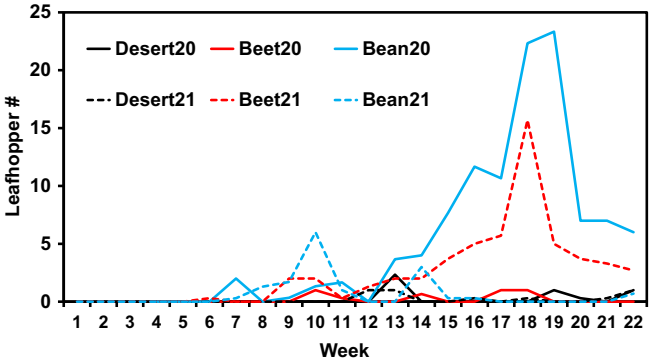

FIGURE 2
The mean number of beet leafhoppers, *Circulifer tenellus*, in Bingham County on 258 cm^2 of yellow sticky cards after 7 days in desert areas or next to sugar beet or common bean fields collected for 22 weeks in 2020 and 2021 beginning April 21 in southern Idaho.

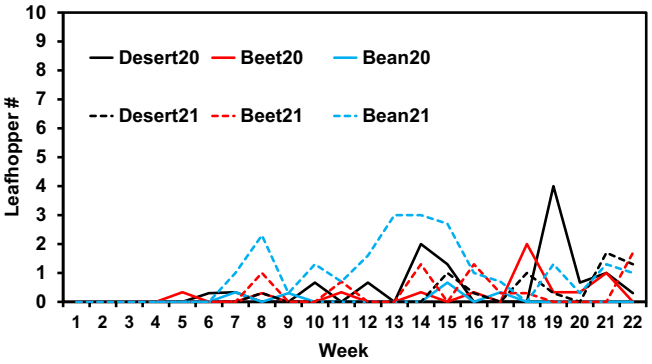

FIGURE 4
The mean number of beet leafhoppers, *Circulifer tenellus*, in Minidoka County on 258 cm^2 of yellow sticky cards after 7 days in desert areas or next to sugar beet or common bean fields collected for 22 weeks in 2020 and 2021 beginning April 21 in southern Idaho.

strains and/or SpCTAV 19% of the time. The PeCT and Svr strains were only found in Elmore and Twin Falls Co. sites, whereas the CA/Logan strain was only found in Twin Falls Co. in 2020.

In 2021, 854 samples were determined to be BLH, with 69% of the samples (589 samples) positive for BCTV CP (Tables 1 and 2). However, the percentage of BCTV CP-positive samples in Elmore Co. in 2021 was much higher (83 to 100% positive) in weeks 7 through 20 (Table 1). The BCTV CP-positive samples contained the following BCTV strains: 83% Wor-like, 29% CO, 6% CA/Logan, 2% PeCT, and 1% Svr (Table 2), which did not differ ($\chi^2 = 7.1$; $P = 0.1330$) from that found the previous year. In addition, 43% of the samples were positive for SpCTAV. The 2021 BLH samples contained virus mixtures of BCTV strains and/or SpCTAV 43% of the time. The Svr strain was only found in Elmore and Owyhee Co. sites, whereas the CA/Logan strain was found in all counties.

The BCTV presence and strain identification was confirmed by sequencing. When amplicons generated for 25 samples (selected randomly from across years, locations, and crops) that were positive for the BCTV CP primers were sequenced (GenBank accessions OR451635-OR451659), they had 98.07 to 100% sequence identity with known BCTV accessions. The sequencing from five amplicons generated with CA/Logan primers were identical (OR451587) and had 99.66% sequence identity with BCTV-CA/Logan genomes KX867029, KX867040, and KX867041. The sequences from eight amplicons generated with the Colorado

TABLE 1

The number of beet leafhopper, *Circulifer tenellus*, samples collected for 22 weeks and found to be positive for the beet curly top virus coat protein primers in a polymerase chain reaction assay in 2020 and 2021 in five southern Idaho counties

	Bingham Co.		Elmore Co.		Minidoka Co.		Owyhee Co.		Twin Falls Co.	
Week-date[a]	Sample number	Positive (%)[b]	Sample number	Positive (%)[b]	Sample number	Positive (%)[b]	Sample number	Positive (%)[b]	Sample number	Positive (%)[b]
2020										
1-Apr 21	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
2-Apr 28	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
3-May 5	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
4-May 12	0	NS	3	0	0	NS	ND	ND	1	0
5-May 19	0	NS	6	50	0	NS	ND	ND	0	NS
6-May 26	0	NS	8	12	1	0	ND	ND	0	NS
7-Jun 2	1	0	17	24	1	0	ND	ND	7	0
8-Jun 9	0	NS	16	88	1	0	ND	ND	6	0
9-Jun 16	0	NS	20	85	0	NS	ND	ND	4	0
10-Jun 23	3	0	20	90	0	NS	ND	ND	8	50
11-Jun 30	2	0	17	100	1	0	ND	ND	8	25
12-Jul 7	0	NS	17	82	1	0	ND	ND	11	55
13-Jul 14	3	0	19	95	0	NS	ND	ND	11	73
14-Jul 21	4	0	19	95	1	100	ND	ND	11	64
15-Jul 28	4	25	15	93	2	0	ND	ND	9	22
16-Aug 4	3	0	21	81	1	0	ND	ND	9	33
17-Aug 11	5	0	17	53	0	NS	ND	ND	8	12
18-Aug 18	5	40	15	53	4	0	ND	ND	9	44
19-Aug 25	2	0	16	12	2	0	ND	ND	9	0
20-Sep 1	4	50	13	38	1	0	ND	ND	8	12
21-Sep 8	5	0	15	47	4	75	ND	ND	8	38
22-Sep 15	5	20	14	57	2	0	ND	ND	9	44
Overall	46	13	288	67	22	18	ND	ND	136	33
2021										
1-Apr 21	0	NS	0	NS	0	NS	0	NS	0	NS
2-Apr 28	0	NS	0	NS	0	NS	0	NS	1	0
3-May 5	0	NS	6	33	0	NS	3	33	2	0
4-May 12	0	NS	2	100	0	NS	2	50	2	50
5-May 19	0	NS	15	67	0	NS	4	75	4	50
6-May 26	0	0	14	64	0	NS	3	33	2	0
7-Jun 2	2	50	22	91	2	0	10	50	6	17
8-Jun 9	2	0	22	100	4	75	10	70	7	57
9-Jun 16	3	33	22	100	1	0	13	85	10	60
10-Jun 23	5	40	23	100	1	100	13	92	7	86
11-Jun 30	3	0	24	100	2	50	15	100	6	83
12-Jul 7	3	0	20	90	2	100	13	77	4	75
13-Jul 14	2	0	43	86	3	0	25	48	3	33
14-Jul 21	4	0	24	83	3	0	15	60	4	0
15-Jul 28	3	33	24	100	6	0	18	67	5	40
16-Aug 4	5	40	24	92	2	0	16	31	6	33
17-Aug 11	3	100	21	95	3	0	13	38	5	40
18-Aug 18	3	0	24	96	4	25	18	72	9	33
19-Aug 25	4	25	20	95	2	0	13	38	7	14
20-Sep 1	3	0	21	86	1	0	14	71	8	12
21-Sep 8	4	25	19	79	3	67	13	85	5	0
22-Sep 15	4	25	19	74	5	40	9	67	5	20
Overall	53	23	409	89	44	27	240	65	108	39

[a] NS = no samples. ND = no data. Sample number = the total number of beet leafhopper (BLH) samples. Each BLH sample could contain from 1 to 5 BLHs. No data were collected the first 3 weeks in 2020 because of both state and federal COVID travel restrictions.

[b] The percentage of samples positive for the beet curly top virus coat protein primers.

primers were evaluated in a BLASTn search; they all had 99.08 to 99.31% sequence identity with BCTV-CO genome KX867015. Seven of the CO sequences had 100% sequence identity (OR451585), whereas one sequence differed (OR451586) by a single nucleotide change. The sequencing of five PeCT strain amplicons (OR451591, OR451592, OR451615, OR451616, and OR451617) had 98.74 to 99.39% sequence identity with one of the PeCT genomes (EF501977, JX487184) in GenBank. The Svr strain amplicons were from faint bands and thus could not be confirmed via sequencing. The Wor-like primers detected both Wor

TABLE 2

Pathogens associated with beet leafhopper (BLH), *Circulifer tenellus*, samples from yellow sticky cards collected for 22 weeks beginning April 21 in five counties in southern Idaho during 2020 and 2021

			BCTV strain (% of CP positive)[b]						
Site[a]	Sample #	CP (%)	CA/Logan	CO	PeCT	Svr	Wor+	SpCTAV	Phyto
2020									
Bingham									
Desert	2	0	0	0	0	0	0	50	0
Sugar beet	6	0	0	0	0	0	0	0	0
Common bean	38	18	0	57	14	0	43	8	0
Across county	46	15	0	57	14	0	43	9	0
Elmore									
Desert	173	62	0	16	6	2	93	25	1
Sugar beet	55	78	0	12	7	2	86	5	0
Common bean	60	77	0	28	0	0	93	23	3
Across county	288	68	0	17	5	2	90	21	1
Minidoka									
Desert	12	25	0	0	25	0	33	0	0
Sugar beet	7	14	0	0	100	0	0	0	0
Common bean	3	0	0	0	0	0	0	0	0
Across county	22	18	0	0	50	0	25	0	0
Owyhee									
Desert	ND	ND	ND	ND	ND	ND	ND	ND	ND
Sugar beet	ND	ND	ND	ND	ND	ND	ND	ND	ND
Common bean	ND	ND	ND	ND	ND	ND	ND	ND	ND
Across county	ND	ND	ND	ND	ND	ND	ND	ND	ND
Twin Falls									
Desert	31	42	8	31	0	0	62	10	0
Sugar beet	77	31	8	4	4	4	79	10	0
Common bean	28	36	0	40	0	0	50	11	0
Across county	136	35	6	19	2	2	68	10	0
Overall	492	51	1	18	5	2	82	16	1
2021									
Bingham									
Desert	5	20	0	0	0	0	100	20	0
Sugar beet	35	29	10	0	10	0	80	54	0
Common bean	13	15	0	0	0	0	50	15	0
Across county	53	25	8	0	8	0	77	42	0
Elmore									
Desert	225	92	2	35	<1	<1	91	87	1
Sugar beet	98	88	6	9	2	2	88	28	0
Common bean	86	85	8	41	0	0	84	31	1
Across county	409	89	5	30	1	1	89	61	1
Minidoka									
Desert	12	17	50	0	0	0	0	17	0
Sugar beet	7	29	0	50	0	0	100	43	0
Common bean	25	32	0	0	0	0	62	12	0
Across county	44	27	8	8	0	0	58	18	0
Owyhee									
Desert	69	57	3	23	5	3	87	33	3
Sugar beet	78	72	7	46	4	2	82	19	5
Common bean	93	62	2	29	3	0	83	27	0
Across county	240	63	4	34	4	1	84	26	2
Twin Falls									
Desert	32	38	0	0	8	0	42	12	0
Sugar beet	65	34	27	18	5	0	45	29	0
Common bean	11	64	14	29	0	0	100	27	0
Across county	108	38	17	15	5	0	54	24	0
Overall	854	69	6	29	2	1	83	43	1

[a] Beet leafhopper (BLH) samples were collected from yellow sticky cards on a weekly basis from three sites (desert, sugar beet, and common bean) in five southern Idaho counties. Sample # = the total number of BLH samples. Each BLH sample could contain from 1 to 5 BLHs. CP = percentage of the BLH samples that were positive for the beet curly top virus (BCTV) coat protein (CP) primers. BCTV strains = percentage of samples amplified with a strain-specific primer pair. ND = no data.

[b] The percentage of CP-positive samples that had this specific BCTV strain based on strain specific primers: California/Logan (CA/Logan), Colorado (CO), pepper curly top (PeCT), Severe (Svr), and Worland (Wor). The Wor+ primers detected both Wor and CO strains based on sequencing. SpCTAV = the percentage of beet leafhopper samples positive for spinach curly top Arizona virus based on species-specific primers. Phyto = the percentage of beet leafhopper samples positive for the universal phytoplasma primers.

and CO strains across all locations. The sequencing from 13 Wor-like amplicons from BLH samples collected in June or July were 85% Wor and 15% CO, which differed ($\chi^2 = 64$; $P < 0.0001$) from 17 Wor-like amplicons from BLH samples collected in August or September (71% CO and 29% Wor). The 16 BLH Wor sequences (OR451618-OR451632) had 98.31 to 100% identity with Wor genomes MW182244 and KX867053. The 14 BLH CO sequences (OR451601-OR451613) had 96.91 to 99.72% identity with one of the following CO genomes: KT276901, KX867018, KX867047, KX867057, MK803280, and MW182244. The sequencing of SpCTAV amplicons from 14 BLH samples had 99.00 to 99.52% sequence identity (OR451593-OR451599) with the SpCTAV genome sequence (HQ443515).

Phytoplasmas were detected in 1% of the BLH samples in both years (Table 2). Sequencing of the amplicon generated with the universal phytoplasma primers revealed that '*Candidatus* Phytoplasma trifolii' and '*Candidatus* Phytoplasma pruni' were present. Based on results from BLAST searches, the six '*Ca.* P. trifolii' samples (OR428569) had 100% sequence identity with the '*Ca.* P. trifolii' sequences OQ597724, MF092789, and KY047615. Four other '*Ca.* P. trifolii' samples (OR428568) had 99.92% sequence identity with these same three accessions. The RFLP pattern for these 10 samples via *i*PhyClassifier (Zhao et al. 2009) was identical to the 16SrVI-A accession AY390261.

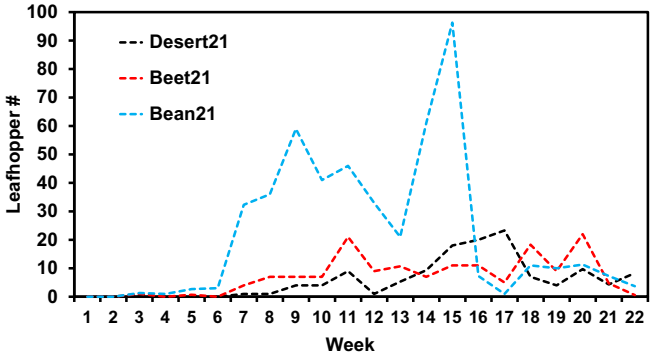

FIGURE 5
The mean number of beet leafhoppers, *Circulifer tenellus*, in Owyhee County on 258 cm^2 of yellow sticky cards after 7 days in desert areas or next to sugar beet or common bean fields collected for 22 weeks in 2021 beginning April 21 in southern Idaho.

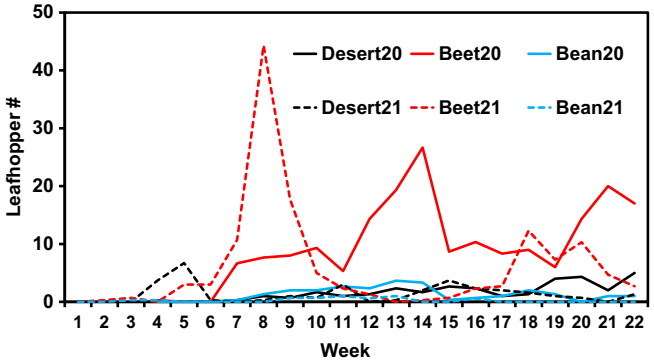

FIGURE 6
The mean number of beet leafhoppers, *Circulifer tenellus*, in Twin Falls County on 258 cm^2 of yellow sticky cards after 7 days in desert areas or next to sugar beet or common bean fields collected for 22 weeks in 2020 and 2021 beginning April 21 in southern Idaho.

Isolate LH21-522-CaPtrifolii (OR428571) had 99.68% sequence identity with the three GenBank accessions mentioned above. The RFLP pattern for LH21-522-CaPtrifolii was different from all reference patterns but most similar (0.93) to 16SrVI-A accession AY390261. Isolate LH21-362-CaPtrifolii (OR428570) had 99.84% sequence identity with the three GenBank accessions mentioned above. The RFLP pattern for LH21-362-CaPftrifolii was identical to the reference pattern for a 16SrVI accession X83431, which is a subgroup D strain and previously shown to be closely related to the '*Ca.* P. trifolii' sequence of AY390261 (Gopala et al. 2018).

The '*Ca.* P. trifolii' samples were all from Elmore and Owyhee Co. in southern Idaho, which were below 944 m in elevation, and all but one sample was collected in late June or July. The two '*Ca.* P. pruni' samples (LH20-380-CaP_pruni from Bingham Co. and LH21-848-CaP_pruni from Elmore Co.) had 100% sequence identity (OR428572) with the '*Ca.* P. pruni' 16SrIII-Q accession AF302841. Based on RFLP results from *i*PhyClassifier, both sequences were also identical to the reference pattern for 16SrIII-Q accession AF302841. Spiroplasmas were not detected in the plant samples.

Sequencing of the BLH COI gene identified 194 haplotypes (a set of DNA variants along a single chromosome that tend to get inherited together; the variation may only be a single nucleotide) over the two years. The sequencing for the 23 haplotypes that occurred more than twice was deposited in GenBank (OR433969-OR433991). In both years, the same two haplotypes were dominant, whereas 45% of the BLH samples were haplotype 1 (OR433969) and 28% were haplotype 2 (OR433970) and widely distributed throughout southern Idaho (Table 3). Haplotypes 1 and 2 only differed by a single-nucleotide polymorphism. BLASTn analysis found that 96% of haplotypes (representing 99.4% of the samples) had 98.46 to 99.66% sequence identity with the *C. tenellus* NCBI accession HM462269. Although closely related, HM462269 did not have complete sequence identity with 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. 7).

Pathogen identification in plant samples

The incidence of sugar beet plants with curly top symptoms in 2020 was at trace levels (<1%) at all sites except for the edge of the Twin Falls County field (>80% at the edge but rapidly dropped to trace levels moving toward the center of the field). In 2021, the incidence of sugar beet plants with curly top symptoms was at trace levels in Elmore and Minidoka Co. fields, whereas the Owyhee Co. field had about 2% incidence by the edge of the field. The Bingham and Twin Falls Co. fields had more than 80% incidence of plants with symptoms at the edge of the field, but the incidence rapidly dropped to trace levels moving toward the center of the fields. In 2020, 10% of the sugar beet plant samples were positive for CP, and the virus was not detected with the strain-specific primers (Table 4). In 2021, 44% of the sugar beet samples were positive for CP, and the following strains were detected: 55% Wor-like, 32% CA/Logan, 9% CO, 9% Svr, and 5% undetectable with strain-specific primers.

The incidence of plants with curly top symptoms in 2020 bean fields was at trace levels except for the edge of the Bingham Co. field, which was near 100% (rapidly dropped to trace levels toward the center of the field). In 2021, the incidence of plants with curly top symptoms in bean fields was at trace levels at all sites except for the Owyhee Co. field. In the Owyhee Co. field, there was nearly 100% incidence of plants with curly top symptoms at the edge of the field, but this rapidly dropped

to trace levels moving toward the center of the field. In 2020, 28% of the bean samples were positive for CP, and 91% were Wor-like strains and 9% undetectable with the strain specific primers (Table 4). In 2021, 42% of the bean samples were positive for CP, and the following strains were detected: 48% Wor-like, 48% CA/Logan, and 19% Svr. When the sequencing from three amplicons (two from sugar beet and one from common bean) generated with CA/Logan primers (OR451588-OR451590) was evaluated via BLASTn, they had 99.66 to 100% sequence identity with at least one of the following BCTV-CA/Logan genomes: KX867029, KX867034, KX867040, and KX867041. The sequences from three Svr strain amplicons (two from common bean and one from sugar beet) were identical (OR451600) and had 99.51% sequence identity with BCTV-Svr genomes KX867028, KX867027, KT276916, KT276906, and U02311. Three amplicons were generated with the Wor-like primers from sugar beet leaf tissue. One amplicon (OR451614) had 96.09% sequence identity with CO genome KX867047. The other two amplicons (OR451633 and OR451634) had 98.31 to 98.59% sequence identity with Wor genome KX867053. When samples that were positive for CP primers but failed for strain-specific primers were investigated further, they were found to have too little DNA to allow for sequencing of the CP amplicon. Thus, these samples were not investigated further. The PeCT strain of BCTV, SpCTAV, phytoplasmas, and spiroplasmas were not detected in the plant samples either year.

Parasitoids in BLH samples

When amplifying a region of the COI gene in 2020, 10 BLH samples were found to contain parasitoid species of the order Diptera (true flies): *Chalarus* sp. Walker (Diptera:Pipunculidae; one sample), *Eudorylas* sp. Aczél (Diptera:Pipunculidae; four samples), *Tomosvaryella* sp. Aczél (Diptera:Pipunculidae; four samples), and *Voria* sp. Rob.-Des. (Diptera:Tachinidae; one sample). The *Chalarus* sample (OR433993) had 100% sequence identity with *Chalarus* sp. accession KR651558. One of the *Eudorylas* samples (OR433995) had 100% sequence identity with *Eudorylas subopacus* Loew accessions KM627941 and KM649142. Two of the *Eudorylas* samples (OR433994) had 95.5 to 99.8% sequence identity with at least one of the following *Eudorylas* sp. accessions: MF852369, MF854488, and KR682774. *Eudorylini* sample LH20-421 had 87% sequence identity with *Eudorylas* sp. accession MF857553. The four *Tomosvaryella* samples had 95.9 to 99.8% sequence identity (OR433996-OR433998) with *Tomosvaryella* sp. accessions JF871394 and MG299054. The *Voria* sample (OR434999) had 99.8% sequence identity with *Voria ruralis* Fallén accession KP899671. In 2020, no parasitoid wasps were found in the BLH samples. In 2021, four BLH samples were found to contain parasitoid fly species: *Eudorylas* sp. (one sample) and *Tomosvaryella* sp. (three samples). The *Eudorylas* sample (OR434001) had 99.3% sequence identity with the *Eudorylas* sp. accession MF850556. Two of the *Tomosvaryella* samples (OR434002, OR434003) had 96.1 to 99.8% sequence identity with accession JF871394. The *Tomosvaryellini* sample had 86.5% sequence identity with accession MG294051. In 2021, five BLH samples were found to contain parasitoid wasps: *Dryinidae* sp. (four samples) and *Mymaridae* sp. (one sample). Three of the *Dryinidae* samples had 99.5 (OR433992), 97.3, and 90.1% sequence identity with the *Dryinidae* accessions MF903468 and MF905962. The closest GenBank accessions identified to the species level to these three samples was an *Aphelopus* sp. Dalman (Hymenoptera:Dryinidae) accession MZ626344 with 90.1 to 92.0% identity. The fourth *Dryinidae* sample had 89.8% sequence identity with *Gonatopus clavipes* Thunberg (Hymenoptera:Dryinidae) accession MZ628772. The *Mymaridae* sample had 89.8% sequence identity with the *Cosmocomoidea* sp. Howard (syn. *Gonatocerus* sp.; Hymenoptera:Mymaridae) accession KR898520.

Discussion

Aside from plant resistance, historically, the timing and BLH movement in relation to plant growth was considered the most important factor for controlling beet curly top in the western United States (Blickenstaff and Traveller 1979; Rojas et al. 2018). The initial BLH movement was highly correlated with above-normal temperatures from February through May (Harries and Douglass 1948). From 1936 to 1961, the average initial spring migration of BLH in southern Idaho started on May 25 and reached their peaks on June 23, with most of the BLH entering the fields within 7 to 10 days before the peak (Blickenstaff and Traveller 1979). During 1935 to 1944 in southern Idaho, the initial spring migration ranged from May 12 to June 5, with May 25 as the average (Douglass et al. 1946). The period between the initial and peak movement ranged from 18 to 56 days, with 31 days as the average (Douglass et al. 1946). During the current study in Elmore Co., the BLH populations in the desert peaked on May 19, 2020, and June 2, 2021, which is about 1 month earlier than that mentioned in historical data for southern Idaho. The early-season Elmore Co. desert peaks in the current study are closer to when BLH migrations used to begin in the historical literature (Blickenstaff and Traveller 1979; Douglass et al. 1946). In Owyhee Co. in 2021, the BLH population in the desert peaked late in the season on August 11, which was likely because rainfall was not adequate to support the growth of annual weeds over the winter and early spring. By late spring/early summer, rains

TABLE 3

Percentage of the two dominant *Circulifer tenellus* cytochrome oxidase haplotypes found in samples from yellow sticky cards collected for 22 weeks beginning April 21 in five counties in southern Idaho during 2020 and 2021

	Haplotype 1						Haplotype 2					
	Desert		Sugar beet		Common bean		Desert		Sugar beet		Common bean	
County[a]	2020	2021	2020	2021	2020	2021	2020	2021	2020	2021	2020	2021
Bingham	0	25	50	37	29	31	0	50	33	37	32	23
Elmore	51	47	56	53	29	52	26	27	35	22	38	22
Minidoka	8	42	57	14	67	40	17	25	29	29	33	12
Owyhee	ND	33	ND	56	ND	46	ND	38	ND	17	ND	27
Twin Falls	39	26	48	36	43	36	26	35	32	31	18	9
Overall	47	42	51	44	33	46	25	30	33	25	31	22

[a] Total sample number for desert in 2020 and 2021 was 218 and 343, respectively. The total sample number for sugar beets in 2020 and 2021 was 145 and 283, respectively. The total sample number for common beans in 2020 and 2021 was 129 and 228, respectively. ND = no data.

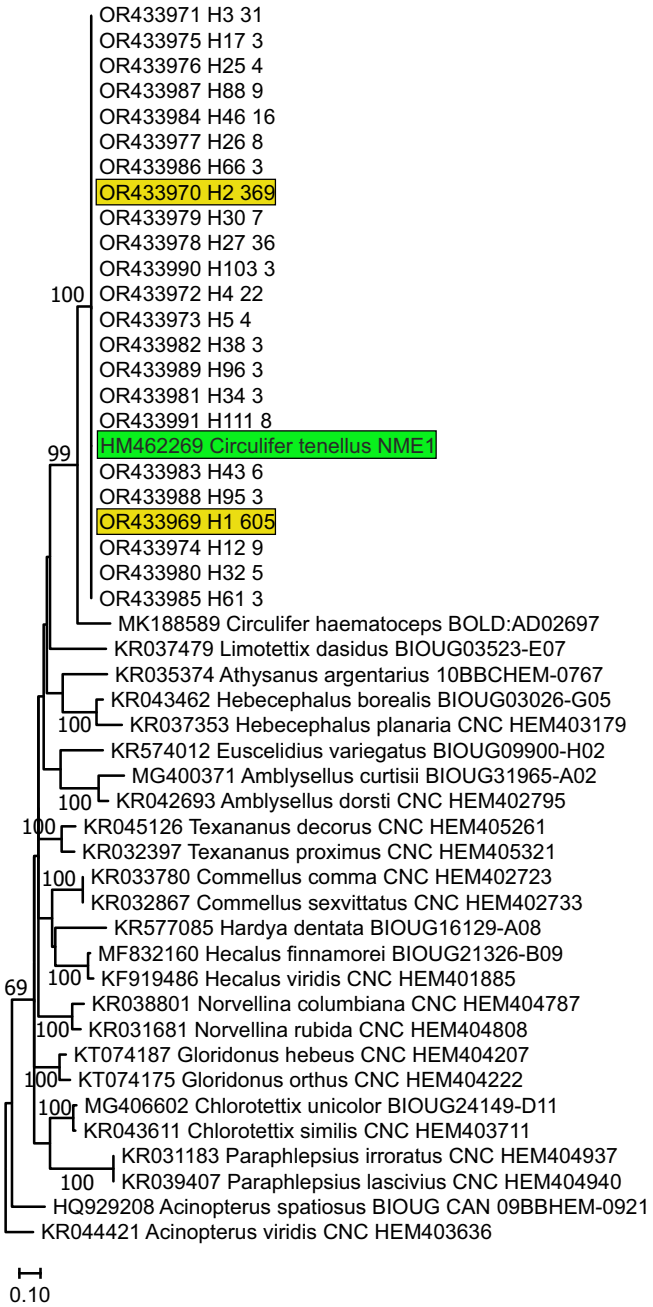

FIGURE 7

Phylogenetic relationships among the 23 most frequently identified *Circulifer tenellus* haplotypes from yellow sticky card samples collected in southern Idaho and compared with sequences from GenBank for genera from the *Deltocephalinae* subfamily based on a 585-bp region of the cytochrome oxidase gene. Numbers on the nodes of the maximum-likelihood (ML) tree represent the statistical support for ML (1,000 replicates). The tree is drawn to scale, with the branch lengths measured in substitutions per site. The tree is rooted to *Acinopterus viridis*. Haplotypes are identified by their accession number followed by haplotype designation (H number) and the number of samples they represent, whereas the GenBank accessions have the accession number followed by the species and sample identification. 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).

in this desert area did allow for weed growth, which likely led to the late-season peak in that desert area. The BLH desert populations in the other locations were low to nonexistent, making peak identification difficult or impossible. With shifting weather patterns and drought conditions occurring in southern Idaho, the desert areas monitored in Minidoka and Twin Falls counties had no winter annual weeds during the start of the winter and at the time of initial card placement in the spring. During the winter and early spring at the Minidoka desert site, all desert vegetation was dead in both years, whereas sagebrush (*Artemisia tridentata* Nutt.) was the only green vegetation present at the Owyhee and

TABLE 4

Beet curly top virus (BCTV) strains associated with sugar beet and common bean plant samples collected in five counties in southern Idaho during 2020 and 2021

			BCTV strain (% of CP positive)[b]			
Site[a]	Sample #	CP (%)	CA/Logan	CO	Svr	Wor+
2020						
Bingham						
Sugar beet	10	0	0	0	0	0
Common bean	10	60	0	0	0	100
Across county	20	30	0	0	0	100
Elmore						
Sugar beet	10	0	0	0	0	0
Common bean	10	10	0	0	0	100
Across county	20	5	0	0	0	100
Minidoka						
Sugar beet	10	20	0	0	0	0
Common bean	10	40	0	0	0	75
Across county	20	30	0	0	0	50
Owyhee						
Sugar beet	ND	ND	ND	ND	ND	ND
Common bean	ND	ND	ND	ND	ND	ND
Across county	ND	ND	ND	ND	ND	ND
Twin Falls						
Sugar beet	10	20	0	0	0	0
Common bean	10	0	0	0	0	0
Across county	20	10	0	0	0	0
Overall	80	19	0	0	0	66
2021						
Bingham						
Sugar beet	10	20	0	0	0	50
Common bean	10	30	100	33	0	0
Across county	20	25	60	20	0	20
Elmore						
Sugar beet	10	90	11	0	22	78
Common bean	10	40	75	0	25	25
Across county	20	65	31	0	23	62
Minidoka						
Sugar beet	10	20	50	0	0	50
Common bean	10	20	100	0	50	0
Across county	20	20	50	0	25	25
Owyhee						
Sugar beet	10	50	60	0	0	40
Common bean	10	50	0	0	0	100
Across county	20	50	30	0	0	70
Twin Falls						
Sugar beet	10	40	50	50	0	25
Common bean	10	70	29	0	29	57
Across county	20	55	36	0	18	45
Overall	100	43	37	7	14	51

[a] In mid-August to early September 2020 and late July 2021, plant samples were collected from stunted plants with curled leaves from sugar beet and common bean fields in five southern Idaho counties. CP = percentage of the BLH samples that were positive for the beet curly top virus (BCTV) coat protein primers. ND = no data.

[b] The percentage of coat protein-positive samples that had this specific BCTV strain based on strain-specific primers for the California/Logan (CA/Logan), Colorado (CO), Severe (Svr), and Worland (Wor) strains. The Wor+ primers detected both Wor and CO strains based on sequencing.

Twin Falls desert sites at initial card placement. In Elmore and Bingham Co., winter annual weeds were present in both years. Winter annual weeds in desert areas were historically thought to be the primary source of BLH (Cook 1967; Douglass and Hallock 1957; Douglass et al. 1946; Wallace and Murphy 1938). However, given that the BLH population peaks found next to crop fields differed from what was found in desert areas, local weed populations seemed to be the primary source of BLH in most if not all locations in this study. The 2021 Owyhee bean site was near the Snake River, which was supporting considerable weed growth along its banks, which likely led to the large BLH peaks (C. A. Strausbaugh, *personal observation*). Sprinkler irrigation during windy periods led to drift and weed populations around other crop fields in the study, which led to local BLH populations in these areas (C. A. Strausbaugh, *personal observation*). Other poorly or unmanaged weedy areas near these fields may have also contributed to BLH numbers.

If fall rains are not sufficient to promote germination of overwintering annual weeds, the BLH must pass some time on shelter hosts; high mortality rates occur on such hosts (Cook 1967). In Idaho, males rarely survive the winter, but fertilized female leafhoppers are highly resistant to low temperatures and overwinter on annual plants and then were reported to lay their eggs in March or early April on these plants based on historical reports (Bennett 1971; Cook 1967; Douglass et al. 1942). This first generation has been reported to mature in May or June, adults of the second generation (summer generation) in July or early August, and the third generation (overwintering generation) in September or October (Cook 1967; Douglass et al. 1942). Given the earlier peak BLH captures in the present study, perhaps the timing of the generations may be changing as well in Idaho. There is considerable overlapping of generations, especially in the fall. In Oregon and Washington, three generations were observed with the following timing: early May to mid-June, early July to mid-August, and mid-August to late September (Munyaneza et al. 2008). In both years, the BLH population in the Elmore desert area seemed to match this timing. The BLH population peaks in other counties and sites were hard to identify because of low numbers.

Historically, eastern Idaho has had less beet curly top than western Idaho because the incidence of infected sugar beet plants in the eastern Oregon-western Idaho area was 1.5 to 2.0 times higher than in the Burley-Rupert area (Cassia and Minidoka Co. in Fig. 1) and 6.0 times greater than that in the Idaho Falls area (Bingham and Bonneville Co.) (Blickenstaff and Traveller 1979). In the western end of the study area (also lower elevation), the Elmore desert sites (average of 222 and 401 BLHs on a yellow card in one week) and the Owyhee bean field (average 96 BLHs per card) had the highest peak values in the study. The Twin Falls sugar beet sites (in the central part of the valley) had low to intermediate peaks (average of 27 to 44 BLHs per sugar beet card, but only zero to seven at other sites), whereas on the eastern side of the valley, the Bingham field sites peaked at an average of 16 to 23 BLHs at field sites. The Bingham desert sites and all Minidoka sites had very low BLH numbers (average of zero to four BLH). Thus, the BLH population data collected during the 2020 and 2021 growing seasons fit with what has been historically observed in southern Idaho for beet curly top pressure.

The number of BLHs entering fields can vary tremendously depending on area and season. The population of migrating BLHs varied from 79 adults/9.29 m^2 in 1939 to 2,939/9.29 m^2 in 1937 (Douglass et al. 1946). The higher population would equate to about 30 BLHs per sugar beet plant (Bennett 1971). In 2020, the average number of BLHs on 258 cm^2 of yellow card after 1 week peaked at 22 in the Bingham bean field, at 27 in the Twin Falls sugar beet field, and at 24 and 69 in the Elmore bean and sugar beet fields, respectively. In 2021, the average number of BLHs on 258 cm^2 of yellow card after 1 week peaked at 6 and 16 in the Bingham bean and sugar beet fields, at 44 in the Twin Falls sugar beet field, at 54 and 55 in the Elmore bean and sugar beet fields, and at 22 and 96 in the Owyhee sugar beet and bean fields, respectively. Fortunately, most of these peaks occurred later in the season when plants were larger in size and had developed age-related resistance (Wang et al. 1999). Based on our experience running curly top nurseries in southern Idaho, if commercial sugar beet plants are infected with BCTV by six viruliferous BLHs per plant (the inoculation level for resistance screening in curly top nurseries) at the four- to six-leaf growth stage, the plants will not survive to the end of the growing season. In years with normal spring rainfall, early movement of BLH can be expected, but with late spring and above normal rainfalls, the movement of BLH will be delayed because weed hosts such as Russian thistle [*Kali tragus* (L.) Scop. (syn. *Salsola kali* and *Salsola tragus*)] will still be green and reasonably healthy (Blickenstaff and Traveller 1979).

Idaho desert areas used to be sprayed with insecticides targeting BLH using an action threshold of 50 BLHs or more per 9.29 m^2 (Blickenstaff and Traveller 1979). From 1949 through 1969, Idaho sprayed 83,366 ha of weedy host plant desert areas for direct control of BLH, but Idaho has since discontinued this practice (Blickenstaff and Traveller 1979; Strausbaugh et al. 2006). Aside from environmental concerns, part of the reason for not spraying desert areas was the realization that within cultivated areas, there were many other areas able to support a large spring BLH generation that could not easily be sprayed with insecticide. These areas included intermittently farmed ground, livestock corrals, lava outcrops, idle or waste land areas, roadsides, river and ditch banks, pastures, overgrazed areas, and other sites supporting wild mustard (principally flixweed [*Descurainia sophia* (L.) Webb ex Prantl; syn. *Sophia parviflora* (Lam.) Standl.], perfoliate pepperweed [*Lepidium perfoliatum* L.], and tumblemustard [*Sisymbrium altissimum* L.; syn. *Norta altissima* (L.) Britton]) (Bennett 1971; Blickenstaff and Traveller 1979). In more recent times with the switch to sprinkler irrigation (Panella et al. 2014), drift during wind events can also lead to problematic weedy areas. There is some historical evidence for an occasional spring migration of BLH into southern Idaho from the southern desert, but such movements are rare (Cook 1967). Movements into crop fields in southern Idaho are predominantly from local breeding grounds and only a few miles at most rather than long-distance flights (Bennett 1971; Carter 1930; Cook 1967; Douglass et al. 1942, 1946). COI sequencing from the current study is consistent with BLH populations coming from local sources. None of the 194 COI haplotypes matched the *C. tenellus* NCBI accession HM462269, which originated in New Mexico, suggesting that our populations differ. However, the same two haplotypes dominated the BLH population in both 2020 and 2021 and were widely distributed in the southern Idaho study areas (Table 3). The selection pressure leading to the haplotypes being dominant is unknown.

Historical data suggest a low percentage of the BLH moving into sugar beet fields had virus in the spring, but by the summer, nearly 100% of the BLHs moving through the same fields carried viruses based on bioassays on sugar beet plants (Wallace and Murphy 1938). When collected from pure stands of Russian-thistle in 1941 and 1947, 29 and 31% of the BLHs were viruliferous, whereas those collected from Russian-thistle in sugar beet fields were 61 and 43% viruliferous the previous springs (Hallock and Douglass 1956). When BLHs were collected in laboratory studies (utilizing flowerpots, flats, and cages) from sugar beets and kochia, 90 and 82% were viruliferous, respectively

(Hallock and Douglass 1956). Laboratory studies indicated that Halogeton and kochia were much more suitable than Russian-thistle and smotherweed [*Bassia hyssopifolia* (Pall.) Kuntze] as reservoirs of BCTV (Hallock and Douglass 1956). In Oregon, of the 800 BLHs tested, 19% were found positive for BCTV (Rondon et al. 2016). Of the 460 BLH samples collected from Washington and one location in Idaho in 2021, 27% tested positive for BCTV (Cooper et al. 2023). In the present study, 51% of the BLHs were viruliferous for BCTV in 2020 and 69% in 2021, which is higher than the two recent studies just mentioned. The percentage of BLHs with virus was as high as 100% in June and July, but early and late in the season, the percentage of BLHs with virus was lower, which fits with historical data. If researchers intend to find positive BCTV samples in Idaho, sampling in late June or July is ideal.

The incidence of BCTV strains in sugar beet has been shown to vary between samplings in southern Idaho (Strausbaugh et al. 2008, 2017). In 2006 to 2007 samples, the BCTV strain incidence was 87% Svr, 60% Wor-like, 7% CA/Logan, and 59% mixed infections, whereas in 2012 to 2015 samples, the incidence was 2% Svr, 87% Wor-like, 30% CA/Logan, and 16% mixed (Strausbaugh et al. 2008, 2017). Thus, there was a transition from Svr and Wor-like strains and mixed infections in sugar beet plants in 2006 to 2007 to primarily Wor-like strains (mostly CO and Wor) and single infections in the 2012 to 2015 samples (Strausbaugh et al. 2008, 2017). The BCTV strain data in the BLH study are similar to those found in 2012 to 2015 sugar beet plants, except that the PeCT strain was also detected. The PeCT strain was not detected in the plant samples and has not been detected previously in Idaho.

The incidence of BCTV strains in common bean has not been thoroughly investigated. In a recent study focused on host resistance in Washington state, the Wor-like primers did detect BCTV in common bean tissue naturally infected in the field (Soler-Garzón et al. 2023). In agro-inoculation assays, CA/Logan was also shown to infect common bean (Soler-Garzón et al. 2023). During the current study in Idaho, we established that the CA/Logan, CO, Svr, and Wor strains of BCTV could be detected in common bean leaves.

The Wor primers (BMCTV-C1 2213F and 2609R) utilized in previous studies were found to detect more than just the Wor strain (Strausbaugh et al. 2017). Based on sequencing the genomes of 39 Wor-like primer-positive samples, 56% of these samples were established to be CO, whereas 39 and 5% were Wor and Kim1, respectively (Strausbaugh et al. 2017). In the current study, we utilized Wor primers (BMCTVv2825 and BGc396) developed by Chen et al. (2010), and the results were similar to those of the 2017 study. However, one should note that all these primers were developed prior to 2014, when some of the mild strains were considered to be the same strain, and have since been recognized as separate strains (Varsani et al. 2014a). Thirty samples positive for the Wor primers in the current study were sequenced and found to vary depending on the time of the season collected. The 13 samples collected in June or July were 85% Wor and 15% CO, which differed ($\chi^2 = 64$; $P < 0.0001$) from the 17 samples collected in August or September (71% CO and 29% Wor). Why the strains present would vary between early and late in the season is unknown.

When trying to detect other curly top species, the only positive was SpCTAV. SpCTAV was initially detected in spinach plants originating from Arizona in 2009 (Hernández-Zepeda et al. 2013) and is a member of the *Becurtovirus* genus. The only other member of this genus is BCTIV, which can be transmitted by *Circulifer haematoceps* Mulsant & Rey (syn. *Neoaliturus haematocepts* Baker; Hemiptera:Cicadellidae) (Gharouni Kardani et al. 2013; Heydarnejad et al. 2013). However, *C. haematoceps* is not known to occur in North America. There is no direct proof that *C. tenellus* can transmit SpCTAV, but our BLH data show that it can at least be detected in *C. tenellus* samples. The SpCTAV was widespread in Idaho in BLH samples but not in plant samples. These data suggest that SpCTAV is likely infecting plant species other than sugar beet and common bean. This is the second study to identify this virus outside of Arizona. A recent publication determined that SpCTAV could be detected in red table beet being grown in Idaho (Ramachandran et al. 2023).

Giddings (1950) found that BLH may carry two or more strains simultaneously and that such strains can be introduced into susceptible plants singly or in combination. In the present study, there were mixed infections in 18% of the 2020 BLH samples and 43% of the 2021 BLH samples. In recent studies, the mild strains of BCTV have also been found in the western United States. In the Hermiston-Umatilla region of northeastern Oregon, viruses closely related to the Wor strain were predominant in BLHs (Rondon et al. 2016). In Oregon hemp, Rivedal et al. (2022) found Wor and CO strains of BCTV. In California hemp, Melgarejo et al. (2022) found that plants were infected with Wor or CO strains, and 43% of samples had mixed Wor and CO infections. Similar results for hemp were found in Arizona and Colorado (Chiginsky et al. 2021; Giladi et al. 2020; Hu et al. 2021). In the present study when the CO strain of BCTV was found, they were typically coinfected (plants) or mixed (BLHs) with the Wor strain. The CO and Wor strains have also been reported in mixed infections in tomato, sugar beet, and coriander (Chen et al. 2010; Strausbaugh et al. 2017; Swisher Grimm et al. 2021) as well as in mixtures in BLHs (Chen et al. 2010; Rondon et al. 2016).

The BLH is also capable of transmitting phytoplasmas such as '*Ca.* P. trifolii' (16SrVI), which can infect potato and other vegetables; this pathogen was initially referred to as beet leafhopper transmitted virescence agent (BLTVA; Crosslin et al. 2006, Munyaneza et al. 2007). In the Columbia Basin and Yakima Valley in Washington, BLTVA averaged 18% incidence in BLHs from potato areas and 23.5% from weedy areas in 2005 (Munyaneza et al. 2008). In 2006 from the same areas, BLTVA had an average incidence of 36% from potato areas and 21% from weedy areas (Munyaneza et al. 2008). Of the 460 BLH samples collected from Washington and one location in Idaho in 2021, 23% tested positive for phytoplasmas (Cooper et al. 2023). In the present study, phytoplasmas were found in 1% of the BLH samples in both years. Phytoplasmas were not detected in the sugar beet or common bean plant samples. All BLH '*Ca.* P. trifolii'-positive Idaho samples originated from the lower elevation Treasure Valley sites (below 944 m in Elmore and Owyhee Co.). Most if not all the sites in this study were at higher elevation than areas of the Columbia Basin, which may explain the lower percentage of positive samples in this study. Two samples were identified as '*Ca.* P. pruni', which is the X-disease phytoplasma and a considerable problem in stone fruits but is not known to affect sugar beet or common beans (Harper et al. 2023). The 16S sequencing for the '*Ca.* P. pruni' samples had 100% sequence identity with a strain (AF302841) isolated from black raspberries in Oregon, which was used to establish new subgroup Q within 16SrIII (Davis et al. 2001). The BLH can also transmit spiroplasmas such as *Spiroplasma citri* (Chen and Gilbertson 2016; Liu et al. 1983), but none was detected either year in southern Idaho.

Biocontrol of BLH was considered throughout the early to mid-1900s (Bennett 1971). Some of the parasitoids investigated included the internal and egg parasitoids, such as the *Eudorylas subopacus* and *Pipunculus* sp. Latreille (Diptera:Pipunculidae); *Gonatopus* sp. Ljungh (Hymenoterpa:Dryinidae); *Aphelinoidea*

sp. Gir. (Hymenoptera:Trichogrammatidae) and *Paracentrobia subflava* Gir. (Hymenoptera:Trichogrammatidae); and *Anagrus* sp. Haliday, *Anaphes* sp. Haliday, *Gonatocerus* sp. N. v. Esen., and *Polynema* sp. Haliday (all Hymenoptera:Mymaridae). There are also lacewings such as *Chrysopa* sp. Leach (Neuroptera:Chysopidae) and several groups of predatory bugs, including *Geocoris* sp. Fallén (Hemiptera:Geocoridae), *Neides* sp. Fabricius (Hemiptera:Berytidae), *Nabis* sp. Latreille (Hemiptera:Nebidae), and *Zelus* sp. Fabricus (Hemiptera: Reduviidae) as having possibilities in controlling BLH (Britt et al. 2022; Hartung 1919; Henderson 1955; Knowlton 1932; Krugner et al. 2008; Severin 1933; Stahl 1920; Walker et al. 2005). In the present study, parasitic flies (*Chalarus* sp., *Eudorylas* sp., *Tomosvaryella* sp., and *Voria* sp.) and wasps (*Dryinidae* and *Mymaridae* sp.) were detected at relatively low prevalence. To our knowledge, this is the first survey of the parasitoid species attacking BLH in Idaho since reports mentioned by Bennett (1971). No effective biological controls for BLH have been found when evaluated previously (Munyaneza et al. 2008).

Control of BCTV in sugar beet and common beans has historically centered around host resistance (Larsen and Miklas 2004; Soler-Garzón et al. 2023; Strausbaugh et al. 2017). Common bean contains effective resistance from the *Bct* allele in some cultivars; current commercial sugar beet cultivars are considered to have only low to intermediate resistance to BCTV (Larsen and Miklas 2004; Panella et al. 2014; Soler-Garzón et al. 2023). With the change in curly species and strains in the western United States, utilizing host resistance in sugar beet will continue to be a challenge because some sources of resistance appear to be strain specific, resistance is quantitatively inherited, and resistance is difficult to maintain in parental lines used to create the commercial hybrids (Gillen et al. 2008; Kaffka et al. 2002; Montazeri et al. 2016; Panella et al. 2014; Strausbaugh et al. 2007). Thus, although neonicotinoid seed treatments have some environmental concerns, they will need to be the primary supplement to host resistance until better host resistance can be introduced into sugar beet cultivars or alternative control measures can be found.

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Literature Cited

Bennett, C. W. 1971. The Curly Top Disease of Sugarbeet and Other Plants, Monograph No. 7. American Phytopathological Society, St. Paul, MN.

Blickenstaff, C. C., and Traveller, D. 1979. Factors affecting curly top damage to sugarbeets and beans in southern Idaho, 1919-77. Science and Education Administration, Agricultural Reviews and Manuals, Western Series, No. 8. USDA-ARS, Oakland, CA.

Bonmatin, J.-M., Giorio, C., Girolami, V., Goulson, D., Kreutzweiser, D. P., Krupe, C., Liess, M., Long, E., Marzaro, M., Mitchell, E. A. D., Noome, D. A., Simon-Delso, N., and Tapparo, A. 2015. Environmental fate and exposure; neonicotinoids and fipronil. Environ. Sci. Pollut. Res. 22:35-67.

Britt, K. E., Meierotto, S., Morelos, V., and Wilson, H. 2022. First year survey of arthropods in California hemp. Front. Agron. 4:901416.

Carter, W. 1930. Ecological studies of the beet leafhopper. USDA Tech Bull. 206.

Chen, L.-F., Brannigan, K., Clark, R., and Gilbertson, R. L. 2010. Characterization of curtoviruses associated with curly top disease of tomato in California and monitoring for these viruses in beet leafhoppers. Plant Dis. 94:99-108.

Chen, L.-F., and Gilbertson, R. L. 2009. Curtovirus-cucurbit interaction: Acquisition host plays a role in leafhopper transmission in a host-dependent manner. Phytopathology 99:101-108.

Chen, L.-F., and Gilbertson, R. L. 2016. Transmission of curtoviruses (*Beet curly top virus*) by the beet leafhopper (*Circulifer tenellus*). Chapter 17. Pages 243-262 in: Vector-Mediated Transmission of Plant Pathogens. J. K. Brown, ed. American Phytopathological Society, St. Paul, MN.

Chen, L.-F., Natwick, E., Cabrera, S., and Gilbertson, R. L. 2014. First report of curly top disease of basil caused by *Beet severe curly top virus* in California. Plant Dis. 98:286.

Chiginsky, J., Langemeier, K., MacWilliams, J., Albrecht, T., Cranshaw, W., Fulladolsa, A. C., Kapuscinski, M., Stenglein, M., and Nachappa, P. 2021. First insights into the virus and viroid communities in hemp (*Cannabis sativa*). Front. Agron. 3:778433.

Cook, W. C. 1967. Life history, host plants, and migrations of the beet leafhopper in the western United States. USDA Technical Bulletin.

Cooper, W. R., Walker, W. B., III, Angelella, G. M., Swisher Grimm, K. D., K., D., Foutz, J. J., Harper, S. J., Nottingham, L. B., Northfield, T. D., Wohleb, C. H., and Strausbaugh, C. A. 2023. Bacterial endosymbionts identified from leafhopper (Hemiptera: Cicadellidae) vectors of phytoplasmas. Environ. Entomol. 52:243-253.

Creamer, R. 2020. Beet curly top virus transmission, epidemiology, and management. Pages 521-527 in: Applied Plant Virology. L. P. Awasthi, ed. Academic Press, Cambridge, MA.

Creamer, R., Luque-Williams, M., and Howo, M. 1996. Epidemiology and incidence of beet curly top geminivirus in naturally infected weed hosts. Plant Dis. 80:533-535.

Crosslin, J. M., Vandermark, G. J., and Munyaneza, J. E. 2006. Development of a real-time, quantitative PCR for detection of the Columbia Basin potato purple top phytoplasma in plants and beet leafhoppers. Plant Dis. 90:663-667.

Davis, R. E., Dally, E. L., and Converse, R. H. 2001. Molecular identification of a phytoplasma associated with witches'-broom disease of black raspberry in Oregon and its classification in group 16SrIII, new subgroup Q. Plant Dis. 85:1121.

Deng, S., and Hiruki, C. 1991. Amplification of 16S rRNA genes from culturable and nonculturable Mollicutes. J. Microbiol. Methods 14:53-61.

Dewar, A. M. 2022. Integrated pest management in action – derogation for use of the neonicotinoid, thiamethoxam in sugar beet in 2022. Outlooks Pest Manag. 33:37-40.

Douglass, J. R., and Cook, W. C. 1954. The beet leafhopper. USDA Circular 942.

Douglass, J. R., Dorst, H. E., and Peay, W. E. 1942. Beet leafhopper populations in southern Idaho and northern Utah during the seasons of 1940 and 1941. Proc. Amer. Soc. Sugar Beet Technol. 3:438-451.

Douglass, J. R., and Hallock, H. C. 1957. Relative importance of various host plants of the beet leafhopper in southern Idaho. USDA Technical Bulletin 1155.

Douglass, J. R., Hallock, H. C., Fox, D. E., and Hofmaster, R. N. 1946. Movement of spring-generation beet leafhoppers into beet fields of south-central Idaho. Proc. Amer. Soc. Sugar Beet Technol. 4:289-297.

Folmer, O., Black, M., Hoeh, W., Lutz, R., and Vrijenhoek, R. 1994. DNA primers for amplification of mitochondrial cytochrome *c* oxidase subunit I from diverse metazoan invertebrates. Mol. Mar. Biol. Biotechnol. 3: 294-299.

Gharouni Kardani, S., Heydarnejad, J., Zakiaghl, M., Mehrvar, M., Kraberger, S., and Varsani, A. 2013. Diversity of *Beet curly top Iran virus* isolated from different hosts in Iran. Virus Genes 46:571-575.

Giddings, N. J. 1950. Combination and separation of curly-top virus strains. Proc. Amer. Soc. Sugar Beet Technol. 6:502-507.

Giladi, Y., Hadad, L., Luria, N., Cranshaw, W., Lachman, O., and Dombrovsky, A. 2020. First report of beet curly top virus infecting *Cannabis sativa* in western Colorado. Plant Dis. 104:999.

Gillen, A. M., Strausbaugh, C. A., and Tindall, K. V. 2008. Evaluation of *Beta corolliflora* for resistance to curly top in Idaho. J. Sugar Beet Res. 45:99-118.

Golino, D. A., Oldfield, G. N., and Gumpf, D. J. 1987. Transmission characteristics of the beet leafhopper transmitted virescence agent. Phytopathology 77:954-957.

Gopala, Khasa, E., Rao, A., Madhupriya, and Rao, G. P. 2018. Molecular characterization of 'Clover proliferation' phytoplasma subgroup-D (16SrVI-D) associated with vegetable crops in India. Physiol. Mol. Biol. Plants 24:203-210.

Greenway, G. 2022. Estimating economic impacts of beet leafhopper on potato and carrot crops in the Columbia Basin region of Oregon and Washington. Southwest Entomol. 47:89-96.

Haegele, R. W. 1927. The beet leafhopper (*Eutettix tenellus* Baker); A survey in Idaho. University of Idaho, Agricultural Experiment Station Bulletin 156.

Hall, T. A. 1999. BioEdit: A user-friendly biological science sequence alignment editor and analysis program for Windows 95/98/NT. Nucleic Acids Symp. Ser. 41:95-98.

Hallock, H. C., and Douglass, J. R. 1956. Studies of four summer hosts of the beet leafhopper. J. Econ. Entomol. 49:388-391.

Harper, S. J., Northfield, T. D., Nottingham, L. R., DuPont, S. T., Thompson, A. A., Sallato, B. V., Serban, C. F., Shires, M. K., Wright, A. A., Catron, K. A., Marshall, A. T., Molnar, C., and Cooper, W. R. 2023. Recovery plan for X-disease in stone fruit caused by '*Candidatus* Phytoplasma pruni'. Plant Health Prog. 24:258-295.

Harries, F. H., and Douglass, J. R. 1948. Bionomic studies on the beet leafhopper. Ecol. Monograph 18:45-79.

Hartung, W. J. 1919. Enemies of the beet leafhopper. Natural foes of *Eutettix tenella* in California and their usefulness. Facts Sugar 8:470-471.

Henderson, C. F. 1955. Overwintering, spring emergence and host synchronization of two egg parasites of the beet leafhopper in southern Idaho. USDA Circular 967.

Hernández-Zepeda, C., Varsani, A., and Brown, J. K. 2013. Intergenic recombination between a new, spinach-infecting curtovirus and a new geminivirus belonging to the genus *Becurtovirus*: First New World exemplar. Arch. Virol. 158:2245-2254.

Heydarnejad, J., Keyvani, N., Razavinejad, S., Massumi, H., and Varsani, A. 2013. Fulfilling Koch's postulates for beet curly top Iran virus and proposal for consideration of a new genus in the family *Geminiviridae*. Arch. Virol. 158:435-443.

Hu, J., Masson, R., and Dickey, L. 2021. First report of *Beet curly top virus* infecting industrial hemp (*Cannabis sativa*) in Arizona. Plant Dis. 105: 1233.

Jensen, A. 2008. Beet leafhopper monitoring with yellow sticky cards. Potato Prog. 8:1-5.

Kaffka, S. R., Wintermantel, W. M., and Lewellen, R. T. 2002. Comparisons of soil and seed applied systemic insecticides to control *Beet curly top virus* in the San Joaquin Valley. J. Sugar Beet Res. 39:59-74.

Knowlton, G. F. 1932. The beet leafhopper in northern Utah. Utah Agricultural Experiment Station Bulletin 234.

Krugner, R., Johnson, M. W., Groves, R. L., and Morse, J. G. 2008. Host specificity of *Anagrus epos*: A potential biological control agent of *Homalodisca vitripennis*. BioControl 53:439-449.

Kumar, S., Stecher, G., and Tamura, K. 2016. MEGA7: Molecular evolutionary genetics analysis version 7.0 for bigger datasets. Mol. Biol. Evol. 33:1870-1874.

Larkin, M. A., Blackshields, G., Brown, N. P., Chenna, R., McGettigan, P. A., McWilliam, H., Valentin, F., Wallace, I. M., Wilm, A., Lopez, R., Thompson, J. D., Gibson, T. J., and Higgins, D. G. 2007. Clustal W and Clustal X version 2.0. Bioinformatics 23:2947-2948.

Larsen, R. C., and Miklas, P. N. 2004. Generation and molecular mapping of a sequence characterized amplified region marker linked with the *Bct* gene for resistance to *Beet curly top virus* in common bean. Phytopathology 94:320-325.

Liu, H.-Y., Gumpf, D. J., Oldfield, G. N., and Calavan, E. C. 1983. The relationship of *Spiroplasma citri* and *Circulifer tenellus*. Phytopathology 73:585-590.

Lorenz, K.-H., Schneider, B., Ahrens, U., and Seemüller, E. 1995. Detection of the apple proliferation and pear decline phytoplasmas by PCR amplification of ribosomal and nonribosomal DNA. Phytopathology 85: 771-776.

Melgarejo, T. A., Chen, L.-F., Rojas, M. R., Schilder, A., and Gilbertson, R. L. 2022. Curly top disease of hemp (*Cannabis sativa*) in California is caused by mild-type strains of *Beet curly top virus* often in mixed infection. Plant Dis. 106:3022-3026.

Meyerdirk, D. E., and Oldfield, G. N. 1985. Evaluation of trap color and height placement for monitoring *Circulifer tenellus* (Baker) (Homoptera:Cicadellidae). Can. Ent. 117:505-511.

Montazeri, R., Shams-Bakhsh, M., Mahmoudi, S. B., and Rajabi, A. 2016. Evaluation of sugar beet lines for resistance to beet curly top viruses. Euphytica 210:31-40.

Munyaneza, J. E., Crosslin, J. M., and Lee, I.-M. 2007. Phytoplasma diseases and insect vectors in potatoes of the Pacific Northwest of the United States. Bull. Insectol. 60:181-182.

Munyaneza, J. E., Jensen, A. S., Hamm, P. B., and Upton, J. E. 2008. Seasonal occurrence and abundance of beet leafhopper in the potato growing region of Washington and Oregon Columbia Basin and Yakima Valley. Am. J. Pot. Res. 85:77-84.

Panella, L., Kaffka, S. K., Lewellen, R. T., McGrath, J. M., Metzger, M. S., and Strausbaugh, C. A. 2014. Sugarbeet. Pages 357-396 in: Yield Gains in Major U.S. Field Crops. S. Smith, B. Diers, J. Specht, and B. Carver, eds. Crop Science Society of America (CSSA) Special Publication 33. CSSA, Madison, WI.

Ramachandran, V., Wyatt, N. A., Santiago, E. R., Neher, O. T., Weiland, J. J., and Bolton, M. D 2023. First report of pepper yellow dwarf strain of beet curly top virus and Spinach curly top Arizona virus in red table beet in Idaho, United States. Plant Dis. 107:2571.

Rivedal, H. M., Funke, C. N., and Frost, K. E. 2022. An overview of pathogens associated with biotic stresses in hemp crops in Oregon, 2019 to 2020. Plant Dis. 106:1334-1340.

Rojas, M. R., Macedo, M. A., Maliano, M. R., Soto-Aguilar, M., Souza, J. O., Briddon, R. W., Kenyon, L., Bustamante, R. F. R., Zerbini, F. M., Adkins, S. Legg, J. P., Kvarnheden, A., Wintermantel, W. M., Sudarshana, M. R., Peterschmitt, M., Lapidot, M., Martin, D. P., Moriones, E., Inoue-Nagata, A. K., and Gilbertson, R. L. 2018. World management of Geminiviruses. Annu. Rev. Phytopathol. 56:637-677.

Rondon, S. I., Roster, M. S., Hamlin, L. L., Green, K. J., Karasev, A. V., and Crosslin, J. M. 2016. Characterization of *Beet curly top virus* strains circulating in beet leafhoppers (Hemiptera: Cicadellidae) in northeastern Oregon. Plant Dis. 100:1586-1590.

Schneider, B., Seemüller, E., Smart, C. D., and Kirkpatrick, B. C. 1995. Phylogenetic classification of plant pathogenic mycoplasma-like organisms or phytoplasma. Pages 369-380 in: Molecular and Diagnostic Procedures in Mycoplasmology, Vol. 1. S. Razin and J. G. Tully, eds. Academic Press, San Diego, CA.

Severin, H. H. P. 1933. Field observations on the beet leafhopper, *Eutettix tenellus*, in California. Hilgardia 7:281-360.

Singh, S. P., and Schwartz, H. F. 2010. Breeding common bean for resistance to diseases: A review. Crop Sci. 50:2199-2223.

Soler-Garzón, A., Goldoff, D., Thornton, A., Swisher Grimm, K. D., Hart, J. P., Song, Q., Strausbaugh, C. A., and Miklas, P. N. 2023. A robust SNP-haplotype assay for *Bct* gene region conferring resistance to *beet curly top virus* in common bean (*Phaseolus vulgaris* L.). Front. Plant Sci. 14:1215950.

Stahl, C. F. 1920. Studies in the life history and habits of the beet leafhopper. J. Agric. Res. 20:245-252.

Strausbaugh, C. A., Eujayl, I. A., and Foote, P. 2010. Seed treatments for the control of insects and diseases in sugarbeet. J. Sugar Beet Res. 47: 105-125.

Strausbaugh, C. A., Eujayl, I. A., and Wintermantel, W. M. 2017. *Beet curly top virus* strains associated with sugar beet in Idaho, Oregon, and a western U.S. collection. Plant Dis. 101:1373-1382.

Strausbaugh, C. A., Gillen, A. M., Camp, S., Shock, C. C., Eldredge, E. P., and Gallian, J. J. 2007. Relationship of beet curly top foliar ratings to sugar beet yield. Plant Dis. 91:1459-1463.

Strausbaugh, C. A., Gillen, A. M., Gallian, J. J., Camp, S., and Stander, J. R. 2006. Influence of host resistance and insecticide seed treatments on curly top in sugar beets. Plant Dis. 90:1539-1544.

Strausbaugh, C. A., Wenninger, E. J., and Eujayl, I. A. 2012. Management of severe curly top in sugar beet with insecticides. Plant Dis. 96: 1159-1164.

Strausbaugh, C. A., Wenninger, E. J., and Eujayl, I. A. 2014. Control of curly top in sugar beet with seed and foliar insecticides. Plant Dis. 98:1075-1080.

Strausbaugh, C. A., Wenninger, E. J., and Eujayl, I. A. 2016. Length of efficacy for control of curly top in sugar beet with seed and foliar insecticides. Plant Dis. 100:1364-1370.

Strausbaugh, C. A., Wintermantel, W. M., Gillen, A. M., and Eujayl, I. A. 2008. Curly top survey in the western United States. Phytopathology 98:1212-1217.

Swisher, K. D., Munyaneza, J. E., Velásquez-Valle, R., and Mena-Covarrubias, J. 2018. Detection of pathogens associated with psyllids and leafhoppers in *Capsicum annuum* L. in the Mexican states of Durango, Zacatecas, and Michoacán. Plant Dis. 102:146-153.

Swisher, K. D., Velásquez-Valle, R., and Mena-Covarrubias, J., and Munyaneza, J. E. 2016. Occurrence and molecular detection of *Sprioplasma citri* in carrots and its insect vector, *Circulifer tenellus*, in Mexico. J. Plant Pathol. 98:355-360.

Swisher Grimm, K. D., Crosslin, J. M., Cooper, W. R., Frost, K. E., du Toit, L. J., and Wohleb, C. H. 2021. First report of curly top of *Coriandrum sativum* caused by *Beet curly top virus* in the Columbia basin of Washington State. Plant Dis. 105:3313.

Thompson, H., Vaughan, S., Mahlein, A.-K., Ladewig, E., and Kenter, C. 2022. Is there a risk to honeybees from use of thiamethoxam as a sugar beet seed treatment? Integr. Environ. Assess. Manag. 18:709-721.

Varsani, A., Martin, D. P., Navas-Castillo, J., Moriones, E., Hernández-Zepeda, C., Idris, A., Zerbini, F. M., and Brown, J. K. 2014a. Revisiting the classification of curtoviruses based on genome-wide pairwise identity. Arch. Virol. 159:1873-1882.

Varsani, A., Navas-Castillo, J., Moriones, E., Hernández-Zepeda, C., Idris, A., Brown, J. K., Zerbini, F. M., and Martin, D. P. 2014b. Establishment of three new genera in the family *Geminiviridae*: *Becurtovirus*, *Eragrovirus* and *Turncurtovirus*. Arch. Virol. 159:2193-2203.

Walker, G. P., Bayoun, I. M., Triapitsyn, S. V., and Honda, J. Y. 2005. Taxonomy of *Aphelinoidea* (Hymenoptera: Trichogrammatidae) species attacking eggs of the beet leafhopper, *Circulifer tenellus* (Hemiptera: Cicadellidae), in California. Zootaxa 1068:1-25.

Wallace, J. M., and Murphy, A. M. 1938. Studies on the epidemiology of curly top in southern Idaho, with special reference to sugar beets and weed hosts of the vector *Eutettix tenellus*. USDA Technical Bulletin 624.

Wang, H., Gurusinghe, P. de A., and Falk, B. W. 1999. Systemic insecticides and plant age affect beet curly top virus transmission to selected host plants. Plant Dis. 83:351-355.

Warren, L. M. 2021. To bee or not to beet with neonicotinoids. Environ. Law Rev. 23:103-109.

Woodcock, B. A., Ridding, L., Pereira, M. G., Sleep, D., Newbold, L., Oliver, A., Shore, R. F., Bullock, J. M., Heard, M. S., Gweon, H. S., and Pywell, R. F. 2021. Neonicotinoid use on cereals and sugar beet is linked to continued low exposure risk in honeybees. Agric. Ecosystems Environ. 308:107205.

Yazdi, H. R. B., Heydarnejad, J., and Massumi, H. 2008. Genome characterization and genetic diversity of beet curly top Iran virus: A geminivirus with a novel nonanucleotide. Virus Genes 36:539-545.

Yokomi, R. K., Mello, A. F. S., Saponari, M., and Fletcher, J. 2008. Polymerase chain reaction-based detection of *Spiroplasma citri* associated with citrus stubborn disease. Plant Dis. 92:253-260.

Zhao, Y., Wei, W., Lee, I.-M., Shao, J., Suo, X., and Davis, R. E. 2009. Construction of an interactive online phytoplasma classification tool, *i*PhyClassifier, and its application in analysis of the peach X-disease phytoplasma group (16SrIII). Int. J. Syst. Evol. Microbiol. 59:2582-2593.