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SCIENTIFIC NOTE

First report of *Leptoglossus occidentalis* (Hemiptera: coreidae) feeding on *Rubus* (Rosaceae) fruit

Joey B. Tanney[1], Annie Dicaire[1], Esme John[1], Josie Roberts[2], and Ward Strong[3]

[1]Pacific Forestry Centre, Canadian Forest Service, Natural Resources Canada, 506 Burnside Road West, Victoria, British Columbia, V8Z 1M5, Canada, [2]Centre for Plant Health, Canadian Food Inspection Agency, 8801 East Saanich Road, North Saanich, British Columbia, V8L 1H3, Canada, and [3]Independent Researcher, 7811 Westkal Road, Coldstream, British Columbia, V1B 1Y4, Canada
Corresponding author: Joey B. Tanney; Email: Joey.Tanney@nrcan-rncran.gc.ca

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Abstract
The western conifer seed bug, *Leptoglossus occidentalis* Heidemann (Hemiptera: Coreidae), feeds extensively on the seeds of conifer trees within both its endemic range west of the Rocky Mountains and its expanding introduced range throughout eastern North America, South America, Europe, and Asia. Its damage to conifer seed challenges seed production in natural stands and in orchards that produce seed for forest regeneration and food. In summer 2023, hundreds of *L. occidentalis* adults were observed feeding on the berries of two cultivated blackberry, *Rubus fruticosus* Linnaeus (Rosaceae), vines on southern Vancouver Island, British Columbia, Canada. The adults were observed over a period of approximately six weeks, and their stylets were seen penetrating drupelets and causing subsequent damage. Identification was confirmed based on morphological and molecular (*CO*1 DNA barcoding) methods. To our knowledge, this is the first verifiable report of *L. occidentalis* feeding on a Rosaceae host, suggesting the insect's potential for damaging agricultural systems.

The western conifer seed bug, *Leptoglossus occidentalis* Heidemann (Hemiptera: Coreidae), is widely distributed within its natural range west of the Rocky Mountains in North America, where it feeds on the seeds of conifer hosts, especially Douglas-fir (*Pseudotsuga menziesii* (Mirbel) Franco) (Pinaceae) and both native and introduced pine (*Pinus* spp.) (Pinaceae). Nymphs and adults feed on developing and mature cones by inserting their stylets through cone scales and penetrating individual seeds, injecting salivary enzymes, and sucking out the resulting contents. Feeding damage results in seed loss and conelet abortion (Bates *et al.* 2002). In British Columbia, Canada, large populations of western conifer seed bugs may cause severe damage to seed crops in Douglas-fir and pine; for example, seed losses as high as 83% in lodgepole pine (*Pinus contorta* Douglas) seed orchards have been reported (Forest Genetics Council of British Columbia, n.d.; Strong 2015). Bates and Borden (2005) estimated that a hypothetical density of one *L. occidentalis* per lodgepole pine tree will result in an expected seed loss of approximately 310 seeds.

The invasive range of the western conifer seed bug now continues into eastern North America (McPherson *et al.* 1990) and Europe. It was first found in northern Italy in 1999 (Tescari 2001), from where it spread rapidly throughout Europe in less than 15 years (Lesieur *et al.* 2019). There it impacts, among other species, the stone pine (*Pinus pinea* Linnaeus), which is used for pine nut production. In Italy, pine nut production declined by approximately 95% after the insect's

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Figure 1. *Leptoglossus occidentalis* adults feeding on fruit of *Rubus fruticosus*: **A**, an adult perched on a ripe berry; **B**, arrow highlights the proboscis inserted into a drupelet; and **C**, the proboscis is inserted into a drupelet, and an adjacent pale, collapsed drupelet shows feeding damage.

introduction (Roversi *et al.* 2011). The western conifer seed bug has also invaded South America (*e.g.*, Faúndez *et al.* 2018; Kun and Masciocchi 2019), Asia (*e.g.*, Ishikawa and Kikuhara 2009), and Africa (*e.g.*, Ben Jamâa *et al.* 2013).

On 12 August 2023, western conifer seed bug adults were observed perching on the ripe fruit of two cultivated thornless *Rubus fruticosus* Linnaeus (Rosaceae) vines (Fig. 1) in Langford, British Columbia. The three-year-old vines were planted in a single row occupying a length of 4 m within a suburban yard with a southwest-facing aspect and full sun exposure. The berry vines were located approximately 170 m from a mature *Pseudotsuga menziesii* forest. Western conifer seed bugs were found concurrently with a notable population of conchuela stink bugs, *Chlorochroa ligata* Say (Heteroptera: Pentatomidae), which had caused moderate damage to the blackberry fruit and were subjected to control efforts including hand picking and spraying with a solution of dish detergent and water. Observations were repeatedly made of western conifer seed bugs feeding on fruit, with their proboscis inserted between or through individual drupelets (Fig. 1). Drupelets that were fed on became discoloured (pink to pale) and collapsed. The population on both vines was estimated to be at least 200 individuals. Continuous observations were made until the older canes bearing fruit were pruned and disposed of on 23 September 2023. Casual surveys of wild *Rubus armeniacus* Focke and cultivated *Rubus* spp. within 500 m of the observed western conifer seed bugs did not yield any other detections. Western conifer seed bugs were also not observed on the grapes of a cultivated *Vitis* vine planted within 1 m of the blackberry vines.

Thirty-six adults were collected and stored in vials containing 70% ethanol. Species identification was confirmed by morphology and by amplifying and sequencing the mitochondrial cytochrome *c* oxidase subunit 1 gene (*CO*1). Two specimens were submitted to the National Identification Service (Agriculture and Agri-Food Canada, Ottawa, Ontario, Canada) and subsequently confirmed as *L. occidentalis* and then deposited into the Pacific Forestry Centre Arthropod Reference Collection, Victoria, British Columbia (PFC-2024-00013, PFC-2024-00014). Total genomic DNA was extracted from a fresh specimen using a Qiagen DNeasy® Blood and Tissue Kit (Qiagen, Hilden, Germany) following the manufacturer's protocol, with the modification that grinding the specimen was replaced with piercing the abdomen with a sterilised scalpel. The specimen was kept in the extraction buffer until after the incubation and preserved in 70% ethanol. The partial *CO*1 sequence was amplified and sequenced using the primer pair LepF1 and LepR1 (Hebert *et al.* 2004). Polymerase chain reaction was conducted in a total volume of 25 µL containing 12.5 µmol of each primer, 4 µg BSA, 5 µmol dNTPs mix, 0.2 µL AccuStart II Taq DNA Polymerase (Quantabio, Beverly, California, United States of America), 60 µmol $MgCl_2$, 2.5 µL 10X PCR Buffer II, 4.1 µL water, and 16 ng of template DNA. The thermocycler conditions were 94 °C for 2 minutes, then five cycles of 94 °C for 40 seconds, 45 °C

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for 40 seconds, and 72 °C for 1 minute, followed by 35 cycles each of 94 °C for 40 seconds, 51 °C for 40 seconds, and 72 °C for 1 minute, with a final extension at 72 °C for 5 minutes. Polymerase chain reaction product was verified by gel electrophoresis and submitted to the SANGER sequencing platform at Centre de recherche du CHU de Québec–Université Laval (Québec City, Québec, Canada) for sequencing using the BigDye Terminator sequencing kit, version 3.1 (Applied Biosystems, Foster City, California) and analysed with the ABI 3730xl Data Analyzer (Applied Biosystems). The resulting sequences were assembled and manually checked using Geneious Prime, version 2020.2.5 (https://www.geneious.com). Based on a BLASTn query with the National Center for Biotechnology Information GenBank, our *CO*1 sequence shared a 99.39–99.85% similarity with available *L. occidentalis* sequences – for example, identities = 657/658 (99%) with no (0) gaps (*L. occidentalis* 21_BHS_05, OP686468.1). The *CO*1 sequence was deposited into GenBank (accession number PP301970).

To our knowledge, there are no published reports of *L. occidentalis* feeding on *Rubus* or other Rosaceae hosts. *Leptoglossus occidentalis* appears to preferentially feed on conifer seeds of *Pseudotsuga menziesii* and *Pinus* species within its home range, but it is also reported on a broad range of conifers, including *Abies* (Pinaceae), *Calocedrus* (Cupressaceae), *Cedrus* (Pinaceae), *Cupressus* (Cupressaceae), *Juniperus* (Cupressaceae), *Larix* (Pinaceae), *Picea* (Pinaceae), and *Tsuga* (Pinaceae) (Barta 2009; Lindelöw and Bergsten 2012; Grozea and Muntean 2019), and it is even described as a pest of pistachio seeds, *Pistacia vera* Linnaeus (Anacardiaceae) (Rice *et al.* 1985; Uyemoto *et al.* 1986). The western conifer seed bug has been reported to create holes with its stylets in a variety of natural and nonnatural substrates, including cross-linked polyethylene (PEX) plumbing pipes (Bates 2005), plastic Petri dishes and Tygon tubing, needles of *Pinus monticola* Douglas ex D. Don and *P. contorta* (both Pinaceae), and the frothy spittle mass of the cone spittlebug, *Aphrophora canadensis* Walley (Hemiptera: Aphrophoridae) (W. Strong, unpublished data).

Leptoglossus occidentalis was clearly observed feeding on *Rubus* fruit over a period of approximately six weeks, but it is not known whether it can complete its life cycle on this host or if this was a rare opportunistic behaviour. We were unable to visually ascertain whether the insects were feeding on seeds, the liquid contents of the mesocarp, or the receptacle. In the future, damage to berries should be studied more thoroughly to elucidate feeding behaviour, which presumably involves seeds but may include berry juice, as our observations were made during a period of severe drought that may have limited water availability. Western conifer seed bugs produce an aggregation pheromone, which may explain why the observed insects occurred in such a localised area and not on nearby *Rubus* or other potential host fruit plants (Blatt and Borden 1996; Millar *et al.* 2022). This first observation of *L. occidentalis* feeding on a rosaceous host suggests that the western conifer seed bug has the potential to act as an agricultural pest, thus expanding its phytosanitary significance.

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Competing interests. The authors declare that they have no competing interests.

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