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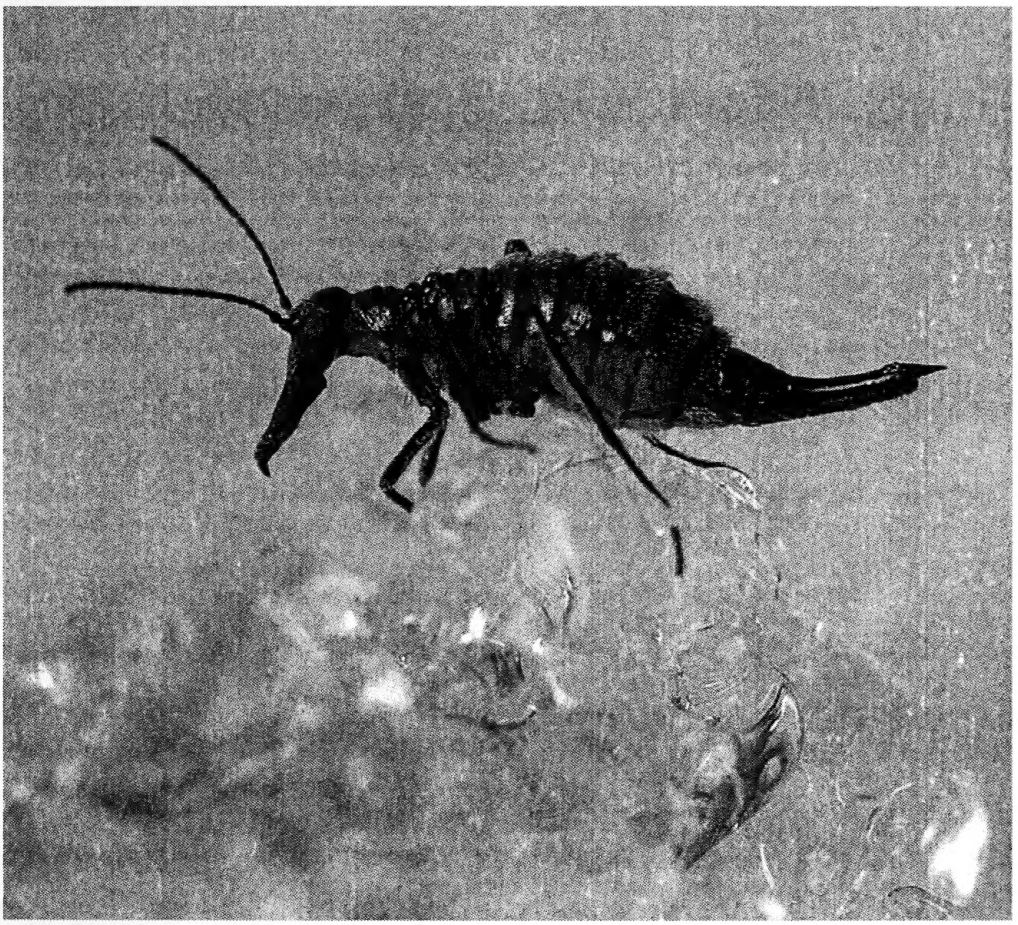

ESBC

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COVER: *Boreus reductus* Carpenter (Mecoptera: Boreidae)

Boreids (order Mecoptera) are interesting for two traits, they are often found wandering the surface of snow from late November until May, and they can jump. A feat they achieve using metatibial extension and resilin to store energy in the thorax before a leap. Their common name of "snow fleas" is both apt and prescient, given that recent molecular studies have placed them as close relatives of the true fleas. Their biology is poorly known and most texts state that both adults and larvae "live in moss". This example is a female of the species *Boreus reductus* (Carpenter 1933). The image was taken a few hundred metres from the junction of the Coquihalla Highway and Highway 1 at the west end of Kamloops in March 2007. An Olympus E-1 DSLR with a Zuiko ZD 50mm macro lens coupled to two 25mm extension tubes was used. The magnification on the sensor (18mmx14mm) was X1 .5 . This image is a crop of the original image, the length of the insect from frons to tip of ovipositor is approximately 4 mm. Lighting was with a 1980's vintage Vivitar 283 flash with a homemade 10cm by 15cm cloth diffuser. The most important part of the field equipment was a set of chest waders.

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New provincial and state records for Heteroptera (Hemiptera) in Canada and the United States

G.G.E. SCUDDER[1]

ABSTRACT

New provincial records are provided for 52 species of Heteroptera in Canada. *Dichaetocoris piceicola* (Knight) is also reported from Alaska, and *Pagasa nigripes* Harris from Washington State.

INTRODUCTION

Since the publication of the checklist of the Hemiptera of Canada and Alaska (Maw *et al.* 2000), new Heteroptera from Canada, and new provincial records have been published by Barnes *et al.* (2000), Henry *et al.* (2008), Jansson (2002), Kenner and Needham (2004), Paiero *et al.* (2003), Roch (2007), Schuh (2000a, 2000b, 2001, 2004a, 2004b), Schuh and Schwartz (2004, 2005), Schwartz and Scudder (2001, 2003), Schwartz and Stonedahl (2004), Scudder (2000, 2004, 2007), Scudder and Foottit (2006), Scudder and Schwartz (2001), Wheeler and Hoebeke (2004), and Wheeler *et al.* (2006). Wright (1989) and Kerzhner (1993) also published records not included in Maw *et al.* (2000).

Recent research has revealed additional new provincial records for 52 species. *Dichaetocoris piceicola* (Knight) has been found in Alaska, and *Pagasa nigripes* Harris in Washington State.

These are reported below, with Museum abbreviations as follows:

AAFCL: Agriculture and Agri-Food Canada, Lethbridge, AB (J.R. Byers).

CNC: Canadian National Collection of Insects, Agriculture and Agri-Food Canada, Ottawa, ON (R.G. Foottit).

LM: Lyman Entomological Museum, Macdonald College, McGill University, Ste.-Anne-de-Bellevue, QC (T. Wheeler).

MU: Memorial University. St. John's , NF (D. Larson).

NSM: Nova Scotia Museum of Natural History, Halifax, NS (A. Hebda and C. Majka).

RBCM: Royal British Columbia Museum, Victoria, BC (R.A. Cannings).

SM: Saskatchewan Provincial Museum, Saskatoon, SK (R. Hooper).

UBC: Spencer Entomological Museum, Department of Zoology, University of British Columbia, Vancouver, BC (K.M. Needham).

UG: Department of Environmental Biology, University of Guelph, Guelph, ON (S.A. Marshall).

UM: J.B. Wallis Collection, University of Manitoba, Winnipeg, MB (R.E. Roughley).

UPEI: Department of Biology, University of Prince Edward Island, Charlottetown, PE (K.A. Campbell and D. Giberson).

NEW PROVINCIAL RECORDS

The systematic order of families and higher taxa listed below, follows Maw *et al.* (2000).

Infraorder NEPOMORPHA

Family CORIXIDAE

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Arctocorisa chanceae Hungerford

A subarctic species known from Alaska and across northern Canada from Yukon to Newfoundland and Labrador (Scudder 1997; Maw *et al.* 2000). The species can be keyed using Hungerford (1948) and Brooks and Kelton (1967).

New record. **BC**: 1♂, Atlin, 10 mi S, 18.viii.1980 (R.J. Cannings) [UBC].

Sigara compressoidea (Hungerford)

An eastern Nearctic species, in Canada reported from Ontario east to Newfoundland (Maw *et al.* 2000), but not previously reported from Prince Edward Island. *Sigara compressoidea* is keyed in Hungerford (1948) and Tinerella and Gundersen (2005), and a dorsal colour photograph is given in the latter reference, showing the typically effaced membrane patterning.

New record. **PE**: 1♂ 1♀, Deroche Natural Protected Area, 46.42114°N 62.94082° W, kicknet, small wetland, 27.ix.2007 (K. Alexander Campbell) [UPEI].

Family NOTONECTIDAE

Notonecta spinosa Hungerford

A Cordilleran species, so far in Canada, recorded only from British Columbia (Scudder 1977; Maw *et al.* 2000). The species can be keyed using Hungerford (1933).

New records: **AB**: 1 specimen, Waterton Lakes Nat. Park, grassland pond with mudstone, substrate, near Buffalo Paddock, 49° 07'44"N 113°51'11"W, 9.vii.2005 (R.E. Roughley & R.D. Kenner) [UBC]; 1 specimen, Waterton Lakes Nat. Park, spring-fed *Carex* marsh, near West Entrance on Hwy. 5, 49°07'12"N 113°50'53"W, 9.vii.2005 (R.E. Roughley & R.D. Kenner) [UBC].

Family PLEIDAE

Neoplea striola (Fieber)

This pygmy backswimmer, so far in Canada is recorded only from Manitoba, Ontario and Quebec (Maw *et al.* 2000). The species is keyed in Brooks and Kelton (1967).

New record. **BC**: 1 specimen, Edgewood, F11, edge forest by beaver pond, 25.viii.-5.ix.1988 (H. Knight) [CNC].

Infraorder GERROMORPHA

Family VELIIDAE

Microvelia pulchella Westwood

This cosmopolitan species is recorded from Alaska, and in Canada from most provinces from British Columbia to Newfoundland (Maw *et al.* 2000). The species is keyed in Brooks and Kelton (1967).

New record. **SK**: 1♂ 2♀, Cowan Dam at Hwy. 55, 54°11'49"N 107°27'0"W, *Typha* pond, 22.vii.2003 (R.E. Roughley) [CNC].

Family GERRIDAE

Gerris incognitus Drake & Hottes

A species reported across Canada from British Columbia to Labrador (Maw *et al.* 2000), but not previously recorded from Prince Edward Island. The species is keyed by Drake and Harris (1934).

New record. **PE**: 1♂ 1♀, Millburn, 8.vi.1970 (Ray Wenn) [UPEI].

G. pingreensis Drake & Hottes

This species is known from Alaska and across Canada from Yukon to Labrador (Maw *et al.* 2000), but to date there are no published records for New Brunswick, Prince Edward Island, Nova Scotia and Newfoundland. The species is keyed by Drake and Harris (1934) and Brooks and Kelton (1967).

New record. **NF**: 2♂ 1♀, Plum Point, ponds, 11.ix.1999 (T. Huxley) [CNC].

Limnoporus notabilis (Drake & Hottes)

This western water strider in Canada is known from British Columbia and Alberta (Maw *et al.* 2000; Andersen and Spence 1992). The species is keyed by Andersen and Spence (1992).

New record. **SK**: 1♂, Cypress Hills, 15.vii.1977 (R. Hopper) [SM].

Family SALDIDAE

Micracanthia humilis (Say)

A Nearctic-Neotropical species, widely distributed in North America, and in Canada reported from British Columbia and Northwest Territories east to Newfoundland, but not previously recorded from Nova Scotia (Maw *et al.* 2000). The species

is keyed by Schuh (1967) and Polhemus and Chapman (1979).

New records. **NS**: 2♀, Halifax Co., Lawrencetown Beach, viii.1979 (B. Wright) [NSM]; 1♀ Halifax Co., Petpeswick Harbour, wet sandy upper shore, 29.vii.1971 (Barry Wright) [NSM].

Salda lugubris (Say)

A widely distributed Nearctic species that also occurs in Mexico, and in Canada is reported from Yukon to Newfoundland, but not previously from New Brunswick and Nova Scotia (Maw *et al.* 2000). The species is keyed by Brooks and Kelton (1967) and Schuh (1967).

New records. **NB**: 1♂, French Lake, 2.vii.1928 (W.S. Brown) [CNC]. **NS**: 1♀, CBI, Chiticamp, vi.-vii.1917 (F. Johansen) [CNC]; 2♀, Halifax Co., Port Wallis, 17.viii.1952 (D.C. Ferguson) [NSM].

Saldula ablusa Drake & Hottes

This is an eastern Nearctic species, keyed and reported from Ontario by Schuh (1967). Polhemus (1988) noted that references to *Saldula xanthochila* (Fieber) from the northeastern United States almost certainly refer to *S. ablusa* or *S. pallipes* (Fabricius). Wright (1989) reported *S. xanthochila* from Sable Is., Nova Scotia, and examination of 2♂ 1♀ specimens in the Nova Scotia Museum of Natural History, labeled "NS. Sable Is., brackish ponds east of station, 26.vii.1976 (Barry Wright)" shows these to be *S. ablusa*. As pointed out by Polhemus (1985), *S. ablusa* lacks a distinct dark distal streak on the ventral side of the hind femora, and as noted by Schuh (1967) typically has pale lateral margins to the pronotum, whereas these are never present in the *Saldula "pallipes – palustris"* group.

New record. **PE**: 7♂ 1♀, Can. Nat. Park, Dalvay House, 19.vii.1940 (G.S. Walley) [CNC].

S. bouchervillei (Provancher)

This species was previously reported from Nova Scotia as *Salda bouchervillei* (Provancher) by Wright (1989).

Material examined: **NS**: 2♀, Sable Island, brackish ponds east of station, 26.vii.1976 (Barry Wright) [NSM].

S. laticollis (Reuter)

Lindskog (1981) clarified the identity of this species, distinguishing it from the closely related *S. pallipes* (Fabricius) and *S. palustris* (Douglas) by the presence of long, curved, semi-recumbent or suberect setae on the head dorsally, and noted that *S. fernaldi* Drake is a synonym. In Old World populations of both *S. pallipes* and *S. palustris*, the pubescence on the head and dorsum generally is uniformly short and recumbent. However, as observed by Schuh (1967), the *Saldula "pallipes – palustris"* species complex has confused systematic heteropterists for some time, because of the extreme variability of the "species". Nevertheless, coastal populations from western North America and Newfoundland, previously identified as *S. palustris* have been shown to be *S. laticollis* (Lindskog 1981; Polhemus 1988), the intertidal biology of which has been described by Stock and Lattin (1976) under *S. palustris*.

Wright (1989) reported *S. palustris* from Sable Is., Nova Scotia, but examination of specimens in the Nova Scotia Museum of Natural History shows these to be *S. laticollis*. This species is also now known from New Brunswick and Prince Edward Island.

New records. **NB**: 1♂ 1♀, Kouchibouguac Nat. Park, 14.vi.1977 (S.J. Miller) [CNC]. **NS**: 2♂ 3♀, Digby Co., Sandy Cove, 4.viii.1971 (Barry Wright) [NSM]; 2♀, Guysborough Co., Liscombe, marshy area above beach, 13.viii.1971 (Barry Wright) [CNC]; 2♂ 1♀, Sable Island, West Light, 23.vii.1976 (Barry Wright) [NSM]; 1♂ 1♀, Sable Island, freshwater pond at West Light, 13.vi.1977 (Barry Wright) [NSM]. **PE**: 1♂, Brackley Beach, 5.vii.1966 (L.A. Kelton) [CNC]; 3♂, Green Gables, Cavendish Beach, 22.vii.1967 (J.E.H. Martin) [CNC].

Infraorder CIMICOMORPHA

Family ANTHOCORIDAE

Anthocoris tomentosus Péricart

This western Nearctic, and Beringian species occurs in the western United States south to Arizona (Lewis *et al.* 2005), and Alaska to Manitoba, but has not previously been reported from Saskatchewan (Maw *et al.* 2000). The species is keyed in Kelton (1978) under the name *A. melanocerus* Reuter, and characteristically has the hemelytra entirely shiny, and the pronotum and antennae completely black. Lewis *et al.* (2005) reported *A. tomentosus* being regularly collected from *Alnus*, *Populus*, *Pyrus* and *Salix* growing in and near fruit-growing regions in Washington State, as well as on *Rumex* and psyllid-infested *Sheperdia argentea* (Pursh) Nutt.

New record. **SK**: 1♀, Fort Qu'Appelle, 17.vi.1967 (R. Hooper) [SM]; 1♀, Regina, on elm, 6.x.1986 (K. Roney) [SM].

Dufouriellus ater (Dufour)

This species was described from Europe, and was first reported from America north of Mexico by Van Duzee (1916). It is now known to be widely distributed in North America, with published records in the USA for New York (Van Duzee 1917), North Carolina (Blatchley 1926), California and Kentucky (Blatchley 1928), Idaho (Harris and Shull 1944), Oregon (Lattin 2004) and Hawaii (Lattin 2005, 2007a), whereas in Canada it has previously been reported from British Columbia (Anderson 1962) and Ontario (Kelton 1978).

Dufouriellus ater is usually collected under the bark of trees, but also is often associated with stored products (Awadallah *et al.* 1984; Arbogast 1984; Lattin 1999). As a result, it is a useful predator of some economic importance (Lattin 2000).

The species is keyed by Kelton (1978), who noted that in Canada it is rare in collections, and probably introduced into British Columbia and Ontario. Although *D. ater* was not included as an alien in Canada by Scudder and Foottit (2006), Lattin (2004, 2007b) considered the species to be non-indigenous to America north of Mexico.

New record. **NS**: 1♂, Halifax, Grain Elevators, from dust and debris samples, 17.vii.1991 (J. Hulton) [NSM].

Family NABIDAE

Nabis inscriptus (Kirby)

This Holarctic species, with somewhat abbreviated wings is difficult to distinguish from shorter winged specimens of *N. americoferus* Carayon. Reliable separation is based on the shape of the male parameres and structure of the copulatory pouch of the female, as illustrated in Vinokurov (1988) and discussed by Kerzhner (1963). *Nabis inscriptus* is recorded from Alaska and across northern Canada (Maw *et al.* 2000), but as noted by Henry and Lattin (1988), reported distribution records need to be verified.

New records. **NS**: 1♀, Antigonish Co., Pomquet Beach, 31.v.1978 (B. Wright) [NSM]; 1♂, Halifax, found on corpse, Path. Lab., 1.xi.1979 (B. Wright) [NSM].

N. roseipennis Reuter

This species is recorded from British Columbia to Nova Scotia in Canada (Maw *et al.* 2000), but has not previously been reported from Prince Edward Island. The species is keyed in Blatchley (1926) and Harris (1928) and characteristically has black spots on the hind tibiae.

New record: **PE**: 6♂ 8♀, Charlottetown, UPEI, nr. East edge, 46°15'25"N 63° 08'08"W, sweeping, 20.ix.2004 (G.G.E. Scudder) [CNC, UPEI].

Pagasa nigripes Harris

Kerzhner (1993) raised *Pagasa fusca* var. *nigripes* Harris to specific rank, and recorded the species in Canada from Alberta, Quebec and Saskatchewan. He also reported *P. nigripes* from Alaska, Colorado, Massachusetts, New Hampshire, New Mexico, New York, Pennsylvania, Vermont and Wyoming. He distinguished it from *Pagasa fusca* (Stein) by differences in the male and female genitalia, and noted that the legs tend to be brown or brownish yellow in *P. nigripes*, whereas they are yellow in *P. fusca*, with the femora often orange or reddish. However, the legs in *P. fusca* can

sometimes be partly brownish or entirely black. Hence, the genitalic characters are the most reliable. The parameres in *P. fusca* are relatively large with the outer margin rounded, whereas in *P. nigripes* they are slightly smaller than in *P. fusca* and distinctly angulate on the outer margin.

Pagasa nigripes is here recorded for the first time from British Columbia, Northwest Territory and Yukon, where the species was previously reported as *P. fusca* in Maw *et al.* (2000): *P. fusca* does not occur in Alaska, Northwest Territory and Yukon. However, in British Columbia, *P. fusca* co-occurs with *P. nigripes*, the species having been collected together at Merritt (23 km E, Hamilton Commonage), Osoyoos (Mt. Kobau in Montane Spruce habitat), Vaseux Creek (Kennedy bench), and the Windermere Valley.

New records. **BC**: 1♂ , Canal Flats, 10.8 km S., 31.viii.1998 (G.G.E. Scudder) [CNC]; 1♂, Fairview, White L., BGxh1, SWm, pan trap WL/P-1, 4.vii.-11.vii.1995 (J. Jarrett) [UBC]; 3♀, Merritt, 23 km E, Hamilton Commonage, Upper Fescue grassland, early seral, 1250 m, 16.ix.2000 (G.G.E. Scudder) [CNC, UBC]; 1♀, Merritt, 35 km S, 14.viii.1988 (G.G.E. Scudder) [CNC]; 1♂, Nicola, 24.vii.1932 (G.J. Spencer) [UBC]; 1♂, Osoyoos, East Bench, *Artemisia/Purshia* assoc., pitfall trap, 15.vii. 17.viii. 1990 (G.G.E. Scudder) [CNC]; 1♂, Osoyoos IRI, Inkaneep, BGxh1, AN, pitfall trap T1-1, 6.vii.-9.viii.1995 (G.G.E. Scudder) [CNC]; 1♂, *id.*, 9.viii.-9.ix.1995 [CNC]; 1♀, *id.*, 9.ix.-6.x.1995 [CNC]; 1♀ *id.*, T2-1, 9.viii.-9.ix.1995 [CNC]; 1♂, *id.*, T2-5, 4.vii.-7.ix.1994 [CNC]; 1♂, *id.*, T4-1, 9.viii.-9.ix.1995 [CNC]; 1♀, *id.*, T4-3, 6.vii.-9.viii.1995 [CNC]; 1♀, *id.*, T4-5, 4.viii.-7.ix.1994 [CNC]; 1♀, *id.*, T5-2, 9.viii.-9.ix.1995 [CNC]; 1♂, Osoyoos, Mt. Kobau, MSxh, VK, Pitfall trap K4A-5, 10.viii.-8.ix.1995 (J. Jarrett) [UBC]; 1♀, *id.*, K4A-3, 18.viii.-28.ix.1997 [UBC]; 1♂, Osoyoos, Mt. Kobau Rd., IDFdk1, pitfall trap K3B-5, 18.viii.-28.ix.1996 (J. Jarrett) [UBC]; 1♂, *id.*, IDFxh1, pitfall trap K2B-4, 18.viii.-28.ix.1996 [UBC]; 1♂, *id.*, PPxh1, pitfall trap K1A-2, 28.vii.-18.viii.1997 [UBC]; 1♂ 1♀, Tatlayoka L., 16.vii.1978 (G.G.E. Scudder) [CNC]; 1♀, Vaseux Cr., CWS bench, BGxh1, AN, pitfall trap Y1-4, 6.ix.-4.x.1995 (G.G.E. Scudder) [CNC]; 1♀, *id.*, Y2-5 [CNC]; 1♂, *id.*, Y1-4, 8.vii.-3.viii.1994 [CNC]; 1♀, *id.*, Y2-5, 12.viii.-6.ix.1995 [CNC]; 1♀, *id.*, Y2-5, 6.ix.-4.x.1995 [CNC]; 1♀, *id.*, Y3-3, 5.vii.-12.viii.1995 [CNC]; 2♂, *id.*, Y3-3, 6.ix.-4.x.1995 [CNC]; 1♂, *id.*, Y3-5, 5.vii.-12.viii.1995 [CNC]; 1♂, *id.*, Y4-1, 12.viii.6.ix.1995 [CNC]; 1♂, *id.*, Y4-5, 3.viii.-6.ix.1994 [CNC]; 1♂, *id.*, Y4-5, 5.vii.-12.viii.1995 [CNC]; 1♂, Vaseux Cr., 'Kennedy bench', 49°16'N 119°30'W, BGxh1, AN, pitfall trap Z2-4, 3.vi-8.vii.1994 (G.G.E. Scudder) [CNC]; 1♀, *id.*, Z2-5, 12.viii.-6.ix.1995 [CNC]; 1♂, Vaseux Cr., 'Kennedy flats', 49°15'N 119° 31'W, BGxh1, AN, pitfall trap X2-1, 6.ix.-4.x.1995 (G.G.E. Scudder) [CNC]; 1♀, *id.*, X3-4 [CNC]; 1♂, X3-5, 12.vii.-6.ix.1995 [CNC]; 1♀, *id.*, X4-1 [CNC]; 1♂. Vaseux L., Wildlife Res., BGxh1, AN:F, pitfall trap VL1-2, 27.vii.-17.viii.1997 (J. Jarrett) [UBC]; 1♂, Westwick L., Cariboo, 28.vi.1961 (J. Scudder) [UBC]; 1♀, *id.*, 17.viii.1962 (G.G.E. Scudder) [CNC]; 3♂ 4♀, *id.*, 18.viii.1962 [CNC, RBCM, UBC]; 1♀, White L., BGxh1, SWm, pitfall trap WL2-3, 17.viii.-28.ix.1996 (J. Jarrett) [UBC]; 1♂ 2♀, Windermere Valley, pitfall trap No. 5, 26.vii.-17.ix.2000 (R. Sargent) [CNC, UBC]. **NT**: 1♀, Fort Smith, 27.v.1950 (W.G. Helps) [CNC]; 1♀, Fort Smith, 6.viii.1950 (J.B. Wallis) [CNC]. **YT**: 2♀, Alaska Hwy. km 1768, Duke R., 9.vii.1983 (G.G.E. Scudder) [CNC]; 1♂, Alaska Hwy. mi 1054, Kluane L., 16.vii.1962 (G.G.E. Scudder) [CNC];1♂, *id.*, 7.vii.1983 [CNC]; 2♂, Canyon, Aishihik R., 9.vii.1983 (G.G.E. Scudder) [CNC]; 3♂ 3♀, Carcross, 8.vii.1983 (G.G.E. Scudder) [CNC, UBC]; 1♂, Lapie R., 1 km E on Campbell Hwy., 28.vii.1981 (C.S. Guppy) [UBC]; 1♀, Pelly Crossing, 17.vii.1983 (G.G.E. Scudder) [CNC]; 1♀, Tatchun Cr., 62°17'N 136°17'W, 17.vii.1983 (G.G.E. Scudder) [CNC]; 1♀, Whitehorse, 17.vii.1959 (R. Madge) [CNC]; 1♀, White-

horse, 31.vii.1981 (C.S. Guppy) [UBC].

I have also collected *P. nigripes* in Washington State, USA, as follows: **WA**: 1♀, Oroville, E. Osoyoos L., 48°53'N 119° 25'W, *Purshia* assoc., AN BGxh1, pitfall trap O2-1, 10.ix.-4.x.1995 (G.G.E. Scudder) [CNC].

Family MIRIDAE

Ceratocapsus modestus (Uhler)

A widely distributed eastern Nearctic species, previously reported from Saskatchewan east to Quebec in Canada (Maw *et al.* 2000). Recorded hosts in West Virginia are *Quercus alba* L. and *Vitis* sp. (Wheeler *et al.* 1983). The species is keyed by Knight (1941), Henry (1979), Kelton (1980) and Larochelle (1984).

New records. **NS**: 1♀, Grand Pre, on *Picea*, 10.viii.1966 (L.A. Kelton) [CNC]; 1♀, Grand Pre, *Pinus sylvestris*, 10.viii.1966 (L.A. Kelton) [CNC].

Conostethus americanus Knight

To date this species in Canada has been reported only from Alberta, Northwest Territories and Saskatchewan (Maw *et al.* 2000). On the prairies *C. americanus* occurs on grasses (Kelton 1980). It is keyed and illustrated in Kelton (1980).

New record. **YT**: 4♂ 7♀, Whitehorse, Dillabough's graze lease, 8V 6754911 490889, 12.vii.2005 (G.E. Hutchings) [RBCM].

Cyrtorhinus caricis (Fallén)

A Holarctic species, reported to occur on sedge (*Carex* spp.) across Canada (Kelton 1980), and also recorded from Alaska and Minnesota, with an apparent relict population occurring in Colorado (Wheeler and Henry 1992). The species is keyed and illustrated by Kelton (1980).

New record. **NS**: 1♂, Lake Egmont, 18.vii.1991 (B. Wright) [NSM].

Deraeocoris quercicola Knight

An eastern Nearctic species, widely distributed and in Canada reported to date from Saskatchewan east to Quebec (Maw *et al.* 2000). Recorded hosts include *Carya* sp., *Quercus alba*, *Q. ilicifolia* Wangenh and *Tilia americana* L. (Wheeler *et al.* 1983), as well as *Quercus macrocarpa* Michx. where it preys on aphids (Kelton 1980). The species is keyed by Knight (1921), Kelton (1980) and Larochelle (1984).

New record. **NB**: 1♀, St. Johns, Rockwood Pk., 5.viii.1954 (J.F. Brimley) [CNC].

D. triannulipes Knight

This Nearctic species in Canada has previously been reported from British Columbia east to Quebec (Maw *et al.* 2000). *Deraeocoris triannulipes* is reported to feed on aphids on *Populus tremuloides* Michx. and *Alnus* spp. (Kelton 1980). It is keyed by Knight (1921), Kelton (1980) and Larochelle (1984).

New records. **NS**: 1♂, Exfern, on apple, 4.vii.1950 (F.T. Low) [CNC]; 1♀, Halifax, *Pyrus*, 22.vii.1976 (L.A. Kelton) [CNC]; 2♀, Kentville, on apple, 10-14.vii.1976 (L.A. Kelton) [CNC]; 1♂, Kentville, *Tilia cordata*, 15-17.vii.1976 (L.A. Kelton) [CNC].

Dichaetocoris piceicola (Knight)

A western Nearctic species, known from Colorado in the USA, and in Canada recorded from Alberta, British Columbia and Yukon (Maw *et al.* 2000). The genus is keyed by Knight (1968). The species *D. piceicola* is distinguished from the only other northern species (*D. gillespiei* Schwartz and Scudder) by Schwartz and Scudder (2003). It is recorded from *Picea engelmanni* Parry (Polhemus 1994), and in British Columbia has been collected on *Abies lasiocarpa* (Hook.) Nutt., *Picea* sp., *Pinus contorta* Dougl. and *Tsuga heterophylla* (Raf.) Sarg. (Scudder, unpublished).

New record. **AK**: 1♀, Mosquito L., 59° 27'N 136°02'W, 6.vii.1983 (G.G.E. Scudder) [CNC].

Labops verae Knight

A western Nearctic and Beringian species distributed from Alaska to Manitoba and south to Washington State (Henry and

Wheeler 1988; Maw *et al.* 2000), but not previously reported from Saskatchewan. The species is keyed in Slater (1954) and Kelton (1980). The host plants are unknown (Kelton 1980).

New record. **SK**: 1♂, Stony Rapids, 30.vi.1975 (R. Hooper) [SK].

Lygidea salicis Knight

This Nearctic species in Canada is previously reported from Alberta east to Newfoundland (Maw *et al.* 2000), and in the United States from New York to Minnesota, Colorado, and California (Henry and Wheeler 1988). It has not previously been recorded from British Columbia. *Lygidea salicis* Knight is a small species with the average length in the male of 5.8 mm, and in the female 6.2 mm. The species is keyed by Kelton (1980), who notes that the pilosity on the second antennal segment is shorter than the thickness of this segment. *Lygidea salicis* is usually collected on *Salix* spp. (Kelton 1980; Wheeler *et al.* 1983).

New records. **BC**: 2♂ 3♀, Fernie, goldenrod, 23.vii.1959 (L.A. Kelton) [CNC]; 2♂ 2♀, Mt. Revelstoke Nat. Pk., *Salix*, 17.vii.1970 (L.A. Kelton) [CNC].

Megalopsallus femoralis Kelton

This species so far has been reported from Alberta, Manitoba, Saskatchewan, Colorado, South Dakota and Wyoming (Schuh 2000b). *Megalopsallus femoralis* has been collected on *Salicornia rubra* Nels. (Kelton 1980). It is keyed by Kelton (1980) and Schuh (2000b), and is illustrated in colour in the latter reference.

New record. **BC**: 3♂ 2♀, Kamloops, Ironmask L., 10U 6804 56152, saline flats, *Salicornia/Plantago*, 730 m., 14.vi.1995 (S.G. Cannings) [RBCM].

Orthotylus alni Knight

This Nearctic species is distributed from Yukon to Newfoundland, and south to New York and Minnesota in the eastern United States (Henry and Wheeler 1988; Maw *et al.* 2000). However, it has not previously been recorded from Nova Scotia. *Orthotylus alni* is keyed by Kelton (1980), and has been collected on *Alnus rugosa* (DuRoi) Spreng. (Kelton 1980), as well as *A. tenuifolia* Nutt., *Betula glandulosa* Michx., *Lupinus* sp. and *Salix* sp. (Scudder 1997).

New records. **NS**: 1♂, Chester, 10.vii.1969 (B. Wright) [NSM]; 1♀, Chester, 16.vii.1969 (B. Wright) [NSM].

O. nyctalis Knight

Described originally from Minnesota (Knight 1927), this species has been reported in the USA also from Iowa, Illinois, New York and Wisconsin (Henry and Wheeler 1988). It has not previously been recorded from Canada under this name.

According to Knight (1927), *O. nyctalis* can be recognized chiefly on the structure of the male genital claspers. The left clasper is slender with two short dorsal prongs, and the right clasper decurved on the apex and devoid of spines, but the dorsal margin has a prominent spine at the basal third, and two other spines just before the decurved apex.

The record of *O. candidatus* Van Duzee from Saskatchewan (Kelton 1980; Maw *et al.* 2000; Roch 2007) is evidently referable to *O. nyctalis*, as is the record of *O. candidatus* from Ontario (Maw *et al.* 2000; Roch 2007). The recorded occurrence of *O. candidatus* in Quebec (Henry and Wheeler 1988; Roch 2007) may also refer to *O. nyctalis*. Some specimens from Dawson and Moose Creek in the Yukon, listed as *Orthotylus* sp. in Scudder (1997) are actually *O. nyctalis* (see below), but it may be noted that *O. candidatus* also occurs at both these localities. *Orthotylus nyctalis* evidently occurs on *Populus tremuloides* (Kelton 1980).

New records. **AB**: 1♂, Stettler, 3.viii.1957 (A. & J. Brooks) [CNC]: 1♀, Vermilion Provincial Park, Beaverdam Loop Trail, *Populus tremuloides* Michx., 22.viii.1993 (M.D. Schwartz) [CNC]. **MB**: 1♂, Falcon L., 5.viii.-10.viii.1978 (L.A. Kelton) [CNC]; 1♀, *id.*, 6.viii.1978 (L.A. Kelton) [CNC]; 1♀, Rennie, 16.viii.1961 (F.I.S.) [CNC]. **ON**: 1♂, One Sided Lake, *Salix* sp., 1.viii.1960 (Kelton and Whitney) [CNC]; 1♀, Tillsonburg, 18.vii.1962

(Kelton and Thorpe) [CNC]. **SK**: 1♀, Cypress Hills Prov. Park., *Sheperdia canadensis*, 19.ix.1951 (L.A. Konotopetz) [CNC]. **YT**: 1♂, Dawson, 14 mi E, *Populus* sp., 29.vii.1962 (R.E. Leech) [CNC]; 1♀, Dawson, *Salix* sp., 23.vii.1983 (L.A. Kelton) [CNC]; 1♀, Moose Creek, *Salix* sp., 28.vii.1983 (L.A. Kelton) [CNC].

Phytocoris buenoi Knight

An eastern Nearctic species, in Canada previously reported from Ontario and Quebec (Maw *et al.* 2000). The species is keyed by Blatchley (1926). Knight (1920) reported it to occur on Norway spruce (*Picea abies* (L.) Karst.) in the eastern United States, and Wheeler *et al.* (1983) added *Picea glauca* (Moench) Voss and *P. rubens* Sarg.

New records. **NS**: 1♂, Chester, 29.viii.1968 (B. Wright) [NSM]; 1♀, Sandy Cove, 4.viii.1971 (B. Wright) [NSM]; 1♀, Baddeck, 28.viii.1972 (B. Wright) [NSM]; 1♂, Kemptville, 24.viii.1982 (Agriculture Canada) [NSM].

P. procteri Knight

This species, which is a member of the *P. junceus* Knight group, was described from Maine (Knight 1974), and is reported from Quebec (Roch 2007). The frons has definite transverse red lines, the pronotum is pallid with a basal submarginal strong black band, and the propleura are brownish black. The clavus is more or less fuscous external to the claval vein, with insect length of 8.0 mm. The first antennal segment is pallid, but clothed with black, recumbent setae, and without distinct dark spots. The first antennal segment is longer than the width of the vertex, but does not exceed the width of the pronotum. The second antennal segment is without annuli or coloured bands. There are no records of a host for this species.

New records. **NS**: 1 specimen (abdomen missing), Lake Kejimkujik, 13.vii.1961 (D.C. Ferguson) [NSM]; 1♂, Chester, 24.vii.1968 (B. Wright) [NSM].

Pilophorus neoclavatus Schuh & Schwartz

This eastern Nearctic species to date in Canada has been reported from Alberta east to Quebec (Maw *et al.* 2000), and is keyed by Schuh and Schwartz (1988). Hosts include *Alnus rugosa*, *Quercus ilicifolia*, *Q. palustris*, *Q. stellata* Wangenh and *Salix longifolia* Muhl. (Schuh and Schwartz 1988).

New record. **NS**: 1♀, Kentville, 8.viii.1952 (C.R. McL.) [LM].

Rhinocapsus rubricans (Provancher)

An eastern Nearctic species, in Canada to date reported from Saskatchewan east to Quebec (Maw *et al.* 2000). The species is keyed and illustrated in Kelton (1980), who reports it collected on *Kalmia polifolia* Wang.

New record. **NS**: 1♂, Chester, 4.vii.1969 (B. Wright) [NSM].

Sixeonotus deflatus Knight

An eastern Nearctic species, in Canada previously only reported from Quebec (Larochelle 1984; Maw *et al.* 2000). The species is keyed by Larochelle (1984) as *S. insignis* Reuter, who records the host as *Symplocarpus foetidus* (L.).

New records. **NB**: 1♂, St. John, 9.viii.1954 (J. Brimley) [CNC]. **ON**: 8♂ 5♀, St. Catherines, 22.vi.1961 (Kelton and Brampton) [CNC].

Family TINGIDAE

Acalypta lillianis Torre-Bueno

This Nearctic tingid is widely distributed in North America and Beringia (Drake and Lattin 1963; Scudder 1997). It occurs in Alaska, and from Yukon to Newfoundland, but has not previously been reported from Manitoba (Maw *et al.* 2000). It is keyed by Drake and Lattin (1963), who note that host records are mosses.

New record. **MB**: 1♂, Bird Cove, 4 km NE, Churchill North Studies Centre, 58° 46'14"N 93°50'33"W, pit trap in tundra zone, 18.viii.2006 (Boreal & Arctic Entomol.) [UM].

Family ARADIDAE

Aradus uniannulatus Parshley

A Nearctic and Beringian species, distributed from Yukon to Quebec, and in the United States, south to New York and Colorado (Scudder 1997). The species is keyed by Matsuda (1977) who notes that it is reported to be associated with *Pinus contorta murrayana* (Balf.) Critchfield elsewhere.

New records. **BC**: 1♀, Fernie, 22.viii.1934 (Hugh Leech) [CNC]; 1♀, Lorna, *Picea engelmanni*, 17203 Lot 1, 29.vi.1929 (R. Hopping) [CNC]; 1♂, *id.*, 17203 Lot 2, 10.vii.1924 [CNC]; 1♀, *id.*, *Pinus contorta*, 17203 Lot 8, 12.vii.1924 [CNC]; 1♂, *id.*, *Picea engelmanni*, 17203 Lot 14, 12.vii.1924 [CNC]; ♂, *id.*, 17203 Lot 25, 25.vii.1924 [CNC]; 1♂, *id.*, 17203 Lot 26, 27.vii.1924 [CNC] 1♀, Midday Valley, Merritt, *Pinus ponderosa*, Exp. 17501 Lot 1035, 30.v.1923 (R. Hopping) [CNC]; 1♀, Pine Pass, *Picea*, 11.vii.1972 (D.E. Bright) [CNC].

Infraorder PENTATOMOMORPHA

Family COREIDAE

Leptoglossus occidentalis Heidemann

This leaf-footed bug, commonly called the western conifer seed bug, feeds on numerous conifer species (Koeber 1963; Krugman 1969; Hedlin *et al.* 1981; Schaefer and Mitchell 1983; Gall 1992; Mitchell 2000) although the apparently strong reliance on Pinaceae as a food source is not absolute (Mitchell 2000). However, it is of considerable economic importance because it can cause significant losses in conifer seed orchards (Koeber 1963; Schowalter and Sexton 1990; Blatt and Borden 1996; Mitchell 2000; Strong *et al.* 2001; Bates *et al.* 2002; Strong 2006). The species is keyed in Allen (1969) and McPherson *et al.* (1990), and illustrated by Koeber (1963) and Ruth *et al.* (1982).

Originally considered a western Nearctic species, *L. occidentalis* has naturally expanded its range eastwards in the past few decades (Schaffner 1967; McPherson *et al.* 1990; Marshall 1991; Gall 1992; Ridge-O'Connor 2001), and has invaded Europe (Taylor *et al.* 2001; Gogala 2003; Tescari 2004; Hilpold 2005; Rabitsch and Heiss 2005; Ribes and Oleguer 2005; Foldessy 2006; Moulet 2006). It is now known to occur in Nova Scotia.

New records. **NS**: 1♀, Kings Co., Middleton, in house, 20.ix.2006 (J. Parks) [NSM]; 1♀, Halifax Co., Halifax, on house, 3.x.2006 (B. Fay) [NSM]; 2♀, Kings Co., Lakeville, 1 of 4, 12.iii.2007 (J. Morton) [NSM 36185]; 1♀, Halifax Co., Halifax, in dwelling, 15.x.2007 (John Sherwood) [NSM Cat. 36219].

Family RHOPALIDAE

Liorhyssus hyalinus (Fabricius)

This cosmopolitan species is widely distributed in North America, and in Canada has so far been reported from British Columbia, Manitoba and Ontario (Maw *et al.* 2000). It is keyed by Blatchley (1926), Slater and Baranowski (1978), and Hoebeke and Wheeler (1982), and illustrated in Slater and Baranowski (1978). Schaefer and Chopra (1982) report *Abutilon*, *Euphorbia*, *Lactuca* and *Sonchus* as host plant genera.

New record. **QC**: 1♀, Terrebonne Co., Lac Carre, Lot 31, Range 8, 19-23.viii.1968 (W. Boyle and R. La Conde) [LM].

Family LYGAEIDAE

Kleidocerys ovalis Barber

This widely distributed Nearctic species has so far in Canada only been recorded from British Columbia and Ontario (Maw *et al.* 2000), as well as Quebec (Roch 2007). It is keyed in Barber (1953) and Scudder (1962). In British Columbia, *K. ovalis* has been collected on *Abies lasiocarpa*, *Alnus* sp., *Betula occidentalis* Hook., *B. papyrifera* Marsh., *Malus* sp., and *Pinus ponderosa* Dougl. (Scudder, unpublished).

New records. **AB**: 5♂ 2♀, Drumheller, 18.vi.1957 (Brooks, MacNay) [CNC]; 7♂ 6♀, *id.*, 11.viii.1957 (A.R. & J.E. Brooks) [CNC]; 2♂ 3♀, Empress, 7.vi.1957 (Brooks, MacNay) [CNC]; 1♀, Lethbridge, 3.vii.1929 (J.H. Pepper) [CNC]; 1♂, Lundbreck, 7.viii.1930 (J.H. Pepper) [CNC]. **MB**: 1♀, Aweme, 5.vii.1920 (H.A. Robertson) [CNC]; 1♀, *id.*, 8.vii.1920 (P.N.

Vroom) [CNC]; 3♀, *id.*, *Betula*, 30.vi.1922 (N. Criddle) [CNC]; 1♀, *id.*, 3.v.1923 (N. Criddle) [CNC]; 1♂ 1♀. Carberry, 9.v.1953 (Brooks, Kelton) [CNC]; 1♂ 3♀, Ninette, *Betula glandulosa*, 21.vi.1958 (J.F. McAlpine) [CNC]; 1♂, *id.*, 14.vii.1958 (R.B. Madge) [CNC]; 1♂, *id.*, 15.vii.1958 (R.L. Hurley) [CNC]; 1♂, Onah, 10.v.1923 (R.M. White) [CNC]; 8♂ 1♀, *id.*, *Betula papyrifera*, 10.ix.1930 (R.M. White) [CNC]; 1♀, Turtle Mtn., 22.vii.1953 (Brooks, Kelton) [CNC]. **SK**: 1♀, Punnichy, 21.v.1965 (R. Hooper) [SM].

K. resedae (Panzer)

This Holarctic species is widely distributed in North America, and occurs in Alaska and from Yukon to Newfoundland and Labrador, but has not previously been recorded from Prince Edward Island. It is keyed in Barber (1953) and Scudder (1962). *Kleidocerys resedae* usually occurs on *Alnus* spp. and *Betula* spp. (Scudder 1997).

New records. **PE**: 3♂ 1♀, Blooming Point, 46°24'33"N 62°58'07"W, sweeping, 20.x.2004 (G.G.E. Scudder) [CNC, UPEI]; 2♂ 4♀, Charlottetown, UPEI, nr. NE point, 46°15'39"N 63°08'19"W, sweeping, 20.x.2004 (G.G.E. Scudder) [CNC, UPEI].

Melacoryphus lateralis (Dallas)

A widely distributed Nearctic species, so far only recorded from British Columbia and Saskatchewan in Canada (Maw *et al.* 2000). The species is keyed by Slater (1988). Specimens of *M. lateralis* collected at light in Wyoming contained cardenolides in the body (Scudder and Duffey 1972), and thus showed evidence of feeding on Asclepiadaceous host plants.

New record. **ON**: 1♂, Guelph, 3.viii.1977 (W.A. Attwater) [UG].

Family RHYPAROCHROMIDAE

Antillocoris minutus (Bergroth)

An eastern Nearctic species, in Canada previously recorded from Ontario east to Newfoundland (Maw *et al.* 2000). The genus is keyed in Blatchley (1926), Slater and Baranowski (1978), and Larochelle (1984), with key to species given by Barber (1952) and Larochelle (1984). The biology of *A. minutus* in New England has been described by Sweet (1964), who notes the species typically occurs on the ground and usually is found in forest litter, most frequently found beneath gray birch (*Betula populifera* Marsh) and white birch (*B. papyrifera*), but also occurs under hemlock (*Tsuga canadensis* (L.) Carr.), and in sphagnum bogs.

New record. **MB**: 1♀, Winnipeg, St. Charles Rifle Rge., Block B Refuge, Pitfall trap, 6-13.x.1999 (D.A. Pollock, J.K. Diehls and R.E. Roughley) [UM].

Drymus unus (Say)

An eastern Nearctic species, in Canada so far recorded from Saskatchewan east to Nova Scotia (Maw *et al.* 2000). The species is keyed by Blatchley (1926) and Larochelle (1984), and illustrated by Slater and Baranowski (1978). Sweet (1964) described the biology *D. unus* in New England, and noted that this is a ground-dwelling species, most abundant in subclimax forests, particularly where black birch (*Betula lenta* L.) and red maple (*Acer rubrum* L.) are associated with oak (*Quercus* spp.) and hickory (*Carya* spp.).

New record. **NF**: 1♂, St. John's, Long Pond, ix.-x.2001 (Biology 4150) [MU].

Perigenes constrictus (Say)

This eastern Nearctic species is distributed throughout the northern and central United States, and in Canada so far reported from Nova Scotia, Ontario and Quebec (Maw *et al.* 2000). The species is keyed by Blatchley (1926) and Larochelle (1984), and illustrated by Blatchley (1926) and Slater and Baranowski (1978). Sweet (1964) described the biology of *P. constrictus* in New England and noted that it typically occurs in temporary habitats, such as vacant lots, roadsides and newly fallow fields.

New record. **SK**: 1♂, Big Beaver, 9.vii.1974 (R. Hooper) [SM].

Plinthisus americanus Van Duzee

In Canada previously reported from Alberta east to New Brunswick (Maw *et al.*

2000). *Plinthisus americanus* is distinguished by the hemelytra of the female being densely pilose as noted by Sweet (1964), whereas in the closely related *P. compactus* (Uhler) the hemelytra of the female are glabrous. Sweet (1964) noted that *P. americanus* is a forest species in New England, most abundant in *Tsuga* litter.

New record. **BC**: 1♀, Attachie, 32 km W of Charlie L., 5.viii.1982 (R.A. Cannings) [RBCM].

Trapezonotus arenarius (Linnaeus)

A Holarctic species with a wide distribution in both the Nearctic and Palearctic, in Canada recorded from British Columbia and Yukon east to Quebec (Maw *et al.* 2000), and in the United States evidently restricted to the highlands of New England and northern New York (Sweet 1964). The species is keyed by Blatchley (1926) and Larochelle (1984), and illustrated by Slater and Baranowski (1978). Sweet (1964) reported that in New England *T. arenarius* is a species of open upland habitats, particularly well-drained and rather dry sites.

New record. **NS**: 1♂, Lunenberg, 7.viii.1991 (B. Wright) [NSM].

Family CYDNIDAE

Amnestus pusillus Uhler

A widely distributed species in North America, with recorded occurrence also in Mexico and Guatemala (Froeschner 1960). In Canada, so far reported only from Ontario and Quebec (Maw *et al.* 2000). The species is keyed in Froeschner (1960), McPherson (1982), and Larochelle (1984). McPherson (1982) reported that elsewhere *A. pusillus* has been collected from vegetation along streams and margins of roadsides and cultivated fields, as well as beneath rubbish in sandy places.

New record. **NB**: 1♀, Woodstock, 22.v.1966 (L.A. Kelton) [CNC].

Family PENTATOMIDAE

Acrosternum hilare (Say)

A widely distributed Nearctic species, in Canada so far recorded only from British Columbia, Ontario and Quebec (Maw *et al.* 2000). This species has been collected from numerous plants (McPherson 1982) and can damage some crops (Panizzi *et al.* 2000). The species is keyed in Blatchley (1926), McPherson (1982), Rolston (1983), and Larochelle (1984).

New record. **NS**: 1♀, Debert, 1.ix.1952 (R.L. Horsburgh) [LM].

Cosmopepla intergressus (Uhler)

A widely western Nearctic species, in Canada so far recorded only from British Columbia (Maw *et al.* 2000). *Cosmopepla intergressus* is keyed by McDonald (1986), who reported records of the species on "currants", *Rubus parviflorus* Nutt. and *Ribes* sp.

New record. **AB**: 3♂ 3♀, Lethbridge, black current, 21.ix.2005 (J.R. Byers) [AAFCL].

Euschistus servus euschistoides (Vollenhoven)

A Nearctic species widely distributed in North America, and in Canada recorded from British Columbia to Nova Scotia, but not previously reported from New Brunswick (Maw *et al.* 2000). It has been recorded from numerous host plants (McPherson 1982), and the species has caused yield and quality losses to several crops (Panizzi *et al.* 2000). The species is keyed in McPherson (1982) and Larochelle (1984).

New records. **NB**: 1♀, Jonah Mt., 3.vi.1976 (P. Kevan) [LM]; 1♀, Whittier Ridge, 30.v.1976 (P. Kevan) [LM].

Menecles insertus (Say)

Widely distributed in North America, and in Canada previously recorded from Nova Scotia, Ontario and Quebec. *Menecles insertus* is nocturnal, arboreal, and phytophagous, and has been collected on deciduous trees (McPherson 1982). The species is keyed in Blatchley (1926), Rolston (1973), McPherson (1982) and Larochelle (1984).

New record. **SK**: 1♀, Buffalo Pound Park, 5.v.1975 [SM].

Zicrona caerulea (Linnaeus)

This Holarctic species is widely distributed in North America, Europe and Asia, and occurs in the Oriental region (De Clercq 2000). In Canada it is reported from British Columbia east to New Brunswick (Maw *et al.* 2000). This predaceous species attacks only small prey (De Clercq 2000), and is keyed by McPherson (1982), Larochelle (1984), and Thomas (1992).

New record. **NF**: 1♂, Red Indian L., Winddrift Lot 5, 25.vi.1980 (Brennan and Larson) [MU].

Family THYREOCORIDAE

Corimelaena pulicaria (Germar)

A widely distributed species in North America, also reported from Mexico and Guatemala. In Canada recorded from British Columbia to Nova Scotia, but not previously reported from New Brunswick (Maw *et al.* 2000). The species has been collected on many plants (McPherson 1982), and is keyed in Blatchley (1926), McPherson (1982), and Larochelle (1984).

New record. **NB**: 1♀, Fredericton, French Lake, 10.vi.1931 (C.W. Maxwell) [LM].

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First Canadian records of *Lampropteryx suffumata* ([Denis & Schiffermüller], 1775) (Geometridae: Larentiinae)

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ABSTRACT

The first Canadian records of the Holarctic species *Lampropteryx suffumata* ([Denis & Schiffermüller], 1775) are documented, based on collections from Alberta and British Columbia. Widespread and common throughout much of Eurasia, the larvae feed on *Galium* species (Rubiaceae). Diagnostic descriptions and images are provided to aid in future recognition of this species. The specimens were originally detected while constructing a DNA barcoding library for western North American Geometridae, and provide a good example of how genetic methods can enhance the construction of regional inventories and aid in surveillance for invasive species.

Key Words: *Lampropteryx suffumata*, black-banded carpet, DNA barcoding, invasive species

INTRODUCTION

The genus *Lampropteryx* Stephens includes ten species, most of which are restricted to Asia, with two species also occurring in Europe (Scoble 1999). The black-banded carpet *Lampropteryx suffumata* ([Denis & Schiffermüller], 1775), described from Vienna, Austria, occurs from western Europe and the northern Mediterranean region to northern Scandinavia, east through the Tien Shan and Altai mountain ranges of south-central Asia to the Kamchatka Peninsula, Russia and Hokkaido, Japan (Skou 1986; Beljaev and Vasilenko 2002). Previously known in North America only from Alaska (Choi 2000), we report here historical and contemporary records in British Columbia and Alberta, flagged by DNA barcoding.

MATERIALS AND METHODS

During the course of documenting the molecular diversity of western Canadian geometrid moths from museum and field collections using standard DNA barcoding methods (Hajibabaei *et al.* 2005; deWaard *et al.* 2008), it became evident that a number of specimens variously identified as *Antepirrhoe* Warren or *Xanthorhoe* Hübner were highly divergent compared to other congeners. Using the identification engine of the Barcode of Life Database (BOLD) (Ratnasingham and Hebert 2007), their cytochrome oxidase I (COI) sequences were a nearly identical match to those of *Lampropteryx suffumata* ([Denis & Schiffermüller], 1775) specimens from Bavaria, Germany

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(Figure 1). Although many larentiines are very similar in habitus and are difficult to identify when the wing pattern is worn, subsequent genitalic examination of the suspect *Antepirrhoe* and *Xanthorhoe* specimens showed unequivocally that they are in fact *L. suffumata.*

To determine the Canadian distribution, and whether or not the species is likely native, we examined historical and contemporary *Antepirrhoe* and *Xanthorhoe* specimens from various Canadian collections. We identified specimens in the Royal British Columbia Museum, Victoria, BC (RBCM), the E.H. Strickland Entomological Museum, University of Alberta, Edmonton, AB (UASM) the Canadian National Collection of Insects, Agriculture and Agri-Food Canada, Ottawa, ON (CNC), and the Biodiversity Institute of Ontario, University of Guelph, Guelph, ON (BIOUG) as *L. suffumata.* The collections of the Pacific Forestry Centre, Canadian Forest Service, Victoria, BC (PFCA), the Spencer Entomological Museum, University of British Columbia, Vancouver, BC (UBCZ), and the Northern Forestry Centre, Canadian Forest Service, Edmonton, AB (NFRC) do not contain any specimens of *L. suffumata.*

RESULTS

Specimens examined (all specimens are single, pinned adults; the BOLD accession number (italicized) is provided for specimens that have been barcoded).

AB: Hillcrest, 49.568N 114.377W, 20-vi-1919 (K. Bowman) [UASM, UASM10792]; West Castle River, W Castle R. Rd., 15 km SW, 49.294N 114.273W, 23-v-1999 (B.C. Schmidt) [CNC, CNCLEP00033310, *GWNC311-07*]; **BC:** Elkford, 35 km north, 50.266N 114.921W, 12-Jun-1988 (C.S. Guppy) [RBCM, ENT991-006550, *GWNR470-07*]; Glacier National Park, Abandoned Rails Trail west of Rogers Pass Centre, 51.2902N 117.516W, 04-Jul-2005 (K. Pickthorn) [BIOUG, HLC-20568, *LBCA568-05*]; Glacier National Park, Glacier National Park Compound at Rogers Pass, 51.3032N 117.519W, 28-Jun-2005 (K. Pickthorn) [BIOUG, HLC-20320, *LBCA320-05*]; Glacier National Park, Illecillewaet Campgrounds west of Rogers Pass, 51.2648N 117.494W, 24-Jun-2005 (K. Pickthorn) [BIOUG, HLC-20175, *LBCA175-05*]; Glacier National Park, Glacier National Park Compound at Rogers Pass, 51.3032N 117.519W, 16-Jun-2005 (K. Pickthorn) [BIOUG, HLC-20022, *LBCA022-05*]; Trinity Valley Field Station, 50.400N 118.917W, 18-May-1961 (W.C. McGuffin) [CNC, CNCLEP00054030].

Identification. A medium-sized, broad-winged moth with a wingspan of 2.5–3.2 cm (Figure 2a.). The forewing basal and median bands are dark, varying from red-brown to black, being dark brown in most specimens. The median band has a jagged proximal and distal margin, with the distal margin extending towards the base just below the median area, such that the median band is narrower along the anal third than on the upper half. The apex is also darkened, divided by a white apical dash. There is a subterminal line of white spots or wedges, and the fringe is checkered. It is very similar to *Antepirrhoe semiatrata* (Hulst), but can be readily separated by the following characters: forewing pale antemedian band faintly bordered with two whitish lines both proximally and distally (only one pale border line in *A. semiatrata*); forewing subapical dark patch bordered towards costal margin by contrasting pale line (indistinctly so in *A. semiatrata*). The dorsal markings on the abdomen are the most reliable external features for diagnosing *L. suffumata*, which has a row of black triangles along the midline (Fig. 2a), whereas *Antepirrhoe* species have two black dots broken at the midline by a pale line/spot. Some specimens may be melanic and lack the contrasting white forewing bands present in most specimens. *Xanthorhoe* species are superficially similar, but lack the combination of broad, dark

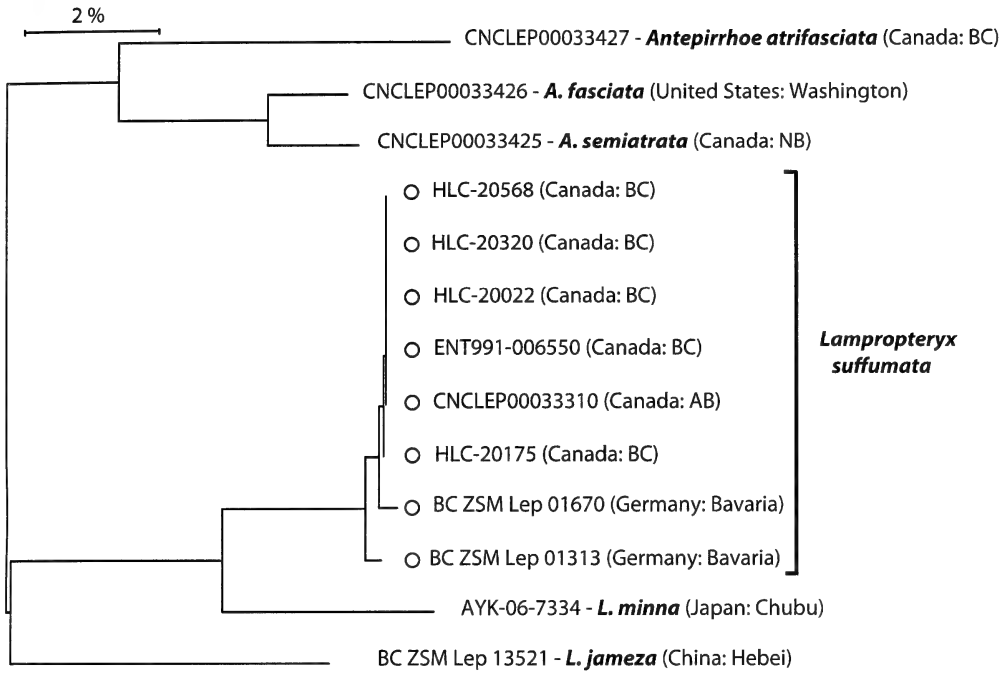

Figure 1. Neighbour-joining tree of *Lampropteryx suffumata* and related species. Tree was reconstructed with the barcode fragment of the COI gene. Sequences shaded in grey are derived from specimens previously misidentified as *Antepirrhoe* or *Xanthorhoe* spp. The 13 sequences are publicly available in the Barcode of Life Database and GenBank (accession nos. FJ376631–FJ376643).

basal and median bands with a contrastingly bordered subapical dark patch that extends to the distal wing margin. Genitalic examination of *L. suffumata* will easily segregate this species: the male valve is simple and lobe-shaped, costa lacking apical process; socii prominent, about half as long as valve, with bundle of apical setae as long as socius; aedeagus uncurved, vesica with two cornuti (Figure 2b,c.). Identification through genitalic examination of males can usually be made by brushing away the terminal abdominal scales to reveal the apical portion of the valve which lacks the pointed, dorsally projecting costal process of *A. semiatrata*, in addition to the long tubular socii (stout and triangular without apical hair pencils in *A. semiatrata*). Male genitalic structure of *Xanthorhoe* species is very different, with a comparatively massive costal process that extends beyond the valve apex and is variously enlarged, broadened and/or armed with spines.

Distribution and Habitat: Great Britain and northern Europe east to southern Siberia, Kamchatka and Japan (Skou 1986; Beljaev and Vasilenko 2002); in North America, known from two areas: Alaska (Choi 2000) and southwestern British Columbia and adjacent Alberta (Figure 3). It is likely that this species occurs in intervening regions of northern British Columbia and the Yukon, but these areas have not been adequately surveyed. The single historical collection from Hillcrest, Alberta, coupled with the fact that *L. suffumata* occurs in relatively remote, mountainous habitats but has not been recorded near the international shipping ports of the coastal Pacific Northwest, suggests that *L. suffumata* is native to Canada. Furthermore, it likely expanded over Beringia during the Pleistocene, a common pattern in the western Canadian arthropod fauna, as evident by present ranges and fossil evidence of past ranges (Danks et al. 1997). Its habitat appears to be open wooded areas, edges and meadows.

Life History and Notes: There is a sin-

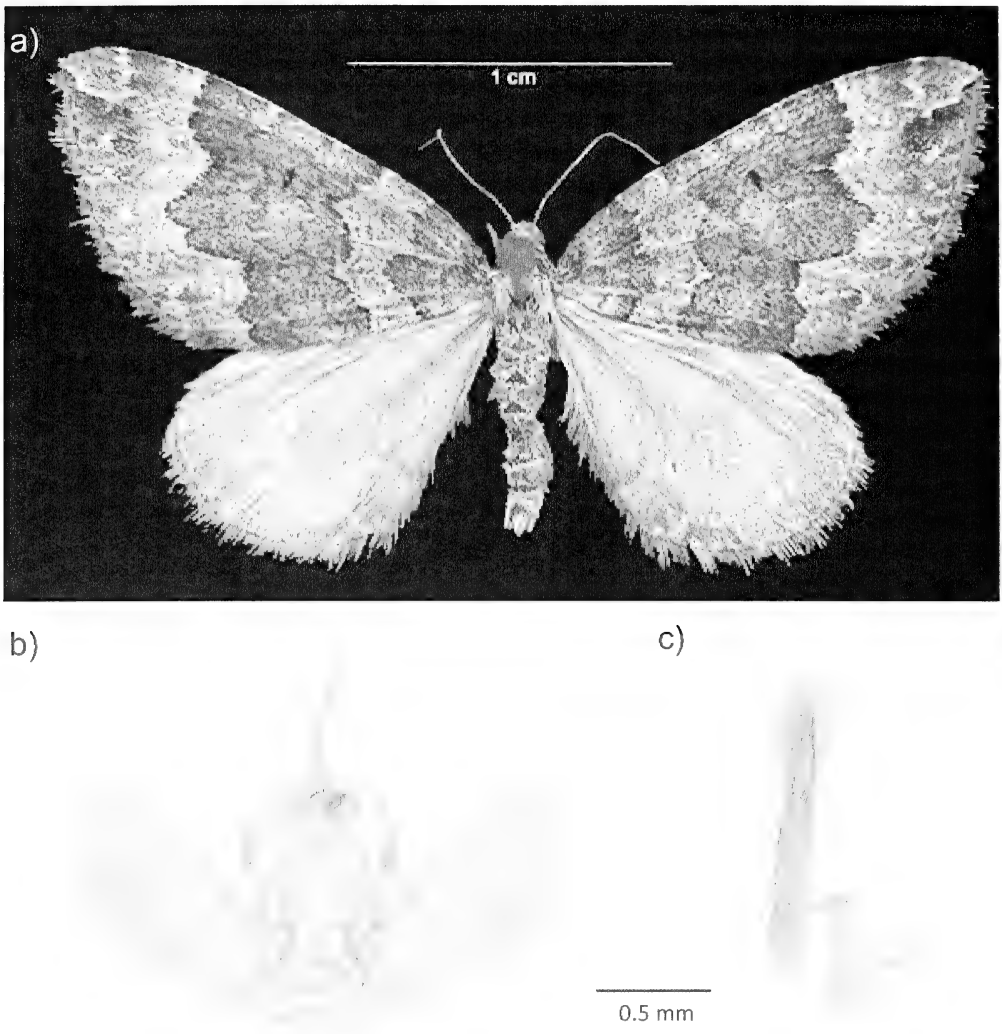

Figure 2. Adult male of *Lampropteryx suffumata*: a) dorsal view b) genital capsule c) aedeagus.

gle annual brood, with adults in late May to early July. Adults are nocturnal and come to light. The only reported larval hosts are bedstraw species (*Galium* sp.), particularly *G. aparine* Linnaeus (Skou 1986). The pupa overwinters underground (Skou 1986). Based on the scarcity of specimens in Canadian collections, we conclude the species is rarely collected and likely rare. The COI barcode sequences are publicly available in the Barcode of Life Database and GenBank (accession nos. FJ376631–FJ376643).

DISCUSSION

The late discovery of a relatively large and conspicuous native macromoth in Western North America is surprising, but we believe it can be explained simply by the paucity of taxonomic expertise and literature on the group. The Canadian larentiines are notoriously hard to discriminate, due in part to the lack of a treatment of this

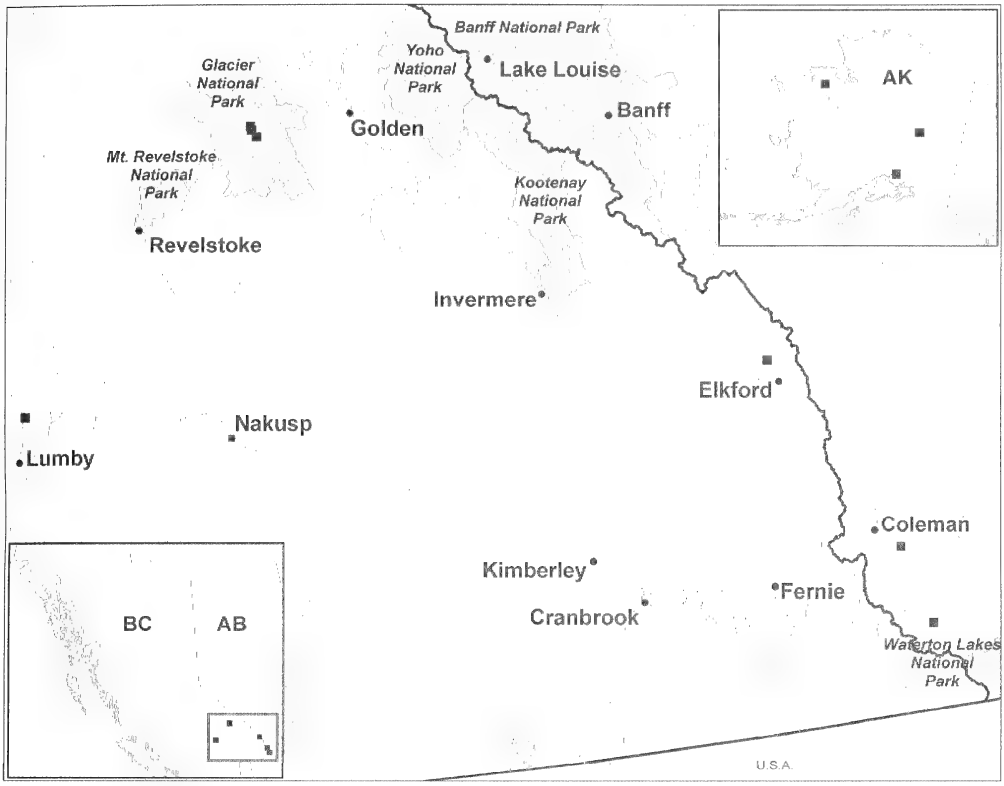

Figure 3. Distribution of *Lampropteryx suffumata* in North America. Black squares are locations of records.

subfamily in McGuffin's 'Guide to the Geometridae of Canada' series (1967, 1972, 1977, 1981, 1987, 1988). While a few larentiine genera have been revised (*Hydriomena* Hübner: McDunnough 1954; *Eupithecia* Curtis: Bolte 1990; *Entephria* Hübner: Troubridge 1997), most are in dire need of revision, and the Xanthorhoini in particular contain a number of genera that need attention, with cryptic and previously unrecognized species awaiting description (e.g., *Psychophora* Kirby, *Xanthorhoe* and *Zenophleps* Hulst: B.C.S. unpublished data; *Antepirrhoe*: J.R.D. *et al.* unpublished data). It is reasonable to assume that the few specimens of this rarely collected (and presumably rare) species could go unnoticed due to the lack of reliable guides and keys for the group.

Although *L. suffumata* is in all likelihood native, its discovery clearly illustrates how DNA barcoding can assist in the detection and surveillance of nonindigenous organisms (Armstrong and Ball 2005; Chown *et al.* 2008). A monitoring program that incorporates DNA barcoding can flag potential introduced species in one of two ways. First, as in this study, a barcode match is made with one or more specimens collected from the native range. The potential nonindigenous specimens can then be verified by morphological examination or further genetic analysis. At that point, national and regional collections can be examined for historical and contemporary specimens in the new range to determine if the species is native or introduced. Secondly, with a barcode library for a regional fauna complete (e.g., Geometridae of British Columbia – J.R.D. *et al.* unpublished data), any barcoded specimens that do not match the database are flagged as potentially nonindigenous and again warrant further examination. Using genetic methods for this initial screening has numerous advantages, most notably the ability to differentiate species objectively across all life stages as well as using damaged specimens. It is also ap-

parent that with the current costs of genetic analysis steadily dropping and new technologies emerging (Hajibabaei *et al.* 2007), genetic screening may soon be more cost- and time-efficient than current morphological methods of biodiversity monitoring.

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Identification of new aphid vector species of *Blueberry scorch virus*

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ABSTRACT

Transmission of *Blueberry scorch virus* (BlScV) by the aphid species *Ericaphis fimbriata* (Richards), *Aphis spiraecola* (Patch), *Aphis pomi* DeGeer, *Acyrthosiphon pisum* (Harris), *Myzus ornatus* Laing, *Aphis helianthi* Monell, *Myzus persicae* (Sulzer), and *Rhopalosiphum padi* (L.), was studied in the laboratory using timed aphid acquisition feeding periods and known numbers of aphid vectors. Successful infection of *Nicotiana occidentalis* Wheeler (Solanaceae), a newly identified herbaceous host, and highbush blueberry, *Vaccinium corymbosum* L. (Ericaceae), following brief virus-acquisition feeds lasting less than 5 min, demonstrated that BlScV was transmitted in a non-persistent, non-circulative manner. Based on transfer of 10 aphids per plant, the most efficient vector of BlScV from infected to healthy *N. occidentalis* was *M. ornatus*. Compared with this herbaceous host, infection rates for blueberry were much lower even though higher numbers of aphids (25/plant) were used. The highest rate of infection for blueberry (20%) was achieved when the green colour form of *E. fimbriata* was used to transmit the virus. The relatively low rate of transmission from infected to healthy blueberry suggests that BlScV would spread slowly in the field. Planting of certified virus-free nursery material and aggressive removal of infected plants should help control this economically important disease of highbush blueberries.

Key Words: *Blueberry scorch virus*, aphid vectors, virus transmission

INTRODUCTION

Blueberry scorch virus (BlScV) was first reported in New Jersey in the late 1970's as Sheep Pen Hill disease of highbush blueberry, *Vaccinium corymbosum* (L.) (Ericaceae) (Podleckis and Davis 1989). Several distinct strains infect highbush blueberry in the northeastern and northwestern United States and southwestern British Columbia (Cavileer *et al.* 1994, Catlin and Schloemann 2004, Bernardy *et al.* 2005, Wegener *et al.* 2006). BlScV has also been recently reported from Europe (Ciuffo *et al.* 2005). Depending on the virus strain and blueberry cultivar, infection can result in a wide range of symptoms. While some varieties are tolerant to certain strains and display no visible symptoms, infection with other strains can result in severe necrosis of new leaves, twigs and flower clusters and almost complete loss of yield over time (Martin and Bristow 1988, Catlin and Schloemann 2004, Wegener *et al.* 2006). The latent period between infection and development of symptoms for established plants is thought to be one to two years (Caruso and Ramsdell 1995).

There are relatively few previous studies on BlScV; these mostly relate to detection, symptomology and strain differentiation. Although little is currently known about the insect vectors of BlScV, carlaviruses as a group are transmitted primarily by aphids in a non-persistent, non-circulative manner (Ng and Perry 2004). Non-persistent virus transmission is characterized by short acquisition and inoculation feeding times,

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lasting from several seconds to a few minutes in duration (Raccah 1986). In uncontrolled cage studies, Hillman *et al.* (1995) were the first to demonstrate aphid transmission of BlScV. An unidentified aphid collected from blueberry and placed on infected *Chenopodium quinoa* Willd. (Chenopodiaceae), an alternate herbaceous host for the New Jersey strain of BlScV, was shown to transmit the virus to uninfected *C. quinoa*. In a similar manner, Bristow *et al.* (2000) were able to demonstrate infection of containerized highbush blueberry plants in cages supplied with diseased blueberry leaves infested with *Ericaphis fimbriata* (Richards). In the same study, transfer of individual aphids from infected blueberry leaves to containerized potted test plants resulted in a very low rate of infection, less than one percent. These previous studies were not designed to determine if BlScV was transmitted by aphids in a semi-persistent or non-persistent manner. Two carlaviruses vectored by aphids are thought to be transmitted in a semi-persistent manner (Bristow *et al.* 2000).

A better understanding of BlScV epidemiology will aid in the development of effective control measures. To this end, the purpose of our study was to determine the mode of transmission of BlScV and compare aphid transmission efficiencies of *E. fimbriata*, a species that colonizes blueberry, with transmission by several non-colonizing aphid species. Identification of effective aphid vectors will also assist in future laboratory investigations to determine biological differences between the various strains of BlScV.

MATERIALS AND METHODS

Plant and aphid culture. Large highbush blueberry plants from two commercial fields near Abbotsford, British Columbia (BC), that had previously tested positive for BlScV by ELISA using polyclonal antibodies (Agdia, Elkhart, Indiana) were potted into large (~ 60 cm x 43 cm deep) plastic pots and moved to a greenhouse at the Pacific Agri-Food Research Centre, Summerland, BC. These plants also formed the basis for the isolation and molecular characterization of two major strains of BlScV (Bernardy *et al.* 2005).

Nicotiana occidentalis Wheeler, recently identified as a herbaceous host for BlScV (Lowery *et al.* 2005), was grown in the greenhouse in 20-cm plastic containers in a 1:1:5 mixture of steam-sterilized field soil, perlite, and commercial potting soil (Pro-Mix BX, Premier Horticulture Ltd., Dorval, Quebec). Temperatures were variable and ranged from daytime highs of 25 °C to nighttime lows of 15 °C, with supplemental lighting supplied by sodium vapour lamps to provide a 16-h photophase. Plants were used at the four- or five-true-leaf stage. Small BlScV-free blueberry plants cv 'Berkeley' were acquired from a commercial supplier (Fall Creek Nurseries, Lowell, Oregon) and grown in the greenhouse in 3.8-litre plastic pots under the same conditions.

Aphids were maintained in vented, Plexiglas ® cages (50 cm x 50 cm x 33 cm wide) in a growth room (18 °C, 16-h photophase) on suitable host plants as follows: red and green forms of *E. fimbriata* on strawberry, *Fragaria x ananassa* Duchesne (Rosaceae); spirea aphid, *Aphis spiraecola* (Patch) and apple aphid, *A. pomi* DeGeer, on apple, *Malus domestica* L. (Rosaceae); pea aphid, *Acyrthosiphon pisum* (Harris), on garden pea, *Pisum sativum* L. (Fabaceae); violet aphid, *Myzus ornatus* Laing, and *Aphis helianthi* Monell on sunflower, *Helianthus annuus* L. (Asteraceae); green peach aphid, *Myzus persicae* (Sulzer), on bok-choi, *Brassica rapa* L. (Brassicaceae); and the bird cherry-oat aphid, *Rhopalosiphum padi* (L.), on barley, *Hordeum vulgare* L. (Poaceae). Host plants were reared in the greenhouse under conditions outlined above.

Except for *E. fimbriata* that were originally collected from commercial fields of highbush blueberry in the Fraser Valley and provided by Dr. D.A. Raworth (Agriculture and Agri-Food Canada, Pacific Agri-Food

Research Centre, Agassiz, BC), all of the aphid species used in these studies, other than *A. pisum*, were collected in Summerland, BC, from the hosts on which they were reared. *Acyrthosiphon pisum* was collected from garden peas in Armstrong, BC. Aphids were identified by Dr. R.G. Foottit (Agriculture and Agri-Food Canada, Eastern Cereals and Oilseeds Research Centre, Ottawa, Ontario).

Aphid transmission studies. Fourth instar and adult apterous aphids from the laboratory colonies were placed in small self-sealing petri dishes containing moistened filter paper for a 2- to 3-h pre-acquisition starvation period. Aphids were allowed to feed for 5 min on BlScV-infected leaf pieces in groups of 10 aphids/petri dish, and then transferred, 25 aphids/plant for blueberry and 10 aphids/plant for *N. occidentalis*, to BlScV-free test plants, which were then sealed in plastic bags to prevent the aphids from escaping. Fine, moistened natural fibre brushes were used to transfer aphids. At least 1 h after the final transfer, plants were sprayed with the aphicide pirimicarb (Pirimor 50WP, Chipman Chemicals Ltd., Stoney Creek, Ontario) to kill any remaining aphids. Plants were held in the bags for a further 24 h to ensure that all aphids were dead. *Nicotiana occidentalis* were then moved to a growth chamber at 20 °C under fluorescent and incandescent lights (approx. 185 μmol m^{-2} s^{-1} PAR) and a 16-h photophase. After 6 to 8 wk, plants were tested for BlScV infection by enzyme-linked immunosorbent assay (ELISA) using polyclonal antibodies (Agdia, Elkhart, Indiana). Blueberry plants were held in the greenhouse under the conditions outlined above and tested for infection approximately 3 mo later. Plants were then moved to cold storage at 4 °C for 3 mo and then re-tested 2 to 3 mo after being returned to the greenhouse. Virus transmission studies for all species of aphids and both species of host plant were conducted concurrently.

ELISA analysis. The double-antibody sandwich (DAS) ELISA method used was a modification of the protocol described by Clark and Adams (1977). All reagents were added at 100 μl per well in microtitre plates. Microtitre plates (EIA Microplate, ICN Biomedicals, Irvine, California) were coated with purified immunoglobulin (IgG) (Agdia, Elkhart, Indiana) diluted (5 μl ml^{-1}) in phosphate-buffered saline (PBS) for 4 h at 37 °C. Plates were washed three times with PBS. Plant samples (0.25 g) were thoroughly ground in Bioreba bags (Bioreba AG, Reinach, Switzerland) with 1.5 ml borate buffer (0.1 M boric acid, 0.01 M sodium borate, 2% polyvinylpyrolidine (PVP 44,000), 0.2% non-fat milk powder, 0.05% Tween-20, 0.5% nicotine), and the bags briefly centrifuged at 2000 rpm to aid pipetting. The liquid extract (25 μl) and borate buffer (75 μl) were added to the microtitre plates, which were covered in cellophane and placed overnight on an orbital shaker at 600 rpm. After washing the plates with PBS-Tween and adding a dilute (5 μl ml^{-1}) IgG-enzyme conjugate in PBS-Tween-BSA-polyvinylpyrolidine, plates were incubated at 37 °C for 2 h. After plates were washed with buffer, a dilute (0.5 mg ml^{-1}) solution of p-nitrophenyl phosphate buffer was added. Plates were incubated at room temperature on an orbital shaker (600 rpm) for about 1 hr and absorbance was read at 405 nm. A subset of healthy blueberry nursery plants was tested by ELISA to verify that they were free of BlScV.

In order to verify BlScV infections, a subset of blueberry and *N. occidentalis* plants that had tested positive by ELISA were also tested by reverse transcriptase polymerase chain reaction (RT-PCR) as described in Bernardy *et al.* (2005).

Statistical analysis. Differences in rates of transmission of BlScV by the various aphid species were determined by contingency table analysis and multiple comparisons for proportions, analogous to a Tukey's test (Zar, 1984). Data were analyzed separately for each combination of infected source and healthy test plants. Infection rates were not included in the analysis if fewer than ten test plants had been inoculated.

RESULTS

By using *N. occidentalis* for both BlScV-infected and healthy test plants, we were able to compare transmission rates for several species of aphids not previously known to vector this disease (Table 1). Several species, including *A. pomi*, *M. persicae*, and *R. padi*, were inefficient vectors that were able to infect *N. occidentalis* only at low transmission rates ranging from 2% to 4%. The highest rate of transmission from infected to healthy *N. occidentalis* occurred when *M. ornatus* (average transmission rate 69%) or *A. helianthi* (data not shown) were used as vectors. Unfortunately, the latter species was not included in the statistical analysis due to the death of the colony from a fungal infection before the tests could be completed. Both the green and red forms of *E. fimbriata* transmitted BlScV between *N. occidentalis* at intermediate rates of 10% and 8%, respectively. *Acyrthosiphon pisum* and *A. spiraecola* did not transmit BlScV from infected to healthy *N. occidentalis*.

The highest rate of infection of highbush blueberry (20%) was recorded for the green form of *E. fimbriata*, whereas infection rates for *M. ornatus* and *A. spiraecola* were both 7% (Table 1). *Aphis helianthi* was not included in the data analysis, as we were only able to inoculate six blueberry plants with BlScV using this species before the colony collapsed due to a fungal infection. However, the infection rate for this species, which does not colonize blueberry, appeared to nearly equal that for the colonizing species *E. fimbriata*.

Virus transmission tests from infected blueberry to *N. occidentalis* were conducted to evaluate the acceptability of *N. occidentalis* as a trap plant in field studies of BlScV epidemiology. No plants became infected when *M. persicae* was used to vector the virus from infected blueberry to *N. occidentalis* (Table 1), but use of the green form of *E. fimbriata* resulted in an infection rate of 27%.

The utility of *N. occidentalis*, a recently identified herbaceous host of BlScV (Lowery *et al.* 2005), for laboratory studies of aphid transmission efficiencies was demonstrated in this study. Even though fewer aphids (10/plant) were used to inoculate *N. occidentalis* than blueberry (25/plant), overall infection rates were similar. Blueberry plants had to be held for many months to demonstrate virus transmission, and approximately half the plants tested positive only after an intervening 3-month cold period. This was expected since virus titres are generally low in blueberry compared with herbaceous hosts, the virus is often distributed unevenly within blueberry plants, and detection may vary seasonally (Martin and Bristow 1988, Wegener *et al.* 2006). In comparison, unequivocal ELISA results could be obtained for infected *N. occidentalis* within 6 to 8 wk after infection and plants then retained a high virus titre over a period of several months.

DISCUSSION

Carlaviruses were, until recently, one of the largest and least studied of the plant virus groups (Foster 1992). Diseases caused by these viruses often result in latent infections or they cause indistinct, mild symptoms, which resulted in carlaviruses being largely ignored by pathologists. BlScV is an exception to this general condition, with infections resulting in significant loss of yield and eventual death of certain cultivars of highbush blueberry. For this reason, a number of recent studies have investigated the molecular characteristics, epidemiology, and aphid transmission of BlScV.

Carlaviruses are transmitted largely by aphids in a non-circulative, non-persistent manner (Foster 1992). Certain of them are thought to be transmitted in a semi-persistent manner, however, and at least one member of the group, *Cowpea mild mottle virus*, is transmitted by whiteflies (Harris 1983, Ng and Perry 2004). In the

Table 1.
Aphid transmission of *Blueberry scorch virus* from infected *Nicotiana occidentalis* or highbush blueberry, *Vaccinium corymbosum*, to healthy test plants.

Aphid Species	Infected Source	Test Species	Infected/ Total	% Infection
Acyrthosiphon pisum	*Nicotiana occidentalis*	*Nicotiana occidentalis*	0/41	0d[1]
Aphis pomi	*N. occidentalis*	*N. occidentalis*	1/48	2c
Aphis spiraecola	*N. occidentalis*	*N. occidentalis*	0/33	0d
Ericaphis fimbriata, green form	*N. occidentalis*	*N. occidentalis*	4/40	10b
Ericaphis fimbriata, red form	*N. occidentalis*	*N. occidentalis*	3/40	8bc
Myzus ornatus	*N. occidentalis*	*N. occidentalis*	11/16	69a
Myzus persicae	*N. occidentalis*	*N. occidentalis*	2/48	4bc
Rhopalosiphum padi	*N. occidentalis*	*N. occidentalis*	1/40	3bc
Acyrthosiphon pisum	blueberry	blueberry	0/18	0b
Aphis pomi	blueberry	blueberry	0/15	0b
Aphis spiraecola	blueberry	blueberry	1/14	7a
Ericaphis fimbriata, green form	blueberry	blueberry	5/25	20a
Ericaphis fimbriata, red form	blueberry	blueberry	0/7	-
Myzus ornatus	blueberry	blueberry	1/14	7a
Myzus persicae	blueberry	blueberry	0/24	0b
Rhopalosiphum padi	blueberry	blueberry	0/23	0b
Ericaphis fimbriata, green form	blueberry	*N. occidentalis*	4/15	27a
Myzus persicae	blueberry	*N. occidentalis*	0/30	0b

[1] For each combination of infected source and healthy plant species, infection rates followed by the same letter are not significantly different based on contingency table analysis and multiple comparisons for proportions (Zar 1984).

present study, the results of earlier uncontrolled cage studies that demonstrated transmission of BlScV by *E. fimbriata* (Bristow *et al.* 2000) were confirmed. Utilizing timed acquisition feeding periods, we found that BlScV is indeed transmitted by aphids in a non-persistent manner, as might be expected for a member of the carlavirus group. Aphids were able to acquire the virus during brief acquisition-feeding periods lasting less than 5 minutes. Additionally, aphids that do not colonize blueberry, such as *M. ornatus*, were equally efficient virus vectors compared to the colonizing species *E. fimbriata*. A pre-acquisition fasting period and short virus-acquisition probes increase transmission of non-persistent viruses, while prolonged feeding leads to greatly reduced transmission rates (Maramorosch 1963). For this reason, under field conditions, non-colonizing aphids are often more important vectors of viruses such as BlScV. Due to low virus titres, however, a slightly longer acquisition feeding period might improve transmission efficiencies when BlScV is acquired from highbush blueberry. Although *E. fimbriata* was not the most efficient vector of BlScV from *N. occidentalis* to *N. occidentalis*, it was the best vector when the virus was ac-

quired from highbush blueberry, possibly because this species was observed to settle and feed more readily on this host plant. Thus, *E. fimbriata* might contribute significantly to the spread of this virus within infected fields, particularly in years with large numbers of these colonizing aphids.

Infection of *N. occidentalis* by viruliferous aphids in this study occurred at a level comparable with that for infections of herbaceous plants with non-persistently transmitted potyviruses. In similar controlled studies, transmission of *Potato virus Y* from infected to healthy sweet pepper by *M. persicae* resulted in an 89% infection rate (Lowery *et al.* 1997), whereas in another study involving several species of aphids the maximum rate of infection of rutabaga with *Turnip mosaic virus* was 55% (Lowery 1997). The highest rate of infection of *N. occidentalis* with BlScV falls within these range of values (Table 1). Successful transmission of BlScV from blueberry to *N. occidentalis* by *E. fimbriata* has also been used successfully to help purify and amplify virus in strain determination studies (Bernardy *et al.* 2005). Compared with highbush blueberry, this herbaceous host should prove useful as a trap or sentinel plant in studies of BlScV epidemiology. It will be necessary, however, to first show that *N. occidentalis* is uniformly susceptible to all strains of BlScV.

During a two year study, Raworth *et al.* (2006) captured alate aphids of 87 species in water pan traps placed in commercial blueberry fields in the Fraser Valley, BC. Our results suggest that many of these species are likely vectors of BlScV. Future virus transmission studies involving aphid species that were captured in large numbers from the middle of June to the middle of July when most trap plants became infected (Raworth *et al.* 2008), which would include several species such as *Euceraphis betulae* (Koch) that develop on trees (Raworth *et al.* 2006), might help identify some of the other major vectors contributing to the spread of BlScV and suggest possible management strategies.

Based on our laboratory results, BlScV is transmitted between highbush blueberry at a rate similar to that for other non-persistent, aphid-borne viruses of woody perennial plants. In comparable transmission tests using 50 *M. persicae* per plant a 'D' strain of *Plum pox virus*, a member of the *Potyviridae*, was transmitted from infected peach, *Prunus persicae* L., to healthy peach seedlings at an average infection rate of 22% (D.T.L. unpublished data). In the present study with blueberry, but using only 25 aphids per plant, a maximum infection rate of 20% was recorded for *E. fimbriata* (Table 1). The relatively low rate of transmission from blueberry to blueberry as compared with infection of herbaceous hosts suggests that a number of years would be required for BlScV to spread throughout a blueberry field from an initial infection locus. Accordingly, mapping of disease incidence in three commercial blueberry fields in the Fraser Valley, BC, showed that BlScV spread only slowly (Wegener *et al.* 2006). Similarly, spread of BlScV throughout two commercial fields of blueberry in the northwestern United States required between 5 to 8 years (Bristow *et al.,* 2000), and Raworth *et al.* (2008) recorded a low rate of BlScV infection for highbush blueberry and *N. occidentalis* bait plants placed weekly throughout the summer in highly infected commercial blueberry fields, indicating a low rate of natural spread. Given the relatively slow spread of the virus under field conditions, these findings suggest that planting only certified virus-free nursery material and aggressive removal of diseased plants might provide an effective means of control of BlScV under field conditions.

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Sex attraction in *Polistes dominulus* (Christ) demonstrated using olfactometers and morphological source extracts

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ABSTRACT

Y-tube and parallel tube olfactometers were used to test for attraction between and within genders of the European paper wasp, *Polistes dominulus* (Christ). In the Y-tube olfactometer, unmated females were attracted to male odour, while males were repelled by unmated female odour. Males and females were not attracted to the odour of the same sex in this experiment. In the parallel tube olfactometer, females were attracted to male odour, while males were not attracted to female odour. Morphological sources of potential sex attractants were tested using an arena bioassay design. Males rubbed their mandibles and gaster on the substrate when exposed to extracts of unmated female or male tagmata, female or male legs, or the male seventh gastral sternite. We did not see overt behavioural responses by females to male or female extracts.

Key Words: attractant, pheromone, *Polistes dominulus*, paper wasp

INTRODUCTION

Sex pheromones are chemicals that elicit behaviour related to mate-finding, mate-selection, and copulation in insects, including vespid wasps (Wilson 1971; Shorey 1977; Landolt et al. 1998). Close-range attractants and copulatory incitants or aphrodisiacs have been demonstrated between males and females of the social wasps *Polistes exclamans* Viereck (Post and Jeanne 1984; Reed and Landolt 1990a), *Polistes fuscatus* (F.) (Post and Jeanne 1983a, 1984), *Belonogaster petiolata* Degeer (Keeping et al. 1986), *Vespula squamosa* Drury (Reed and Landolt 1990b), and *Vespa* spp. (Batra 1980; Ono and Sasaki 1987). Despite such demonstrations, no vespid sex pheromone chemical structure has been identified. Sexual behaviour of *Polistes dominulus* (Christ) (Hymenoptera: Vespidae) has not been described or quantified in controlled experiments. Knowledge of behavioural responses to putative pheromones is necessary for accurate pheromone characterization.

Behavioural evidence in *Polistes* paper wasps suggests that sex pheromones from exocrine glands in the mandibles, legs, and gastral sterna may be involved in mate attraction (Landolt and Akre 1979; Jeanne *et al.* 1983; Beani and Turillazzi 1988; Beani and Calloni 1991a,b; Beani *et al.* 1992). In several of these species, mating often occurs away from the nest (Noonan 1978) on perching substrates that are at prominent locations such as on hilltops (Beani and Turillazzi 1988; Mathes-Sears and Alcock 1986). Males, in some species of *Polistes*, scent-mark by dragging their posterior gastral sternites (Post and Jeanne 1983b; Reed and Landolt 1991) and by rubbing their mandibles (Wenzel 1987; Reed and Landolt 1991) on the perching substrate. Four spe-

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cies, including *P. dominulus*, are shown to drag their hind legs on the substrate (Beani and Calloni 1991a). *Polistes dominulus* males have ducted class III gland cells that open onto the cuticle in their legs, as well as onto the seventh abdominal sternite (Downing *et al.* 1985; Beani and Calloni 1991a). Although females of *Polistes* spp. are not known to show overt scent-marking behaviour, *P. exclamans* females possess a pheromone in the venom that elicits sexual behaviour in both conspecific and heterospecific males (Post and Jeanne 1984). Additionally, a surface pheromone on the thoracic and gastral cuticle in *P. fuscatus* is important for male recognition of conspecific females (Post and Jeanne 1984).

The objective of this study was to investigate orientation and behavioural responses of *P. dominulus* to potential sex odours from a variety of morphological sources. Klinotaxic (turning orientation) and orthotaxic (forward orientation) (Fraenkel and Gunn 1940; Wyatt 2003) responses to male odour and unmated female odour were tested using Y-tube and parallel tube olfactometers, respectively. An arena-type bioassay was used to test for behavioural responses to extracts of male and unmated female tagmata and glands. These studies are foundational in aiding the overarching objectives of determining sex pheromone signaling systems in this species, and determining sources of those sex pheromones.

MATERIALS AND METHODS

Colony Collection and Maintenance. *Polistes dominulus* nests with pupae were collected in the field, and placed in plastic screened cages (30.5 x 30.5 x 30.5 cm) during late August and early September in 2003 and 2004. Nests were collected at this time because males were abundant, indicating that reproductive females would be emerging from nests, and not worker females. Collected nests were monitored daily to remove and segregate male and female adults that emerged, in order to minimize encounters between the sexes and exposure to sex pheromones. Male and female wasps obtained in this manner were assumed to be sexually inexperienced since mating is reported to occur away from the nest (Noonan 1978). These wasps were used for the preparation of extracts and for behavioral assays conducted in Pullman and Yakima, Washington.

In Pullman (Whitman County), Washington, USA, the newly emerged wasps were kept in a laboratory at 24°C, 40% RH, under a natural light regime (14 hours of light and 10 hours of dark), until testing in the Y-tube olfactometer and in the parallel tube olfactometer. All Y-tube olfactometer tests were conducted in Pullman, while one half of bioassay replicates for each experiment conducted with parallel tube olfactometers were conducted in Pullman and the other half at the USDA, ARS Yakima Agricultural Research Laboratory near Yakima (Yakima County), Washington, USA. Wasps used in olfactometer tests and the arena bioassays conducted in Yakima were kept in a glass greenhouse under natural lighting at 30 ± 3 °C and 35% RH. At both sites, wasps in cages were provided water and a 1:10 molasses:water solution on cotton balls for nutrition. Water and the solution of molasses were refreshed or replaced daily. Wasps used in all assays were between 2 and 14 days old; they were randomly selected for each trial and were not reused in other trials for at least 48 hours.

Y-tube Olfactometer Bioassay. Unmated male and female wasps were tested for klinotaxic responses to unmated female odour and male odour in the Y-tube olfactometer. The inside diameter of the glass Y-tube was 2.5 cm and the length of the tube, from stem base to Y-juncture, was 18 cm. Airflow of 100 ml/min was measured with a flowmeter (Aalborg Instruments, Monsey, NY) before and after passing through the 480 ml jars housing the treatments. Air passed through the treatment jar containing 3 "bait wasps" of the sex being assayed, then through one arm of the Y-tube, and out the stem of the Y-tube. Simultaneously, air

passed through the empty control jar, through the other arm of the Y-tube, and out the stem of the Y-tube.

A paper wasp was placed in the stem of the Y-tube and observed for a maximum of five minutes. If the wasp moved upwind to the Y-juncture and then moved completely beyond the juncture into either of the arms (with treatment airflow or with control airflow), that assay was ended and the response was recorded. Ten wasps were tested individually and in succession using the same "bait wasps" in the treatment jar. To eliminate a potential left or right turning bias, the positioning of the treatment and control was switched after the first 5 wasps had been tested. A clean olfactometer system was then set up and a fresh set of bait wasps was placed in the treatment jar. This experimental protocol was conducted four times to provide a total of 40 wasps (5 wasps in series x 2 treatment positions x 4 = 40) tested for responses. Wasps entering the treatment arm, the control arm, or neither arm of the Y-tube were recorded. For each experiment, the numbers of wasps that entered the treatment arm or the control arm were compared using the Chi-square goodness-of-fit test with Yates correction for continuity at $P \leq 0.05$ (Zar, 1974).

The olfactometer system was placed horizontally 50 cm beneath two 1.2 m long, 34W fluorescent bulbs (Osram Sylvania Corp., Danvers, MA) and one 160W mercury vapour bulb (Osram Sylvania Corp., Danvers, MA). Temperature at the olfactometer surface was 31°C. Air moving through the Y-tube olfactometer was supplied by an aquarium air pump, purified through a hydrocarbon trap (Alltech Associates Inc., Deerfield, IL), and humidified with a gas diffusion bottle. All glassware (Ace Glass, Inc. Vineland, NJ) and steel tubing were washed in hot water with Micro-90 cleaning solution (International Products Corp., Burlington, NJ), and then rinsed serially with deionized water, acetone, and then hexane. Glassware was subsequently placed in a drying oven at 180°C overnight before used again in assays.

Parallel Tube Olfactometer Bioassay. Unmated females were tested for orthotaxic responses to male odour and males were tested for orthotaxic responses to unmated female odour in a parallel or "straight tube" olfactometer design. This design is based on that of Tobin *et al* (1981) and was reported by Landolt *et al.* (1988). The olfactometer set up was the same as the Y-tube set up, except for the replacement of the Y-tube with two straight glass tubes. Each straight tube was supplied 100 ml/min of metered, purified humidified airflow that was passed through a glass jar housing an odour source, separate from the other tube and odour source. This setup was placed on a laboratory table with fluorescent lighting above and natural lighting from windows. A wasp was placed in a straight glass tube, 2.5 cm diameter and 18 cm long, downwind from the treatment airflow (3 bait wasps) and another wasp was placed in an identical tube downwind from the control airflow (empty). For each wasp, the time it took to cover the full 18 cm distance of the tube was recorded, if indeed it completed the full distance upwind. This assay was conducted with 10 pairs of wasps (treatment and controls paired), and the glassware for treatment and control were switched after 5 pairs of wasps were tested. This experiment was then replicated eight times, (N = 80) and treatment mean times were separated from control mean times using a paired *t*-test at $P \leq 0.05$. Also, the mean percents of those that travelled the entire lengths of the treatment and control tubes, within the five minute time limit, were separated using a paired *t*-test at $P \leq 0.05$.

One half of the parallel tube olfactometer replicates were conducted under the same conditions as the Y-tube olfactometer bioassays, in Pullman. The other replicates, in Yakima, were conducted as they were in Pullman, with these slight modifications: (1) the bioassays were conducted in a controlled environment room at 24 °C and 65% RH; (2) airflow was from a compressed air source; (3) light was supplied by two, 1.2 m long, 34 W fluorescent light bulbs (Osram Sylvania Corp., Danvers, MA) 50 cm above the olfactometer. A J16 Digital Photometer

(Tektronix Inc., Beaverton, OR) measured at 27,663 lux (lumens/m^2) at the olfactometer surface; (4) after cleaning, the olfactometer glassware and tubing were placed in a drying oven for 24 h. Data from the parallel tube olfactometer bioassays in Pullman and Yakima were pooled and analyzed together. We did not expect the minor differences in assay conditions to alter the behaviour of the wasps and a preliminary analysis of the results indicated similar responses in the assays.

Tagmata and Extract Preparations. Dissecting and grinding tools and equipment were washed in hot water with Micro-90 cleaning solution, and then rinsed with deionized water, acetone, and methylene chloride. Samples of 40 female heads, 40 male heads, 40 female thoraces, 40 male thoraces, 40 female gasters, and 40 male gasters, all from freshly freeze-killed wasps, were each ground with a mortar and pestle in methylene chloride. Additionally, 40 female venom sacs with acid sting glands, 40 female alkaline glands, legs of 40 females, 40 male mandibles with ectal glands, 40 male seventh gastral sternites with glands, and legs of 40 males were dissected or removed and then extracted with methylene chloride. All tagmata and gland extracts were reduced to 4 ml under a N_2 stream and kept in a freezer at -15 °C, providing concentrations of one wasp-equivalent per 100 µL of extract.

Tagmata and Gland Bioassay. Arena bioassays were conducted in the same greenhouse environment in which the wasps were housed. The assays occurred over the course of 3 weeks in September between 10:00 and 16:00 hr. Light intensity at the table was 16,758 ± 795 (mean ± S.E.) lux, measured at 20 different times throughout the bioassays.

On a table covered with white paper, a wasp was placed under the bottom half of an upside down, plastic, 8.5 cm diam. Petri dish for one minute before experiencing extract odour. Petri dishes and paper were discarded after each bioassay. Immediately prior to conducting the assay, 100 µl of the treatment extract or 100 µl of the methylene chloride control were applied to ¼ wedges of 5.5 cm diameter, #3 Whatman Filter Paper (Whatman International Ltd., Maidstone, England). The methylene chloride was evaporated before the filter paper was placed under the Petri dish with the wasp. Wasps were observed for two minutes while in the presence of the extract or solvent blank, after which they were placed into holding cages to ensure they were not used again in the assay. At the end of the assay period on any given day, all wasps were returned to cages that constituted the general pools of male and female wasps from which random selections were made for subsequent experiments.

Male and female wasps were tested for responses to extracts of tagmata and glands from both sexes. Each of the tests was replicated 20 times. In bioassays of male and female tagmata, the sequence was: blank, head, thorax, and gaster. In bioassays of female gland bioassay the sequence was: blank, venom, legs, and alkaline gland. In bioassays of male glands the sequence was: blank, mandibles, legs, and seventh sternal gland. For each assay, a record was kept of continuous movement, no movement, and the number of times a wasp showed stop & go movement, antennal contact with the filter paper, grooming fore legs through mandibles and then rubbing antennae, grooming fore legs through mandibles then rubbing thorax, grooming gaster with hind legs, rubbing hind legs together, rubbing mandibles on substrate, and rubbing gaster on substrate. For each behaviour and each sex, a 2x2 contingency table was constructed to make comparisons of the number of times a behaviour was observed for the control versus each of the tagmata and gland extracts. For each behavior, contingency tables were analyzed using the Chi-square statistic with Yates correction for continuity at $P \leq 0.05$ (Zar, 1974).

RESULTS

Y-tube Olfactometer Bioassays (Table 1). Females significantly more often turned toward male odour and away from the control ($P < 0.001$). Males significantly more often turned away from female odour and toward the control ($P < 0.05$). Neither males nor unmated females turned toward odour from males ($P > 0.10$) and females ($P > 0.5$), respectively, compared to the control.

Parallel Tube Olfactometer Bioassays. The mean time (± S.E.) of female movement toward male odour (33.7 ± 4.4 sec, df = 79, $P = 0.002$) was significantly lower than toward the control (49.1 ± 4.1 sec). There was no significant difference in the mean percent (± S.E.) of females that traveled the length of the treatment (89.0 ± 4.3%, df = 7, $P = 0.087$) and control (81.0 ± 4.3%) tubes within five minutes. The mean time (± S.E.) for male movement toward female odour (88.2 ± 14.3 sec, df = 79, $P = 0.141$) was not statistically different compared to the control (68.1 ± 12.0 sec). This was also true for the mean percent (± S.E.) of males that travelled the length of the treatment (70.0 ± 7.3%, df = 7, $P = 0.361$) and control (78.8 ± 6.9%) tubes within five minutes.

Tagmata Experiments. Females did not behave differently in the presence of extracts of female or male tagmata compared to the blank. Males rubbed their mandibles on the substrate more often when treated with extract of ♀ thorax ($P < 0.05$) compared to the blank (Table 2). They also rubbed their gaster on the substrate more often when treated with extracts of ♀ thorax ($P < 0.005$) or ♀ gaster ($P < 0.05$) compared to the blank (Table 2). When presented with extracts of ♂ thorax and ♂ gaster, males rubbed their mandibles on the substrate more often ($P < 0.05$) compared to the blank (Table 2).

Gland Experiments. Unmated females did not show any significant behavioural differences in the presence of extracts of female or male glands compared to the blank. Males rubbed their mandibles ($P < 0.05$) and gaster ($P < 0.005$) on the substrate more often when exposed to ♀ leg extract (Table 3). When treated with ♂ leg extract, males continuously moved ($P < 0.001$) and rubbed their mandibles on the substrate ($P < 0.05$). They also rubbed their mandibles ($P < 0.005$) and gaster ($P < 0.005$) on the substrate when exposed to ♂ seventh sternite with gland extract (Table 3). Lastly, males rubbed their gaster on the substrate ($P < 0.005$) more often when exposed to ♂ mandibles with ectal mandibular gland extract (Table 3).

DISCUSSION

Polistes males are known to use mating strategies such as marking behaviour by rubbing their mandibles, gaster, and legs on perch sites (Beani and Calloni 1991a; Polak 1993). It is hypothesized that such scent-marked perch sites attract females (Landolt and Akre 1979; Post and Jeanne 1983b,c; Wenzel 1987; Reed and Landolt 1990a). Our orientation bioassay results using olfactometers support the hypothesis that females are attracted to a pheromone of males since male odour elicited positive turning and increased speed of forward movement from females. Female attraction to males in olfactometers has also been shown for the paper wasp species *P. fuscatus* (Post and Jeanne 1983a) and *P. exclamans* (Reed and Landolt 1990a).

The observed repellency of females to males in the Y-tube, but not the parallel-tube olfactometer, assays is puzzling and calls for possible explanation. It is assumed that females would control release of any sex attractant or courtship pheromone and we have no means of knowing if and when females were "calling" when they were tested in olfactometers. Thus, females may be attractive to males under circumstances not met by our assay conditions. Also, female venom may possess alarm pheromone (Bruschini et al. 2006), which could complicate assay results when females have

Table 1.

Numbers of European paper wasps, *Polistes dominulus*, turning towards airflow from over conspecific wasps compared to control airflow, in a Y-tube olfactometer[1].

Bioassay	$N_{treatment}$	$N_{control}$	$\chi^2_{experimental}$	*P*-value
♀ to ♀	22	18	0.225	$P > 0.5$
♀ to ♂	31	9	11.025	$P < 0.001$
♂ to ♀	13	27	4.225	$P < 0.05$
♂ to ♂	16	24	1.225	$P > 0.10$

[1] Analyzed using Chi-square goodness-of-fit test with Yates correction for continuity where $\chi^2_{theoretical(1, 0.05)} = 3.841$.

Table 2.

Numbers of unmated male European paper wasps, *Polistes dominulus*, responding to unmated male and female tagmata in an arena type assay[1].

Behaviour[2]	Blank	♀Head	♀Thorax	♀Gaster	♂Head	♂Thorax	♂Gaster
A	12	14	15	17	11	11	14
B	3	4	0	2	4	2	2
C	5	2	5	1	5	7	4
D	5	8	10	10	4	4	10
E	12	8	13	8	7	10	7
F	5	0	7	3	4	6	8
G	2	4	4	3	3	3	3
H	4	0	5	1	0	4	4
I	2	7	9*	7	4	9*	9*
J	0	2	9	6*	2	3	4

[1] Within a row, treatments compared only to blank in a 2x2 contingency table analyzed using the Chi-square statistic with Yates correction for continuity are significant at $P \leq 0.05$. Numbers with an asterisk are significantly different from the blank.

[2] A – continuous movement, B – no movement, C – stop & go, D – antennate paper, E – groom fore legs in mandibles; rub antenna, F – groom fore legs in mandibles; rub thorax, G – groom gaster with hind legs, H – rub hind legs together, I – rub mandibles on substrate, J – rub gaster on substrate.

been handled and disturbed. Caution must then be exercised in interpreting negative or apparently conflicting results.

Female *P. dominulus* in this study showed neither a positive nor negative orientation response toward female odour. Overwintering females in search of hibernation sites might be expected to respond to aggregations of females (Reed and Landolt 1991). Heterospecific aggregations of paper wasp gynes in hibernacula have been reported (Rau 1930, 1938; Bohart 1942; Hermann *et al.* 1974; Gibo 1980; Reed and Landolt 1991) as well as purely conspecific aggregations (Rau 1938; Strassmann 1979). *Polistes dominulus* queens appear to overwinter in multi-colony groups (Starks 2003). However, there may be multiple factors (such as temperature and day length) that stimulate females to seek out hibernacula and other overwintering females. The wasps used in these bioassays were housed under summer-like temperatures and day length. Hence, we can make no conclusions regarding the presence or absence of a female aggregation pheromone.

Venom is thought to include a sex pheromone that elicits copulatory behaviour in males of *P. fuscatus* and *P. exclamans*

Table 3.

Numbers of unmated male European paper wasps, *Polistes dominulus*, responding to unmated male and female glands in an arena assay[1].

Behaviour[2]	Blank	♀Venom	♀Legs	♂Mandibles	♂Legs	♂Sternite	Blank	♀Alkaline
A	8	13	14	15	19**	15	11	12
B	3	1	2	1	0	2	2	5
C	9	6	4	4	1	3	7	3
D	6	11	11	13	12	13	5	7
E	9	8	15	14	9	9	11	12
F	1	0	3	2	2	1	4	2
G	6	2	10	7	6	6	7	5
H	4	2	7	6	4	5	4	6
I	2	6	9*	6	9*	12*	4	3
J	1	4	10*	9*	5	10*	3	5

[1] Within a row, treatments compared only to blank in a 2x2 contingency table analyzed using the Chi-square statistic with Yates correction for continuity (significant at $P \leq 0.05$, $df = 1$). Numbers of responses with an asterisk are significantly different from the blank.

[2] A – continuous movement, B – no movement, C – stop & go, D – antennate paper, E – groom fore legs in mandibles; rub antenna, – groom fore legs in mandibles; rub thorax, G – groom gaster with hind legs, H – rub hind legs together, I – rub mandibles on substrate, J – rub gaster on substrate

(Post and Jeanne 1983a; 1984). Odorants on the female cuticle are thought also to be a conspecific sex pheromone in these species (Post and Jeanne 1983a, 1984). Our olfactometer bioassays did not evaluate copulatory behaviour, but rather orientation behaviour. Males of *P. dominulus* did not orient by turning or increasing their speed of movement toward female odour in either the Y-tube or parallel tube bioassays, respectively. We did observe behavioural responses such as increased movement and possible scent marking by males to extracts of female gasters and venom glands in an arena assay, but again we did not study copulatory responses. Although venom seems to play a role in mediating courtship or copulation in some paper wasps, it is not known to serve as a sex attractant. In our experiments, we cannot rule out possible female release of alarm chemicals due to handling stress, complicating the interpretation of behavioural assay results, although Freisling (1943) was unable to demonstrate an alarm pheromone in *P. dominulus*.

Polistes dominulus females have ducted type III gland cells in their legs (Beani and Calloni 1991a). *Polistes fuscatus* males responded to female tagmata extracts in a wind tunnel, but the greatest response was to female thorax extract (Reed and Landolt 1990a). During our behaviour bioassays, males responded to extract of ♀ legs in the same manner as to extract of the entire thorax, by rubbing their mandibles and gaster on the substrate. Additionally, males rubbed their gaster on the substrate when exposed to ♀ gaster extract. These behavioural responses have been observed in males of other *Polistes* species and described as scent-marking (Post and Jeanne 1983b; Wenzel 1987). Female odour may not elicit orientation responses, but female extracts of thoraces and legs do appear to elicit scent-marking in males.

Scent-marking behaviour in males is believed to attract females for mating purposes, and to signal territorial ownership to other males (Post and Jeanne 1983b; Beani and Calloni 1991a). Male odour may deter other males of the same species since territorial males will actively chase intruders away and, afterwards, increase grooming and scent-marking behaviour (Post and

Jeanne 1983b; Beani and Calloni 1991a). Males of other *Polistes* species are known to patrol and defend territorial perches near hibernacula and nest sites by gaster dragging and mandible rubbing on the substrate (Post and Jeanne 1983b; Wenzel 1987; Reed and Landolt 1991). The dragging of the male gaster has also been in observed in *Mischocyttarus* spp. (Litte 1979, 1981). Nineteen of 20 males that were presented with ♂ leg extract continuously moved for the two-minute assay; they also rubbed their mandibles on the substrate more often than when presented with the blank. Males rubbed their mandibles on the substrate when presented with ♂ gaster extract. When the seventh sternal gland was dissected and presented to males, they performed gaster dragging in addition to rubbing their mandibles on the substrate. Although males did not respond to ♂ head extract, they did drag their gaster on the substrate when presented with ♂ mandible extract. Therefore, we quantified and report behaviours that are consistent with previous behavioural studies of different *Polistes* species.

The orientation and behavioural evidence reported herein supports the previously stated hypothesis that males use scent-marking for mating and territorial defense purposes by depositing pheromone from well-developed exocrine glands in the gaster, mandibles, and legs onto the perch substrate (Landolt and Akre 1979; Jeanne *et al.* 1983; Beani and Calloni 1991b). The quantification of orientation and behavioural responses to potential sex attractant odours between and within genders is foundational to the larger work of isolating, identifying, and testing sex pheromones in Vespidae.

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Efficacy of Isomate-CM/LR for management of leafrollers by mating disruption in organic apple orchards of western Canada

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ABSTRACT

Results of a three-year study demonstrated that Isomate-CM/LR, a polyethylene, single tube-type pheromone dispenser releasing an incomplete mixture of several species' multi-component pheromones is an effective management tool that provides multiple-species mating disruption for *Choristoneura rosaceana* (Harris) and *Pandemis limitata* (Robinson). When applied at a rate of 500 dispensers / ha within the orchard and the equivalent of 2000 dispensers / ha to trees on the orchard perimeter, levels of control were adequate for production of organic apples in British Columbia, Canada. Trap catches with synthetic pheromone lures were reduced by 79 -99% and mating of females on mating tables was reduced by 87 - 98% in these species. At harvest, damage from leafrollers in pheromone-treated organic orchards was below organically-acceptable economic levels (5%) and similar to damage levels observed in insecticide-treated conventional orchards (2%). Over three years, total trap catches of these two leafrollers and their damage decreased in four of five orchards treated with pheromone, but catches and damage from leafrollers increased in one orchard. These indices remained relatively unchanged in paired insecticide-treated conventional orchards over the same three-year period. In pheromone-treated orchards, levels of damage from leafrollers at harvest were positively correlated with total leafroller catches in pheromone monitoring traps. Use of Isomate-CM/LR as a supplemental pest management tactic in organic orchards will help to reduce damage and economic losses from leafrollers that have been increasing under the area-wide codling moth sterile insect programme ongoing in this semi-desert, montane apple production region.

Key Words: leafrollers, multiple-species mating disruption, organic apples

INTRODUCTION

Over the last decade a new paradigm for integrated pest management in pome fruits has emerged in western North America. This transformation was driven by implementation of area-wide programmes to control codling moth, *Cydia pomonella* (L.), using sterile insect technique (SIT) in Canada (Dyck and Gardiner 1992) and pheromone-based mating disruption in the United States (Calkins 1998). Application of these species-specific controls for codling moth has resulted in increasing damage from secondary pests (Brunner *et al.* 1994, Knight 1995, Gut and Brunner 1998). Consistent with earlier prediction (Madsen and Morgan 1970), several species of leaf-rolling caterpillars (Lepidoptera: Tortricidae) have become an increasing problem when broad-spectrum insecticides that target codling moth, but which provide partial control of leafrollers, are removed from the production system (Madsen and Proctor 1985). Although the species complex varies across production regions, increasing damage from leafrollers in orchards using mating disruption for codling moth is a recur-

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ring problem seen around the world (Charmillot 1990, Wearing *et al.* 1995, Walker and Welter 2001).

The British Columbia (BC) organic apple industry is concentrated in the Similkameen Valley (Judd *et al.* 1997, Mullinix 2005). Although orchards in this region have been receiving sterile codling moth since 1994 as part of an area-wide SIT control programme (Dyck and Gardiner 1992), producers of organic fruit found it necessary to supplement this programme with codling moth mating-disruption technology in spring (Judd and Gardiner 2005). Because codling moth is presently under control but profit margins are shrinking under increased foreign competition, organic apple producers in BC are seeking alternative controls for leafrollers to improve quality of graded export fruit. Before 2005, *Bacillus thuringiensis* Berliner (Bt) was the only organic material available for controlling leafrollers in Canada. Although Bt is effective, its use is often limited by inclement spring weather in montane areas of BC, and while fairly benign to beneficial species, it can have a significant effect on parasites of leafrollers if applied at the wrong time (Cossentine *et al.* 2003).

Organic pome-fruit producers in BC have been interested in pheromonal control of leafrollers ever since pheromones were first applied to control codling moth (Judd *et al.* 1997). The idea of using one mating-disruption system to simultaneously control codling moth and leafrollers has been around for some time (van Deventer *et al.* 1992) but few commercial products exist. Isomate-CM/LR, a multiple-species mating-disruption product designed to control codling moth and leafroller species important in western North America, was registered in the United States in 1997 and in Canada in 2004 (Don Thomson, personal communication). When Isomate-CM/LR was used in conjunction with insecticides (Knight *et al.* 1997, Knight *et al.* 2001), apple orchards had 41% less leafroller damage and received one less spray per season. These same orchards consistently had less codling moth damage than orchards receiving Isomate-C^{+} and supplemental insecticides. Whether leafrollers can be controlled effectively, or sufficiently, with Isomate-CM/LR when no supplemental insecticides are used remains untested.

Judd and Gardiner (2005) reported on the use of mating disruption as a supplementary tactic for spring control of codling moth in organic orchards that were part of the area-wide SIT programme. Herein we report results using Isomate-CM/LR to control damage from leafrollers while at the same time supplementing codling moth control in those same organic apple orchards. The primary objective of this study was to conduct season-long assessments on disruption of pheromonal communication and mating in the leafrollers, *Choristoneura rosaceana* (Harris) and *Pandemis limitata* (Robinson) in commercial, organic apple orchards where Isomate-CM/LR was used, and to report on levels of leafroller damage in the absence of insecticide inputs. Second, we wanted to collect data on the relationship between different measures of disruption and relative density of adult leafrollers as measured by pheromone traps, because questions about the efficacy of mating disruption and population density are rarely addressed experimentally. Third, we wanted to determine if pheromone trapping of *C. rosaceana* and *P. limitata* has the potential to be a predictive tool of potential damage in orchards under pheromone-based mating disruption (Walker and Welter 2001).

MATERIALS AND METHODS

Test orchards. All apple orchards used in this study are located at Cawston, BC, in the Similkameen Valley (latitude 49° 10.8′ N, longitude 119° 46.2′ W, elevation 401 m). The five organic orchards treated with Isomate-CM/LR and five paired conventional orchards treated with insecticides (Table 1) were described in detail by Judd

Table 1.

Management, monitoring and fruit sampling details for the leafrollers *Choristoneura rosaceana* and *Pandemis limitata* in each organic (O1 - O5) and paired conventional (C1 - C5) apple orchard studied in Cawston, BC, Canada, 1997 - 1999.

	No. of pheromone dispensers / ha applied each year[2]			Yearly insecticide treatments for leafrollers[3]			Yearly number of traps for each leafroller species[4]			Number of fruit sampled for damage at harvest each year		
Orchard[1]	1997	1998	1999	1997	1998	1999	1997	1998	1999	1997	1998	1999
O1	500	500	500	0	0	0	4	4	4	3368	2145	2000
O2	500	500	500	0	0	0	6	6	6	5006	1756	2000
O3	500	500	500	0	0	0	4	4	4	4298	2100	2000
O4	500	500	500	0	0	0	2	2	2	4300	2223	2000
O5	500	500	500	0	0	0	4	4	4	3031	1939	2000
C1	0	0	0	0	1	1	–	2	2	2270	1698	2000
C2	0	0	0	3	3	4	–	2	2	2000	2000	2000
C3	0	0	0	2	2	3	–	2	2	2000	2000	2000
C4	0	0	0	1	1	0	–	2	2	2100	2000	2000
C5	0	0	0	2	3	4	–	2	2	2000	1883	2000

[1] Orchards O1 to O5 were cited by Judd and Gardiner (2005) as orchards A1 - A4 and B1, respectively, and orchards C1 - C5 remain the same across studies. Six untreated, organic, comparison orchards not listed were monitored but not sampled for damage (Cossentine *et al.* 2004).

[2] All trees on the perimeter of each orchard received the equivalent of 2000 dispensers / ha.

[3] Leafroller sprays consisted of spring (April) sprays of methidathion in dormant oil and summer (July - August) sprays of Confirm ® (tebufenozide).

[4] No leafroller monitoring was done in conventional comparison orchards in 1997 but they were sampled for damage at harvest. Six untreated, organic, comparison orchards (Cossentine *et al.* 2004) were each monitored with two traps in 1998 and 1999. Each species was monitored with a septal lure loaded with 3 mg of a multi-component blend described by Deland *et al.* (1994).

and Gardiner (2005). Six organic apple orchards that received no treatments for control of leafrollers were described in detail by Cossentine *et al.* (2004) as part of a study on parasitism of leafrollers in 1998 and 1999. The latter untreated orchards were used to compare relative trap catches of leafrollers only, as no damage data were collected in the original study. Briefly, all orchards ranged in size from 0.5 - 2 ha and were composed of mixed apple varieties planted at densities of 267 - 938 trees/ha with tree x row spacing of 2.4 - 6.1 x 3.0 - 6.1 m, respectively. Trees ranged in height from 2.5 - 3.5 m and were pruned using a pyramid shape training system.

No synthetic insecticides were applied to any of the organic orchards examined in this study (Table 1), but one orchard (O3) received Bt sprays (Dipel 2X DF) in 1998 and this is noted in the Discussion. All but one conventional orchard also received at least one application of Guthion® (azinphosmethyl at 0.84 kg a.i./ha) for codling moth control in 1997, but in later years growers used Confirm® (tebufenozide) during both spring and summer usually timed for control of leafroller larvae. One caveat is that conventional orchards did not necessarily receive similar or optimal insecticide

spray programmes because these orchards were chosen by Judd and Gardiner (2005) as local comparisons not controlled treatments.

Pheromone disruption treatment. Isomate-CM/LR is a brownish red, single-tube, polyethylene dispenser, manufactured by the Shin-etsu Chemical Company Ltd. (Tokyo, Japan) and marketed by Pacific Biocontrol Corporation (Vancouver, Washington). Each Isomate-CM/LR dispenser contains a 285 mg blend of 36.9% (*E,E*)-8,10-dodecadien-1-ol (codlemone), 1.8% isomers of codlemone, 6.0% dodecanol and 1.2% tetradecanol for disruption of codling moth, and 43.5% (*Z*)-11-tetradecenyl acetate (Z11-14:Ac) and 2.4% (*E*)-11-tetradecenyl acetate (*E*11-14:Ac) for disruption of leafrollers, plus 8.2% inert ingredients and pheromone stabilizers (Don Thomson, personal communication). Isomate-CM/LR dispensers were deployed in five organic apple orchards at a rate of 500/ha, however each perimeter tree received the equivalent of 2000 dispensers/ha (Table 1). Dispensers were attached to branches in the upper third of tree canopies ca. 1 m below the top of the central leader, or on the first lateral branch beneath the central leader. All pheromone dispensers were deployed a few days before codling moths were expected to emerge and no later than 8 May each year (Judd and Gardiner 2005).

Monitoring seasonal flight activity of moths. In all orchards, seasonal flight activity and capture of adult leafrollers were assessed using traps baited with synthetic pheromones. Disruption of pheromone communication in leafrollers was calculated by expressing catches of moths in pheromone-treated organic orchards as a percentage of catch in either insecticide-treated conventional orchards or untreated organic orchards in 1998 and 1999. No comparisons were made in 1997 as the latter sets of orchards were not monitored that season.

Depending on orchard size and shape, 2 - 6 Pherocon 1-C style open (5-cm side spacers) wing traps (Pherotech International, Delta, BC) baited with each species' multi-component pheromone blend (Deland *et al.* 1994; described below) were deployed evenly throughout each orchard (Table 1). One trap for each species was hung ca. 1.5 - 2.0 m above ground on different sides of the same tree in 2 - 6 separate trees. Positions for all traps remained fixed within and across years. In 1997 only the five Isomate-CM/LR-treated organic orchards were monitored with pheromone traps. On 30 May 1997, wing traps were deployed in each of these organic orchards and checked weekly from 6 June until 18 September. In 1998, wing traps were deployed in these same five Isomate-CM/LR-treated organic orchards, five paired insecticide-treated conventional orchards and six untreated organic orchards on 8 May, and checked weekly from 15 May until 25 September. In 1999, wing traps were deployed in all orchards on 27 May and checked weekly from 3 June until 30 September. Moths were counted and removed weekly with trap bottoms replaced as needed and pheromone lures changed every three weeks.

Synthetic multi-component pheromone lures for each leafroller species were prepared with chemical components (Aldrich Chemical Company Inc., Milwaukee, Wisconsin) of known purity, as confirmed by gas chromatographic analysis (*Z*11-14:Ac, 98% with 2% *E*11-14:Ac; (*Z*)-11-tetradecenal 96%; (*Z*)-11-tetradecanol; 97%, (*Z*)-9-tetradecenyl acetate, 96%) using published ratios (Roelofs *et al.* 1976, Vakenti *et al.* 1988). In making pheromone lures for each species, 200 µl of each multi-component pheromone blend was dissolved in dichloromethane and 3 mg of the pheromone blend was loaded into each red rubber septum (Aldrich Chemical Company Inc., Milwaukee, Wisconsin). After loading, septa were air-dried for ca. 18 h at 23 °C in a fume hood and stored at 0 °C until pinned to the inner side of trap lids in the field.

Assessment of mating in leafrollers. Mating was assessed using laboratory-reared, virgin, female moths placed in Teflon®-lined mating tables described by McBrien and Judd (1996). Both *C.*

rosaceana and *P. limitata* were reared on a modified pinto bean-based diet (Shorey and Hale 1965) at 24 °C and 16:8 h L:D photoregime. Female pupae of each species were placed individually in 150-ml plastic cups provided with moist cotton wicks until eclosion. Female moths aged 24 - 72 h were immobilized at 0.5 °C and one forewing and a tarsal tip were removed with fine forceps to prevent escape from mating tables. Females were kept chilled and transported to field sites in small ice chests.

In 1998, mating activity of both leafroller species was assessed weekly from 2 June until 3 September in each pheromone- and insecticide-treated orchard. During each weekly assessment, one female of each species was placed in 5 or 10 separate trees in each, insecticide- or pheromone-treated orchard, respectively, on Tuesdays, Wednesdays and Thursdays (n = 15 or 30 females/species/orchard/week). Two mating tables, each containing an individual female of each species, were placed in opposite sectors of the same tree, in the upper third of the canopy, several rows and trees distant from any pheromone traps. Females were placed in the field during the afternoon and removed the following morning to minimize predation and escape. Females recovered from the field <24 h after deployment were returned to the laboratory and each was dissected and examined for the presence of a spermatophore in the *bursa copulatrix* which indicates females have mated. Any females that were dead when collected from the field were omitted from the data.

In 1999, we conducted an experiment during flight of the first summer generation to determine if the probability of mating increased with the length of time (24 - 96 h) females were exposed in the field in either pheromone- or insecticide-treated orchards. Starting on Monday, 21 June, 100 female *P. limitata* were deployed in one pheromone-treated orchard and another 100 females were placed in an insecticide-treated orchard. At 24-h intervals for four consecutive days, 25 females were recovered from each orchard and returned to the laboratory where their mating status was assessed as before. This procedure was repeated for several consecutive weeks.

Fruit damage sampling. Depending on year and orchard, we examined fruit from 18 to 48 trees in the paired, pheromone- and insecticide-treated orchards (Table 1) using a stratified, cluster sampling procedure, where the outer row of trees and all interior trees represent two strata, and each tree represents a cluster of fruit, respectively (Judd *et al.* 1997). Sample trees were chosen systematically by crossing each orchard from corner to corner and edge to edge, ensuring that each variety and stratum was sampled. It was necessary to sample irregular numbers of trees and fruit from year to year because biennial bearing in organic orchards resulted in large annual differences in fruit set. All orchards were sampled during normal periods of harvest for each variety as fruit maturity and growers dictated. In most cases, a minimum of 100 fruit were removed from each sample tree by picking 50 low and 50 high fruit from south side branches. If there were fewer than 100 fruit on a tree, all fruit were removed from the sample tree. Early- or late-season leafroller damage, caused by either overwintering or summer larvae, respectively, can be distinguished by the degree of surface tunneling and scarring of fruit. Only late-season damage caused by summer-feeding larvae is scored in this study. All leafroller damage observed in this study was caused by either *C. rosaceana* or *P. limitata*, as no other species were previously found in this region (Madsen and Madsen 1980, Judd and Gardiner 2004). Our damage comparisons were limited to pheromone *vs.* insecticide-treated orchards because no damage samples were taken in the untreated organic orchards examined by Cossentine *et al.* (2004).

Statistical analyses. For each species, moth captures from all traps in the same orchard were pooled and transformed ($\log_{10}$ [x +1]) to normalize the data. Annual mean total number of moths caught per trap in untreated, Isomate-CM/LR-treated, and insecticide-treated orchards were compared

using an analysis of variance (ANOVA) followed by a Student Neuman Keuls' multiple comparisons test (Zar 1984), where orchards are treated as replicates (n = 5 or 6). Mean weekly and seasonal total percent mating of each species in pheromone- and insecticide-treated orchards in 1998 were compared using two-sample *t*-tests (n = 5). Linear regression analysis was used to relate mean weekly percent mating and mean weekly trap catches in 1998. The frequency of mating among females placed in the field for varying lengths of time in either a pheromone-treated or insecticide-treated orchard was compared weekly and seasonally using contingency tables and χ^2 tests or a binomial *Z*-test (Zar 1984). Mean percent leafroller damage at harvest for pheromone- and insecticide-treated orchards was compared annually using a two sample *t*-test following an arcsine $\sqrt{p}$ transformation of raw data. Linear regression analysis was used to relate mean percent damage at harvest in pheromone-treated orchards with mean seasonal cumulative moth catches using all three years of data (n = 15 data pairs) and to examine changes in moth catches and damage over time in organic orchards. All statistical tests were performed using SigmaStat® (Version 3.0.1, SPSS Software Inc., San Jose, California) and an experimental error rate of $\alpha = 0.05$.

RESULTS

Seasonal flight activity of leafrollers. Mean weekly catches of *C. rosaceana* and *P. limitata* in orchards under different treatment regimes in 1999 are shown in Fig. 1. Similar weekly catches were seen in 1997 and 1998 but for brevity data are not shown. Catches of both species reflect two adult flight periods representing the first and second generations in this region, respectively. Weekly catches of *C. rosaceana* under all treatment regimes tended to be smaller during second generation than those during first generation, but this trend was often reversed in *P. limitata* (Fig. 1). *P. limitata* appeared to be the most abundant leafroller species in conventional orchards, but the relative species makeup reversed itself annually in untreated and pheromone-treated organic orchards (Table 2).

Disruption of leafroller pheromone trap catches. Weekly catches of *C. rosaceana* were reduced 70 - 100% in the Isomate–CM/LR treatment relative to both untreated and insecticide-treated orchards in 1999 (Fig. 1), and relative reductions averaged 90.4 and 92.1%, respectively, across years in 1998 and 1999 (Table 2). Weekly catches of *P. limitata* were reduced 67 - 100 % by treatment with Isomate-CM/LR compared to untreated and insecticide-treated orchards (Fig. 1), and relative reductions averaged 92.5 and 87.2 %, respectively, across years (Table 2). Catches of *P. limitata* were always higher in insecticide-treated conventional orchards compared with untreated organic orchards (Table 2).

Mating in leafrollers. During the entire 1998 season, 35.1% of female *C. rosaceana* (n = 757 females recovered) and 57.4% of female *P. limitata* (n = 702) mated on mating tables hung in the insecticide-treated orchards (Fig. 2). If 1 August is used as an approximate starting point for second-generation flight activity in both species (Fig. 2), then mating of both species tended to increase during the second-generation flight period. In the insecticide-treated orchards mating of *C. rosaceana* during the first (26.8%) and second generation (54.3%) was significantly different (χ^2 = 23.01, df = 1, $P < 0.001$). However, mating of *P. limitata* during first (53.6 %) and second generation (65.3%) was not significantly different (χ^2 = 2.05, df = 1, P = 0.152).

During the entire 1998 season only 1.6% of female *C. rosaceana* (n = 1610) and 7.4% of female *P. limitata* (n = 1522) mated on mating tables hung in the Isomate-CM/LR-treated orchards (Fig. 2). As seen in the absence of pheromone disruption (Fig. 2), significantly more *C. rosaceana* mated during second generation (3.5%) than during first generation (0.75%)

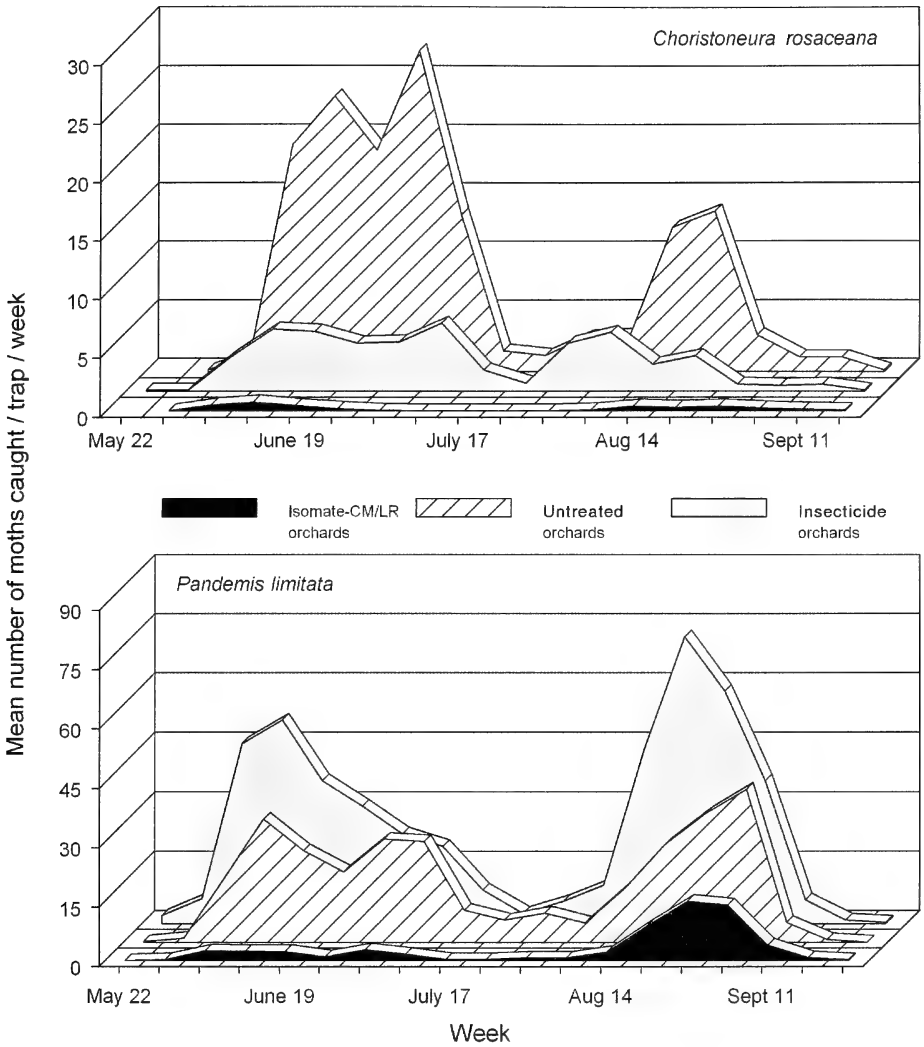

Figure 1. Mean weekly catches of two leafroller species in their species-specific synthetic pheromone-baited traps hung in untreated organic apple orchards (n = 6), Isomate-CM/LR-treated organic apple orchards (n = 5) and insecticide-treated conventional apple orchards (n = 5) in 1999.

(χ^2 = 14.74, df = 1, P < 0.001) and mating of *P. limitata* was also significantly greater in the second generation (15.4%) than in the first generation (2.9%) in pheromone-treated orchards (χ^2 = 65.14, df = 1, P < 0.001).

Regression analyses (Fig. 3) indicate that weekly differences in mating of *C. rosaceana* and *P. limitata* in the insecticide-treated orchards were partially correlated with the differences in weekly catches of males in pheromone-baited traps, respectively. In the Isomate-CM/LR-treated orchards, weekly differences in mating of female *P. limitata* were explained (see r^2 values) by differences in weekly catches of males (Fig. 3), but both mating and catches of *C. rosaceana* were too low in pheromone-treated orchards to ascribe any significant relationship to these variables (P = 0.06).

Estimates of mating in *P. limitata* did not appear to increase with increasing time deployed in the orchard (Table 3). Comparing results in Table 3 (1999) and Fig. 2 (1998), it appears that percent mating of

Table 2.
Seasonal total number of male leafroller moths, *Choristoneura rosaceana* and *Pandemis limitata*, caught in synthetic pheromone-baited traps placed in untreated organic orchards (n = 6), insecticide-treated conventional orchards (n = 5) and Isomate-CM/LR-treated organic orchards (n = 5) and relative percent disruption of trap catches.

Year	Species	Mean ± SE total number of moths / trap / year / treatment[1]			Relative % trap catch reduction[2]	
		Untreated	Insecticide	Isomate-CM/LR	Untreated	Insecticide
1997	*C. rosaceana*	–	–	124.2 ± 187.6	–	–
	P. limitata	–	–	84.7 ± 103.7	–	–
1998	*C. rosaceana*	137.0 ± 45.1a	56.3 ± 21.6b	3.8 ± 2.1c	93.3	97.2
	P. limitata	282.6 ± 33.5b	460.3 ± 173.7a	66.2 ± 35.3c	85.6	76.6
1999	*C. rosaceana*	211.7 ± 37.1a	150.1± 73.1a	27.8 ± 11.3b	87.5	86.9
	P. limitata	101.7 ± 28.5b	386.8 ± 155.2a	2.3 ± 0.8c	99.4	97.7
Mean 1998-1999	*C. rosaceana*	174.4 ± 37.4a	103.2 ± 46.9a	51.9 ± 36.8b	90.4 ± 4.6	92.1± 5.1
	P. limitata	192.2 ± 90.5b	423.6 ± 36.7a	51.1 ± 24.9c	92.5 ± 4.8	87.2 ± 10.6

[1] Means within a row a followed by different letters are significantly different ($P < 0.05$) by Student Neuman Keuls' multiple comparisons test following significant ($P < 0.05$) ANOVA.
[2] Percent trap catch reduction in Isomate-CM/LR-treated organic orchards relative to catches in untreated organic orchards or insecticide-treated conventional orchards.

female *P. limitata* in the insecticide-treated orchards was higher during the first generation in 1999 (82.9%) compared with 1998 (53.6%), while percent mating (2.9%) during the first generation in the Isomate-CM/LR-treated orchards was identical in 1998 (Fig. 2) and 1999 (Table 3).

Disruption of mating in leafrollers. When the entire 1998 season is considered (Fig. 2), Isomate-CM/LR significantly reduced mating of *C. rosaceana* by 95.4% (χ^2 = 378.47, df = 1, $P < 0.001$) and mating of *P. limitata* by 87.1% (χ^2 = 376.78, df = 1, $P < 0.001$) relative to their mating in insecticide-treated orchards, respectively. Disruption of mating in both species tended to be lower during the second-generation flight period, dropping to 93.5% in *C. rosaceana* and 76.4% in *P. limitata*, respectively. Even though percent mating of *P. limitata* in the insecticide-treated orchards was lower during the first generation of 1998 (Fig. 2) than 1999 (Table 3), disruption of mating by treatment with Isomate-CM/LR was similar in 1998 (95.7%) and 1999 (96.4%).

Fruit damage. Summer leafroller damage in pheromone-treated organic orchards ranged from 1.7 - 24.8% in 1997, 0.4 - 4.2% in 1998, and 0.4 - 13.7% in 1999 (Fig. 4A). From 1997 - 1999 leafroller damage declined in 4 of the 5 organic orchards and in 12 out of 15 orchard-years damage was less than 5% at harvest under Isomate-CM/LR treatment. Downward trends in leafroller damage over three years appeared correlated with downward trends in total catches of leafrollers in each orchard, respectively (Fig. 4B). There was a significant correlation (r = 0.65, $P < 0.009$) between total leafroller trap catches and damage at harvest, but catches of leafrollers only explained 42.3% of the variation in harvest damage (Fig. 4C). Comparison of leafroller damage in five organic orchards under management with Isomate-CM/LR, and five conventional orchards under various insecticide programmes is shown in Fig. 5. Damage levels were not significantly different between the two groups of pheromone- and insecticide-treated orchards in 1998 and 1999 (Fig. 5).

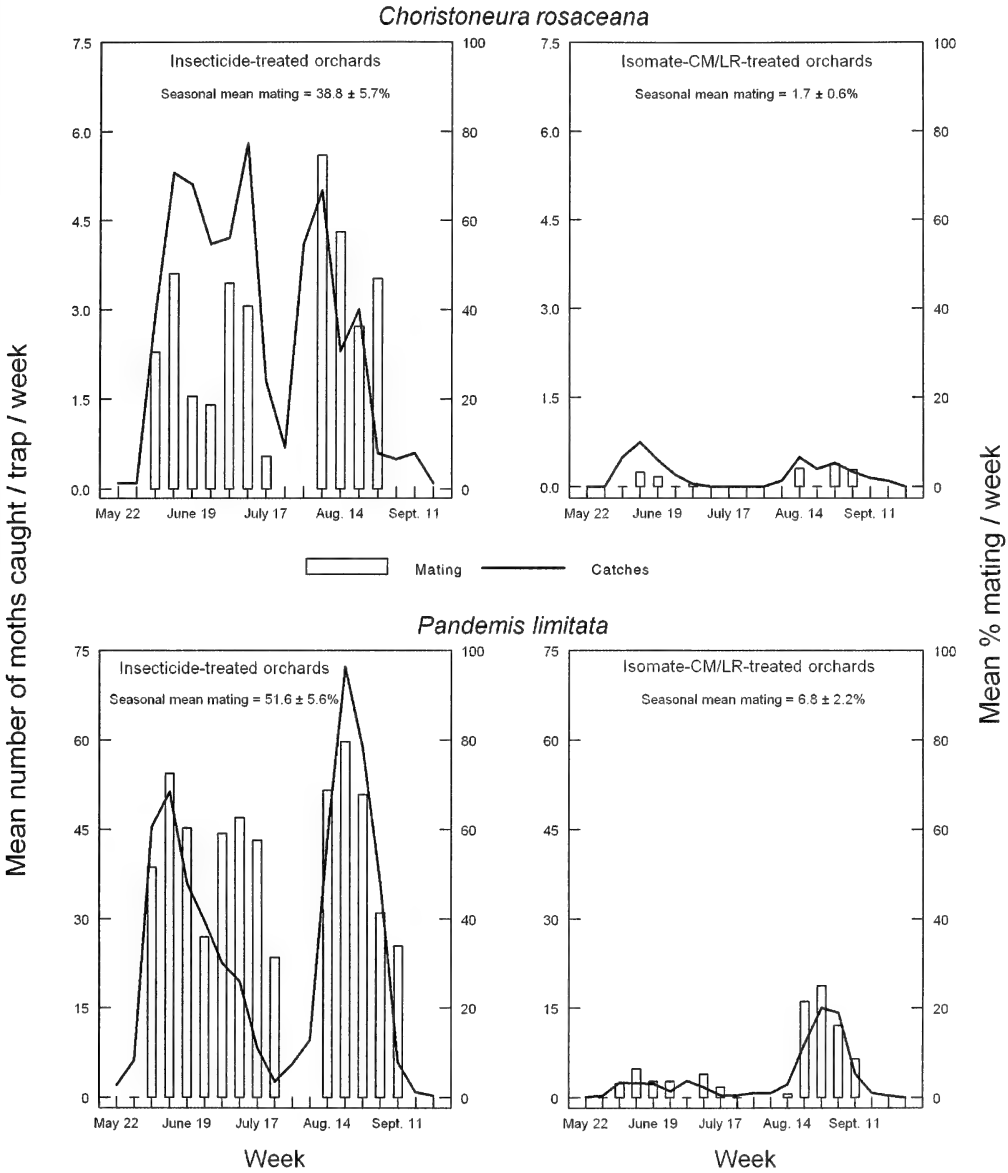

Figure 2. Observed mean weekly percent mating of two leafroller species on mating tables hung in Isomate-CM/LR-treated organic apple orchards (n = 5) and insecticide-treated conventional apple orchards (n = 5) and mean weekly catches of moths in synthetic pheromone traps in the same orchards in 1998.

DISCUSSION

Previously we showed that Isomate-CM/LR was a useful spring-time supplement for the codling moth SIT programme in BC (Judd and Gardiner 2005), but its additional benefits as a supplement for control of leafrollers in organic orchards was not described. This study has demonstrated that pheromone communication and mating in sympatric leafroller moths commonly found infesting organic apples in the Similkameen Valley of BC can be effectively and simultaneously disrupted by releasing a mixture

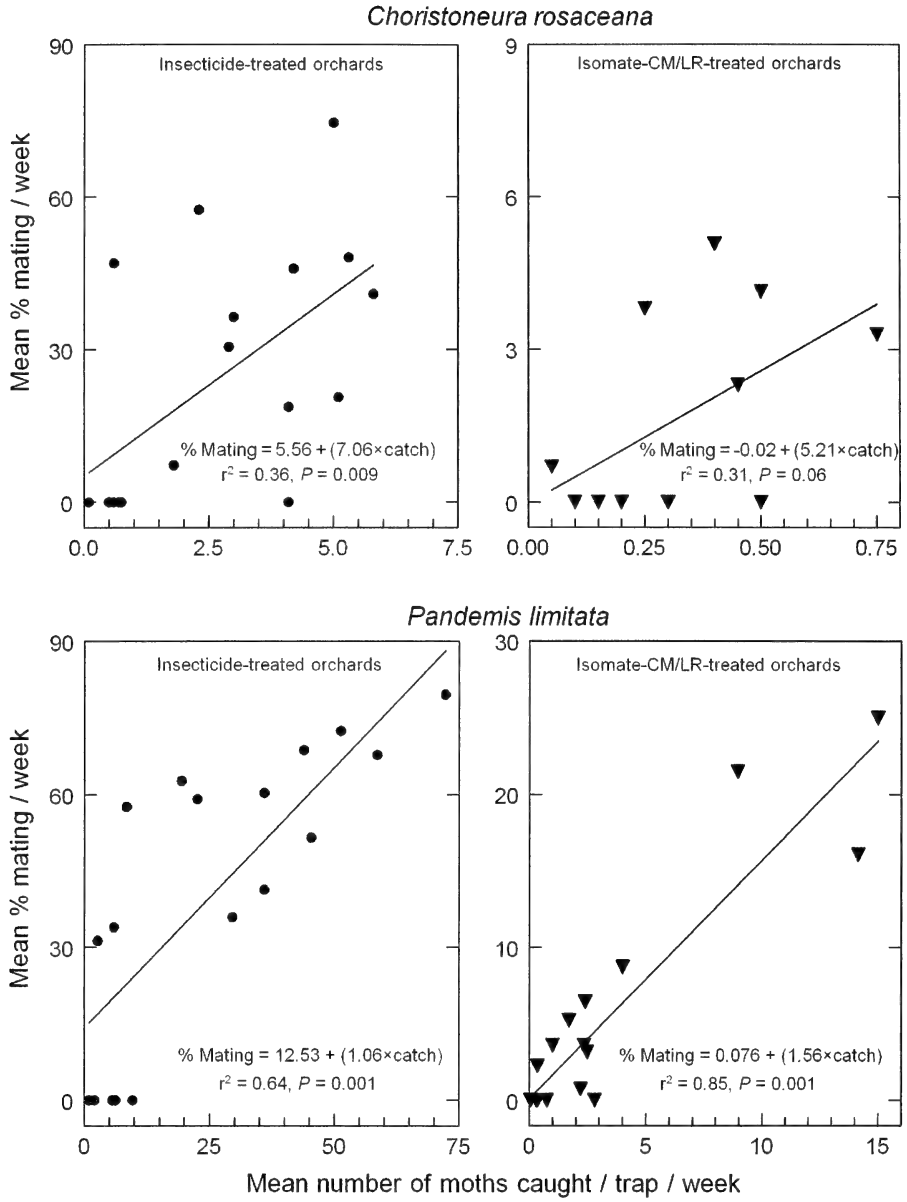

Figure 3. Regression analyses showing relationships between relative moth density (trap catches) and mating in two leafroller species in Isomate-CM/LR-treated organic apple orchards (n = 5) and insecticide-treated conventional apple orchards (n = 5) in 1998.

of their major pheromone components from Isomate-CM/LR. Season-long reductions of pheromone trap catches of both *C. rosaceana* and *P. limitata* with Isomate-CM/LR were comparable to levels seen in several studies examining each species individually (Deland *et al.* 1994, Agnello *et al.* 1996, Knight *et al.* 1998, Knight and Turner 1999, Trimble and Appleby 2004) and greater than levels observed in other studies (Lawson *et al.* 1996). There is no generally accepted level for the reductions in pheromone trap catches often observed using mating disruption that correlate with crop protection, but the observation has been made that this reduction is usually 97 - 100% in species where disruption appears to be an effective crop-protection tool

Table 3.

Percentage of female *Pandemis limitata* mating in mating tables when placed in insecticide-treated conventional and Isomate-CM/LR-treated organic apple orchards for increasing lengths of time during first-generation flight in 1999.

Hours females were in field	Insecticide-treated orchard		Isomate-CM/LR-treated orchard		% mating reduction[3]
	n females[1]	% mated[2]	*n* females[1]	% mated[2]	
24	274	86.5a	246	2.03a	97.6*
48	274	75.9a	252	4.36a	94.3*
72	242	80.6a	224	3.12a	96.1*
96	220	79.5a	220	2.27a	97.1*
Total (24 - 96 h)	1010	82.9	942	2.97	96.4*

[1] *n* = total number of live females recovered from field in test period.

[2] Percentages within a column followed by the same letter are not significantly different ($P > 0.05$) based on a χ^2 test of the null hypothesis of equal mating frequencies across time categories.

[3] Asterisk indicates a significant ($P < 0.001$) reduction in mating based on a comparison of paired proportions of mating within a row using a binomial *Z*-test (Zar 1984).

(Trimble and Appleby 2004). In many mating-disruption studies on *C. rosaceana* (Reissig *et al.* 1978, Deland *et al.* 1994, Agnello *et al.* 1996, Lawson *et al.* 1996, Knight *et al.* 1998, Trimble and Appleby 2004), pheromone treatments have resulted in less than 97% reduction in pheromone trap catches relative to catches with the same traps in insecticide-treated orchards. In our study, a reduction of this magnitude was achieved in 1998 when fewer than 4 moths were caught / trap / year (Table 2) and a similar reduction was observed for *P. limitata* in 1999 when catches of this moth averaged fewer than 3 moths / trap / year. Knight and Turner (1999) found a significant negative relationship between mean catches of *Pandemis* spp. / trap and percent reduction of catches in synthetic pheromone traps. A similar relationship has not been reported for *C. rosaceana*, but our data (Table 2) reflect this type of trend for both species.

Presumably, reductions in pheromone trap catches are correlated with reductions in mating, but this is almost never confirmed in mating-disruption studies because measures of mating are often missing. Actual reductions in female mating should be more directly correlated with reductions in damage from larvae than reductions in males caught in pheromone traps. We found relatively large correlations between trap catches, a relative measure of population density, and mating of *P. limitata* on mating tables (Fig. 3), but only a weak correlation was found for *C. rosaceana* in the insecticide-treated orchards, and no correlation was found for this species in the pheromone-treated orchards (Fig. 3). Given that catches of *P. limitata* represented about 50% of the total leafroller catch in organic orchards during 1997 - 1999, and total catches of leafrollers declined in Isomate-CM/LR-treated orchards each year (Table 2), a significant, albeit weak, relationship between total leafroller catches and damage may be expected (Fig. 4C). A better relationship might be observed in orchards having populations of *P. limitata* only. Nevertheless, the relationship shown in Fig. 4C is consistent with the view that the reduction in trap catches needs to be close to 99% (ca. 6 moths / trap / season) to ensure damage from leafrollers is 1% or less, an acceptable level for organic apple producers in BC.

We acknowledge that the relationship we have shown between trap catches in pheromone-treated organic orchards and damage from summer-feeding leafrollers may not hold true for conventional orchards in this region, because there are significant

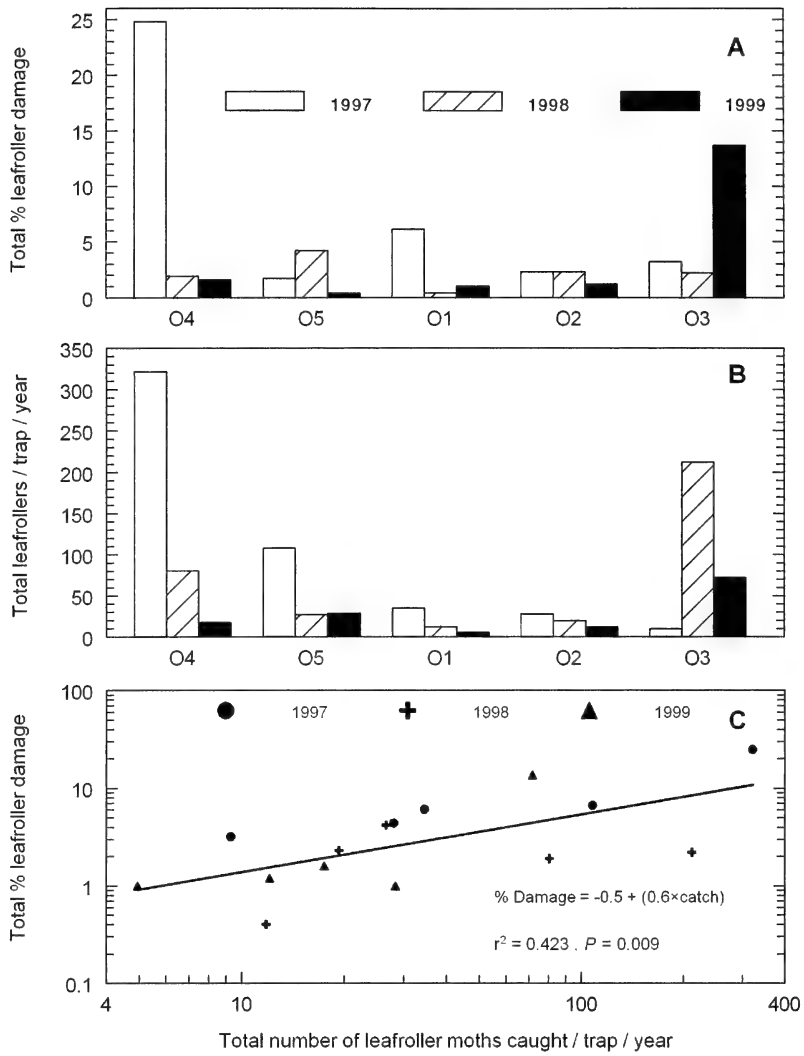

Figure 4. Percentage of damage at harvest caused by summer-feeding leafroller larvae in each Isomate-CM/LR-treated organic apple orchard by year (A), total number of leafroller moths (*Choristoneura rosaceana* and *Pandemis limitata*) caught in synthetic pheromone traps in each Isomate-CM/LR-treated organic apple orchard by year (B), and linear regression of the observed relationship between damage and total leafroller catches (log scales) in 1997 - 1999 (C).

differences in the levels of biological control in these different production systems. In a study running parallel to ours, Cossentine *et al.* (2004) found that summer larval populations of both *C. rosaceana* and *P. limitata* in untreated organic apple orchards in the Similkameen Valley experienced parasitism rates as high as 68% during 1998 and 1999, and higher levels of parasitism were observed as leafroller populations declined. Given these observations, if mating disruption was causing leafroller populations to decline then it might be expected to increase the impact of parasitoids and reduce damage. The general absence of these natural control agents in local conventional orchards (Joan Cossentine, personal communication) may invalidate any application of an established relationship between trap catches and damage from organic orchards where natural controls are also acting. Mating disruption of leafrollers in conventional orchards may have to be augmented with insecticides,

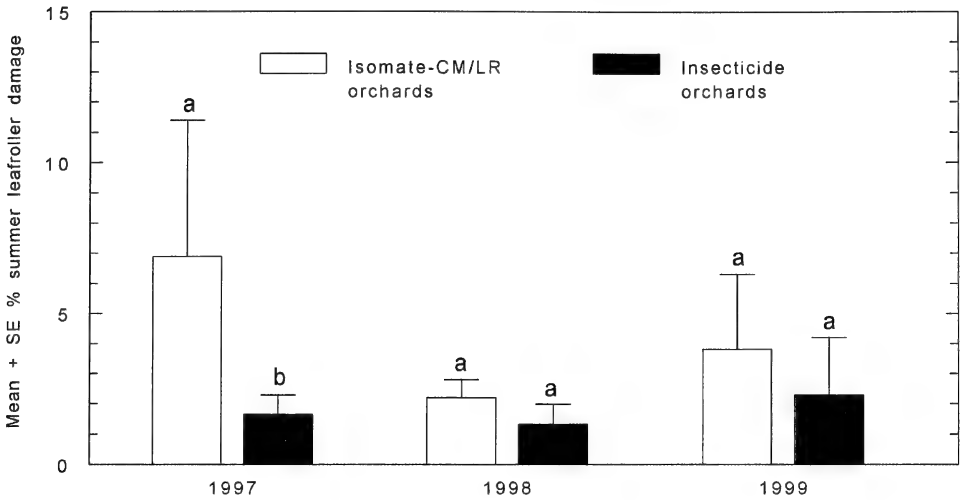

Figure 5. Comparison of mean (+ SE) percent damage at harvest caused by summer-feeding leafroller larvae in Isomate-CM/LR-treated organic apple orchards (n = 5) and insecticide-treated conventional apple orchards (n = 5) in 1997 -1999. Paired means with the same letter superscript are not significantly different ($P > 0.05$) by two-sample t-test.

particularly where *C. rosaceana* is the dominant species. With one exception (Trimble and Appleby 2004), limited use of selective insecticides in combination with mating disruption has provided a small improvement in the control of *C. rosaceana* over mating disruption alone (Agnello *et al.* 1996, Lawson *et al.* 1996, Knight *et al.* 1998, Knight *et al.* 2001).

To the best of our knowledge this study is the only evaluation of mating disruption of *C. rosaceana* and *P. limitata* with Isomate-CM/LR in organic production systems where no insecticides were applied. This technique holds promise for organic pome fruit producers in the Similkameen Valley, especially if *P. limitata* is the dominant species. Our results are in sharp contrast to a failure of mating disruption to keep damage from *C. rosaceana* below economic levels in other regions even when used in conjunction with pesticides (Agnello *et al.* 1996, Lawson *et al.* 1996). This failure has been attributed to high population density and potential immigration of adults and larvae into treatment areas. Population density should be an important factor in limiting the efficacy of mating disruption, but this has seldom been shown experimentally. Our data certainly point to a strong relationship between relative adult numbers, mating success and harvest damage, but moth catches in our pheromone-treated orchards were often greater than those reported elsewhere so it is difficult to reconcile our results on the basis of adult population differences alone. Four of the five organic orchards in this study were somewhat isolated by having other orchards located on only one border. Orchard O3 was the only one that was surrounded by adjacent orchards, particularly cherries, which were not treated for leafrollers, and it was the one orchard in which we saw a significant increase in catches of *P. limitata* late in 1998 and damage in 1999 (Fig. 4). Interestingly this was the only orchard that received a petal-fall spray of Bt in spring of 1998. As noted by Knight *et al.* (1998), immigration may be an important constraint on use of mating disruption for leafrollers, but monitoring may help to allay some of this concern if it can predict immigration of adults, as it did in orchard O3.

Although damage in the organic orchards was comparable to that seen in some comparison insecticide-treated orchards (Fig. 5), we make no claim that mating disruption is as effective as an optimal insecticide-based control programme. However, we are of the opinion that the efficacy of mating disruption against any species is

best evaluated over several years. Mating disruption is certainly a less robust control technique than most insecticides, and more constrained by population density than the latter. Suppression of codling moth populations using mating disruption often takes several years and this will probably be true of leafrollers (Fig. 4). Depending on the comparison orchards chosen, it is possible for mating-disruption technology to look very effective, or highly ineffective; a better approach may be to examine its long-term effects in the same locations over several years and compared with standard systems as has been used for codling moth (Charmillot 1990, Judd *et al.* 1997). The long-term impact of mating disruption on biological control of leafrollers in orchards (Cossentine *et al.* 2004) relative to standard controls also needs to be considered.

In areas of BC outside the Similkameen Valley, control of leafrollers using pheromone-based mating disruption requires a multi-species approach if different complexes of sympatric leafrollers are to be controlled effectively (Judd and McBrien 1995). For example, in the Okanagan and Creston Valleys of BC, eye-spotted bud moth, *Spilonota ocellana* (Denis and Schiffermüller), European leafroller, *Archips rosanus* (L.) and fruit-tree leafroller, *Archips argyrospilus* (Walker) are also important pests of apple. The latter two species use *Z*11-14:Ac as the major component in their multi-component pheromone blends (Arn *et al.* 1982) and small-plot studies (Deland 1992) demonstrated that pheromone communication and mating in *A. rosanus* and *A. argyrospilus* could be disrupted effectively with a pheromone blend similar to that used in Isomate-CM/LR. *Spilonota ocellana*, however, uses (*Z*)-8-tetradecenyl acetate as its major pheromone component (Arn *et al.* 1982) and would not be controlled by Isomate-CM/LR, but can be controlled by mating disruption (McBrien *et al.* 1998). An Isomate dispenser containing this added ingredient is currently under study as a mating-disruption system for control of the entire leafroller complex found in organic pome fruit orchards in BC. With an organic formulation of spinosad (Entrust®) registered in Canada during 2005, the combined use of this insecticide and mating disruption in organic orchards also warrants study, because the impact of spinosad on parasites of leafrollers in these systems needs to be considered carefully.

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Climate change and potential selection for non-diapausing two-spotted spider mites on strawberry in southwestern British Columbia

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ABSTRACT

A validated model of the timing of post-diapause oviposition in the two-spotted spider mite, *Tetranychus urticae* Koch, was used to predict when 50% of strawberry leaflets with *T. urticae* also have *T. urticae* eggs ($IO_{0.5}$) in each year from 1954 to 2006 at Langley, British Columbia. This timing was studied in relation to hours of frost occurring before and after oviposition. Historically, $IO_{0.5}$ occurred before there were frost-free days, but there was a clear threshold at 390 h with temperatures < 0 °C after $IO_{0.5}$, which was not exceeded. This suggests that there is selection pressure for early oviposition, but also a limit to the extent of selection. The subzero temperature profile ~1 month before oviposition was clearly different from that after $IO_{0.5}$. The number of hours with subzero temperatures 1 month before oviposition, and the standard deviation of those estimates, were negatively correlated with year and indicated that there could be oviposition in January - rather than February - by 2015. Cumulative hours with temperatures < 0 °C between 27 November (the empirical estimate of the time when *T. urticae* begins accumulating degree-days for post-diapause oviposition) and 30 April was negatively correlated with year, and extrapolation of a linear regression suggested that there could be selection for continuous annual oviposition by 2050. There was considerable variation in the data, but considered in combination with published evidence for climate change, these results will be important in developing pest management strategies, and furthermore, will impact many aspects of agriculture in the Fraser Valley of British Columbia.

INTRODUCTION

Global warming (Intergovernmental Panel on Climate Change 2007) will probably affect arthropod ranges (Gray 2004; Logan and Powell 2004; Gutierrez et al. 2006; Musolin 2007) assuming fixed biological tolerances to environmental conditions. At the same time, within a home range, it will probably also affect the life history characteristics of arthropods through selection (cf. Bradshaw *et al.* 2004). Winter diapause is a key feature of temperate arthropods (Danks 2006). Diapause characteristics will probably be affected by global warming, and changes may become evident first in areas that have a relatively mild but temperate climate, such as the Fraser Valley of British Columbia (B.C.), Canada. This study considers the effects of climate change on the timing of initial, post-diapause oviposition (IO) by two-spotted spider mites, *Tetranychus urticae* Koch (Acari: Tetranychidae), on strawberry (*Fragaria* x *ananassa* Duch. Rosaceae).

T. urticae females have a facultative reproductive diapause. Diapause is induced in the pre-imaginal stages, particularly at the end of the protonymphal instar, by short-day photoperiods (Veerman 1977a). Termination is dependent on duration of cold rest, temperature, and photoperiod during the first few months of diapause (Veerman 1977b). During winter, after photoperiodic sensitivity is gone, diapause

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would be sustained and prolonged by low temperatures. Under long days at relatively high temperatures, diapause can be terminated without a cold rest period (Veerman 1977b), but under short days a cold rest period is required.

Given a warmer climate there should be selection for individuals who reproduce continuously, despite short days, and there may be selection for individuals who do not respond to the initial photoperiodic induction cues. This idea is supported by the fact that there is considerable variation in diapause induction and termination characteristics among *T. urticae* strains, probably associated with adaptation to local conditions (Takafuji *et al.* 1991; Koveos *et al.* 1999).

Raworth (2007) developed and validated a FORTRAN program (ProgIO) that determined the timing of post-diapause oviposition – the day when 50% of strawberry leaflets with *T. urticae* also had *T. urticae* eggs ($IO_{0.5}$). The oviposition model for the program was based on field samples from inland and coastal sites, collected during 1988 to 2006; estimates of $IO_{0.5}$ from 10 populations during 1988 to 2003 were used to calibrate the model, and six independent populations during 2004 to 2006 were used for validation.

Here, ProgIO and historical weather data are used to examine 1) where, historically, *T. urticae* has placed $IO_{0.5}$ with respect to frost, and 2) the annual historical trends in hours of frost. These trends are then used to predict when *T. urticae* can be expected to commence oviposition in January – rather than the current timing in February – and when *T. urticae* may go through the winter with no reproductive diapause. It may be objected that viable leaves must be available for feeding and oviposition; however, in this system mature green leaves overwinter, and the mites usually initiate feeding and oviposition on these leaves in February and March (Raworth 2007).

MATERIALS AND METHODS

ProgIO and meteorological data were used to predict $IO_{0.5}$ each year from 1954 to 2006. ProgIO first calculated the temperature under a strawberry leaf in a commercial field at Langley, B. C. based on hourly temperature and cloud opacity data measured by Environment Canada at Abbotsford, B. C. since 1953, and calibration equations (Raworth 2007):

1. Cloud opacity = 1 (full cloud)

a. 0800-1600 h: $y = -8.756 + 1.009\ t + 1.731\ h - 0.013\ t^2 - 0.076\ h^2 + 0.015\ t \times h$

$R^2 = 0.92$, $P < 0.0001$, 425 d.f.

b. 1700-0700 h: $y = -1.104 + 0.864\ t - 0.025\ h + 0.010\ t^2$

$R^2 = 0.92$, $P < 0.0001$, 862 d.f.

2. Cloud opacity = 0 (full sun)

a. 0800-1600 h: $y = -54.250 + 1.116\ t + 9.515\ h - 0.370\ h^2$

$R^2 = 0.85$, $P < 0.0001$, 241 d.f.

b. 1700-0700 h: $y = -3.848 + 1.153\ t + 0.066\ h + 0.023\ t^2$

$R^2 = 0.92$, $P < 0.0001$, 554 d.f.

where: y = temperature experienced by the mites; t = temperature in a Stevenson Screen in the field ($t = -1.141 + 1.043\ tec$, where tec = air temperature at the Environment Canada station); and h = hour (where: 0800-1600 h = 8, 9,...16; and 1700-0700 h = 17, 18,...24, 25, 26,...31). Temperatures under a leaflet in intermediate cloud conditions were determined by linear interpolation between the results provided by the equations for full cloud and full sun. The timing of $IO_{0.5}$ was then determined from thermal summations > 9.4 °C starting on an empirically-derived day, 27 November (Raworth 2007), and a thermal requirement (y) that was negatively correlated with accumulated cold-rest hours < 4 °C (x) summed from 27 November (Raworth 2007):

3. $y = 78.3 - 0.0279\ x$; $r^2 = 0.83$, $P = 0.01$, 4 d.f.

Equation 3 implies that, for equivalent rates of thermal summation, the spider mites will stay in diapause longer if they have had insufficient cold rest. The timing

of $IO_{0.5}$ was compared graphically with the cumulative daily hours of frost below 0 °C summed between 27 November and 30 April of the following year.

To determine what temperature conditions the mites have avoided, annual frequency was plotted against the number of hours of frost below 0, -1, -2,...-10 °C that remained after $IO_{0.5}$ (3-d plot), and the hours of frost below 0, -1, -2,...-10 °C that occurred one month before mite eggs would be observed in the field, 10 January to 10 February (3-d plot). To determine when conditions would be suitable for earlier oviposition by *T. urticae*, the annual sum of hours of frost below 0, -2, -4,...-10 °C that occurred between 10 January and 10 February were regressed (SAS Institute 2004) against year and the regression was extrapolated beyond 2006. Changes in variability as a function of year were determined by pooling the latter data for each decade and regressing 1 SD for mean hours below a given temperature in that decade against the median year. Finally, a similar technique was used to determine when conditions would be suitable for *T. urticae* to pass through the winter without reproductive diapause based on the annual sum of the hours of frost below 0 °C between 27 November and 30 April.

RESULTS AND DISCUSSION

Cumulative hours of frost below 0 °C between 27 November and $IO_{0.5}$, from 1954 to 2006 varied between 400 and 1600 h (Fig. 1B, triangles). Despite this variation, $IO_{0.5}$ always occurred before frost-free days had begun (Fig. 1A, B), but with a clear upper threshold of 390 h of frost remaining after $IO_{0.5}$. This indicates sensitivity to frost, because all the estimates of $IO_{0.5}$ are clustered below 390 h; it also indicates some selection pressure for early oviposition, because the mites do not wait until frost-free conditions occur. As long as a female's progeny can survive and go on to reproduce, a female that initiates reproduction early in the season should have a numerical advantage over one that initiates reproduction later. This result should be qualified. The 390 h threshold was determined from a model of post-diapause oviposition in southwestern B. C.; it would not be expected to apply to *T. urticae* populations that are adapted to different local conditions in other temperate regions. Such a generalization would require further research.

Comparison of the subzero temperature profiles before and after $IO_{0.5}$ suggests that *T. urticae* in southwestern B. C. is able to tolerate >200 h at -2 °C, but only 20 h at -6 °C (Fig. 2). Lower temperatures for longer periods (Fig. 3) have been avoided. However, the number of hours with temperatures below 0, -2, ... -10 °C, 1 month before *T. urticae* normally commences oviposition, have declined significantly since 1954 (Fig. 4). Taking -8 °C as a critical temperature associated with no oviposition (Fig. 2), the data indicate that an average year will have zero hours <-8 °C by 2015 (Fig. 4). At this time there could be reduced selection pressure against emergence, and hence oviposition in January rather than February. Because the regression predicts hours at a given temperature in an average year, one would expect some hours at temperatures <-8 °C in some years, and possible mortality of early-emerging mites. However, the variation in subzero temperatures during this 1 month period has also declined since 1954 (Fig. 5) so that there will be less uncertainty about subzero conditions in 2015 than there was in 1957, and reduced selection pressure against early emergence. The regression of total hours of frost below 0 °C from 27 November to 30 April against year suggests that the number of hours of frost will decrease to the threshold of 390 h by ~2050 (Fig. 6). At this point, selection for early oviposition could result in reproduction by some individuals right through the winter in southwestern B.C.

The objective of this study was to examine general patterns in the initiation of post-

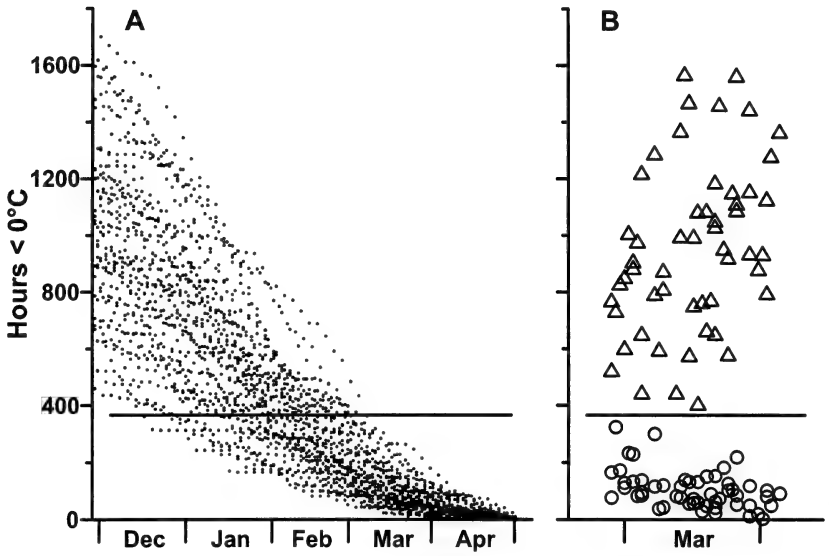

Figure 1. Cumulative hours with temperatures < 0 °C under a strawberry leaflet in a commercial field from: (A) start date to 30 April versus start date (dots); (B) 27 November to the date when 50% of strawberry leaflets with *T. urticae* also have *T. urticae* eggs ($IO_{0.5}$) versus $IO_{0.5}$ (triangles); and (B) $IO_{0.5}$ to 30 April versus $IO_{0.5}$ (circles), each year from 1954 to 2006. The horizontal line represents a threshold number of hours < 0 °C remaining after $IO_{0.5}$.

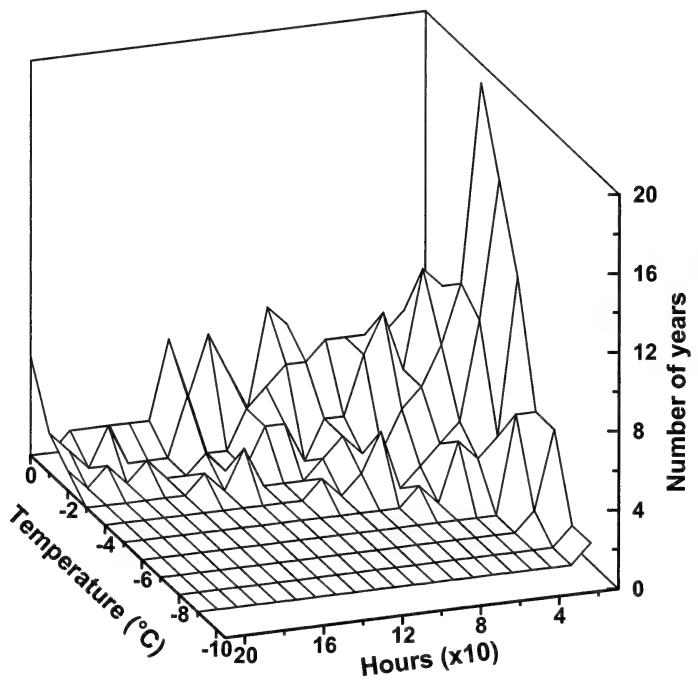

Figure 2. Number of years with a given number of hours at subzero temperatures under a strawberry leaflet in a commercial field, after the date when 50% of strawberry leaflets with *T. urticae* also have *T. urticae* eggs.

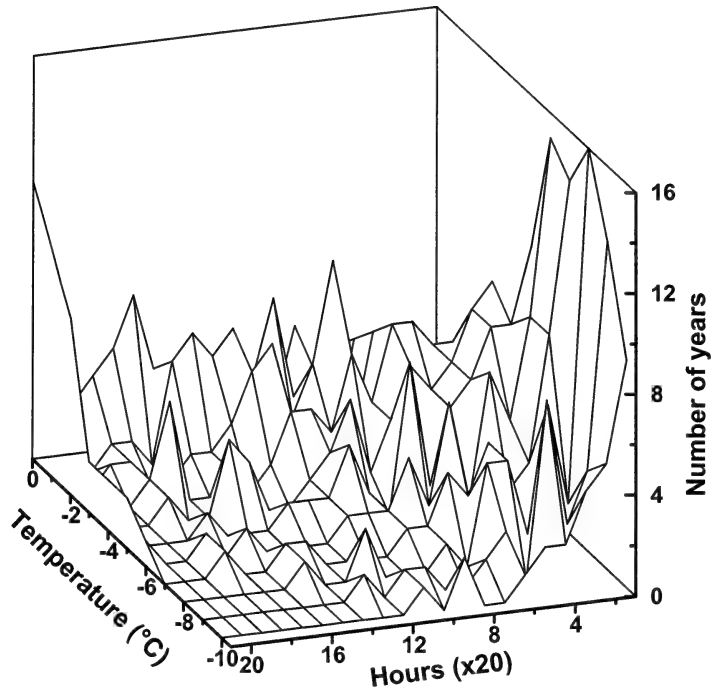

Figure 3. Number of years with a given number of hours at subzero temperatures under a strawberry leaflet in a commercial field, ~1 month before the date when *T. urticae* commences oviposition (10 January to 10 February).

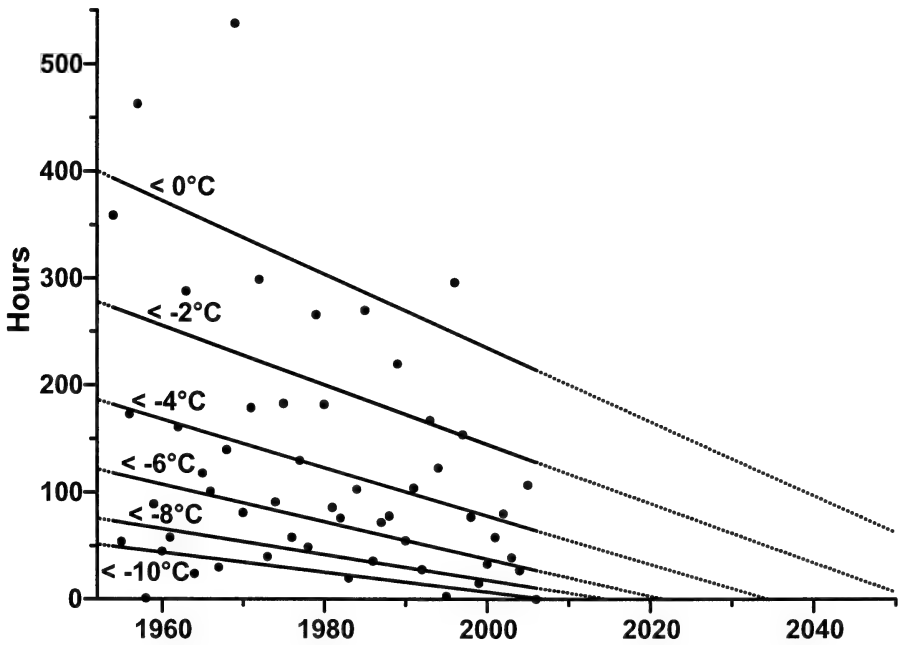

Figure 4. Hours with subzero temperatures under a strawberry leaflet in a commercial field, between 10 January and 10 February, versus year. Solid lines are linear regressions (r^2 = 0.10, 0.08, 0.08, 0.09, 0.10, 0.14 and P = 0.02, 0.03, 0.03, 0.02, 0.02, 0.004) for temperatures < -10, -8, … 0, respectively; data for temperatures <-4 °C are shown.

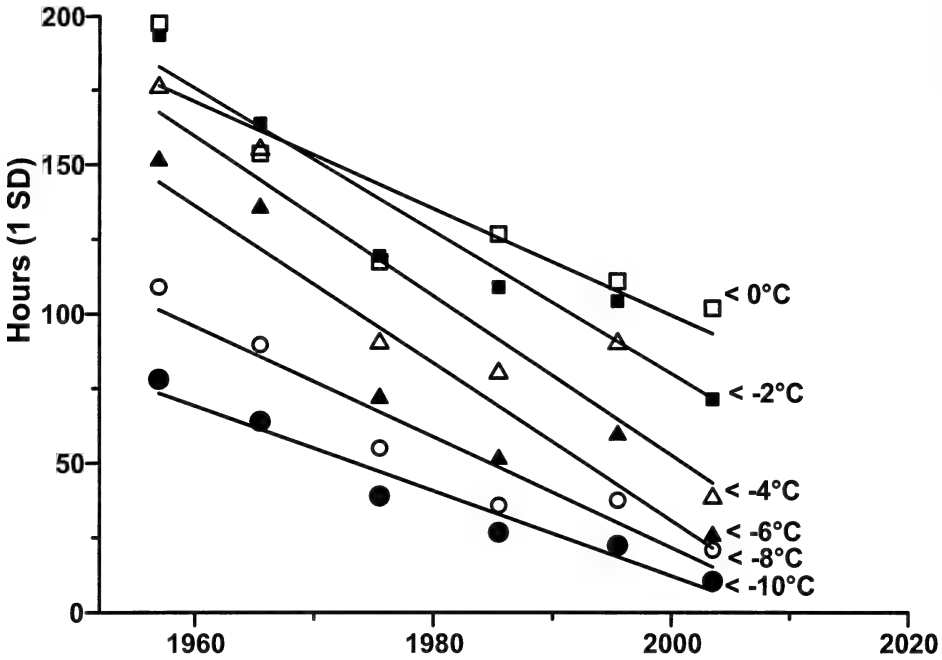

Figure 5. One SD of mean hours with subzero temperatures under a strawberry leaflet in a commercial field, between 10 January and 10 February, versus median year. Solid lines are linear regressions (r^2 = 0.95, 0.92, 0.89, 0.87, 0.93, 0.80 and P = 0.0008, 0.002, 0.005, 0.007, 0.002, 0.02) for temperatures < -10, -8, … 0, respectively.

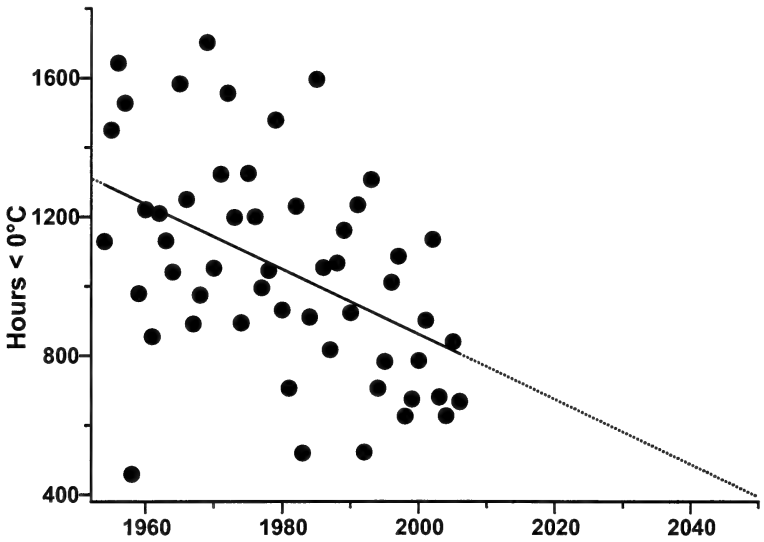

Figure 6. Hours with temperatures < 0 °C under a strawberry leaflet in a commercial field, from 27 November to 30 April, versus year. Solid line is a linear regression y = 19600. – 9.37 x; r^2 = 0.22; P < 0.001.

diapause oviposition in *T. urticae* in relation to field temperatures. A number of thresholds were observed for spider mite populations in southwestern B. C.: a maximum of 390 h of frost after $IO_{0.5}$; post-$IO_{0.5}$ tolerance to >200 h at -2 °C, but only 20 h

at -6 °C; and -8 °C was taken as the critical temperature after $IO_{0.5}$ associated with no oviposition. These thresholds could be confirmed by further work, but this was not the point of the study; regardless of the exact value of the thresholds, the patterns are clear. Oviposition is initiated before frost-free days occur – in ProgIO and *ipso facto* in the field data used to build ProgIO; the number of hours of frost 1 month before oviposition, and variation in those estimates among years, has decreased significantly during the last half century; and the number of hours of frost from 27 November through 30 April have also decreased significantly over the years.

It is clearly risky to extrapolate from a linear regression based on data with considerable scatter (Figs. 4, 6); the relationship may be negative but asymptotic at, for example, 600 h frost (Fig. 6). However, there is additional evidence. Although it is difficult to attribute observed temperature changes to natural or human causes at smaller than continental scales because factors such as land use change and pollution complicate the picture (Intergovernmental Panel on Climate Change 2007), the trends observed in the current study are consistent with the global warming scenario. The mechanism driving global warming, namely increasing levels of greenhouse gases (GHG) (N_2O, CH_4, and CO_2) is well established, and 'With current climate change mitigation policies and related sustainable development practices, global GHG emissions will continue to grow over the next few decades' (Intergovernmental Panel on Climate Change 2007); a global temperature change of +0.2 °C per decade is projected. Therefore, the linear extrapolations in Figs. 4 and 6 may be reasonable. Time will tell, however if correct, oviposition in January should be observable within the next decade.

Despite the many uncertainties in this study, there is sufficient evidence for earlier post-diapause oviposition in *T. urticae* within the relatively near future in southwestern B. C. to at least consider it in planning spider mite monitoring and management activities. This would evolve naturally into planning for continuous annual oviposition should that occur in 4 to 5 decades. Furthermore, a relatively rapid reduction in the number of hours of subzero temperatures during the winter will have significant implications for many aspects of agriculture in the Fraser Valley, including arthropod pest and disease management, crop production, and crop selection in both field and greenhouse environments. These effects need to be considered carefully by growers, pest managers, researchers, and government planners at Provincial and Federal levels with studies and approaches to address potential problems evolving as trends become increasingly certain.

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Biology and management of bark beetles (Coleoptera: Curculionidae) in Washington cherry orchards

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ABSTRACT

The biology and management of bark beetles (Coleoptera: Curculionidae, Scolytinae) in Washington cherry orchards was investigated from 2003-2005. Two dominant species were identified attacking cherry (*Prunus* spp.) orchards: the shothole borer, *Scolytus rugulosus* Müller, and an ambrosia beetle, *Xyleborinus saxeseni* Ratzeburg. *S. rugulosus* was the species most implicated in damage to healthy trees. Two distinct periods of *S. rugulosus* activity occur in Washington, with a possible partial third in some locations. The first activity period begins in late April and peaks in late May to early June, with the second beginning in mid-July and peaks in late July to early August. Yellow sticky traps (unbaited apple maggot traps) were effective tools to monitor *S. rugulosus* activity but ethanol-baited intercept-style traps were necessary to monitor *X. saxeseni* activity. Movement of *S. rugulosus* into orchards was closely associated with emergence from outside hosts, generally a pile of recently pruned or cut wood placed outside the orchard. *S. rugulosus* readily moved distances of 10-50 m to attack trees on orchard borders, but did not move more than two or three rows into a healthy orchard. A residue bioassay technique demonstrated that several insecticides caused mortality of *S. rugulosus* adults. A pyrethroid, esfenvalerate, was the most active 21 d after treatment. Azinphos-methyl was acutely toxic to *S. rugulosus*, but for only seven d. Endosulfan and the neonicotinyls, thiamethoxam and acetamiprid, were somewhat toxic to *S. rugulosus*.

Key Words: bark beetles, *Scolytus rugulosus*, Coleoptera, Curculionidae, Scolytinae, ambrosia beetle, *Xyleborinus saxeseni*

INTRODUCTION

Bark beetles (Coleoptera: Curculionidae, Scolytinae) have historically been reported as pests of pome and stone fruit (Kirk 1969, Linsley and MacLeod 1942, Mendel *et al.* 1987, Payne 1977, Smith 1932). They are commonly described as attacking weakened trees and causing limb or even tree death if present in high enough numbers (Lindeman 1978). Nutritionally stressed trees, or those damaged by sun scald or winter freezing may provide points of access into orchards for opportunistic beetles (Bhagwandin 1992). Health of trees is important to the natural plant defense against bark beetle attack. High plant cell turgor pressure through proper soil water availability may allow trees to mechanically flood out or chemically repel potential colonizers through increased sap flow at the site of attack (Rudinsky 1962, Berryman 1972). The use of synthetic organic insecticides has likely mitigated problems with bark beetles in tree fruit orchards, and until recently they have been considered sporadic and localized pests (Beers *et al.* 1993). However, the reported incidence of injury from bark beetles in Washington stone fruit orchards has been increasing (Brunner

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2003). Anecdotal reports attributed injury to beetles moving into orchards, especially cherry orchards, from outside hosts and attacking healthy trees. The damage most often noted is the boring of beetles at the base of buds by pioneer beetles, causing affected buds to die. Continuous and repeated attacks eventually weaken otherwise healthy trees and make them susceptible to secondary attacks from either conspecifics or possibly other wood boring beetles. Especially vulnerable are new plantings of young cherry trees. Economic difficulties in Washington's fruit industry during the late 1990s likely contributed to the bark beetle problem through an increased occurrence of neglected or abandoned orchards providing suitable host material for bark beetle reproduction (Warner 2006, Mendel *et al.* 1987).

Initial observations (2001-02) of bark beetle damage in cherry orchards indicated a need to further explore certain aspects of their biology and management. With little or no published information from Washington, species identifications and verification of life histories were required for all bark beetles infesting Washington cherry orchards. Preliminary observations indicated that the main beetle species was the shothole borer, *Scolytus sp.*, however at least one other species, possibly an ambrosia beetle, was also involved in attacking healthy cherry trees. While many other wood boring beetles were observed in and around infested hosts, their role in damage to healthy trees was either unknown or unlikely based on what was known of their natural history.

Pests invading orchards from an external host represents a significant challenge to the timing of chemical controls. Knowledge of the pest's development in host plants is needed along with its ability to migrate from these hosts into orchards. Adult traps have proven useful for monitoring bark beetles in other orchard or natural systems (Kovach and Gorsuch 1985, Markalas and Kalapanida 1997), but research is needed to identify and optimize monitoring systems for tree fruit pest management programs. Information is also needed on how much of an orchard requires protection from bark beetles, over what time periods, and which insecticides would be effective at providing required protection.

This paper provides new knowledge that will help Washington cherry growers manage bark beetle problems. The key species involved in attacking pome and stone fruit trees were identified along with a clear understanding of their seasonal life history. We also developed methods of monitoring bark beetles, and determined the distance bark beetles moved from a natal food host to attack healthy orchard trees. A bioassay technique was developed for assessing relative toxicity of candidate insecticides, and we documented successful control strategies used to manage bark beetles in heavily infested orchards.

MATERIALS AND METHODS

Bark beetle identification, monitoring, and life history. Bark beetles and other wood boring Coleoptera infesting Washington cherry orchards were identified by a combination of rearing adults from host wood infested with immature larvae in emergence cages and trapping adults near suspected host sites and along orchard borders. All insects collected in the following trials were stored in alcohol and later identified to family (Dr. Christian Krupke, Purdue University; Dale Whaley, Washington State University; Michael Doerr, Washington State University). All Scolytinae and associated parasitoids were sent to Malcom Furniss, (Entomologist *Emeritus*, University of Idaho) for identification. All Coleoptera collected from emergence cages and adult traps were identified in 2003. In 2004 only Scolytinae were submitted for identification. By 2005 it was apparent that *Scolytus spp.* were the dominant bark beetles present in Washington cherry orchards so identification was further limited to those

species.

Emergence cages were used to identify a species:host relationship. Infested wood from four sites was collected during the spring and summer in 2003, placed in opaque cardboard boxes (50 x 50 x 30 cm), and held under laboratory conditions (22 ± 2 °C). One glass vial (2.5 cm diameter x 10 cm) was placed through a hole in each emergence box. Emerging beetles (and Hymenopteran parasitoids) where attracted to the light coming through the opening in the box and entered the vial. Beetles and natural enemies that entered the vial were removed daily.

Adult traps were placed near infested wood piles outside of orchard blocks (referred to below as 'outside hosts') and on the orchard border closest to the outside host at sixteen locations in north-central Washington from 2003-05 (twenty one orchard-yr equivalents). No specific protocols were followed across all sites, but rather an effort was made to ensure that trap placements sufficiently covered the threatened area of each orchard border and encircled outside hosts. Considerations had to be made depending on the size of each location. Generally, traps were placed approximately 10 m apart on orchard borders and hung directly in the trees at a height of 2 m. At least four traps were placed around a suspected outside host. If circling a host was not possible, traps were placed approximately 10 m apart across the length of the host. Most often traps were hung directly from host material, but it was sometimes necessary to hang them from a 2 m tall post that was placed adjacent to the host. Monitoring efforts in 2003 focused on identifying the best available trap and lure system. Commercially available intercept-style traps (Lindgren Funnel Trap, 8-funnnels, Phero Tech, Inc., Delta, British Columbia, Canada; Pane Intercept Trap, IPM Technologies, Inc., Portland, OR), either with or without an ethanol attractant, and un-baited yellow sticky traps (Pherocon AM, Trècè, Inc., Adair, OK) were evaluated in trials replicated across several locations for their ability to monitor adult activity at an outside host and at a nearby orchard. A 12.5 cm^2 DvDP kill strip (Vaportape II insecticidal strips, Hercon Environmental Co, Emigsville, PA) was placed in the collection container of the intercept-style traps to kill beetles and prevent their escape.

In 2003, a direct comparison was made between the Lindgren Funnel Trap and the Pane Intercept Trap at six locations. Each trap type was baited with the respective manufacturer's commercially available ethanol lure. Traps were placed on 15 April and monitored every seven d until 15 Oct. Lures were replaced at six-wk intervals, based on manufacturer recommendations. A direct comparison to evaluate the effectiveness of the ethanol attractant in the Lindgren Funnel Trap was also made at four locations in 2003. Traps were placed on 15 April and monitored every seven d for six weeks. In 2003 and 2004, a direct comparison was made between a Lindgren Funnel Trap baited with an ethanol lure and an unbaited yellow sticky trap at ten locations. In 2004, traps were placed on 1 Mar and monitored every seven d through October. The ethanol lures were replaced at six-wk intervals. Two traps of each treatment were placed in an alternating pattern at all locations. Season-long captures of the dominant Scolytinae species were averaged for the two traps at each location. Due to high variability in populations between locations, trap capture data from the paired comparisons were analyzed by a Wilcoxon Rank Test (P=0.05) (Wilcoxon 1945) using JMP statistical software (JMP v. 5.1.2 2004).

Adult trap data gathered from the locations with the highest populations (15 orchard-yr for *S. rugulosus* and five orchard-yr for *X. saxeseni*) were used to plot cumulative emergence of the dominant species for each of the generations. With no temperature-dependent developmental (degree-day) data available, the only point of reference between years was Julian days. Cumulative emergence at each date was averaged and plotted with the raw data from each orchard site. Julian days were then con-

verted back to calendar days for ease of reference.

***Scolytus rugulosus* migration and damage distribution**. Two orchards in 2004 and two in 2005 were identified where host wood piles that were heavily infested with *S. rugulosus* were threatening nearby healthy orchards (<50 m). Yellow sticky traps were placed by the hosts located outside the orchards to track adult emergence and on the orchard borders to monitor immigration. Cumulative capture percentiles for an entire *S. rugulosus* generation at the outside host and on the orchard border were plotted together for each study site. If cumulative percentiles were identical at the host and the orchard this would suggest that adult dispersal to a suitable feeding or reproductive site occurred immediately after emergence. However, dramatic shifts in cumulative percentiles would indicate either a delay in migration from the outside host or a constant or prolonged immigration into the orchard from multiple outside hosts. Each location could only be monitored for one generation because we allowed growers to remove the natal host and protect their orchard following our observations.

At the same locations described above, damage to healthy trees in the orchard was monitored by visually inspecting trees. Every tree along the border row and then every tree in subsequent rows moving into the orchard away from the outside host were monitored for *S. rugulosus* damage until no further damage was noted. The total number of trees sampled varied at each orchard (Site 1 – 3 rows x 19 trees, Site 2 – 5 rows x 8 trees, Site 3 – 4 rows x 12 trees, Site 4 – 7 rows x 15 trees). Twenty growing shoots were randomly selected from each tree and the total number of shoots exhibiting wilting or flagging foliage (visually confirmed to be caused by *S. rugulosus* burrowing) was recorded. We calculated the total number of damaged shoots at each site, then noted what percentage of that total was found in the row closest to the outside host (row one) and each subsequent row moving away from the outside host.

***Scolytus rugulosus* insecticide screening**. Insecticides were evaluated using newly emerged *Scolytus rugulosus* adults in 2004. The insecticides outlined in the Results and Discussion section included the majority of those recommended for use on cherries in Washington (Smith et al. 2004). Although the insecticides chosen for this trial were those available on cherry, mature Delicious apple trees at WSU-TFREC were the only trees readily available for this test. The trees were treated with various insecticides at the manufacturers' recommended rates. All treatments were applied with a handgun sprayer at 300 psi to drip, simulating a full dilute spray. Treatments were applied to one-tree plots replicated three times in a randomized complete block. A one-tree buffer (unsprayed tree) was left between each replicate to reduce over-spray and drift. Treated apple branches, approximately 15 cm long x 1.25 cm diameter sections of two-yr-old wood, were collected at 1, 7, 14 and 21 d after treatment (DAT), returned to the laboratory and stored at 2 °C until new adults could be collected. Branch sections were placed into 1 L deli cups (Prime Source PS232, Dallas, TX). Untreated apple branches were used as controls for each sample date. Five arenas were prepared for each treatment. Five *S. rugulosus* adults, collected from emergence cages described above, were added to an arena and survival was recorded after 24 h (25 adults/treatment/DAT). It was assumed that the adults appearing in the vials were newly emerged, but that could not be verified. Both males and females were used in the bioassay, with no effort made to segregate by sex. We did not generate enough adults to run the entire screening at one time so adults were added to the insecticide arenas in the following order: one replicate from each treatment followed by an untreated control replicate for the one DAT samples. All replicates from this initial collection date were completed before beginning evaluations on the next series of samples (seven DAT). The process was repeated until all samples were completed. Rearing conditions were 22 °C, 16:8 L:D. Average

survival and standard error of the means were reported for each treatment.

Successful *Scolytus rugulosus* management practices. During the course of this study, we documented *S. rugulosus* control efforts in four heavily infested orchards. In each situation we were contacted by growers who were already experiencing severe injury to cherry orchards. We worked with growers to monitor potential hosts, whether inside or outside of an orchard, with adult traps in an effort to identify the sources of infestation. Dissections of suspected host material (removing bark to expose live larvae) were conducted to verify *S. rugulosus* were currently utilizing the material as a natal host. We also conducted damage evaluations throughout the orchards to isolate the areas that required intervention. Once the *S. rugulosus* situation was completely described, growers implemented their own sanitation programs. We continued to monitor the orchards with adult traps throughout the clean up process and subsequently conducted post treatment damage evaluations to document the efficacy of these efforts. The methods used in these damage evaluations were consistent with those described in the trials above.

RESULTS AND DISCUSSION

Bark beetle identification, monitoring, and life history. A total of 17,116 adult Scolytinae were collected from infested fruitwood, yellow sticky traps, and ethanol-baited intercept traps. The dominant Scolytinae found throughout Washington was the shothole borer, *S. rugulosus* Müller (ver. Malcom Furniss) (Table 1). An ambrosia beetle, *Xyleborinus saxeseni* Ratzburg (ver. Malcom Furniss), was present in high numbers at only one location, a cherry orchard abandoned for several years. More than one species of Scolytinae were detected at each location where identification was not limited to *Scolytus spp*. A second *Scolytus sp*. (*S. multistriatus*) was found infesting a pile of cherry wood at only one site. Cherry has not been reported as a host (Furniss and Johnson 2002) for *S. multistriatus*, and in this case, *S. multistriatus* infested only the pile of cherry wood and was not detected moving into the neighbouring cherry orchard. Many other wood decomposing beetles were reared from infested fruitwood. In fact, the majority of Coleoptera species collected were associated with dry, older wood (dead for more than 18 mo). Buprestid (Buprestidae) and powderpost beetles (Lyctidae) were the primary beetles associated with dry wood (Table 1). *S. rugulosus* and *X. saxeseni* were the primary attackers of weakened trees or recent cuttings (<18 mo). *S. rugulosus* was the species most implicated in damage to healthy orchards, whereas *X. saxeseni* was found attacking only trees that had been previously damaged or weakened. Initial observations from laboratory emergence cages indicated that there was a high rate of parasitism (approximately 50%) of *S. rugulosus* larvae by *Cheiropachus quadrum* (Hymenoptera: Pteromalidae) (ver. Malcom Furniss) based on their relative abundance in vials from emergence cages.

No statistically significant difference was noted between commercially available intercept-style traps in their ability to capture adult *S. rugulosus* (Chi-Square 3.103, df 1, $P = 0.078$) but *X. saxeseni* captures were slightly higher in Lindgren Funnel Trap than Pane Intercept Trap (Chi-Square 4.021, df 1, $P = 0.045$) (Table 2). Both intercept-style traps should be suitable monitoring systems for *S. rugulosus* and *X. saxeseni* adults. The addition of an ethanol lure significantly enhanced captures of both *S. rugulosus* (Chi-Square 5.333, df 1, $P = 0.021$) and *X. saxeseni* (Chi-Square 5.333, df 1, $P = 0.021$). Although ethanol lures significantly increased captures of both species, this may be an area where monitoring systems could be improved. Synergistic plant volatiles (Montgomery and Wargo 1983) and/or aggregation pheromones (Lindgren *et al.* 1983, Pitman *et al.* 1975, Schroeder and Lindelow 1989, Peacock *et*

Table 1.
Wood-boring beetle collections from infested fruitwood, yellow sticky traps, and ethanol-baited intercept traps from Washington, 2003-05.

			Total annual captures in all traps[1]			
Location	**Yr**	**Host Material**	***S. rugulosus***	***X. saxeseni***	**Buprestidae**	**Lyctidae**
Wenatchee	2003	Dead cherry	13	369	5	6
Wenatchee	2003	<1-yr-old cuttings	7	16	27	20
Mallot	2003	1-yr-old pushed over apple	357	11	530	25
Okanogan	2003	<1-yr-old pushed over cherry	8654	4	25	287
Oroville	2003	<1-yr-old cuttings	695	---	---	---
Oroville	2003	Neglected apple	26	12	7	224
E. Wenatchee	2003	<2-yr-old cuttings	247	31	4	357
Wapato	2003	Neglected cherry	141	---	---	---
W. Valley	2003	Neglected cherry	637	---	---	---
Cowiche	2003	Neglected cherry	284	---	---	---
Orondo	2004	<2-yr-old cuttings	125	32	---	---
Wenatchee	2004	Dead cherry	374	196	---	---
E. Wenatchee	2004	<2-yr-old cuttings	1361	1	---	---
Okanogan	2004	<1-yr-old cuttings	218	21	---	---
Bridgeport	2004	<1-yr-old cuttings	57	6	---	---
Bridgeport	2004	<1-yr-old cuttings	6	1	---	---
Tonasket	2004	<1-yr-old cuttings	170	2	---	---
Okanogan	2005	<2-yr-old cuttings	1247	---	---	---
Orondo	2005	<2-yr-old cuttings	743	---	---	---
E. Wenatchee	2005	<2-yr-old cuttings	292	---	---	---
Moses Lake	2005	Neglected cherry	761	---	---	---

[1] ---, Insects not collected for identification.

al. 1972) have been used to improve monitoring systems for some bark beetle species, unfortunately not all species aggregate in response to pheromone production (Macías-Sámano *et al.* 1998). It is unclear if *S. rugulosus* or *X. saxeseni* produce aggregation pheromones.

Adult *S. rugulosus* were more highly attracted to the yellow sticky traps (Chi-Square 9.143, df 1, $P = 0.003$), than the dark coloured ethanol-baited intercept-style traps. Yellow sticky traps proved to be easy to deploy and read, and were relatively economical compared to the intercept-style traps. The ethanol-baited intercept-style traps were necessary to monitor *X. saxeseni* activity (Chi-Square 12.799, df 1, $P = 0.0003$), but our experience has been that this species is a minor contributor to damage in commercial cherry orchards.

Two distinct periods of *S. rugulosus* activity occurred in Washington (Fig. 1). *S. rugulosus* activity was first noted in late April or early May and continued through June. The second adult flight was detected in mid- to late July and continued through August and into late-September. Adult *S. rugulosus* were trapped through the entire growing season from initial adult emergence through the end of October. A slight increase in trap captures occurring at the end of each season suggested the possibility

Table 2.
Mean (± SEM) *S. rugulosus* and *X. saxeseni* captures using commercially available trap and lure systems, 2003.

Mean adults/trap (SEM)[1]				N[2]
S. rugulosus		*X. saxeseni*		
Pane Intercept With Ethanol Lure	Lindgren Funnel With Ethanol Lure	Pane Intercept With Ethanol Lure	Lindgren Funnel With Ethanol Lure	
68.3 (34.7)	121.3 (59.3)	21.3 (16.4)	31.0 (22.2)*	6
Lindgren Funnel With Ethanol	Lindgren Funnel Without Lure	Lindgren Funnel With Ethanol	Lindgren Funnel Without Lure	
109.1 (66.2)*	59.5 (50.3)	33.9 (29.4)*	7.0 (6.3)	4
Lindgren Funnel With Ethanol	Yellow Sticky Card	Lindgren Funnel With Ethanol	Yellow Sticky Card	
34.9 (27.0)	226.6 (188.6)*	22.7 (14.4)*	0.6 (0.4)	10

[1] Means followed by '*' are significantly different (Wilcoxon Rank Test, P = 0.05)
[2] N, number of sites in study

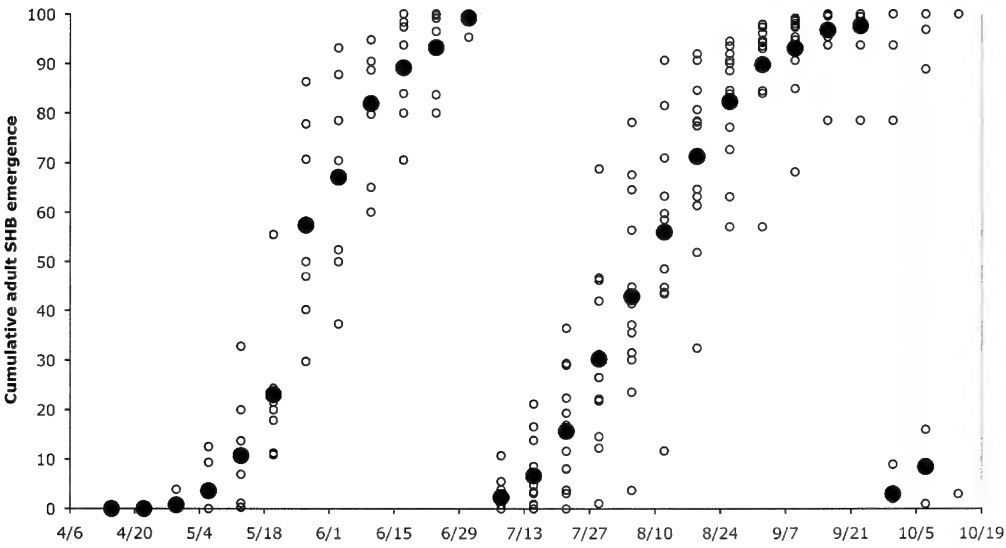

Figure 1. Cumulative emergence of *S. rugulosus* adults (n=5,675) in Washington, 2003-05. Open circles represent cumulative emergence from each location, black dots represent the average emergence on each date. SHB = shothole borer.

of a partial third generation.

Adult *X. saxeseni* activity occurred throughout the entire growing season, with peaks suggesting the presence of three to four generations (Fig. 2). Adult *X. saxeseni* activity was initially noted in late March or early April. A second peak of activity occurred in early June, with a third noted in July and early August. A slight increase *X. saxeseni* activity was observed in September and early October, although at reduced numbers. It is unclear if this activity represented part of a fourth generation or prolongation of the third.

Traps were useful in identifying peak activity periods of *S. rugulosus* but it was not clear if they would be useful in setting thresholds for treatments. We had trouble locating *S. rugulosus* sources with various population sizes near neighboring cherry orchards that were allowed to remain untreated. Since insecticides were applied

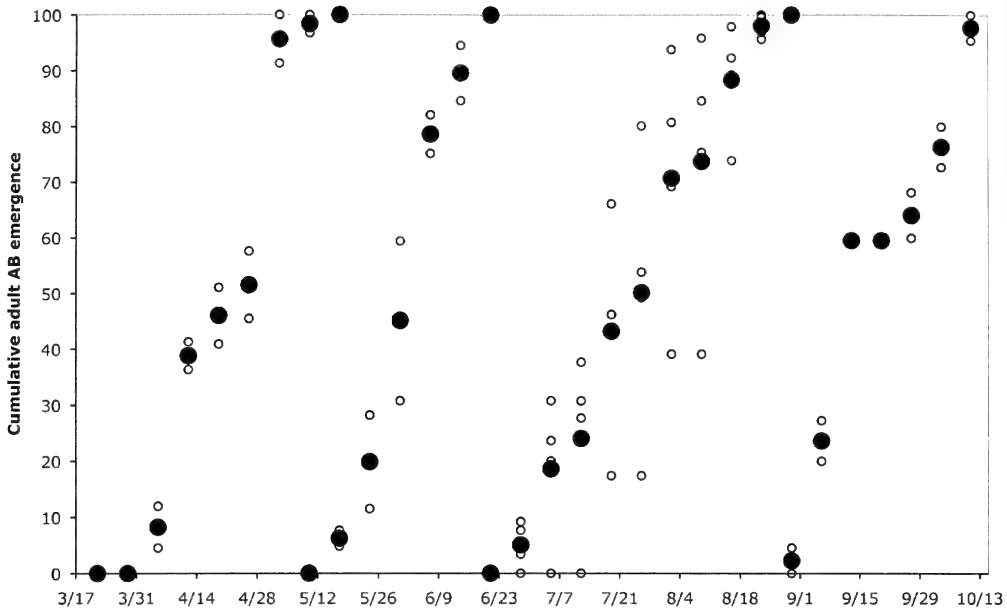

Figure 2. Cumulative emergence of *X. saxeseni* adults (n=805) in Washington, 2003-04. Open circles represent cumulative emergence from each location, black dots represent the average emergence on each date. AB = ambrosia beetle.

frequently in cherry orchards for control of other pests, it was difficult to establish a consistent relationship between trap captures and subsequent damage. However, our observations indicated that if an *S. rugulosus* host was located near a cherry orchard and any significant emergence was detected with yellow traps, some control intervention would be justified to prevent damage.

***Scolytus rugulosus* migration and damage distribution**. Movement of *S. rugulosus* into healthy orchards was closely associated with emergence from a nearby infested host, generally a pile of recently pruned or cut wood placed outside the orchard (Fig. 3). Cumulative captures of adults at the outside host and at the border of the nearby orchard were closely associated at each study site. There was no consistent pattern of either a lag in percentiles, or prolonged captures at the orchard border. In other words, observations at the more heavily infested outside hosts were representative of what was occurring at the orchard borders. Further, *S. rugulosus* activity was easier to monitor at the host than in healthy trees (1819 and 258 total *S. rugulosus* adults, respectively) as a very large number of adults emerged from a relatively small area and dispersed immediately. These data indicated that growers should be able to focus their efforts at locating and monitoring suspected outside hosts, understanding that as adult activity increased at the hosts, immigration to healthy orchards was occurring simultaneously. It appeared that recently emerged *S. rugulosus* adults were highly dispersive and readily moved distances of at least 50 m from infested outside hosts to healthy trees in orchard borders. After dispersal, adult activity in and around suitable natal hosts continued where behaviour appeared to be associated with the construction and care of maternal galleries. This aspect of *S. rugulosus* behaviour needs to be explored further.

S. rugulosus adult feeding damage to healthy trees was most commonly associated with movement from infested hosts. Generally, *S. rugulosus* damage was in close proximity to that host. It appeared that *S. rugulosus* moved readily to and along an orchard border, but did not move more than two or three rows into a healthy orchard. On average, 74% of the total *S. rugulosus* damage that was detected in healthy or-

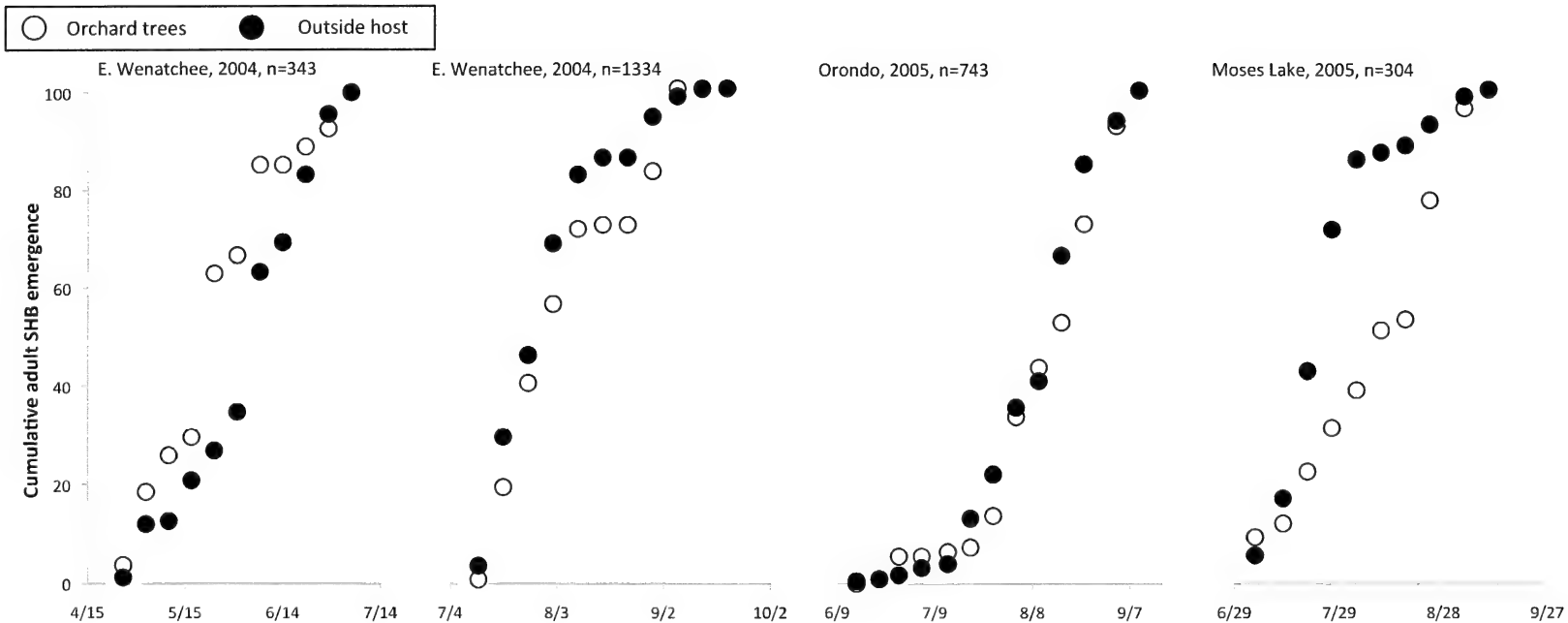

Figure 3. Cumulative captures of *S. rugulosus* on yellow sticky traps from infested host wood piles ('outside host') and the borders of nearby healthy cherry orchards, 2004-05. SHB = shothole borer.

chards occurred on the border row closest to the natal host (Fig. 4). Twenty percent of the total damage was found on the second row. Damage in subsequent rows was minor and scattered. These data indicated that monitoring and control efforts should be focused on determining the natal host responsible for *S. rugulosus* infestation, usually piles of recently cut wood near orchards, and then protecting the area of the orchard immediately adjacent to that host. If control is neglected, trees will become weakened and *S. rugulosus* will successfully colonize and reproduce in the weakened trees. Once this occurs immigration is not the sole source of beetles and damage will spread further into the orchard thereby complicating management efforts.

***Scolytus rugulosus* insecticide screening**. Once introduced into the treatment arenas, adult *S. rugulosus* began feeding or attempting to colonize the limb sections immediately. Although the purpose of this behaviour was not known (feeding or oviposition), the beetles were very active on the treated wood. The average survival of *S. rugulosus* adults on untreated wood was 96% after 24 h. This level of survival indicated any significant mortality could be attributed to pesticide exposure and not a problem with the bioassay method.

Many insecticides caused mortality of *S. rugulosus* in the bioassays (Table 3). A pyrethroid, esfenvalerate, was the most active through 21 d. Azinphos-methyl was acutely toxic to *S. rugulosus*, but for only seven d. Endosulfan and the neonicotinyls, thiamethoxam and acetamiprid, were somewhat toxic to *S. rugulosus*. Malathion, indoxacarb, and spinosad all caused mortality, but not at levels expected to provide adequate control under field conditions. Additional insecticide efficacy trials are necessary to understand the full potential of each insecticide to control *S. rugulosus* under field conditions. The repeated use of these insecticides against other pests, primarily the cherry fruit fly, *Rhagoletis indifferens* Curran (Diptera: Tephritidae), during the early part of the growing season is likely sufficient to suppress damage in most commercial orchards, especially during the first *S. rugulosus* generation. Cherry orchards may become more susceptible to injury in the post-harvest period when insecticide programs for cherry fruit fly and leafroller (Lepidoptera: Tortricidae) have ceased. Second-generation *S. rugulosus* adults would then be able to move into unprotected orchards.

Successful *Scolytus rugulosus* management practices. Sanitation has generally been touted as the key to control, with wood or brush piles identified as contributors of beetles that migrate and attack other trees (Bhagwandin 1992, Beers *et al.* 1993, Payne 1977). In the winter of 2003-04, we monitored an effort to clean up a large infestation of *S. rugulosus* emerging from an outside host that had resulted in significant damage to young, healthy cherry trees. In 2003, approximately 55% of shoots in the trees along the orchard border at this location were damaged despite several insecticide applications, including repeated applications of endosulfan. The outside host was a firewood and brush pile that was replenished each year. While the damage was high, it was fairly well isolated from the orchard border rows adjacent to the host. During the winter of 2003-04, the orchard was pruned heavily, removing all weakened or damaged branches and the grower made a concerted effort to clean up all host material (firewood, brush piles, and current-yr cuttings) and maintain a clean area near the orchard. During the 2004 growing season, the orchard was monitored with yellow sticky traps and ethanol-baited intercept-style traps. Insecticide applications were planned to coincide with increased trap captures. However, a total of only four *S. rugulosus* and nine *X. saxeseni* adults were trapped in five yellow traps and four intercept-style traps and therefore no insecticide applications specifically for control of bark beetles were needed. No *S. rugulosus* damage was noted at any time during the 2004 season demonstrating that the sanitation efforts were the only control measure needed.

In 2005, we monitored efforts at three

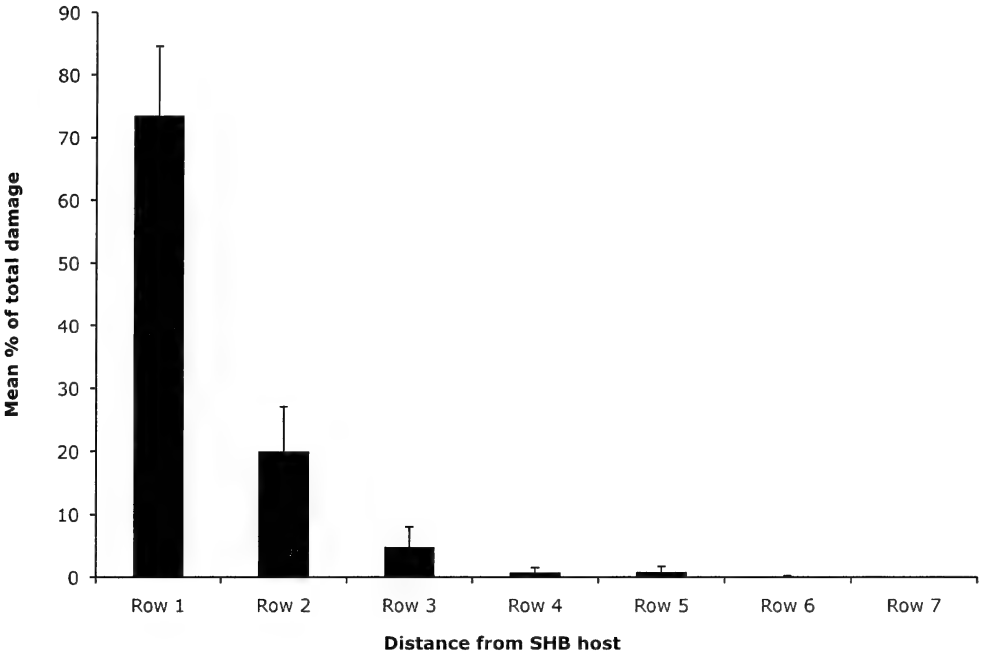

Figure 4. Mean (±SEM) percentage of total damage noted in each orchard (n=4) at the row adjacent to the source of *S. rugulosus* (Row 1), and subsequent rows moving into the orchard, 2004-05. SHB = shothole borer.

Table 3.
Mean (±SEM) *S. rugulosus* survival on field-aged residues of insecticides, 2004.

Insecticide	Active Ingredient	Rate gm ai/ha	Mean no. live *S. rugulosus* (SEM) – 24 h			
			1 DAT[1]	7 DAT	14 DAT	21 DAT
Asana XL 0.66EC	Esfenvalerate	46.8	0.0 (0.0)	0.4 (0.2)	0.0 (0.0)	0.8 (0.4)
Guthion 50WP	Azinphosmethyl	1,135.0	0.0 (0.0)	0.4 (0.2)	2.4 (0.7)	4.0 (0.3)
Actara 25WDG	Thiamethoxam	80.0	0.6 (0.6)	1.0 (0.3)	2.0 (0.9)	1.8 (0.7)
Assail 70WP	Acetamiprid	168.7	0.6 (0.6)	1.4 (0.6)	1.8 (0.6)	0.8 (0.4)
Thiodan 3E	Endosulfan	2,552.5	0.7 (0.3)	1.3 (0.3)	1.7 (0.3)	1.3 (0.9)
Avaunt 30WDG	Indoxacarb	127.8	2.0 (0.5)	2.0 (0.7)	0.8 (0.4)	2.6 (1.1)
Malathion 50%EC	Malathion	300.0	2.2 (0.8)	2.8 (0.6)	4.2 (0.4)	3.8 (0.7)
Success 2SC	Spinosad	106.4	3.8 (0.2)	3.6 (0.7)	4.2 (0.4)	3.6 (0.7)
Untreated			5.0 (0.0)	4.8 (0.2)	5.0 (0.0)	4.4 (0.2)

[1] DAT, Days After Treatment

locations near Okanogan, WA to clean up easily identifiable infested hosts of *S. rugulosus* located just outside of cherry blocks exhibiting signs of recent feeding damage. In addition to the external hosts, weakened limbs and one-yr-old cuttings left in the orchards were also serving as host material for *S. rugulosus* reproduction within the cherry blocks. These sites were brought to our attention after first generation beetles had caused serious damage to trees in the orchard borders. Insecticidal control options were limited as one of the blocks was managed organically, and the conventional blocks were experiencing damage levels of 50% shoot infestation despite a history of

border sprays. Although yellow traps used to determine what host material was serving as the source of the *S. rugulosus* infestations were placed near the end of first generation activity, captures in the first seven d averaged 116 *S. rugulosus* per trap across all three locations. Subsequently, the growers removed all possible host material within the orchard, including the previous winter's cuttings and weakened branches or limbs. This wood was added to the host material located outside the orchard and targeted with an intensive insecticide treatment program. Endosulfan was applied by a handgun sprayer on a 10-14 d retreatment interval for the rest of the season with care taken to thoroughly soak the entire wood pile. Following this action, no second-generation beetle activity was noted at any of the three sites, and no new damage was detected inside the orchard.

Healthy cherry trees can repel initial colonization efforts by *S. rugulosus* adults by flooding attacked sites with resin, but with repeated attacks even healthy trees will eventually become weakened, allowing successful colonization by secondary attacks from conspecifics (Bauernfeind 1996, Payne 1977). Our experience with *S. rugulosus* management indicates orchard sanitation is the most important factor contributing to a reduction in *S. rugulosus* populations and damage to healthy cherry trees. If recent feeding damage is noted on otherwise healthy trees, adult traps can be placed on the orchard borders or in suspected host areas to verify the source of infestation. Sanitation programs must include removing potential host material (weakened limbs or recent cuttings) from within the orchard and eliminating any host material outside the orchard. Beetle host material outside the orchard can be eliminated by burning or by thoroughly soaking the wood with an effective insecticide delivered by a handgun sprayer. The increased volume of water delivered by handgun applications appears to be an important factor in insecticide efficacy. We do not believe growers can rely on traditional insecticide applications via an air-blast sprayer to control infestations that originate from within the orchard, or protect orchard borders from massive immigration originating from a nearby heavily infested host.

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Extracts of *Ginkgo biloba* or *Artemisia* species reduce feeding by neonates of codling moth, *Cydia pomonella* (Lepidoptera: Tortricidae), on apple in a laboratory bioassay

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ABSTRACT

In a simple bioassay, alcohol extracts (91% v/v ethanol, 4% v/v methanol, 5% v/v isopropanol) from *Ginkgo biloba* L., *Artemisia absinthium* L., *A. arborescens* L. x *A. absinthium* L., and *A. ludoviciana* Nutt. "Valerie Finnis", significantly reduced feeding of neonatal codling moth larvae on apple. Extracts from *A. californica* Less. and *A. vulgaris* L. had no effects.

Key Words: insect feeding, larvae, repellent, deterrent

INTRODUCTION

Codling moth, *Cydia pomonella* (L.), is a cosmopolitan pest that primarily attacks apple, *Malus domestica* Borkh., a commodity worth more than US$1 billion per year in the northwestern United States and British Columbia, Canada. Codling moth females oviposit mostly on foliage (Jackson 1979). Newly hatched neonates travel over apple branches and foliage in search of the fruit (Jackson 1982) and finally burrow into it (Tadic 1957). Manipulation of neonate searching behaviour may provide an alternative or complementary approach to current strategies for codling moth management (Pszczolkowski 2007).

Neonate codling moth responses to feeding stimulants have previously been explored (Pszczolkowski 2007). Suomi *et al.* (1986) and Landolt *et al.* (1999) studied deterrent effects of, respectively, plant extracts and plant essential oils. Of the 25 species tested by Suomi *et al.* (1986), five showed promise as feeding deterrents to neonatal codling moth larvae: absinthe wormwood, *Artemisia absynthium* L.; rabbitbrush, *Chrysothamnus nauseosus* (Pall. ex Pursh) Britton; false hellebore, *Veratrum californicum* Durand; garlic, *Alium sativum* L.; and tansy, *Tanacetum vulgare* L. Of the 27 species tested by Landolt *et al.* (1999), the greatest arrestment of neonates was achieved with oils of lavender, *Lavandula officinalis* Chaix ex Vill; pennyroyal, *Mentha pulegium* L.; and cypress, *Cupressus sempervirens* L. Oils of rue, *Ruta graveolens* L.; garlic; patchouly, *Pogostemon cablin* (Blanco) Benth.; and tansy were the most repellent to neonates (Landolt *et al.* 1999). Extracts from the ginkgo tree, *Ginkgo biloba* L., which have deleterious effects on codling moth neonates (Pszczolkowski and Brown 2005), were not studied by Suomi *et al.* (1986) or Landolt *et al.* (1999).

In this paper, we use a simple modification of the assay designed by Suomi *et al.* (1986) to test the effects of extracts of *G. biloba* and of five members of the genus *Artemisia* on feeding by codling moth neonates. Our assay allows small volumes of plant extracts on apple plugs to be presented to individual neonates. We compare

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neonates' response to gingko extract in the apple plug assay with their response to whole apples using an assay previously described in Pszczolkowski and Brown (2005).

MATERIALS AND METHODS

Insects. Codling moths originated from USDA-ARS Yakima Agricultural Research Laboratory in Wapato, WA, the same source as used by Suomi *et al.* (1986). This laboratory has maintained the codling moth colony for about 40 years, comprising more than 480 generations. Moths were held at 25 °C, 70-80% RH, under a 16L:8D light-dark regime, and allowed to oviposit on polypropylene foil. Neonates were collected 0.5-1 h post-hatch, and used for experiments.

Plant extracts. The extracts were prepared from foliage of *G. biloba*, *A. absinthium*, *Artemisia arborescens* L. x *Artemisia absinthium* L., *Artemisia ludoviciana* Nutt. "Valerie Finnis, *Artemisia californica* Less. and *Artemisia vulgaris* L.

Dried *A.absynthium* foliage was purchased from a local pharmacy (London Apothecary, Mansfield, MO). Remaining plant material was collected in the gardens of Missouri State University Research Campus, Mountain Grove, MO, in July 2008.

Dehydration alcohol (91% v/v ethanol, 4% v/v methanol, 5% v/v isopropanol; EMD Chemicals Inc., Gibbstown, NJ) was used to prepare all extracts. Plants were dehydrated using an Open Country food dehydrator (Nesco/American Harvest®, Two Rivers, WI) at 35 °C for 48 hours. The dry foliage was ground in a coffee grinder. Approximately 0.5 ml of dry plant powder was placed in a plastic centrifugation tube, 500 µl of dehydration alcohol added, then the tube was vortexed and left at room temperature for 10 min. The tube was then centrifuged at 2000 G for 10 min and 300 µl of liquid fraction was transferred to a pre-weighed plastic test tube and allowed to air dry overnight. The test tube with the residue was re-weighed the next morning and enough dehydration alcohol was added to make a 10 mg/ml solution of each plant extract. The extracts were prepared immediately before testing.

Modified assay using apple plugs. For each test arena, four plugs were procured from the same Golden Delicious apple, using a length of plastic soda straw (Fig. 1A), such that the straw covered the pulp, but not the epidermis of the apple. The crevice between the plug and the edge of the straw was sealed with paraffin wax applied with a warm spatula (Fig. 1B). The straws were then placed in a holder, apple plug facing up, and 5 µl of test solution were applied to each plug. The plugs were allowed to air dry, and four plugs were placed in a 60 x 15 mm polystyrene Petri dish (Fig. 1C). Small pieces of modeling clay held the plugs in place. New clay was used for each assay. A glass rod (1.3 mm diameter, 25-27 mm long) was positioned such that each end of the rod touched both the control and the treated member of the plug pair (Fig. 1C). One neonate was placed, using a camel-hair artistic brush, in the middle of the glass rod and the Petri dish was covered with a lid. The entire assembly was covered with a half of a white plastic RipBall (TM & Enor Corp., Northvale, NJ) to provide a white, slightly opaque cupola (Fig. 1D) and placed on the testing bench illuminated by fluorescent tubes and Soft White 60 general purpose bulbs (General Electric Canada, Cleveland, OH). Such an arrangement provided dispersed, non-directional light of uniform luminosity (900-920 lux) over each test arena, which was important because codling moth neonates exhibit mild phototropism (Jackson, 1982). Prior to every experiment, glass rods and Petri dishes were washed sequentially in tap water, double distilled water, alcohol, then dried.

Preliminary experiments showed that neonates could be expected to locate a plug and begin feeding within 20 hours, and that

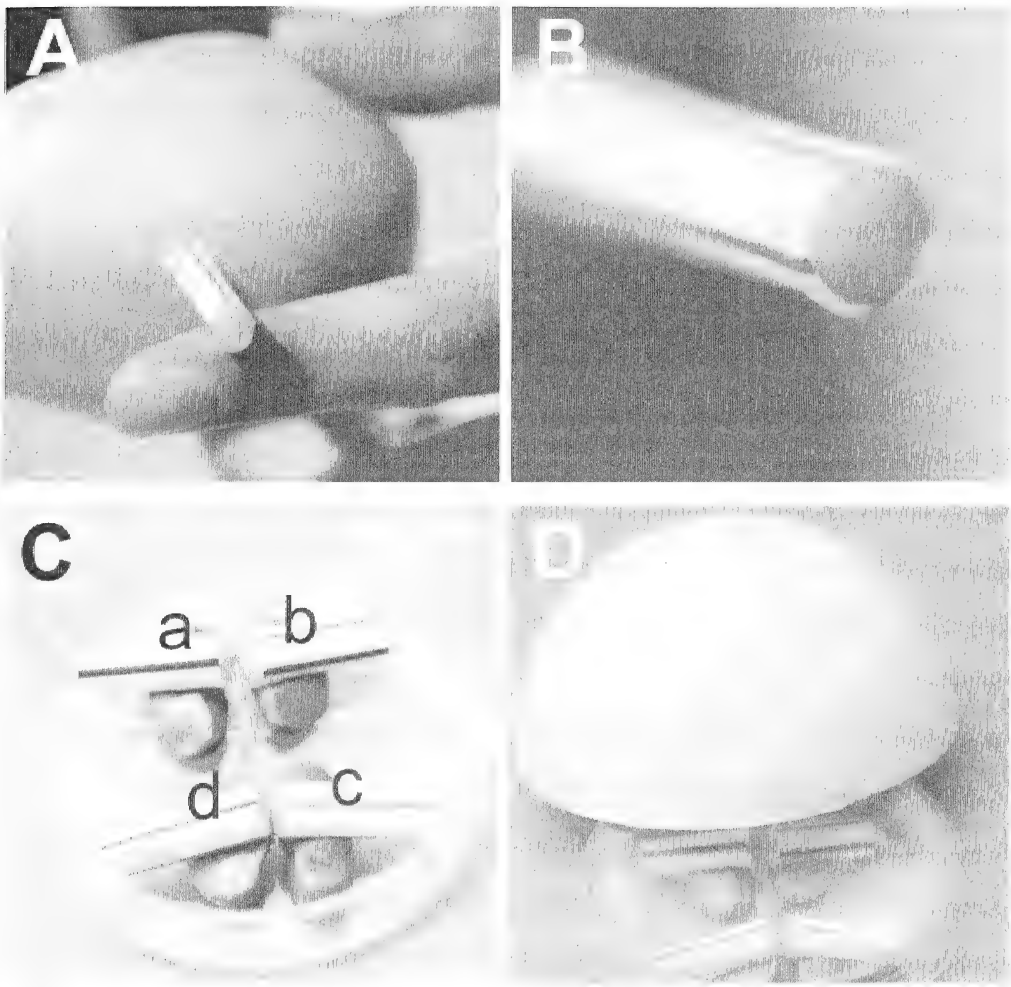

Figure 1. Preparation of test arena (60 mm diameter) for apple-plug assay. A. Cutting the plug out of apple. B. Crevice between the plug and the straw is sealed with paraffin wax. C. a, c. Plugs treated with plant extracts; b, d. control plugs treated with dehydration alcohol. D. Test arena is covered with plastic cupola to disperse light and eliminate possible effects of directional lighting.

neonates showed no preference for any plug position in the arena. In the experiments reported here, 30 neonates were individually exposed to ginkgo extracts, and 18-23 neonates were individually exposed to extracts of each *Artemisia* species. All neonates were tested simultaneously. All apple plugs were examined under a dissecting microscope after 20 h for feeding indicators such as abraded epidermis, presence of excrement, or a feeding cavity. If evidence of feeding was found, the plug was removed from the straw and dissected to reveal the larva.

Whole-fruit assay. To test whether or not neonates behaved similarly when presented with ginkgo extracts on whole fruit, we compared neonate behaviour in the apple-plug assay with behaviour in our previously described whole-fruit assay (Pszczolkowski and Brown, 2005). Uninfested thinning apples (Red Delicious; about 20 mm diameter) from Mountain Grove experiment orchards were used for whole-fruit assays. Apples were submerged in 10 mg/ml extract of ginkgo in dehydration alcohol or in dehydration alcohol only (about 200 µl of test solution per apple),

and allowed to air dry. Two apples (one ginkgo-treated and one alcohol-treated control) were placed 0.5 cm apart in a 70-mm diameter Pyrex glass crystallizing dish. One neonate was gently placed with a fine camel-hair artistic brush in the space between the fruits and the crystallizing dish was covered with a glass Petri dish. To prevent airflow that could bias the results of the assay, the entire assembly was placed in a semi-translucent 473-ml high-density polypropylene container and covered with a transparent lid. The testing bench and test arenas were illuminated as described for the apple-plug assay. Before each test, glassware and polypropylene containers were washed sequentially in tap water, double distilled water, alcohol, then dried.

Thirty neonates were tested individually in this assay. Both apple-plug assay and whole-fruit assay were conducted at the same time. After 24 h, all apples were examined under a dissecting microscope for evidence of feeding, as described above for the apple-plug assay.

Statistical analysis. Exact Fisher's test (α=0.05) was used in all assays to test the null hypothesis that neonates do not discriminate between plugs or apples treated with plant extract and those treated with alcohol (i.e., 50% of the neonates choose treated plugs or apples and 50% of the neonates choose control plugs or apples).

RESULTS

Effects of ginkgo in apple-plug assay and whole-fruit assay. In both the apple-plug and the whole-fruit assays, the majority (29 of 30, and 28 of 30, respectively) of neonates avoided fruit treated with 10 mg/ml of ginkgo extract ($P< 0.001$). For every neonate, feeding indicators such as abraded epidermis, presence of excrement, or feeding cavities were found on only one apple plug or one apple out of two members of one pair. We conclude that this is evidence that each neonate larva, upon arrival at a ginkgo-treated plug or fruit, did not attempt to feed and was deterred or repelled, or both, by ginkgo extract.

Effects of Artemisia extracts in apple-plug assay. Extracts from three of the five *Artemisia* species discouraged neonates from burrowing into apple plugs (Table 1). Extracts from *A. absinthium, A. arborescens x A. absinthium* and *A. ludoviciana* "Valerie Finnis" were active ($P<0.01$). *Artemisia vulgaris* and *A. californica* had no effect. As in the case of ginkgo, for every neonate, feeding indicators were found on only one apple plug out of two members of one pair. We conclude that *Artemisia* extracts were either repellent or deterrent or both.

DISCUSSION

Deterrent or repellent activity of ginkgo extracts toward codling moth neonates is a novel finding. Surprisingly, information about insect deterrent activity of this plant is scarce in the literature, but what exists provides indirect supportive evidence. Two studies showed that extracts from ginkgo foliage reduce feeding by two insect pests of cabbage: *Pieris brassicae* (Fu-Shun *et al.* 1990) and *P. rapae* (Matsumoto and Sei 1987). Addition of anacardic acids (an alkylphenol found in ginkgo) reduced intake of artificial diet in Colorado potato beetle, *Leptinotarsa decemlineata* (Schultz *et al.* 2006). Other biologically active components of ginkgo foliage include flavonoids and there is evidence that some flavonoids have deterrent and antifeedant activity in insects (Simmonds 2001). At the current stage of our study, we do not know what constituents of ginkgo extract discourage codling moth larvae from burrowing into apple plugs.

The finding that *A. absinthium* deters codling moth neonates corroborates the results of Suomi *et al.* (1986). In their experiments, only 9% of larvae bored into apple plugs treated with a 1% extract ob-

Table 1.
Effect of 10 mg/ml extracts obtained from plants in the genus *Arthemisia* on feeding by codling moth neonates.

Plant species used for apple plug treatment	Number of larvae tested	Number of larvae feeding on treated apple plugs[1,2]
Artemisia absinthium	21	2 **
Artemisia arborescens x A. absinthium	16	1 **
Artemisia ludoviciana "Valerie Finnis"	17	1 **
Artemisia vulgaris	16	4 †
Artemisia californica	18	7 †

[1] ** $P<0.01$, Fisher's exact test
[2] † no statistical significance

tained from this plant. Our results showing that extracts from other members of *Artemisia* genus also have deterrent properties against codling moth neonates are novel, but not surprising in the light of other data from the literature. For instance, the essential oil of *A. annua* has repellent activities against two economically important stored-product pests, the red flour beetle *Tribolium castaneum* (Herbst) and the cowpea weevil *Callosobruchus maculatus* (Tripathi *et al.* 2000). The compound 1,8-cineole isolated from the same plant has feeding deterrent activity against *T. castaneum* (Tripathi *et al.* 2001). Essential oils from *A. vulgaris* also repel *T. castaneum* beetles (Wang *et al.* 2006). The fact that different plants from the same genus have different biological activity against codling moth neonates may facilitate isolation of their active components by comparative chemical analysis.

We think that our findings warrant further studies on effects of ginkgo and *Artemisia* extracts on codling moth neonates. Active constituents of these extracts should be identified, and their potential as codling moth feeding deterrents or repellents – assessed. Perhaps, if manufactured on a larger scale, those constituents could be used as organic alternatives to conventional insecticides for management of codling moth on apples.

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Management of dandelion to supplement control of western flower thrips (Thysanoptera: Thripidae) in apple orchards

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ABSTRACT

We evaluated whether management of the broadleaf weed dandelion, *Taraxacum officinale* F.H. Wigg. aggr., affected damage to apples by western flower thrips (*Frankliniella occidentalis* (Pergande)). Four commercial apple orchard blocks in central Washington having high densities of dandelion were monitored over a 3-yr period. Herbicides were applied to the drive rows in one-half of each orchard for each year of the study. A 92% reduction in dandelion densities in the low-weed plots was achieved by the third year of the experiment. The number of thrips per dandelion plant did not change as dandelions became less numerous. This resulted in an overall reduction in western flower thrips per unit area on dandelions throughout the course of the trial. However, the number of western flower thrips in the apple flowers and shoots were not affected by the treatment. Estimated western flower thrips population density per ha on apple and dandelion indicated that dandelions harboured a much smaller pool of western flower thrips in comparison to apple. No significant reduction in fruit injury was detected in any year. Thus, reduction or elimination of dandelion from the orchard floor appears to be of limited value in managing western flower thrips in apple orchards.

INTRODUCTION

Western flower thrips, *Frankliniella occidentalis* (Pergande), is a sporadic, direct pest of apple (Beers *et al.* 1993). The most conspicuous injury consists of an oviposition puncture, which leaves a small, rugose scar, and a series of white spots surrounding it, commonly known as pansy spot (Newcomer 1921). Injury is most apparent on green or blush cultivars (Madsen and Jack 1966). Densities of adult thrips in apple blossom clusters increase in late April and May as the flowers open; oviposition in fruit occurs from the end of bloom until fruit reaches 30 mm in diameter (Cockfield *et al.* 2007a).

Beginning with the earliest investigations of western flower thrips in apple orchards, broadleaf plants in the groundcover have been assumed to greatly influence the population dynamics of this pest. Venables (1925) collected western flower thrips from tumbling mustard (*Sisymbrium altissimum* L.), a common weed, and alfalfa, *Medicago sativa* L., from apple orchards in British Columbia. These host plants harboured the pest throughout the summer. Childs (1927) stated that the choice and management of cover crops is a major component to western flower thrips management. He recommended cultivating or plowing the orchard ground in early spring to eliminate weeds and to disrupt overwintering thrips in the soil. After the practice of growing cover crops such as alfalfa was no longer common, Madsen and Jack (1966) determined that dandelion, *Taraxacum officinale* F.H. Wigg. aggr., was the most abundant understory host of western flower thrips before and after apple bloom, and wild mustard, *Brassica kaber* (DC.) L.C. Wheeler, and asparagus, *Asparagus officinalis* L., were important alternate hosts in the summer. Pearsall and Myers (2000, 2001) found that not only did a number of broadleaf weeds

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harbour western flower thrips, but orchards with the highest population of weeds had some of the highest numbers of western flower thrips caught in sticky traps.

Two recent experiments done on groundcover management have shown mixed results. Hubscher (1983) found that a temporary perturbation, such as mowing dandelions in spring (intended to redirect pollinators to apple blossoms) had no measurable effect on the population of western flower thrips in apple, or damage to fruit. Cossentine *et al.* (1999) showed that elimination of weeds such as dandelion in small plots reduced thrips populations in trees, but the effect was temporary. The magnitude of the effect may have been limited by the temporal and spatial scale of both studies; thus, a more substantial, permanent effect may result from eliminating weeds for a number of seasons in larger orchard blocks. If successful, this method could reduce the need to control western flower thrips around the bloom period, or reduce fruit damage as part of a multi-tactic IPM program. The purpose of this investigation was to determine the effect of a cultural control method, long-term management of dandelion, on populations of western flower thrips in large blocks of commercial apple orchards and to determine the effect of thrips population changes on fruit damage.

MATERIALS AND METHODS

Site Description. This experiment was conducted over the course of three years in four commercial apple orchards near the towns of Bridgeport, Brewster, Pateros, and Quincy in Washington State, U.S.A. The orchards were selected because of a history of significant thrips damage to fruit and for their high densities of dandelions (>10/m^2). Trees in the Bridgeport orchard block were cv. 'Granny Smith', while the Brewster and Pateros blocks were cv. 'Braeburn'. The Quincy block was planted with alternating two rows of cv. 'Fuji' and 'Braeburn'. Trees were 7-15 yr old. Each block was approximately 2-4 ha and was surrounded by other orchards. There were two treatments: 1) low-weed, where herbicide applications were made in the drive rows (the section of the orchard floor between the tree rows left as mixed grass and weed groundcover to facilitate use of equipment) and 2) high-weed (drive rows untreated with herbicides). Each of the four orchards was divided in half, and treatments assigned randomly, with each site serving as a replicate.

Herbicides. A vegetation-free strip about 2 m wide was maintained beneath the trees in both halves of the block. All orchards in the study had a similar program for weed control in the vegetation-free tree rows. Pre-emergent herbicides, and sometimes 2,4-dichlorophenoxyacetic acid (2,4-D), were applied in early spring, followed by individual sprays or mixtures of paraquat, 2,4-D, and glyphosate after bloom. If necessary, repeat applications were made in June.

The drive rows of low-weed plots were sprayed with 1.1-1.6 kg AI/ha of 2,4-D (Weedar 64, Nufarm Inc., St. Joseph, MO) + 370 ml/100 litres of R11 surfactant (Wilbur-Ellis Co., San Francisco, CA). One or two applications were made in the spring with a weed sprayer calibrated to deliver 234 litres/ha. Any surviving weeds were treated individually with a 9% vol:vol solution of 2, 4-D with a 15.1 litre backpack sprayer (Wil-Gro, Wilbur-Ellis Co., San Francisco, CA).

Insecticides. Insecticides applied for apple pests in a given site were the same across the entire block (high-weed and low-weed plots). Densities of lepidopteran pests were generally low in the study orchards, and required minimal treatment. These pests were managed with applications of chlorpyrifos at delayed dormant, spinosad at petal fall, and azinphos-methyl or methoxyfenozide during early summer. Because of the history of fruit injury, growers applied formetanate hydrochloride to control western flower thrips at full bloom every year.

Sampling Methods. Dandelion densi-

ties were sampled by counting the numbers of plants in a marked 1 m^2 area in the drive rows. Ten randomly selected areas were marked in the middle row of each treatment block, with each area separated by about 3-5 m. Within these areas, dandelion plants (both total numbers and those in flower) were counted monthly from March or April through October.

Thrips densities were sampled monthly on four flowering and four vegetative dandelion plants per plot. All thrips samples were taken the same time of mid-day in each of the paired plots, although the time differed between sites. Dandelion plants (outside the m^2 marked areas) were severed at ground level and placed in self-sealing plastic bags.

Thrips were sampled in the blossoms and vegetative shoots of apple trees. Blossom samples were taken during full bloom (April or May) about one day before insecticides were applied. Twenty-five open apple blossoms were selected from eight trees randomly chosen in the middle row of the plot. The blossoms were clipped off and placed in self-sealing plastic bags. The area sampled was at least 30 m away from the edges of the plots. In the third and final year, the sample size was increased to 150-300 flowers per tree. Thrips in vegetative shoots were sampled monthly after the termination of bloom. The tips (3 cm in length) of 10 growing shoots were collected from each of eight trees.

Thrips samples from dandelion and apple were stored under refrigeration and processed the day after collection. Thrips were separated from plant material by filling the bag with water, adding a few drops of liquid detergent, and agitating for several minutes. Thrips and plant material were separated from the soapy water by pouring through two sieves (Hubbard Scientific Co., Northbrook, IL). The larger sieve (#10, 0.25-mm mesh) trapped most of the plant material, and the finer sieve (#230, 0.0014-mm mesh) trapped the thrips. Thrips were then rinsed into a vial of 50% ethanol. Specimens were first examined under a dissecting microscope. All *Frankliniella* adults were slide-mounted in PVA Mounting Medium (BioQuip Products, Inc., Rancho Dominguez, CA) for identification to species. A reference collection was sent to Cheryl O'Donnell, Department of Entomology, University of California, Davis, CA, who confirmed the identity of species. Only numbers of *F. occidentalis* adults were recorded. The means of the samples taken per plot were used in the analyses. All data were expressed as thrips per dandelion plant, apple flower, or apple shoot. Thrips per dandelion plant during peak dandelion flowering were averaged over the three years of the study.

Population Estimates. The monthly population density of western flower thrips per m^2 in dandelions in the drive rows was estimated by multiplying the average number of thrips per sampled nonflowering and flowering dandelions by the average number of dandelions of each type per m^2. Estimates were also summed for each replicate over the three years of the study. The estimate at peak dandelion bloom, just before or during apple bloom, was then used to calculate the number of western flower thrips on dandelion on a per-hectare basis. This was done by multiplying the mean number of thrips per m^2 by the number of m^2 of drive rows in each ha. Similarly, estimates were obtained for western flower thrips per ha in apple flowers. Flower densities were estimated by counting all blossoms on 10 trees per plot. Mean flowers per tree were calculated for each site, then converted to means per ha based on the tree density per ha. Flowers per ha were multiplied by the mean number of thrips per flower.

Data Analysis. The experimental design was a randomized complete block with the four orchards serving as replicates. The effects of herbicide treatment and month on dandelion densities were assessed using a two-way analysis of variance having repeated observations through time (=month). The analyses were done in SAS using PROC MIXED (SAS Institute 2002). Separate analyses were done for each of the three years of the study. We used a square

root transformation on the count data before each analysis to meet ANOVA assumptions. In the event of a significant treatment × month interaction, tests on simple effects of treatment and month were done using the SLICE command. Analyses of flowering dandelions were done only for the sampling date with the highest number of flowering plants. Comparisons of flowering dandelions were made by year using paired t-tests (SAS Institute 2002). Data for thrips densities, whether monthly, averaged, or summed measurements, were analyzed by date using ANOVA. Treatment means were separated using a LSD test (high-weed vs low-weed, α=0.05). Data for proportion (p) of fruit with pansy spot injury were first transformed by arcsine(square root (p+0.001)), then analyzed using ANOVA (SAS Institute 2002).

RESULTS AND DISCUSSION

Dandelion Densities. Densities of total dandelions were high in both high-weed and low-weed treatments at the start of the experiment (Fig. 1A). A few other broadleaf weeds also occurred in the orchard blocks, most notably alfalfa, but population densities of these other weeds were zero in the randomly-sampled areas. All perennial and annual broadleaf weeds were affected by the herbicides and were reduced in the low-weed plots, which resembled manicured lawns. The treatment × month interaction terms were highly significant in 2003 (F=4.10; df=7, 42; P=0.0016) and 2004 (F=3.39; df=8, 48; P=0.0037), and significant in 2005 (F=2.37; df=7, 42; P=0.039), thus we examined the simple effects tests (herbicide effect for each month separately). Treatment differences became significant by July of 2003, with 17.1 (± 11.6 SEM) dandelions/m^2 in the low-weed plots versus 66.1 (± 23.4) in the high-weed plots. These differences were sustained through the remainder of the year. Treatment differences were only marginally significant for the first two months of 2004 (P<0.10), but were re-established by May with lower dandelion densities in the low-weed plots throughout the remainder of the year (P<0.001). Treatment differences were significant throughout 2005 (P<0.001), which was also reflected in the main effect treatment means for this year (low-weed plots, 2.3 ± 0.3; high-weed plots, 29.4 ± 2.4 dandelions/m^2; F=75.79; df=1, 6; P=0.0001). The treatment differences in 2005 reflected a 92% reduction in dandelion densities in the low-weed plots relative to the high-weed plots.

Dandelions flowered primarily in April or May; very few plants flowered in the summer and fall (Fig. 1B). During the peak period, flowering dandelion density was not significantly different in the low-weed (5.5 ± 1.6) and high-weed (7.5 ± 3.4) plots in 2003 (t=0.60, df=3, P=0.59). The effect of the herbicide applications was more clearly seen in 2004 (low weed, 2.8 ± 1.8; high weed, 10.1 ± 3.1; t=3.74, df=3, P=0.03) and 2005 (low weed, 0.3 ± 0.1; high weed, 8.0 ± 1.8; t=4.36, df=3, P=0.02).

Thrips in Dandelions. The estimated population density of thrips on dandelion in the drive rows (on a per m^2 basis) generally increased in the high-weed blocks in late spring, and peaked in April (2005), during which maximum dandelion bloom occured, or June (2003, 2004) (Fig. 1C). Adult thrips decreased in numbers by July and August and remained at low densities in the fall. Estimated densities of thrips were highly variable between sites and monthly comparisons were not significantly different; however, there were higher thrips densities in dandelions in the high-weed plots (33.7 ± 11.0) than in the low-weed plots (2.9 ± 0.7) when the estimates were summed over the three years (F=7.79; df=1,6; P=0.031).

The three-year average of thrips per flowering dandelion plant was 0.22 in high-weed and 0.33 in low-weed blocks, while thrips per vegetative plant was 0.04 in high-weed and 0 in low-weed blocks during peak dandelion flowering (F=0.38; df=1,6; P=0.56,). No difference in thrips density (on a per plant basis) was found in flower-

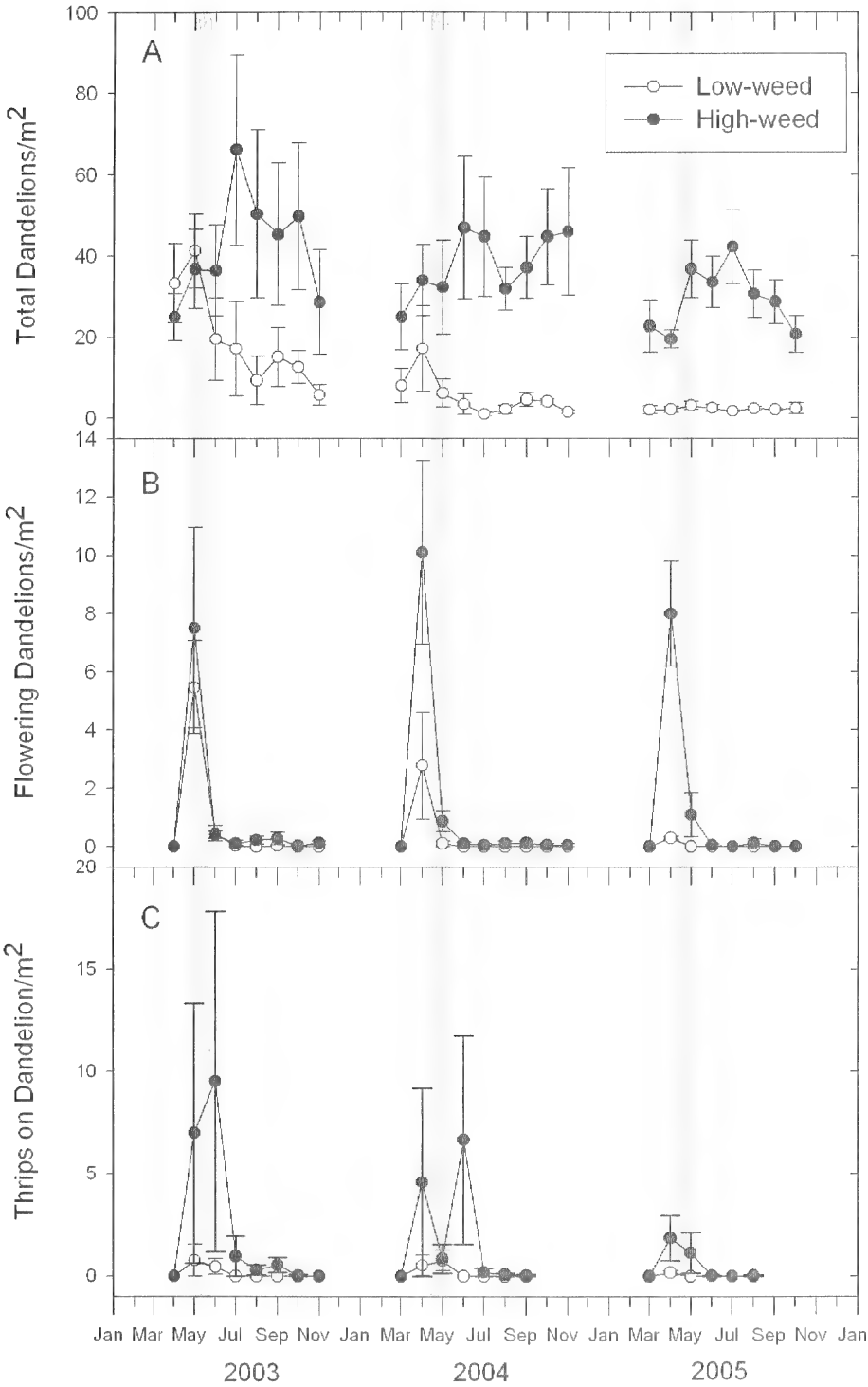

Figure 1. (A) Dandelions per m^2 area of orchard ground in low-weed and high-weed orchard blocks. (B) Blooming dandelions per m^2 area of orchard ground in low-weed and high-weed orchard blocks. (C) Western flower thrips population estimates (adults per m^2) on dandelions in low-weed and high-weed orchard blocks. Symbols are means, error bars are SEM. Grey bars indicate the period of apple bloom.

ing dandelions from either low-weed or high-weed blocks at any sample period. Therefore, thrips population density per dandelion was largely not influenced by dandelion density. The reduction of thrips per area was determined by reduction of dandelions.

Thrips in Apple. All of the thrips specimens collected from apple flowers in the three years of the experiment were western flower thrips, *F. occidentalis*. Western flower thrips has been the dominant thrips species collected from apple flowers in the inland Pacific Northwest (Venables 1925, Childs 1927, Madsen and Jack 1966, Cockfield *et al.* 2007b). In contrast, apple shoots and dandelions contained a mixture of species, including *F. occidentalis*. Western flower thrips accounted for about 38% of the thrips in apple shoots sampled in Washington state (Cockfield *et al.* 2007b). No significant differences in adult western flower thrips populations were found in any of the apple flower or shoot samples from high-weed and low-weed plots, even in 2005, after dandelion numbers had been greatly reduced in the low-weed plots (Table 1). Thrips densities at full apple bloom were average compared with other samples in Washington (Miliczky *et al.* 2007). Thrips in the apple shoots may contribute to fruit injury in late May, but more likely sustain the population in the orchard throughout the summer and from year to year (Cockfield *et al.* 2007a, 2007b). While thrips densities provide a useful measure of the treatment effects, the critical measurement for the purposes of management is fruit damage. In the study orchards, an insecticide treatment was inadequate to prevent fruit injury, indicating the need for an additional management tactic. However, significant reductions in numbers of dandelions did not correspond with a significant reduction in fruit injury in any of the three years (Fig. 2). Thus the effort and time investment needed to manage broadleaf weeds did not provide a substantial benefit to fruit damage reduction.

One possible explanation for the lack of effect is that thrips moved between high-weed and low-weed plots, in spite of the large size of the experimental plots. A second and more likely explanation is that the potential contribution of western flower thrips from dandelion is relatively small during late spring, when fruit injury occurs. While dandelion flowers often harbour large numbers of thrips, western flower thrips may be <10% of the total individuals present (Cockfield *et al.* 2007b). The per-hectare estimates of thrips densities indicated that 45,000 and 4,000 adult western flower thrips per ha (high-weed and low-weed plots, respectively) occurred on dandelion, compared to estimates of 141,000 and 157,000 thrips per ha, respectively, on apple flowers at peak bloom just before the critical period for fruit damage. The attractiveness of flowering plants to western flower thrips is well established (Terry 1991, 1997). During their bloom periods there were 1.9 million apple blossoms per ha compared with 43,000 dandelion blossoms; even assuming they are equally attractive, the sheer number of apple flowers could be expected to dominate this interaction during the bloom period.

There is ample evidence this highly polyphagous species is abundant in many hosts other than apple, including crop and non-crop plants. Recent studies (Pearsall and Myers 2000, Pearsall and Myers 2001, Cockfield *et al.* 2007b) clarify that in the semi-arid interior fruit growing districts of the Pacific Northwest, multiple species of the native vegetation serve as a host for thrips. Further studies indicate that immigration from the native vegetation may affect thrips density and damage in orchard borders (Miliczky *et al.* 2007). Even though the orchards in this study were not adjacent to native vegetation, it is still potentially a very large source of thrips in the industry. Removal of one relatively small source, dandelion blossoms, would constitute only a minor change in local populations. This, coupled with the large resource constituted by apple blossoms, and to a lesser extent, vegetative tissues, effectively negates any benefit of dandelion removal.

Table 1.
Western flower thrips, mean (SEM), sampled per apple flower and per shoot in high-weed and low-weed treatments[1].

	Low-weed		High-weed			
Date	**Flowers**	**Shoots**	**Flowers**	**Shoots**	***F***	***P***
May 2003	0.148 (0.068)a		0.106 (0.051)a		1.42	0.320
June 2003		0.194 (0.034)a		0.181 (0.041)a	0.15	0.721
July 2003		0.056 (0.030)a		0.069 (0.011)a	0.24	0.658
Aug 2003		0.028 (0.020)a		0.028 (0.020)a	0.0	1.000
Apr 2004	0.035 (0.012)a		0.025 (0.006)a		1.04	0.382
May 2004		0.388 (0.229)a		0.316 (0.126)a	0.43	0.557
June 2004		0.203 (0.123)a		0.121 (0.059)a	1.54	0.303
July 2004		0.156 (0.080)a		0.113 (0.053)a	1.01	0.388
Apr 2005	0.074 (0.027)a		0.089 (0.031)a		4.54	0.123
May 2005		0.028 (0.016)a		0.084 (0.028)a	4.96	0.112
June 2005		0.190 (0.112)a		0.200 (0.122)a	6.00	0.092
July 2005		0.058 (0.011)a		0.058 (0.018)a	0.0	1.000

[1] Means within rows followed by the same letter are not significantly different, LSD test, α=0.05. For all analyses, df=1,3.

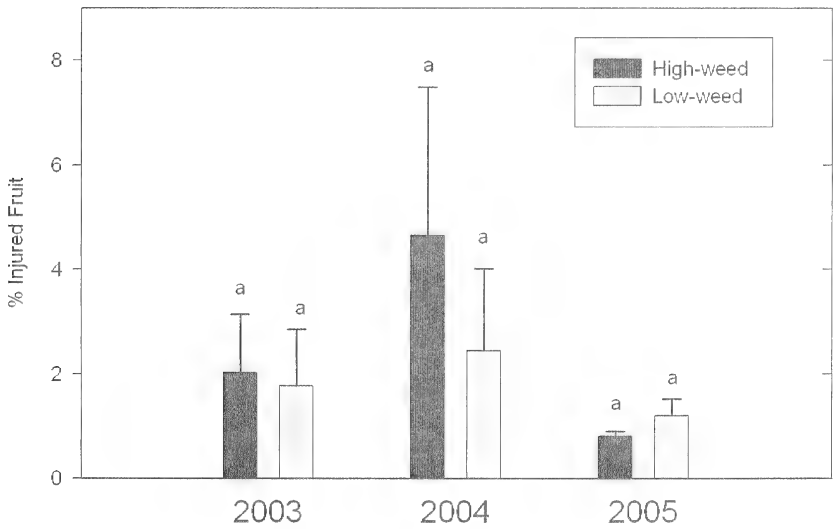

Figure 2. Percentage of fruit with pansy spot in low-weed and high-weed plots. Bars are means, error bars are SEM. Means with the same letter are not significantly different within each year. 2003: F=0.72; P=0.4581. 2004: F=2.33; P=0.2241. 2005: F=1.15; P=0.3625. For all analyses, df=1,3.

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Effect of pesticides on integrated mite management in Washington State

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ABSTRACT

The effect of pesticides used against codling moth, *Cydia pomonella* L., on integrated mite management was studied for three years in five or six commercial apple orchards in central Washington. Phytophagous and predatory mites were counted throughout the season in blocks ranging from 0.4-1.6 ha in size treated with four codling moth insecticides. In one year of the study (2006), five out of six orchards experienced elevated mite densities relative to the standard. In four orchards, novaluron caused a 3.0-16.9× increase in mite populations; acetamiprid caused a 2.6-3.4× increase, and thiacloprid caused a 1.7-13.8× increase. In the fifth orchard, the organophosphate standard had an extremely high mite population, in addition to all three experimental treatments. In 2005 and 2007, only one or two orchards had elevated mite levels in the novaluron, acetamiprid, and thiacloprid treatments. Additive effects of codling moth and thinning programs were evaluated in small plot research trials. Treatments with all three elements [1) codling moth insecticide; 2) calcium polysulfide; 3) carbaryl] produced the highest levels of spider mites. Three sulfur-containing products (calcium polysulfide, ammonium thiosulfate, and dry flowable sulfur) were evaluated for their effect on *Galandromus occidentalis* (Nesbitt) and apple rust mite, *Aculus schlechtendali* (Nalepa). All three materials caused suppressed *G. occidentalis* numbers. Calcium polysulfide caused the greatest reduction in apple rust mite numbers, ammonium thiosulfate the least reduction, with dry flowable sulfur intermediate between the two. Additive effects of codling moth materials, carbaryl, and sulfur-containing products may be causing increased mite levels in Washington orchards.

Key Words: spider mite, integrated mite control, apple, acetamiprid, thiacloprid, novaluron, carbaryl, calcium polysulfide, lime-sulfur, dry flowable sulfur, ammonium thiosulfate

INTRODUCTION

Spider mites are induced pests of pome fruits, generally occurring in orchards which have been disrupted by pesticides. In wild or abandoned trees, spider mites are usually maintained at low densities by predators (Glass and Lienk 1971, Croft 1983). However, in heavily sprayed systems such as commercial apple orchards, perturbations occur regularly based on the need to control direct pests. In Washington State, codling moth, *Cydia pomonella* L., is the key direct pest of apple, and control measures used against it determine the entire pest management program and structure the fauna of the agroecosystem.

The history of spider mite management in apple orchards is characterized by disruption of mite biological control following the introduction of new materials for codling moth control. DDT was introduced following WWII, and its use in tree fruits was accompanied by large scale mite out-

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breaks (Newcomer and Dean 1946, Baker 1952, Clancy and McAlister 1956). As a result of the disruption, acaricide resistance became widespread (Hoyt and Caltagirone 1971). DDT was replaced by organophosphate insecticides, including azinphosmethyl, which initially were toxic to both pest and predatory mites. Spider mites became resistant to the organophosphates, but it was not until the primary predator of spider mites, *Galandromus (=Typhlodromus = Metaseiulus) occidentalis* (Nesbitt) also became resistant that the opportunity for integrated mite control arose in the western US (Hoyt 1969). Integrated mite management was then implemented on approximately 90% of the acreage (Whalon and Croft 1984) by conserving the organophosphate-resistant predatory mites, with growers actively avoiding materials that were toxic to these valuable predators. This opportunity, however, was predicated on continuing efficacy of azinphosmethyl for codling moth control.

The integrated mite control program implemented in the early 1970s in Washington remained largely effective through the 1990s. During this time shifts in the pesticide program occurred, at least for pests other than codling moth. The efficacy of the organophosphates against many of the secondary pests of tree fruit (aphids, leafhoppers, leafminers, and leafrollers) declined steadily through this period, and new materials were substituted for their control. The use of carbaryl for fruit thinning was implemented in the late 1970s; this carbamate insecticide was initially highly toxic to *G. occidentalis*, and its use was restricted to protect predator populations in integrated control programs. However, moderate levels of resistance to this carbamate were documented within a short period of use (Babcock and Tanigoshi 1988). Other carbamate insecticides, similarly toxic to the predator, were also used sparingly, typically only when no other substitute was available. The use of pyrethroids, notoriously disruptive to integrated mite control, was largely avoided in Washington apples in order to protect the integrated mite control program.

However, following almost forty years of reliance on azinphosmethyl for codling moth control, shifts in the codling moth management program began in the mid-1990s. These changes have been driven mainly by either the development of organophosphate resistance in codling moth or by regulatory issues (Beers *et al.* 2005). In the meantime, new control techniques and materials have increased in use. Mating disruption now forms the foundation of codling moth control in about 80% of Washington's apple orchards (J. Brunner, personal communication). Choices for supplementary insecticides include insect growth regulators and neonicotinoids. While the new materials meet the new standard for improved worker safety, their effects on natural enemies and predator/prey dynamics have not been well explored.

One class of alternative insecticide chemistry, the neonicotinoids, demonstrated a tendency to disrupt integrated mite control even in the early phases of testing (Beers *et al.* 2005). These tests, however, were characterized by small plots, high rates, and season-long programs. It remained to be seen if the potential for disruption still existed under commercial use conditions (large acreages, applications against a single generation of codling moth with any given material). Additionally, potential multiple-year effects from continued use of the same products could not easily be examined prior to registration.

Other shifts in the Washington pesticide program occurred during the same period as the change in the codling moth program, especially in the crop load regulation and fungicide programs. The blossom thinner, sodium dinitro-o-cresylate (Elgetol®), was withdrawn from the market in the mid 1990s, and its use was later replaced with calcium polysulfide (lime sulfur). A plant nutrient, ammonium thiosulfate, also became more widely used; its sulfur content is similar to calcium polysulfide. The use of sulfur fungicides increased in part because of the plantings of mildew-susceptible cultivars, and in part as an alternative mode of

action for fungicide resistance management (FRAC 2008).

The goals of this study were to explore the effects on integrated mite control of three newer codling moth insecticides when used in a commercial setting; to examine the additive effects of codling moth insecticides, carbaryl, and calcium polysulfide in a seasonal program; and the comparative effects of three sulfur-containing compounds used as a blossom thinner, fungicide, and plant nutrient, respectively.

MATERIALS AND METHODS

Large-block experiment. This test was conducted in five (2005, 2007) or six (2006) commercial apple orchards from Bridgeport to Royal City, WA. Plot size ranged from 0.4-1.6 ha per treatment at each orchard; treatments were randomly assigned to one of four plots within an orchard, and replicated across the orchards. All treatments were applied at a finished spray volume of 935 litres/ha by the orchard's personnel using their own equipment. The dominant cultivar in the blocks was either 'Delicious' or 'Fuji'. Four of the orchards received the same treatments for all three years of the study (orchards ARR, BAN, QLR, SLH); MZN was treated in 2006-07, while RYL and BTE were treated only in 2005 and 2006, respectively.

Treatments consisted of one of the four insecticides used for codling moth control: acetamiprid, a neonicotinoid (Assail® 70W, Cerexagri, King of Prussia, PA; 0.17 kg AI/ha); thiacloprid, also a neonicotinoid (Calpyso® 4F, Bayer CropScience, Research Triangle Park, NC; 0.21 kg AI/ha); novaluron, a benzoylurea insect growth regulator (Rimon® 0.83EC, Chemtura, Middlebury, CT; 0.23 g AI/ha); and an organophosphate standard. For the organophosphate standard, growers could choose either phosmet (Imidan® 70W, Gowan, Yuma AZ; 3.9 kg AI/ha) or azinphosmethyl (Guthion® 50W, Bayer CropScience, Research Triangle Park, NC; 1.1 kg AI/ha). Applications of acetamiprid, thiacloprid, and novaluron were made only during the first generation of codling moth (May and June). Two applications of acetamiprid and thiacloprid were made per season, the first timed for 250 codling moth degree days, and the second 21 d later. Three applications of novaluron were made (based on its ovicidal activity), the first at petal fall, the second 14 d later, and the third 28 d later.

For the above the treatments, codling moth control for the second generation (July and August) consisted of two applications of the benzoyl hydrazine insect growth regulator methoxyfenozide (Intrepid® 2F, Dow AgroSciences, Indianapolis, IN; 0.28 kg AI/ha), the first timed for 1,250 codling moth degree days, and the second 21 d later.

In the organophosphate standard treatment, two applications were made per generation, using the same timing as for acetamiprid and methoxyfenozide for the first and second generations, respectively.

Mites were sampled every 2-3 wk from late May through mid-September. One hundred leaves per plot were collected from the center portion of the plot and kept cool during transportation and storage. The mites were brushed from the leaves using a mite brushing machine (Leedom, Mi-Wuk Village, CA) and collected on a revolving sticky glass plate. The composite sample on the plate was counted using a stereoscopic microscope. Phytophagous and predatory mites were recorded, including the motile stages of European red mite, *Panonychus ulmi* (Koch); twospotted spider mite, *Tetranychus urticae* Koch; McDaniel spider mite, *Tetranychus mcdanieli* McGregor western predatory mite, *G. occidentalis*; a stigmaeid predatory mite, *Zetzellia mali* Ewing, and apple rust mite, *Aculus schlechtendali* (Nalepa).

Additive effects experiment. This small-plot experiment examined the effect of adding one or two potentially disruptive compounds used for crop load regulation to the same codling moth programs (rates, timing, and materials) described for the

large plot experiment. The compounds used were a blossom thinner, calcium polysulfide (Rex Lime Sulfur®, Or-Cal, Junction City, Oregon), and a fruit thinner, carbaryl (Sevin® 4F, Bayer CropScience, Research Triangle Park, NC). Both compounds were used at their respective recommended timings (Smith *et al.* 2006). Calcium polysulfide was applied three times (pink, 20% and 80% bloom) at a rate of 8% vol:vol. Carbaryl was applied twice, when the fruitlets were 8 and 12 mm in diameter, at rate of 1.7 kg AI/ha.

This test was conducted in a 2 ha block of mature 'Oregon Spur' and 'Red Spur' Delicious apples with 'Golden Delicious' pollenizers. Plots were five rows by five trees. The experimental design was a randomized complete block design with 12 treatments and 4 replicates. All applications were made with an airblast sprayer (Rears Pak-Blast, Eugene, OR) calibrated to deliver 935 litres/ha. Treatment timings and materials for first and second generation codling moth control were the same as described in the large-block experiment.

Mites were sampled every other week from May through September by collecting 40 leaves per plot. The leaves were collected, stored and processed as described above.

Sulfur products experiment. The second small-plot experiment examined the effect of three sulfur-containing products on *G. occidentalis* and apple rust mite. For purposes of comparison, the materials were applied to an existing population of these two species in June, rather than at their normal timing which ranged from prebloom through the early post-bloom period. The three sulfur products were calcium polysulfide (Rex Lime Sulfur®, Or-Cal, Junction City, Oregon; 12% vol:vol), ammonium thiosulfate (a plant nutrient) (Thio-Sul®, Tessenderlo Kerley, Phoenix, AZ; 3.4% vol:vol), and dry flowable sulfur (a fungicide) (Kumulus® 80DF, Micro-Flo, Memphis, TN; 10.8 kg AI/ha).

The experimental design was a randomized complete block (randomized on the basis of a pretreatment count) with seven treatments and four replicates. Each replicate consisted of three trees in a single row, with one untreated buffer row separating the treatment rows. Plots consisted of three cultivars, 'Oregon Spur', 'Goldspur', with 'Red Fuji BC2' in the center; however, only the center tree was sampled. Treatments consisted of either one or three applications of each of the three sulfur-containing compounds plus an untreated check. Treatments receiving a single application were applied 26 June 2006; treatments receiving three applications were made 26 June, 8 July and 19 July. Treatments were applied by airblast sprayer at 935 litres/ha. Mite populations were assessed by collecting 25 leaves per plot and processed using the method described above. Counts were made pretreatment and weekly after treatment through late July.

Data analysis. Cumulative mite days (CMDs) were calculated for tetranychid (*P. ulmi* plus *T. urticae*), predatory (*G. occidentalis* plus *Z. mali*), and apple rust mite. CMDs provide an estimate of population densities integrated over the course of the test, and are calculated as the sums of the average density of mites on two dates multiplied by the number of intervening days:

$$CMD = \Sigma 0.5(P_a+P_b)D_{a-b}$$

where P_a and P_b are the population densities (mean mites/leaf) at times a and b, and D_{a-b} is the number of days between time a and time b.

Data were analyzed using the Statistical Analysis System (SAS 1988). Data were tested prior to analysis for homogeneity of variance using Levene's (1960) test. Variances found to be non-homogeneous were transformed [ln(y+0.5)] before analysis. PROC GLM was used to conduct an analysis of variance, and treatment means were separated using the Waller-Duncan *k*-ratio *t*-test. Single degree-of-freedom contrasts were used to compare groups of treatments in the small plot experiments.

RESULTS

Large-block experiment. Spider mite populations in the experimental blocks consisted primarily of European red mite, with 99, 92, and 78% of the population of motile forms comprised of this species in 2005, 2006, and 2007, respectively. Twospotted spider mite was the next most numerous species; a relatively higher proportion of this species occurred only in one orchard, MZN, in 2007, when 57% of the motile forms were twospotted spider mite. Only trace numbers of McDaniel spider mite were found during the course of the study.

There were no statistical differences among treatment mean CMDs for tetranychids or for predatory mites in any of the three years of the study (Table 1). In 2005 and 2006, rust mite populations were higher in the acetamiprid treatment compared to the standard. In general, the differences in densities among years and orchards were greater than those among treatments.

Despite this variation, some trends in these data are apparent. In 2005, elevated mite densities occurred in only one of five orchards (QLR) (Fig. 1). However, the highest mite levels occurred in the novaluron (peak density 54 mites/leaf), acetamiprid (21 mites/leaf), and thiacloprid (20 mites/leaf) treatments, with only a moderate increase in the organophosphate standard treatment (11 mites/leaf). Not all of the population peaks in the treatments with newer insecticides can be explained by low predator numbers, although predatory mite densities were highest in the organophosphate treatment (peak density 2.3 predators/leaf). The thiacloprid treatment peaked at 0.9 predators/leaf, while the acetamiprid and novaluron treatments never exceeded predator densities of 0.3/leaf. Apple rust mite densities were moderate in most of the treatment (peak density of 150-300 rust mites/leaf), with the exception of the novaluron treatment, which had relatively low rust mite densities (<10/leaf) for most the season.

Mite densities were much higher overall in 2006 than in 2005 (Fig. 1). Five of six orchards experienced elevated tetranychid mite densities in one or more treatments. This may have been due to a cumulative effect of disruptive products in four of the orchards; however, 2006 was characterized by a high frequency of mite outbreaks throughout the central fruit-growing district of the state. The novaluron treatment had elevated tetranychid mite levels in five orchards (20-45 mites/leaf at peak density); the acetamiprid treatment in two orchards (22-34 mites/leaf); and the thiacloprid treatment in three orchards (13-32 mites/leaf). One orchard (QLR) had a high peak mite density in the organophosphate treatment, as well as the other three treatments; however, this was the same orchard that had high levels in several treatments the previous year. Trends in predatory mite densities were again difficult to interpret. Although no statistical differences occurred among treatment means for the entire season, the peak densities of predators occurred too late in the season to prevent the mid-July peak in tetranychid mites (data not shown).

Mite densities in the experimental orchards were much lower in 2007 than in 2006 (Fig. 1), with no treatment exceeding 7 mites/leaf. Only two of the five orchards experienced a moderate increase in tetranychid mite levels, with a slight elevation in the novaluron treatment in one orchard (SLH) (6.2 mites/leaf peak density), and acetamiprid in two orchards (4.4 and 3.5 mites/leaf in MZN and SLH, respectively) and thiacloprid (3.6 mites/leaf) in one orchard (MZN).

Additive effects experiment. The results from the experiment examining the additive effect of several disruptive products during the season showed a distinct trend toward increased tetranychid mite densities when one of the newer codling moth insecticides was used in the same program with both a blossom and fruit thinner (Fig. 2). The lowest tetranychid mite densities occurred in those treatments where only insecticides for codling moth were used. Treatments where all three compounds were used (codling moth insecticide

Table 1.
Seasonal mite densities (cumulative mite days) resulting from four codling moth control regimes, in commercial apple orchards in Washington, 2005-2007

Treatment	Rate (AI/ha)	n	Cumulative mite days[1]		
			Tetranychids (± SEM)	Predators (± SEM)	Apple rust mite (± SEM)
			2005		
Acetamiprid	0.17 kg	5	113 ± 99a	12 ± 2a	10,861 ± 2,690a
Thiacloprid	0.21 kg	5	93 ± 84a	17 ± 4a	8,563 ± 1,718ab
Novaluron	0.23 kg	5	252 ± 244a	11 ± 3a	6,589 ± 2,639b
Standard	---	5	49 ± 38a	26 ± 8a	6,668 ± 1,394b
		F, P	4.32, 0.013	1.90, 0.16	12.03, 0.0001
			2006		
Acetamiprid	0.17 kg	6	267 ± 132a	26 ± 3a	870 ± 372a
Thiacloprid	0.21 kg	6	305 ± 149a	28 ± 5a	634 ± 244ab
Novaluron	0.23 kg	6	520 ± 178a	33 ± 13a	563 ± 301ab
Standard	---	6	401 ± 343a	27 ± 5a	476 ± 197b
		F, P	4.78, 0.0045	1.98, 0.12	19.80, <0.0001
			2007		
Acetamiprid	0.17 kg	5	55 ± 31a	29 ± 13a	719 ± 346a
Thiacloprid	0.21 kg	5	18 ± 10a	35 ± 14a	883 ± 240a
Novaluron	0.23 kg	5	37 ± 23a	16 ± 8a	346 ± 153a
Standard	---	5	23 ± 15a	32 ± 16a	632 ± 229a
		F, P	6.17, 0.0032	5.74, 0.0043	3.43, 0.030

[1] Means within columns not followed by the same letter are significantly different. For 2005 and 2007, df=7, 19; for 2006, df=8, 23.

+ calcium polysulfide + carbaryl) had significantly higher tetranychid mite densities than when codling moth insecticides alone were used (df=1, F=5.54, P=0.02).

Trends in seasonal densities of predatory mites and apple rust mite were less clear (Figs. 3, 4). Comparisons of treatments with or without calcium polysulfide indicated that there was a significant reduction in the seasonal apple rust mite densities where calcium polysulfide was included in the program (df=1, F=5.07, P=0.03), however, there was no effect on predatory mite densities (df=1, F=0.70, P=0.41).

Sulfur products experiment. All three sulfur products used in this study suppressed *G. occidentalis* to about the same extent (Fig. 5). There was a 64-74% reduction in densities of *G. occidentalis* in the treatments containing sulfur products in relation to the check. There was no difference between treatments with one application versus three applications (df=1, F=0.11, P=0.75), likely because most of the mortality had occurred from the first application, without sufficient time for reinfestation between applications.

The effect of the three sulfur products on apple rust mite was more variable. There was a 30-80% reduction in densities of apple rust mite in these treatments. The reduction in apple rust mite numbers was greatest in the calcium polysulfide treatment (Fig. 6), and least in the ammonium thiosulfate treatment. As with *G. occidentalis*, there were no differences in treatment means between treatments with one versus three applications (df=1, F=0.99, P=0.33).

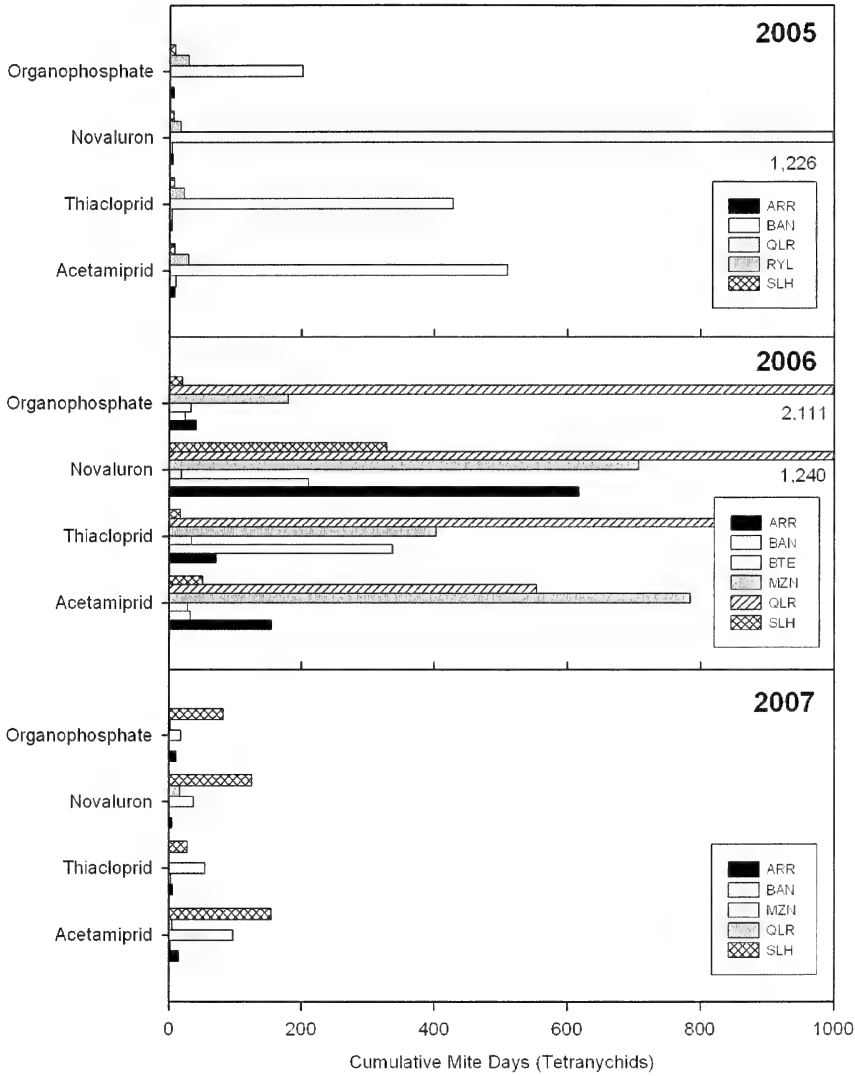

Figure 1. Seasonal tetranychid mite densities (cumulative mite days) in commercial apple orchard blocks treated with four insecticides for codling moth control, 2005-2007.

DISCUSSION

The responses to the two neonicotinoid insecticides used in the large-block study confirms previous work done on small plots (Beers *et al.* 2005). Mite populations in the acetamiprid treatments averaged 2.3, 2.2 and 3.0× higher than the standard organophosphate treatment during 2005-2007, respectively. Mite populations in the thiacloprid treatments averaged 1.3, 3.4 and 2.2× higher than the standard. In addition to the neonicotinoids, this study provides evidence that novaluron also causes disruption of integrated mite management, although this trend was not apparent in small-plot trials (J. Brunner, personal communication). Mite populations in the novaluron treatments were 2.1, 7.6, and 2.7× higher than the standard treatment in the three years of the study.

Although widely observed, the mecha-

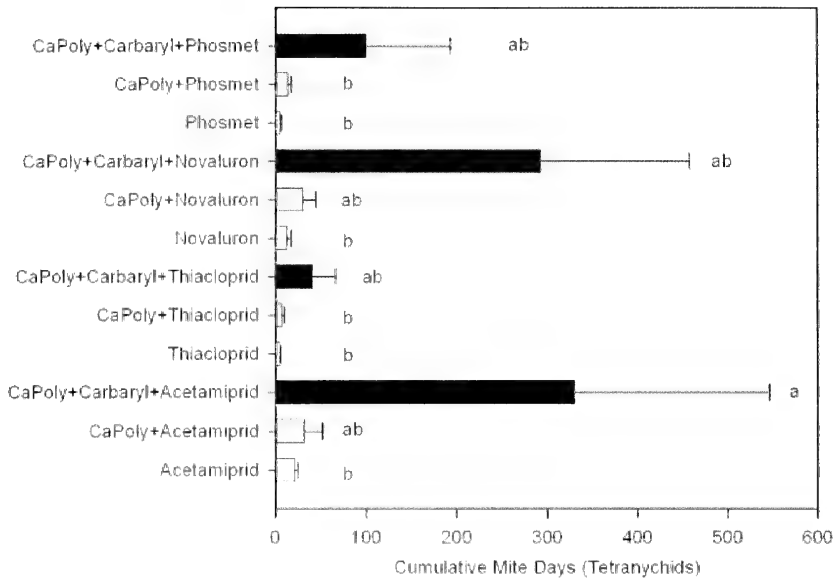

Figure 2. Additive effect of thinning materials and codling moth insecticides on seasonal

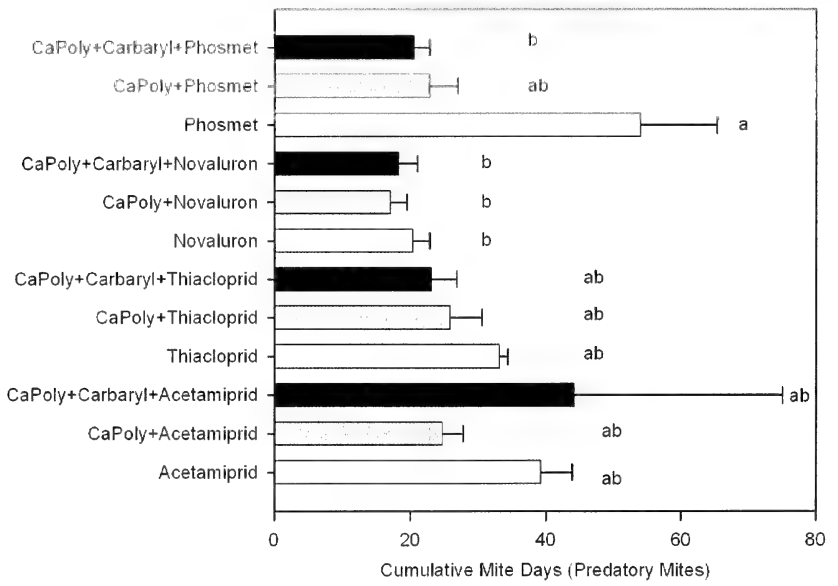

Figure 3. Additive effect of thinning materials and codling moth insecticides on seasonal predatory mite populations. F=2.05, P=0.054, df=11, 47. Data transformed log(x+0.5) prior to

nism for the neonicotinoid effect has never been clearly established. Hormoligosis is thought to play a role in stimulating pest reproduction (James and Price 2002), but other studies have found no hormoligosis effect (Ako *et al.* 2004, Ako *et al.* 2006). Conversely, neonicotinoids have also been found to stimulate reproduction in beneficial arthropods (James 1997). Repellency (James 1997) and suppression of functional

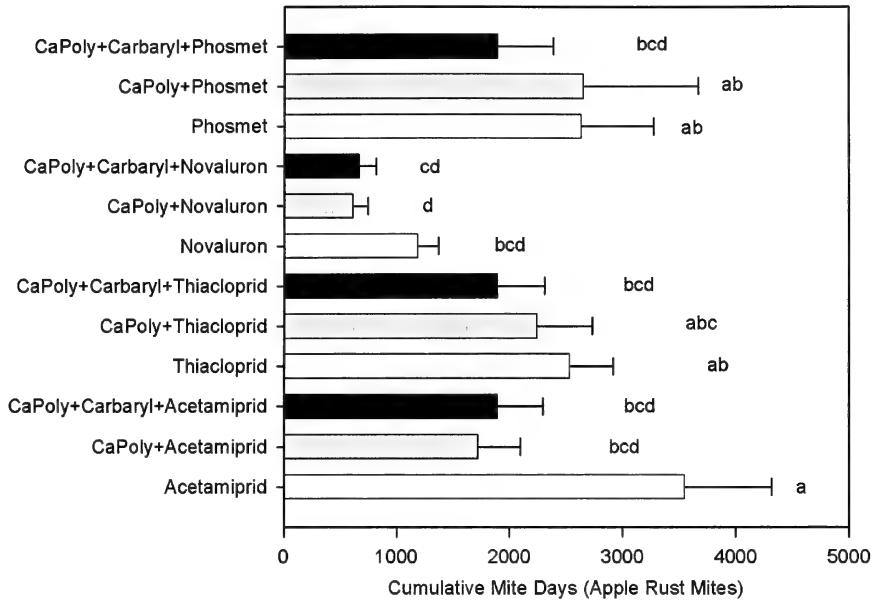

Figure 4. Additive effect of thinning materials and codling moth insecticides on seasonal apple rust mite populations. $F = 3.03$, $P = 0.0067$, df = 11, 47.

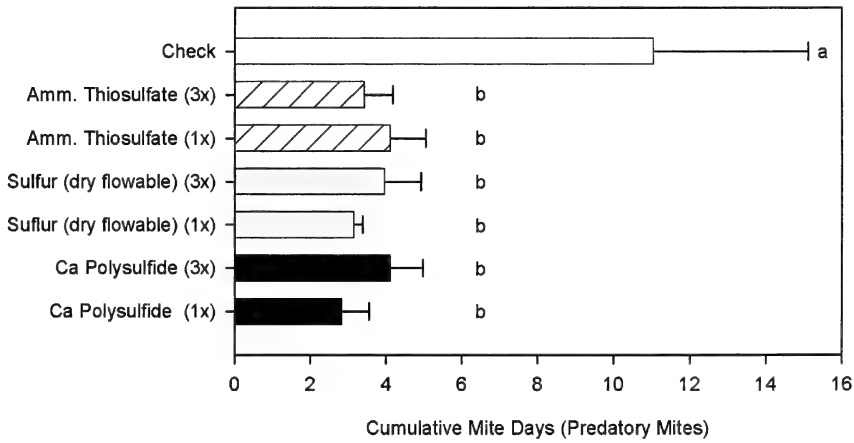

Figure 5. Effect of sulfur-containing products on predatory mites. $F = 2.68$, P=0.0.049, df = 6, 27.

response (Poletti *et al.* 2007) may also play a role in the disruption of biological control.

It is evident from previous studies (Beers *et al.* 2005) that while neonicotinoids can cause mite outbreaks, they would not do so in every case. This makes the role of other disruptive materials more important on a relative scale. In an organophosphate-based pest management program, calcium polysulfide and carbaryl had been used with few apparent deleterious effects; under this program, only about 7% of Washington's apple orchards were treated with acaricides (NASS 1992). The low mite levels documented by the survey are likely typical of acaricide use from the early 1970s, when integrated mite control was first established, until the early 2000s when shifts in codling moth insecticides began. However, there has been a substantial increase in the percentage of Washington apple acreage treated with sulfur fungicides (7.8×) and calcium polysulfide (11×) since

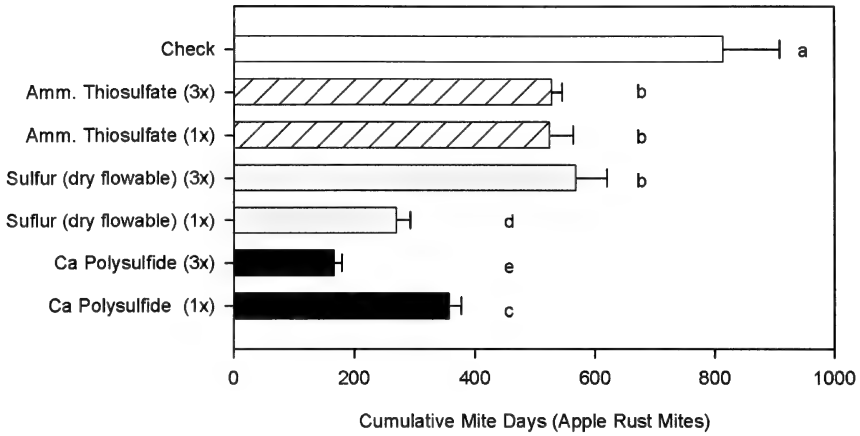

Figure 6. Effect of sulfur-containing products on apple rust mite. $F = 22.3$, $P = <0.0001$, df = 6, 27.

1991 (NASS 1992, 2006). These compounds, which are typically applied early in the season, may predispose the orchard to later disruption by codling moth insecticides.

The toxic effect of carbaryl and sulfur-containing products on mites is well known (McMurtry *et al.* 1970). In the case of carbaryl, moderate levels of resistance in *G. occidentalis* (Babcock and Tanigoshi 1988) may have mitigated the disruptive effect to some extent. Sulfur products have a long history of disruption of integrated mite control, and although resistance in *G. occidentalis* populations had been found in California vineyards, this had apparently not occurred in Washington orchards (Hoy and Standow 1982). Thus it is reasonable to expect that increased use of these materials could contribute to mite outbreaks.

The organophosphate–based programs of the past few decades have provided one of the most stable periods in integrated mite control in Washington orchards. The insecticides that replaced the organophosphates were initially thought to be more selective, but a number have shown nontarget effects on beneficial arthropods. Because of this destabilization, acaricide use has increased in recent years (NASS 1992, 2006), leading to increased production costs and increasing the probability for resistance development in pest mite species. It could be argued that because of widespread resistance to organophosphates in populations of both pests and natural enemies, that many of the organophosphates are now fairly selective from a pest management perspective. While human and environmental health concerns outweigh pest management issues, it will require further study and manipulation to reestablish the highly successful integrated mite control program as the primary means of mite control in Washington apple orchards.

ACKNOWLEDGEMENTS

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SCIENTIFIC NOTE

Additional records for semiaquatic Hemiptera in southwestern British Columbia

R.D. KENNER[1] and K.M. NEEDHAM[2]

Scudder (1977) published an annotated checklist of the aquatic and semiaquatic Hemiptera of British Columbia (BC); subsequently, several more species were added (Kenner and Needham 2004, Maw *et al.* 2000, Scudder 1986). Scudder (1977) lists only a single record for *Hydrometra martini* Kirkaldy: Lytton, 26.vii.1931, W. Downes. For *Mesovelia mulsanti* White, three records are given for southwestern BC, all from Vancouver Island: Elk Lake (Saanich district), ix.1924, W. Downes; Duncan, 4.ix.1926, W. Downes; Malahat, 4.ix.1929, W. Downes; as well as several records for the interior of BC. We report additional records for these two species from southwestern BC. Voucher specimens for all new records were deposited in the Spencer Entomological Museum (SEM) at the University of British Columbia.

Hydrometridae, water measurers or marsh treaders, are small bugs resembling tiny walkingstick insects (Phasmida) and have long, slender heads and bulging eyes at about mid-length on the head. They are often found walking slowly over the water surface searching for prey. There are nine species in North America, all in the genus *Hydrometra* (Polhemus 2008); only one of these, *H. martini*, is known from Canada (and BC) (Maw *et al.* 2000). We collected a single specimen of *H. martini* from a 10 m wide roadside ditch in Pitt Meadows (ditch between Neave Road and the Pitt River dike, 49°20'N 122°38'W, 12.iv.2008, R.D. Kenner and K.M. Needham). The specimen is apterous, strongly suggesting breeding at this site. This appears to be only the second time this species has been collected in this province. The 77 year gap between records, even though knowledgeable collectors have been looking for this species (Scudder pers. comm.), suggests that *H. martini* is truly rare in BC.

Mesoveliidae, water treaders, are small predatory bugs usually found crawling or running over the water surface. There are three species in North America, all in the genus *Mesovelia* (Polhemus 2008), two of which are known from Canada, with only one species, *M. mulsanti* known from BC (Maw *et al.* 2000). We report the first records for *M. mulsanti* from the Lower Fraser Valley: Bowen Island, Killarney Lake, 49°23'N 123°21'W, 20.viii.1998, R.D. Kenner, 1 specimen, apterous; Vancouver, Jericho Park, Main Pond, west end, 49° 16'N 123°11'W, 08.viii.2008, 8 specimens, 1 macropterous, 7 apterous; 22.ix.2008, 4 specimens, all apterous, R.D. Kenner. In addition, we collected this species on Vancouver Island: Hamilton Marsh, 49°24'N 124°38'W, 11.viii.2004, R.D. Kenner and K.M. Needham, 1 specimen, apterous. Searches through the collections of the SEM and Royal British Columbia Museum (Victoria) add the following unpublished records: Saanich, 48°33'N 123°22'W, 15.ix.1924, 30.ix.1925, W. Downes; Galiano Island, north end, 49°0'N 123°34'W, 1 & 2.ix.1984, G.G.E. Scudder. Although the records for *M. mulsanti* from the interior of BC span the range from April to September, those from the coast are only from Au-

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gust and September. This species may not be as rare as these few records suggest since most specimens are apterous and may easily be dismissed as 'unidentifiable' immature bugs.

We thank G.G.E. Scudder for confirmation of identifications and useful discussions, and R.A. Cannings and C.R. Copley for checking the Royal BC Museum collection for additional records.

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SCIENTIFIC NOTE

Podosesia syringae (Lepidoptera: Sesiidae): a new clearwing moth record for British Columbia

VIRGILIU M. AURELIAN[1], MARIO LANTHIER[2] and GARY J.R. JUDD[3]

Podosesia syringae (Harris), commonly known as the ash borer, is a clearwing moth (Sesiidae) whose larvae are borers within the trunks of lilac, *Syringa vulgaris* L., and various ash species, *Fraxinus* spp. This native North American insect (Eichlin and Duckworth 1988) is considered a major pest of wild, cultivated and ornamental ash trees in eastern provinces and states (Appleby 1973, Solomon 1983). In their review of North American Sesiidae, Eichlin and Duckworth (1988) reported collections of *P. syringae* from Washington State and eastern Alberta, but British Columbia (BC) was excluded from their description of its western range. Adults of this species occur in two distinct colour morphs, a black morph that has a dark brown abdomen, and a yellow morph that has a light brown abdomen surrounded by yellow bands. The two morphs are geographically distinct, the yellow morph *fraxini* being restricted to Western North America and the nominate morph being restricted to Eastern North America (Eichlin and Duckworth 1988). A wide hybridization zone exists in the mid-West and the prairies (Eichlin and Duckworth 1988), suggesting the two extreme morphs may be two different subspecies. Future molecular research will be important in elucidating whether a subspecies level is warranted for the ash borer.

In winter of 2006 one of us (ML) discovered many unknown lepidopteran larvae infesting various three-year-old *Fraxinus* spp. nursery stock collected from commercial tree nurseries located in Westbank and Armstrong, BC. In early 2007 cut sections of these infested ashes were brought into the laboratory and the emerging adults were identified by one of us (VMA) as the ash borer, *Podosesia syringae* (Harris). Voucher specimens from BC (CNCLEP00041170 & CNCLEP0041171) were deposited in the Canadian National Collection of Insects, Arachnids and Nematodes, Ottawa, ON and confirmed as the ash borer by J-F. Landry (personal communication).

In spring 2007 we surveyed wild and ornamental lilacs surrounding nursery plantings in Westbank, and found evidence of larval feeding and pupal exuviae characteristic of the ash borer. In June 2007, Pherocon 1-CP style wing traps (PheroTech Int. Inc., Delta, BC) baited with Clearwing Borer lures SC L103 (Scentry Biologicals, Billings, Montana,USA) containing (*Z,Z*)-3, 13 octadecadienyl acetate, a known clearwing male sex attractant and possibly the major component of the female pheromone for this species (Nielsen and Purrington 1978), were deployed in and around six ash nursery plantings in Westbank and Armstrong, BC. In a total of 102 traps deployed in both regions we captured 325 male *P. syringae* adults. No black morphs were captured in BC. Superficially, the yellow morph resembles a paper wasp (*Polistes* spp.) but on a sticky trap it could also be confused with the Western poplar clearwing, *Paranthrene robinae* Hy. Edwards; which is native to BC. However, on closer examination *P. syringae* can be distin-

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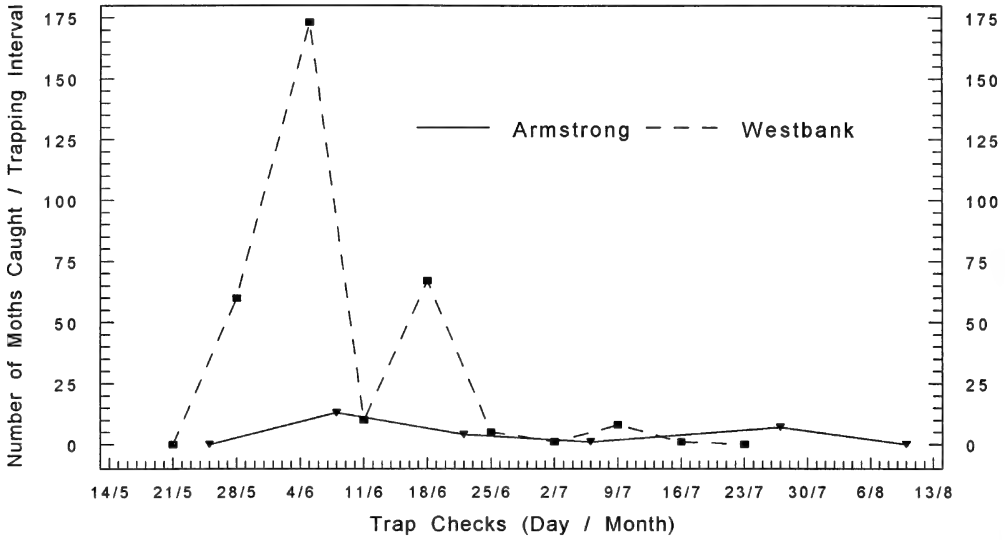

Figure 1. Temporal pattern of seasonal catches of male *P. syringae* adults in sex-attractant baited sticky traps at two locations within the Okanagan Valley, BC, in 2007.

guished from *P. robinae* by the presence of a very long first metatarsal segment and distinctive forewing venation.

Weekly trap checks revealed a male flight period that lasted six weeks in Westbank with peak flight occurring in the first half of June (Fig. 1). Further north in Armstrong, the flight period was more extended, ending in the first half of August, but the smaller peak flight occurred about the same time as it did in Westbank. In eastern North America adult flight begins in April and ends in July (Neal and Eichlin 1983). In summer 2008 we redeployed sex-attractant traps around a fallow nursery field from which all ash trees had been harvested in the fall of 2007. Our catches of *P. syringae* confirmed the presence of this species in the absence of ash, supporting the view that local plantings of lilac on residential properties outside the nursery of initial discovery are now a potential source of this insect. From these results we conclude that *P. syringae* is established within parts of the Okanagan Valley, BC. However, the original source of infestation remains unknown.

We thank Larisa Aurelian for help collecting biological specimens. This research was partially supported with funds from a BC Ministry of Agriculture and Lands Plant Health Grant held by GJ.

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SCIENTIFIC NOTE

Survival of male click beetles, *Agriotes obscurus* L., (Coleoptera: Elateridae) during and after storage at different temperatures

WILLEM G. VAN HERK[1], J. TODD KABALUK[1], VIOLA W.M. LAM[1] and ROBERT S. VERNON[1]

The dusky wireworm, *Agriotes obscurus* L. (Coleoptera: Elateridae) is a significant pest of vegetable and field crops in the Fraser Valley of BC (Vernon *et al.* 2001). Adults emerge from the soil from late March through May and die soon after mating (males) or egg laying (Brian 1947), restricting their availability for research and necessitating storage methods that prolong survival. Here we compare beetle survival during and after storage at various temperatures for various durations.

Male *A. obscurus* beetles were collected at the Pacific Agri-Food Research Centre (Agassiz, BC) during their peak emergence (1st wk of May, 2005) and placed in groups of 10 beetles into 150 ml plastic containers with a freshly-cut apple piece (approx. 2 x 2 x 0.5cm) placed on 70 g of moist sandy clay-loam soil. Containers were put in growth chambers (15 per chamber) (Controlled Environments Ltd., Winnipeg, MB) set at 5, 8.5, 12, and 20 °C (± 0.1 °C), and dead beetles removed, and apple pieces replaced, biweekly. Three containers were removed from each chamber after 2, 4, 6, 8, and 10 wks to determine subsequent survival at room temperature (RT) (21 ±1 ° C), and beetles transferred to 10cm Petri dishes (one per container) placed on 5cm high racks inside Styrofoam boxes. Each box (36 x 26 x 9cm deep) contained 2.5ml water to prevent desiccation; a 1cm gap between the box and its lid permitted air exchange. Beetle feeding and observation continued (as above) for up to 12 wks.

Beetle mortality during storage was highest at 20 °C, and similar at 5, 8.5, and 12° C for the first 6 wks but considerably lower at 8.5 °C thereafter. Beetle mortality was rapid within the first 2 wks of storage, and increased with duration for all temperatures except 8.5 °C (Table 1). Regression (stepwise, backward; Proc REG, SAS 9.1; SAS Institute 2002) of the proportion dead per container during storage (m) to temperature (t) and duration (d) yielded the following: $m = 0.003t^2 - 0.022t + 0.050d$ (SE slopes: 0.0005, 0.012, 0.009 respectively; $P < 0.0001$, 0.06, <0.0001 respectively, d.f. = 3,57, adj. $R^2 = 0.88$), indicating that survival increases as storage temperature decreases.

For post-storage survival analysis, each beetle was considered an experimental unit, and storage temperatures were compared with Kaplan-Meier survival analysis (Proc LIFETEST, SAS 9.1), with strata duration (Cox and Oakes 1984). The survival time of 50% of beetles (ST50) was subsequently estimated by modelling survivorship for each storage temperature-duration combination. Survivorship curves were compared with log-rank tests; ST50 values were compared using 95% confidence intervals.

Post-storage beetle mortality was rapid regardless of previous storage temperature. Beetles stored at 12 °C died more quickly at RT than those stored at 8.5 or 5 °C ($\chi^2 = 12.64$, $P = 0.0004$; $\chi^2 = 19.27$, $P < 0.0001$, respectively). Beetles stored at 5 °C survived longest, but not significantly longer than those stored at 8.5 °C ($\chi^2 = 2.91$, $P = 0.09$). Comparison of ST50 values and survival curves indicated that beetles survived longer if stored at 5 °C than at 8.5 °C, if

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Table 1.

Mortality of *Agriotes obscurus* beetles after storage at various temperature for different durations, and subsequent survivorship at room temperature (RT). N = 30 per storage duration-temperature combination. Post-storage survivorship at RT modelled with Kaplan-Meier survival curves; ST50 = time required for 50% survival.

Storage temperature (°C)	Storage duration (wk)	Mean (SE) number of beetles dead (out of 10) at the end of the storage period	Survival of beetles at RT after storage (d)		
			ST50	Upper 95% CI	Lower 95% CI
5	2	2.33 (0.67)	68.0	54.0	89.0
5	4	2.0 (0.58)	20.0	12.0	33.0
5	6	3.0 (1.53)	19.0	5.0	33.0
5	8	4.0 (2.08)	30.5	19.0	47.0
5	10	5.67 (0.33)	12.0	5.0	26.0
8.5	2	2.0 (0.58)	27.0	26.0	47.0
8.5	4	2.0 (1.0)	26.0	14.0	47.0
8.5	6	3.67 (0.33)	19.0	14.0	33.0
8.5	8	1.33 (0.67)	19.0	12.0	21.0
8.5	10	2.67 (1.67)	12.0	7.0	33.0
12	2	1.33 (0.88)	33.0	12.0	47.0
12	4	2.67 (0.33)	12.0	12.0	14.0
12	6	3.00 (0.58)	12.0	5.0	21.0
12	8	4.67 (2.19)	9.5	5.0	19.0
12	10	8.67 (0.88)	n/a[1]	n/a[1]	n/a[1]
20	2	5.0 (1.15)	5.0	5.0	7.0
20	4	9.67 (0.33)	n/a[1]	n/a[1]	n/a[1]
20	6, 8, 10	10.0 (0)	n/a[1]	n/a[1]	n/a[1]

[1] Not enough beetles survived storage to permit analysis.

stored for 2 wks ($\chi^2 = 15.48$, $P<0.0001$) or 8 wks ($\chi^2 = 6.15$, $P=0.013$; Table 1), but not when stored for 4 or 6 wks ($P>0.05$, Table 1). Surprisingly, beetles stored for 2 wks at 5, 8.5, or 12 °C survived longer at RT than those stored at 20 °C (Table 1).

These results indicate that storage at lower temperatures prolongs male click beetle survival, and that storage at 8.5 °C caused highest overall survival. Future research should investigate how cold storage conditions affects beetle physiology.

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SCIENTIFIC NOTE

Effect of handling and morbidity induction on weight, recovery, and survival of the Pacific Coast wireworm, *Limonius canus* (Coleoptera: Elateridae)

WILLEM G. VAN HERK[1,2] and ROBERT S. VERNON[1]

Wireworms can recover from prolonged morbidity induced by exposure to insecticides (Vernon *et al.* 2008), but it is unclear to what extent wireworm physiology and survival are affected by repeated handling or morbidity inductions, which reduces larval weight and retards development (Nicolas and Sillans 1989) in some insects. It may be that differences in weights or survival in wireworms repeatedly made moribund, similarly handled but not made moribund, or not subjected to handling or morbidity might occur.

Larvae of the Pacific Coast wireworm, *Limonius canus* LeConte, become temporarily moribund after contact with tefluthrin-treated wheat seeds, and recover more quickly when re-exposed (van Herk and Vernon 2007), but it is not known if continued re-exposure further decreases recovery time. Wireworms repeatedly contact insecticide-treated seeds in the soil, thus a continued decrease in the morbidity duration may affect the insecticide's efficacy in the field. Here we discuss if wireworms continue to recover more quickly from tefluthrin-induced morbidity, and if this or handling affects their weight and the time to complete the larval instar.

Wireworms were collected from an organic farm in Kelowna, BC, in June 2007 and stored in soil at 15 °C. Late-instar, feeding wireworms were randomly allocated to one of three treatments (24–50 per treatment), the 'morbidity', 'handled', and 'control' treatments. All observations were made at 21±1 °C. In the 'morbidity' treatment, morbidity was induced, and wireworm health assessed following van Herk and Vernon (2007). Individual wireworms were placed for 2 min in 1.5-ml Eppendorf microcentrifuge tubes (Fisher Scientific) with a single, ungerminated wheat seed (cv. Superb) treated with Tefluthrin 20CS (20% tefluthrin w/v) at 10 g AI/100 kg seed and the fungicide Dividend XLRTA (3.21% difenoconazole, 0.27% mefenoxam) at 13 g AI/100 kg seed. Seeds were treated in 2007 by Syngenta Crop Protection Canada Inc. (Portage la Prairie, MB). After exposure, larvae were placed in individual Petri dishes lined with moistened filter paper to observe the onset of morbidity, and subsequently placed in separate, identical film canisters filled with finely screened soil with 20% moisture by weight. Wireworm 'health' was assessed every 10 min until 30 min after no further symptoms of morbidity were observed (i.e. 7-15 times). Morbidity was induced using this method at 24-h intervals for 4 consecutive days.

Wireworms in the 'handled' treatment were placed for 2 min in Eppendorf tubes with a single untreated wheat seed and, as above, observed for 20 min in a Petri dish, and then transferred to film canisters filled with soil. To expose these wireworms to handling comparable to those in the 'morbidity' treatment, the health of 'handled' wireworms was assessed at 10-min intervals 12 times on the first day and 10 times on the subsequent 3 d. Wireworms in the 'control' treatment were placed in film canisters and weighed, but were not placed in Eppendorf tubes or Petri dishes. Larvae were weighed individually for 5 consecutive d, approx. 6 h before wireworms in the 'handled' or 'moribund' treat-

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ments were placed in Eppendorf tubes the first 4 d, and at the same time of day on day five. Larvae were weighed for final time approx. 2 mo later, at which time the proportion moulted was recorded.

Wireworm recovery durations were compared among treatments with ANOVA, and the % decrease in recovery time between day 1 and days 2, 3 and 4 was calculated. Weight loss over time was compared among treatments with repeated measures ANOVA; orthogonal contrasts were used to compare wireworm weight loss between 'control' and 'handled', and between 'handled' and 'moribund' treatments. Proportions of larvae that moulted were compared with Chi-square analysis.

In the morbidity treatment, the time to recovery on day 1 was 88.8 ± 2.3 (SEM) min, similar to the 86.0 to 87.8 min reported by van Herk and Vernon (2007). Time to recovery on day 1 was significantly longer than on days 2, 3 and 4 (72.4 ± 1.8, 69.4 ± 1.8, 71.8 ± 1.8 min, respectively; $F=20.70$, df=3,196, $P<0.0001$), but recovery durations on the latter 3 d were not different ($P>0.05$). Recoveries on days 2, 3 and 4 were 18.5 – 21.8% more rapid than the initial recovery time. These data suggest that repeated contact with tefluthrin-treated seeds in the field will not likely make wireworms insensitive to the insecticide.

Analysis of individual wireworm weights over time using the first five measurements indicated a weight loss over time ($F=8.35$, df=4,384, $P<0.0001$, Table 1) and an interaction between weight loss and treatment ($F=2.25$, df=8, 384, $P=0.02$). These remained significant ($P<0.05$) when all six weighing days were included in analyses; orthogonal contrasts indicated greater weight loss in 'handled' than in 'control' treatments ($F=3.04$, df=5,480, $P=0.01$), but not between 'moribund' and 'handled' treatments ($F=0.80$, df=5,480, $P=0.55$). This suggests that weight loss in the 'moribund' treatment was due to handling alone, and that extensive handling of wireworms may cause stress or damage, perhaps including elevation of hemolymph sugar or lipids (Woodring *et al.* 1989). It is unlikely the observed weight loss was from desiccation as it continued from days 5 to 68 when wireworms were continuously in moist soil. Thus care must be taken to minimize handling events and/or trauma.

All wireworms were alive on day 68 with no difference in moulting in the treatments by day 68 (0.62, 0.52, 0.42, respectively; $\chi^2 = 2.79$, df=2, $P=0.25$), so that repeated morbidity induction and handling did not affect survival or development.

We thank M. Clodius and C. Harding for technical assistance, S. Reid for permission to collect wireworms, and L. Letkeman for treating wheat seeds.

Table 1.

Mean (SEM) weight (mg) of *Limonius canus* larvae subjected to one of three treatments: 'Moribund': repeated handling plus morbidity induced by insecticide exposure; 'Handled': repeated handling only; 'Control': no handling or morbidity induction.

Treatment	N	day 1	day 2	day 3	day 4	day 5	day 68
Control	24	25.5 (1.2)	25.6 (1.2)	25.5 (1.2)	25.5 (1.2)	25.5 (1.2)	25.7 (1.2)
Handled	25	26.0 (1.1)	25.8 (1.1)	25.8 (1.1)	25.7 (1.1)	25.6 (1.1)	24.9 (1.2)
Moribund	50	25.4 (0.8)	25.5 (0.8)	25.1 (0.8)	25.1 (0.8)	25.1 (0.8)	24.7 (0.8)

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Symposium Abstracts: Biodiversity in Stanley Park

Entomological Society of British Columbia Annual General Meeting, West End Community Centre Auditorium, Vancouver, BC, October 4th, 2008

Parks, Protected Areas and Biodiversity Conservation in B.C.

Geoffrey G.E. Scudder. *Zoology Department, University of British Columbia, Vancouver, B.C.*

Currently over 12% of the land area of British Columbia is categorized as Protected Area. This includes national and provincial parks, ecological reserves, and a few other areas. However, it does not include regional and municipal parks and lands owned by various conservation organizations, although these have definite conservation value. An assessment of the Protected Areas network shows that this not only does not serve as an efficient ecological network, but it has unequal ecosystem representation, and evidently does not adequately protect the biodiversity richness and rarity hotspots in the province. There are few buffer areas and most parks and protected areas have not been adequately assessed for biodiversity. Impending climate change will undoubtedly have a dramatic impact on the species and ecosystems in British Columbia. There is an urgent need to assess current elements of biodiversity in all the parks and protected areas, monitor these, and develop a landscape framework with appropriate connectivity and integrity, that will facilitate the movement of species in the future.

Some first systematic surveys of the insects of Stanley Park in 2007 and 2008

John A. McLean. *Department of Forest Sciences, University of B. C., Vancouver, B. C.*

The winter storms of 2006/07 felled a large volume of large trees throughout Stanley Park. Freshly fallen trees are quickly attacked by ambrosia beetles and bark beetles as part of nature's first phase of recycling the dying tissues. The records of some past outbreaks of insects in Stanley Park will be reviewed. In 2007, baseline surveys were made of moths using light traps in undamaged forest stands near the Aquarium and the Hollow Tree. Semiochemical-baited multiple funnel traps were also set out to evaluate bark beetle and ambrosia beetle populations along with pitfall traps to evaluate ground beetle populations. In 2008, the multiple funnel traps and pitfall traps were set out in areas that had been severely affected by the 2006/07 winter storms after a large portion of the felled trees had been removed and new seedlings planted. A brief overview of some of our results will be given.

A Fascination with Nature or "How big can small get?"

Peter Woods. *Naturalist-photographer, Vancouver, B. C.*

Imagine yourself let loose; able to explore any and every facet of the natural history of a favourite place. Give yourself ten years or so. Where would this journey take you? I have enjoyed just such an opportunity and the rare privilege to observe, listen, touch, imagine, and photograph nature in Stanley Park. This presentation is a sampling of what a naturalist does. It is a blending of curiosity, wonder, fascination, observation, fact, understanding, and shared discovery. It is also about 'small things' and a world of parallel universes. It is about what can happen only when you stop and stand still, in one, magical place. It is about biodiversity with inspiration drawn in equal measure from 'Winnie the Pooh's hundred acre wood', and 'Alice's Adventures in Wonderland'. Here are creatures that have taken me on special journeys captured by a powerful new tool, the 'eye' that sees through the lens of a digital camera.

Challenges of Small Vertebrate Management in Stanley Park

Robyn Worcester. *Conservation Officer, Stanley Park Ecology Society, Vancouver, B. C.*

Although Stanley Park is a fragmented natural area in the heart of a large urban setting, it is home to a diverse array of resident and migratory wildlife and presents unique challenges as one of the oldest and largest urban parks in North America. Following the massive windstorms in the Park in December 2006, the small vertebrates of Stanley Park became recognized as a component of ecologically based forest management and a renewed interest in their protection enabled formal research and inventories to be undertaken, in many cases for the first time. I will discuss the results of the inventories that took place over the last two years, the challenges that were faced throughout the restoration process, and the ongoing efforts of the Stanley Park Ecology Society to maintain and enhance the diversity of the small vertebrates in the Park though data collection and public education.

Stanley Park Restoration Project - Respond/Plan/Restore

Jim Lowden. *Director of Special Projects – Vancouver Board of Parks and Recreation, Vancouver, BC.*

A look at managing a multiple objective program to respond to a natural disaster; balancing competing agendas while attempting to do good. What we learnt from the last 22 months in the trenches.

Hurricanes, invasive beetles, and urban forests: lessons from Point Pleasant Park.

Jon Sweeney. *Natural Resources Canada, Canadian Forest Service, Fredericton, NB.*

Point Pleasant Park, much cherished and heavily used by people (and their dogs) in Halifax, Nova Scotia, has been hit by many disturbances since its inception in 1866, not least of which were the relatively recent arrivals of the brown spruce longhorn beetle and Hurricane Juan. Loss of much of the mature red spruce to wind-throw and the beetle has obviously altered forest age class and stand structure in many areas of the park; arthropod species composition and diversity has likely also changed as a result, but unfortunately our baseline knowledge is limited. I will give a brief history of Point Pleasant Park, focusing on changes in the last 2-3 decades, highlight some of our research on the biology and management of the brown spruce longhorn beetle, and describe plans for park restoration in the aftermath of the Hurricane.

History of Survey and Control Activities for Forest Pests in Stanley Park and Adjacent Forest Environs

Leland Humble. *Natural Resources Canada, Canadian Forest Service, Victoria, B. C.*

Since its opening in 1887, the 400 ha of forest lands of Stanley Park have been subjected to multiple abiotic and biotic disturbances that have impacted the health of its forest stands. The extensive blowdown caused by the 2006/07 winter storms is but the most recent example of an abiotic disturbance. Ninety-nine species of beetles, 122 species of moths and 11 species of sawflies have been recorded from Stanley Park and adjacent forest habitats on the north shore by the Forest Insect and Disease Survey during survey activities between 1949 and 1995. Only a few of these species are damaging. Outbreaks of defoliators such as the western hemlock looper, the green-striped forest looper, and the western blackheaded budworm have caused extensive defoliation or mortality within areas of the park. Aerial control operations to protect the forest resources of the park have been undertaken three times against native defoliators and once for an introduced defoliator. The biology and damage caused by the major forest pests present in Stanley Park and the history of the control operations undertaken in the park are reviewed.

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