

Control of aphid vectored non-persistent viruses for organic seed potato production:
mineral oils

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Project Report To:

Lower Mainland Horticultural Improvement Association
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Fraserland Organics

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EXECUTIVE SUMMARY

Potato virus Y (PVY) is one of the most important viruses affecting potato production. It is also one of the main factors limiting production of organic seed potatoes because of the lack of organic management tools for viruses and their aphid vectors. Soap is the only aphicide currently registered for organic production and it provides insufficient control of aphids to meet seed potato standards (very low tolerance for viruses). Researchers have found ways to prevent the transmission of non-persistent viruses like PVY through the use of mineral oils as foliar sprays. Mineral oils appear to have both insecticidal activity and the potential to significantly reduce the acquisition and transmission of PVY by aphids. This trial looked at two products 13 E and JMS Stylet Oil compared to a water only Control sprayed weekly on potato plants from full emergence until harvest (5-6 weeks). Assessments for this trial consisted of weekly aphid counts and two leaf tests for PVY. There was no significant effect of either product (13E or JMS Stylet Oil) on aphid counts/plot compared to the Control. Also neither of the mineral oil treatments reduced the number of PVY infected plants compared to the Control. Summer 2011 was characterized by cool weather and consequently aphid populations were abnormally low. Therefore, recommendations for future studies are to conduct the trial with larger plots and longer maturing varieties to increase the chances for aphid populations to build up within plots and surrounding fields.

INTRODUCTION

Potatoes are an important vegetable crop for many organic and conventional farms in British Columbia. However, potatoes are susceptible to a wide range of diseases which makes organic production challenging. Potato virus Y (PVY) is one of the most important viruses affecting potato production. For each 1% increase in PVY incidence yield can be reduced by 0.1805 t/ha in commonly grown varieties such as Russet Norkotah, Russet Burbank and Shepody (Nolte *et al.* 2006). PVY, like most potato viruses, is vectored by aphids. Virus diseases can spread rapidly to neighbouring potato fields if aphids are not controlled. In addition to aphid management, the use of certified seed potatoes is an important component for management of virus diseases. A certification program is in place in Canada and other countries to ensure low levels of virus in certified seed potatoes.

While the production of organic certified seed potatoes is a growing industry in BC (Anonymous, 2003), current levels of production cannot meet the demand. Because there is an insufficient supply of organic seed potatoes, organic growers are allowed to use conventionally-produced seed tubers. However, certification bodies are increasingly demanding the use of organic propagation material, and seed potato growers would like to supply this market. One of the main limitations for the production of organic seed potatoes is the lack of organic management tools for viruses and their aphid vectors.

Soap is the only aphicide currently registered for organic production and it provides insufficient control of aphids to meet seed potato standards. In conventional seed potato production, the aphid threshold (for an insecticide application) is 3-5% infested triplets. While insecticides effectively control persistent virus such as potato leaf roll virus, insecticides are not an effective way of controlling non-persistent virus such as PVY (Perring *et al.* 1999). With persistent viruses, there is a longer period of time between the aphid acquiring the virus from one plant and being able to transmit the virus to another plant. So insecticides can kill the aphid prior to a persistent virus being transmitted. In contrast, with non-persistent viruses there is no delay between an aphid acquiring a virus from an infected plant and transmitting the virus to the next plant that is fed upon. Thus for non-persistent viruses, most insecticides do not act fast enough to prevent aphids from transmitting virus (Loebenstein and Raccach 1980). Therefore, an effective tool for organic seed potato production must be able to 1) prevent acquisition or transmission of non-persistent viruses like PVY and 2) kill aphids.

Several studies have shown that mineral oils interfere with virus retention in the aphid mouthparts (stylet) (Powell 1992). A significant reduction in the transmission of etch potyvirus (a non-persistent virus) was obtained by spraying mineral oil on tobacco plants (Wang and Pirone 1996). Similarly, foliar application of mineral oil reduced the transmission of cucumber mosaic virus to pepper plants (Martin *et al.* 2004), PVY to potatoes (Martin *et al.* 2006; Boiteau *et al.* 2008; Wrobel 2009), and PVY to tobacco (Powell, 1992; Powell *et al.* 1998). Together these studies demonstrate that there is a consistent effect of mineral oils on virus transmission which makes mineral oils one of the most effective tools to control non-persistent virus spread (Loebenstein and Raccach

1980). Furthermore, direct insecticidal effect of mineral oils on aphids have been shown in several studies (Martin *et al.* 2004; Martin *et al.* 2006; Iversen and Harding 2007; Kraiss and Cullen 2008; Najar-Rodriguez *et al.* 2008). In order to be effective, a 1% solution of paraffinic mineral oil should be applied weekly from early emergence until harvest (Simons and Zitter 1980; Loebenstein and Racciah 1980; Radcliffe and Ragsdale 2002). At this concentration the risk of phytotoxicity from mineral oil is low (Simons and Zitter 1980). Mineral oils are widely used in Europe and are increasingly used in the United States and Eastern Canada for seed potato production.

In the summer of 2011, Superior 70 Oil (Mineral oil 99%; United Agri Product Canada Inc.) was registered for PVY control on potatoes. However, there is currently no oil registered for organic production of potatoes in Canada. Crop oil 13 E (mineral oil 99%; Petro Canada), is OMRI approved in the US under the trade name Purespray Green. JMS Stylet Oil (paraffinic oil 97.1%; JMS FlowerFarms Inc.) is approved for the control of PVY on organic potatoes in the US. These products have potential both to control aphids and more importantly curtail the spread of PVY for organic seed potato production. The objective of this study is to test the efficacy of 13E and JMS Stylet Oil at controlling aphids and PVY transmission in organic seed potato production.

MATERIALS AND METHODS

Study Site: The trial was conducted in three organic potato (var. Redsen) fields in Delta, BC. Planting and weed control were done by the grower. There were no insect control activities in any of the fields. Each field was sprayed with Parasol (copper hydroxide) weekly for late blight control. There was no artificial irrigation was used in any of the fields. Field 1 was planted on May 22, Field 2 on May 29 and Field 3 on June 09. Fields started to emerge about 3 weeks after planting and took about 2 weeks to fully emerge. The trial was conducted in commercial potato fields, instead of seed potato fields because it allowed for the option to introduce both PVY and aphids to the study plots; inoculating a seed potato field would not have been possible without jeopardizing the certification status of the entire field (i.e. seed potato fields have very low tolerance for PVY).

Treatment Description and Plot Layout: In order to evaluate the efficacy of mineral oil for aphid and PVY control for organic seed potato production, the trial examined two possible solutions 1) 13 E, 2) JMS stylet oil along with a water Control (Table 1). Each of the three treatments was replicated six times, for a total of 18 plots/field. Treatments were randomly assigned to plots resulting in a completely randomized designed. Each plot consisted of one row containing 10 plants (plots ranged in size from 3 to 6 meters long depending on plant emergence). Plots were separated by 1 m buffer. Plots were laid out in a 3 X 6 grid in each field (Appendix I).

Plots were sprayed weekly, starting at full emergence until harvest for a total of 6 sprays in Field 1 and Field 2 and 5 sprays in Field 3 (Table 2). Treatments were applied with a backpack sprayer hand-pumped to maintain full pressure. Treatments were applied either in the early morning or late afternoon. As per the product labels, plants were sprayed to

allow good coverage but to avoid run-off (0.25 L (first two sprays) and 0.42 L (following sprays) of spray solution/plot).

Table 1. Description of the mineral oil treatments evaluated against the water Control for aphid and PVY control in organic potatoes.

Trade Name and Manufacturer	Active Ingredient	Rate	Amount of product/plot First two sprays	Amount of product/plot remaining sprays
13E (Petro Canada)	Mineral oil 99%	1% solution	2.5 ml	4.2 ml
JMS Stylet Oil (JMS FlowerFarms Inc.)	Paraffinic oil 97.1%	1% solution	2.5 ml	4.2 ml
Water			0.25 L	0.42 L

Table 2. Mineral oil and water (for Control) spray schedule for Fields 1, 2 and 3

Field	Spray 1	Spray 2	Spray 3	Spray 4	Spray 5	Spray 6
1	July 1	July 8	July 15	July 22	July 29	Aug 5
2	July 8	July 15	July 22	July 29	Aug 5	Aug 12
3	July 15	July 22	July 29	Aug 5	Aug 12	

Inoculation of plots with PVY and aphids: PVY was present in the commercial potatoes growing in all three fields (about 3% of the plants had PVY). To further insure PVY pressure in our trial plots, one PVY infected plant (collected from the field) was planted in the middle of each plot. This was done on July 08 (Field 1), July 14 (Field 2) and July 21 (Field 3). Furthermore, one winged and 10 apteran (wingless) green peach aphids (collected from the field) were placed on the PVY infected plant in each plot. This was done on July 21 (Field 1), July 22 (Field 2) and August 05 (Field 3).

Assessment and Analysis: Assessments for this trial consisted of weekly aphid counts and two leaf tests for PVY. One pre-treatment aphid count and five post treatment aphid counts were conducted (Table 3). Post treatment counts were done seven days after each set of sprays. In other words data were collected from plots and then plots were sprayed on the same day each week (see Table 2 and 3). A count consisted of one potato triplet taken from the lower portion of the plant (Fig. 1a) from four different plants/plot for a total of four triplets/plot/count. The total number of aphids on all four triplets were recorded for each plot each week. This protocol was based on the E.S. Cropconsult Ltd. seed potato monitoring protocol. Leaf samples, for PVY testing were taken prior to the first spray and at the end of the trial. Leaf sampling consisted of taking one leaf from the mid canopy from each plant the in the plot (10 leaves/plot) (Fig. 1b). Each leaf was placed in a Ziploc bag and refrigerated prior to shipment (in Styrofoam container with icepacks). Leaves were tested for PVY (via ELISA test) by Phyto Diagnostics Lab (North Saanich, BC). PVY testing was done on leaves rather than tubers because leaves will test positive for virus much sooner than tubers (Dr. Peter Ellis, Phyto Diagnostics Lab,

personal communication). The effect of mineral oil treatment on the weekly number of aphids (winged+apteran), winged aphids and aphid infested triplets was analyzed using two-way (treatment X field) repeated measures MANOVA. The effect of mineral oil treatment on the number of PVY positive plants/plot was analyzed using one-way ANOVA. All data were analyzed using JMP-In (Version 5.1).

Table 3. Assessments schedule for Fields 1, 2 and 3 - aphid counts (pre-spray and post weekly spray counts) and leaf collection schedule for PVY testing

Field	Pre-spray aphid + 1st leaf collection	Post spray 1	Post spray 2	Post spray 3	Post spray 4	Post spray 5	<i>Final leaf collection</i>
1	July 1	July 8	July 15	July 22	July 29	Aug 5	<i>Aug 12</i>
2	July 8	July 15	July 22	July 29	Aug 5	Aug 12	<i>Aug 16</i>
3	<i>July 15</i>	<i>July 22</i>	<i>July 29</i>	<i>Aug 5</i>	<i>Aug 12</i>	<i>Aug 19</i>	<i>Aug 19</i>



Figure 1a-b. Triplet for aphid count (left) and leaf from mid-canopy for PVY testing (right).

RESULTS

Total Aphids: There was no significant effect of either product (13E or JMS Stylet Oil) on aphid counts/plot compared to the water only Control (Fig. 2). There was a significant field effect - with more aphids observed in Field 1 than in Fields 2 or 3. Over time aphid populations increased in all fields, although the rate of increase was slightly different among the three fields leading to significant Time and Field X Time interactions (Table 4). For the factor of interest - mineral oil Treatment effects - there were no significant main or interaction effects (Table 4).

Winged Aphids: Overall winged aphid counts were very low in plots over the course of the trial. There was a significant treatment effect - with more winged aphids in JMS Stylet Oil plots than in the Control or 13E plots (Fig. 3, Table 4). However even in JMS

Stylect Oil plots winged aphid counts were extremely low - a mean total over the 5 weeks of post spray counts of less than 1 winged aphid/plot (Fig. 3). While total aphid and infested triplet counts (see below) increased over time, winged aphid counts remained static with no significant effect of time (Table 4).

Infested triplets: As with total aphids, there was no significant effect of the mineral oils in reducing the number of infested triplets/plot; neither Treatment nor any of the interactions terms with Treatment were significant (Table 4). Over time the number of infested triplets increased in all plots and there were more infested triplets in Field 1 than in the other two fields - consistent with the aphid counts. This lead to significant Field, Time and Field X Time interaction effects (Table 4).

Table 4. Statistical results for repeated-measures analysis of mineral oil effects on aphids and infested triplets from potato plots

	Total aphids	Winged aphids	Infested triplets
Treatment	F (2, 45) = 0.67 P = 0.51	F (2, 45) = 3.60 P = 0.04	F (2, 45) = 0.80 P = 0.45
Field	F (2,45) = 15.18 P < 0.0001	F (2,45) = 1.27 P =0.29	F (2,45) = 14.95 P < 0.0001
Treatment X Field	F (4, 45) = 0.54 P = 0.71	F (4, 45) = 0.45 P = 0.77	F (4, 45) = 1.15 P = 0.35
Time	F (5,41) = 24.07 P < 0.0001	F (5,41) = 0.56 P =0.73	F (5,41) = 118.69 P < 0.0001
Time X Field	F (10,82) = 3.96 P = 0.0002	F (10,82) = 0.70 P = 0.72	F (10,82) = 5.26 P < 0.0001
Time X Treatment	F (10,82) = 0.53 P = 0.87	F (10,82) = 1.55 P = 0.14	F (10,82) = 0.35 P = 0.96
Time X Treatment X Field	F (20,137) = 0.80 P = 0.71	F (20,137) = 0.99 P = 0.48	F (20,137)= 1.11 P = 0.35

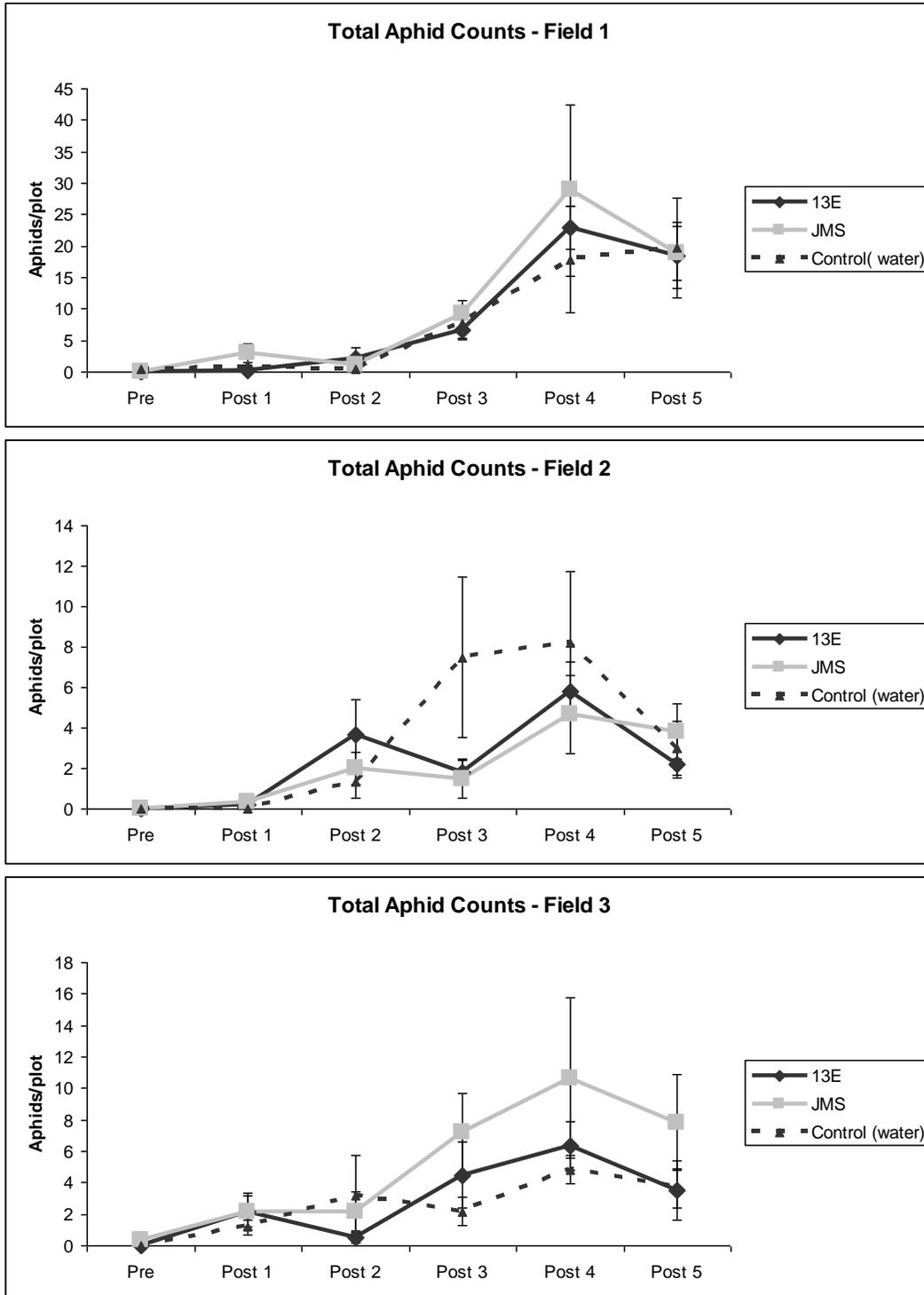


Figure 2. Effect of organic mineral oil formulations on the mean (\pm s.e.) aphid (winged and wingless) populations in plots prior to treatment and then weekly for five weeks following sprays applied seven days prior to each count. Six replicates for each treatment in each field, total N = 54.

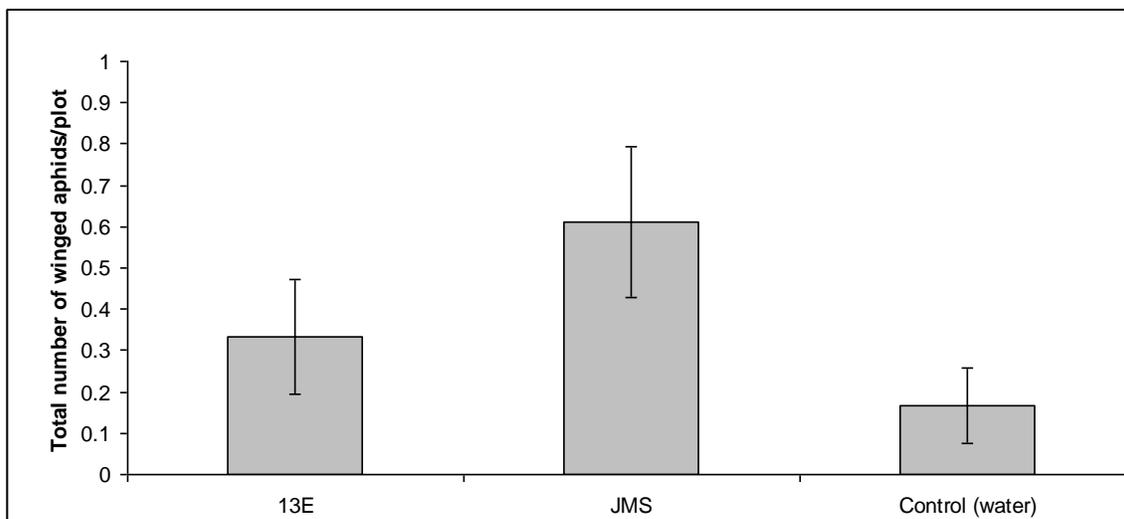


Figure 3. Effect of weekly mineral oil application on the total number of winged aphids in plots over the course of five weeks. Bars represent the mean \pm s.e. of 18 plots/treatment.

PVY infected plants: Neither of the mineral oil treatments reduced the number of PVY infected plants/plot compared to the water only Control (Fig. 4; Treatment: $F(2, 45) = 0.69$, $P = 0.51$; Field: $F(2, 45) = 0.91$, $P = 0.41$; Treatment X Field: $F(4, 45) = 1.46$, $P = 0.23$)

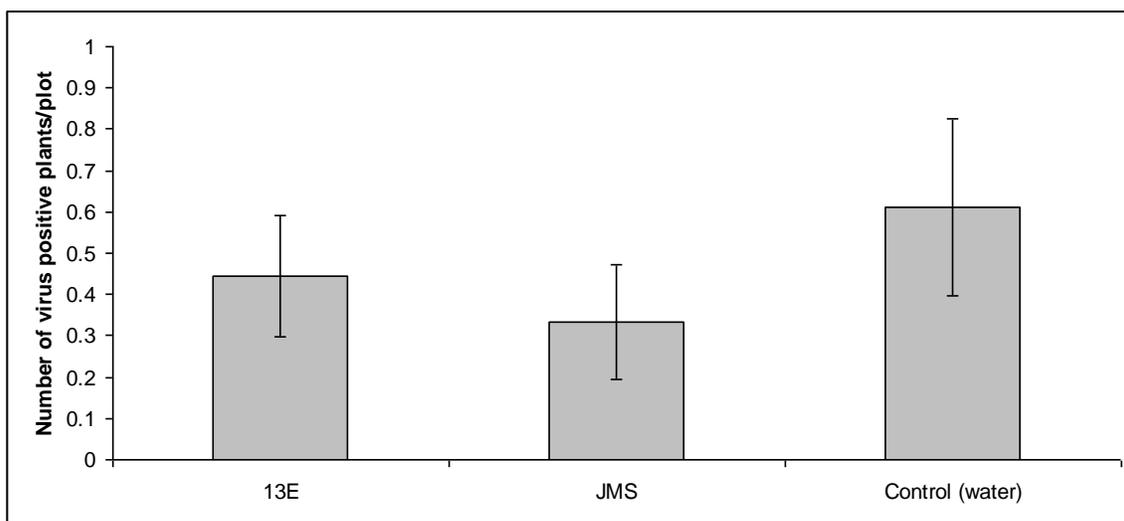


Figure 4. Effect of mineral oil treatments on the mean (\pm s.e.) number of PVY positive plants/plot. For each treatment $N = 18$.

DISCUSSION

The objective of this work was to demonstrate the efficacy of organic formulations of mineral oil for the control of PVY and aphids on potatoes. In an earlier study, Boiteau *et al.* (2008) obtained a 23 to 88 % reduction of PVY transmission on potato using mineral oil. In our study, neither formulation of mineral oil tested caused a reduction in aphids or PVY positive potatoes compared to the water Control. There are several factors that account for the difference between our findings and those of Boiteau *et al.* (2008).

First, there are several methodological differences between our study and that of Boiteau *et al.* (2008). The earlier studies were conducted at Research Station and thus plots were larger (10 m long X 10 rows wide) and a longer maturing potato variety was used so the trials ran for a longer period of time (7 - 15 weeks). In contrast, our study was conducted in a commercial potato field using products not currently registered for potatoes in Canada - so the potatoes produced in our trial area were subject to crop destruct. As a consequence our trial area was only 1 row wide X 3-6 m long, to minimize the number of tubers that had to be destroyed (i.e. minimize impact on grower's yield). Also we conducted our trial with a very fast maturing potato variety so our trial duration was short (5-6 weeks of sprays) compared to the earlier work of Boiteau *et al.* With the larger plot area Boiteau *et al.* (2008) would have increased the chances that the aphid populations in the middle of each plot were not impacted by the sprays in the surrounding plots. With the longer duration of the trial Boiteau *et al.* (2008) would have increased the chances that aphids would have had time to build up in plots, especially Control plots. Also the surrounding virus pressure from potatoes planted adjacent to Boiteau's trial ranged from 22% to 35%, depending on the year of the trial. In contrast, the surrounding virus pressure from the commercial potatoes growing adjacent to our plots was approximately 3% (E.S. Cropconsult Ltd. unpublished data). However, in our trial we also inoculated each plot with a PVY positive potato plant.

The second reason why the results of our study differ from those of Boiteau *et al.* (2008) and others who have demonstrated that mineral oils are effective at reducing PVY transmission (Powell 1992; Powell *et al.* 1998; Martin *et al.* 2006; Boiteau *et al.* 2008; Wrobel 2009) is that 2011 was not a "typical" year for aphids (Fig. 5). The unusually cool start to the field season (May to end of June) resulted in aphids colonizing potato fields later in the season. As a consequence aphid populations were much lower in the commercial potato fields where our trials were conducted. With low overall aphid populations in the fields the production of winged aphids, which migrate within and between fields, was also low. Although our plots were infested with the same number of aphids at the start of the trial, the additional migration of aphids into plots (winged aphids) from the surrounding field did not occur. This would then account for the low levels of PVY positive plants in our Control plots, which we expected would have been infested with PVY both from within the plot and from infected winged aphids migrating into the plots from the field.

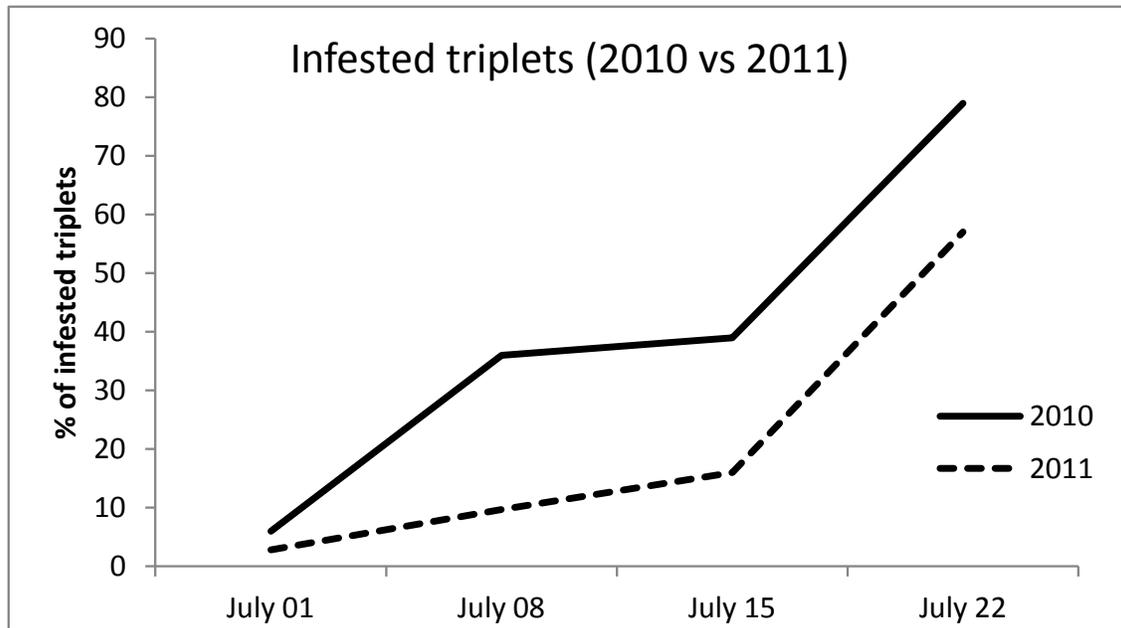


Figure 5. Percentage of triplets infested with aphids in Field 3 in July 2010 compared to 2011.

The final reason why our trial results did not demonstrate an effect of mineral oils on aphids or PVY transmission could be because our trials were conducted in organic potato fields and the aphids that were in our plots were being attacked by natural enemies. For example, on August 19, 25 % of the aphids present on triplets in Field 3 were either parasitized by wasps or entomopathogenic fungi. As a consequence aphid populations started to decline on their own in some of the fields (Fig. 2). Natural enemy populations typically build to high levels in organic potato fields during August and enemies were observed to be active across all plots in all fields of this trial. Enemies would also have further reduced the number of aphids available to migrate into plots from the surrounding field.

The challenges of an unseasonably cool field season (at the start) and of conducting trials in growers' fields most likely account for the lack of efficacy of mineral oils observed in this study. Recommendations for future studies are to conduct the trials with larger plots and longer maturing varieties to increase the chances for aphid populations to build up. Also conducting trials in conventional fields, or in research fields, may help to reduce some of the other factors that made evaluation difficult in this trial, e.g. natural enemies attacking aphids in trial plots. Mineral oils have been shown to be effective for PVY control in other studies and a conventional formulation of mineral oil (Superior 70 Oil) is now registered for PVY control in seed potatoes. Testing should continue to demonstrate efficacy of an organic mineral oil formulation for the same use. As the demand for organic potatoes increases so does the demand for organic seed potatoes; the challenge is to demonstrate that organic mineral oil formulations can be effective for production organic seed potatoes in BC.

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APPENDIX I.

Plot layout in each field

Field 1

13 E	Water	JMS
JMS	Water	13 E
Water	13 E	Water
13 E	JMS	13 E
JMS	JMS	Water
JMS	Water	13 E

→North

Field 2

JMS	JMS	13 E
13 E	13 E	Water
Water	13 E	Water
13 E	JMS	JMS
JMS	Water	13 E
Water	JMS	Water

↓ North

Field 3

JMS	13 E	Water
13 E	JMS	13 E
Water	JMS	Water
Water	13 E	13 E
JMS	Water	JMS
Water	JMS	13 E

←North