

A Literature Review of Insecticide and Mineral Oil Use in Preventing the Spread of Non-persistent Viruses in Potato Crops

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1. PREFACE

The following review forms part of a larger Potato Council-funded R&D project aimed at improving aphid control and minimising potyvirus spread in potato seed crops. This preface provides a brief introduction to the project and the outcomes of the review.

The R&D project was commissioned after a stakeholder consultation which identified knowledge gaps in relation to virus control. The three year project began in July 2009 and brings together expertise from SASA, SAC, SCRI, Scottish Agronomy and Fera. PCL funding totals £240K; however, with work-in-kind contributions from the consortium, the total value of the project is well over £600K.

One of the gaps identified during the consultation was the potential for deploying mineral oils as part of the armoury against aphid-borne virus. This report summarises the available literature on experimental work with oils and also reviews the current range of aphicides which form the major part of current aphid control strategies. Despite the differences in experimental methodology the authors have drawn the conclusion that oils can significantly contribute to the reduction of virus spread.

So, if mineral oils are effective and in commercial use overseas why not in Great Britain? While mineral oils are not strictly speaking prohibited for use in GB seed crops they have not been recommended. There are reports of phytotoxic effects in some crops which can reduce yield and may either mask or mimic virus symptoms in the canopy.

Significant phytotoxic effects from mineral oils, or for that matter any other chemical or combination of chemicals (*eg.* mis-timed herbicide application), can prevent effective in-crop inspections. The certifying authorities can then refuse to inspect the crop and the seed will not be classified and cannot be marketed.

So what happens overseas? In countries such as France and the Netherlands, less intensive in-crop inspections are backed up by routine post-harvest testing. This expensive option on a limited sample of the crop does not give the same level of confidence with regard to the probable level of virus. This is of particular concern in GB, where tolerance levels for virus infection are stricter than our major competitors. These strict GB tolerances are made possible by our system of visual inspections of the growing crop. The GB Certifying Authorities are therefore strongly supportive of visual inspections, on the grounds of both efficacy and costs.

Why, if we have coped up until now without oils, do we need to revisit the issue? There are a number of reasons. Current control strategies are effective in suppressing the build up of colonising aphids within the potato crop. This is essential in combating PLRV spread which requires prolonged feeding for acquisition and transmission between plants. Once acquired, the aphid will remain infective with PLRV for life.

What most aphicides don't appear to do efficiently is to control the potyviruses such as PVY, PVA and PVV. Potyviruses are described as non-persistent since the aphid is infective for a limited period but they can be acquired and spread during the brief probing of plants by many species of aphid.

These aphids can be potential colonisers or any of a range of other species which can migrate through the potato crop in a few hours searching for suitable hosts. Most aphicides may not act quickly enough to stop the acquisition and spread of these viruses. When there is a continuous influx of significant numbers of transient aphids a spray programme may be overwhelmed.

In addition, in the short term it needs to be borne in mind that the proportion of the different strains of virus is not static. Work to understand the ability of aphids to transmit different viruses and virus strains is also being carried out in the research project. PVY^N has become more prevalent than PVY^O in recent years. Being a strain which usually produces mild mosaic symptoms, PVY^N is more difficult to rogue and since the tolerance is higher than for severe mosaic, its incidence has increased. While the proportion of crops downgraded or failed has not increased there is no room for complacency.

Virus strains can change and so can aphid populations. The rationale behind the rotation of aphicide products is to ensure that the same active is not used continuously. This reduces the potential for insecticide resistant aphid clones to multiply in the crop, although insecticide resistance in the UK is only associated with one potato colonising species, the peach potato aphid.

Pesticide use is always under scrutiny. The EU Sustainable Use Directive, in particular, requires that Member States take measures to encourage the development and introduction of integrated pest management (IPM) and of alternative approaches or techniques in order to reduce dependency on the use of pesticides. Measures such as the Potato-Council funded network of Yellow Water Traps (Aphmon)

<u>http://aphmon.csl.gov.uk/levy/</u> allow the potential for virus transmission to be gauged and the subsequent requirement for aphicide use is then demonstrably based on a risk management strategy. It is possible that mineral oils could be an additional measure which could form part of an effective IPM programme though it should be borne in mind that there is little information on their environmental impact/fate.

Next steps: it is stated in the review that the phytotoxic effects mentioned in some of the reports are historical and that newer formulations are less likely to produce the more extreme responses seen in the past. However, the concentration of oil used is always a critical factor in successful control with minimal damage. If oils have a use in GB, then the certifying authorities must be satisfied that their use will not significantly impact on the resources required for inspection or on the reputation of the GB seed sector for quality.

Any follow up research will also need to address concerns regarding the rates and formulations of mineral oils, their environmental impact and the potential for interactions in tank mixes and subsequent sprays.

The Potato Council commissioned the authors (SCRI, Scottish Agronomy Ltd, SAC) to produce the review. The selection of data, its interpretation and views represented in this report remain those of the authors.

THE REVIEW

2. ABSTRACT

The subject of this review is agrochemical approaches that have been used to control non-persistently transmitted viruses in potato crop. Chemical strategies were split between synthetic insecticides and mineral oils. The advantages and limitation for each chemical were considered, and reports about successes and failures were presented. Moreover, combined effects of mineral oil with insecticides, or the synergistic effects of some agricultural practices with chemical application were discussed. The promising repellent and anti-feeding effects of recently introduced insecticide classes on non-persistent virus transmission were considered. Based on the literature a series of recommendations are made.

The aim of this review is to examine the useful characteristics of mineral oils, their application in some countries and their potential for introduction into current UK virus control programmes.

3. KEYWORDS

Potato viruses, non-persistent virus transmission, PVY, PVA, mineral oil, synthetic insecticides, chemical control

4. SUMMARY AND RECOMMENDATIONS

- There is overwhelming evidence that mineral oils work in minimising spread of potyviruses and this is beyond reasonable doubt (see Table 2).
- Prior to mineral oil use in the UK, consideration has to be given to the confidence with which visual inspection methods used by the Certifying authorities can be undertaken. In France and Netherlands, where oil is commonly used, crops are subjected to minimal growing crop inspection and are post harvest tested instead.
- Phytotoxicity and reduced crop yields are reported in many mineral oil studies and this occurs at concentrations of more that 1%.
- A policy on the impact of mineral oils for visual seed inspection has to be developed prior to their use in the UK seed potato industry.
- Further work is required on the environmental fate of mineral oils.
- Oil effectiveness during irrigation and intense rainfall should be established.
- Studies should not be biased by a focus on insecticide resistant *M. persicae* as there are many more abundant aphid species which vector potyviruses that are sensitive to insecticides.
- The majority of literature has examined application of mineral oil as a separate spray. To be practicable the effect of tank mixing oils with fungicides needs to be investigated with respect to biological efficacy of reducing potyvirus and any unintentional increase caused in foliar blight.
- In addition to mineral oils, many reports found that pyrethroids insecticides have good activity in preventing potyvirus spread (see Tables 3 and 4).
- Maintaining a low environmental inoculum is the most effective method of controlling potyviruses.

5. INTRODUCTION

5.1. Seed potato production and pathogens

Potato (Solanum tuberosum) is the fourth most important crop in the world after wheat, maize and rice (Gebhardt & Valkonen, 2001). The World's annual production of potato has increased remarkably during the past few decades, particularly in developing countries. Seed potato production has become an important industry in countries such as Netherlands, Canada, France and Great Britain. British potato seed exports are worth approximately £18million annually and are regularly exported to over 50 countries.

The seed-growing areas of Scotland, Northumberland and Cumbria are designated as an EU Community Grade Region, recognising the high-health status of British seed potatoes, with all British seed potatoes meeting EU and UNICE standards. Thus the importance of the industry stems from the need to use healthy seed potatoes to avoid the build up of diseases that can occur during the multiplication of a vegetatively propagated crop. This is of utmost importance to the GB seed industry to help maintain its recognised high health status.

Potato is affected by many pathogens including fungi, bacteria, viruses and nematodes. In addition, there are many important insects which affect potato by direct feeding or by transmitting viral diseases. Sixty major plant diseases affect potato and 12 of them (20%) are caused by viruses. Plant viruses are therefore one of the most important diseases which threaten potato production worldwide. At least 35 viruses are reported to infect potatoes. However, the most economically important are PLRV, PVY, PVA, PVX, PVM, and PVS. PLRV, PVY, and PVA are spread world-wide and PLRV for example, can decrease potato yield up to 90%. The rest of potato viruses are restricted to some regions of the world, and they can decrease yield by up to 40 % (PVX, PVV, PMTV), 20 % (PVP, PVS, AMV), or 10% (CMV) (Salazar, 2003).

In the last two decades insecticides have been increasingly successful in preventing PLRV epidemics by controlling its aphid vector. However, limited impact has been made on PVY and PVA spread and these viruses have become more dominant than PLRV in potato crops across Europe, Northern United States and Canada. Unlike persistent and semi-persistent viruses, non-persistent viruses are very difficult to control in the field by targeting their insect vector. The reasons for this difficulty stems from the way they are spread between plants that is characterized by both acquisition and transmission during very brief probes by individual aphids, combined with a wide range of efficient aphid vector species. Moreover, there is a need to assess information about the influence of insecticides in non-persistent virus transmission as all the currently used chemical programmes to control potato viruses were primarily developed for PLRV.

5.2. Viruses and their vectors

5.2.1. Types of virus

Plant viruses were initially divided into three main groups according to their relation with their insect vectors. A **persistent** virus is the term used by Watson and Roberts (1939) to describe viruses that can persist in their insect vector for a long time (more than 12 hours). The second transmission pattern was first termed as **non-persistent** by Watson and Roberts (1939) to describe viruses which do not survive for long inside

their vectors (less than 12 hours). Sylvester (1956) first introduced the term **semipersistent** to discriminate between viruses which can be acquired and transmitted briefly be their vectors (non-persistent) and the viruses that require a relatively longer time than the non-persistent viruses to be acquired and transmitted (semi-persistent).

The terminologies used above were based on the retention time of the virus inside its insect vector. The terms "circulative" and "stylet-borne" were introduced by Kennedy (1962) to differentiate persistent viruses that circulate into an insect's blood system from the semi-persistent and non-persistent viruses, which are carried on the stylet.

5.2.2. Mechanism of non-persistent virus transmission

The non-persistent mode of virus transmission is a unique characteristic of the aphids with piercing-sucking mouth parts (reviewed by Pirone and Harris, 1977). As mentioned above this kind of transmission is characterized by brief acquisition and inoculation times. The virus can be acquired with very short probes (5 sec); similarly, the aphid is able to inoculate acquired virus within a similarly short time. However, the virus is apparently lost completely from the vectors after 1-2 hours provided that it is feeding on a healthy plant (Bradley, 1959). Prolonging the acquisition period has an inhibitory effect on the virus acquisition by an aphid vector.

Once virus is acquired, after an optimal feeding time, the aphid vector can make up to 10 infectious probes (Watson and Roberts, 1939). This means that one winged viruliferous aphid can spread a virus infection to 10 healthy plants if it performed a single probe on each one. Alternatively, many infection sites could be initiated if the aphid continued to probe on the same plant. In both situations, this probing behaviour will be harmful by spreading infection to many new healthy plants and or increasing the probability with which individual plants will become infected.

This is true for the colonizing aphid vectors which live on potato plants. However, the majority of the aphid vectors of non-persistent transmitted viruses are non-colonizing species. These aphid species will be much more likely to perform single probes on many plants while searching for a suitable host.

It was first thought that non-persistent transmission was a purely mechanical process, and the **needle like hypothesis** was proposed by Doolittle and Walker, (1928). However, this hypothesis was challenged as early as the 1940s when Watson and Roberts (1939) indicated that non-persistent transmission is a more complicated process and a vector substance is probably involved.

The **ingestion-egestion theory** (Harris, 1977) was one of the most dominant hypotheses in explaining mechanism of non-persistent transmission of plant viruses. It was believed that non-persistent virus was acquired by ingestion and carried on the lining of the alimentary canal until inoculated by egestion. This theory was extensively investigated but its importance declined after the emergence of **the helper virus strategy** (Pirone and Blanc, 1996).

The key factor in the helper hypothesis is a helper component protein factor (HC-Pro), first discovered by Kassanis and Govier (1971a,b). HC-Pro is a viral encoded nonstructural multifunctional protein and one of the most important functions of this viral gene product is to mediate aphid transmission of non-persistent viruses. A lot of work has been done to reveal the mechanism by which the HC-Pro regulates non-persistent virus transmission by aphid with very limited success. The two most important hypotheses in interpreting HC-Pro function were proposed a long time ago; however, there is still no confirmation of the complete involvement of either hypothesis in the transmission process.

The **bridge hypothesis** was first introduced by Kassanis and Govier (1971a,b, Govier and Kassanis, 1974) and proposed by Blanc and Pirone (1996) to suggest that HC-Pro works as a bifunctional molecule by joining the virus particles with putative virus receptors on the aphid's stylet. Most of the evidence supports this proposed action of HC-Pro (Taylor and Robertson, 1974; Pirone and Thornbury, 1984; Ammar *et al*, 1994; Wang et al., 1996; Blanc *et al*, 1997, 1998; Uzet *et al*, 2007).

A second hypothesis suggests the direct involvement of the HC-Pro in mediating the non-persistent virus transmission by aphids, and it is known as the **conformational change hypothesis**. This indirect role of HC-Pro is achieved through conformational changes which eventually lead to direct attachment between the virus particles and the aphid's receptors (Salomon and Bernardi, 1995). There are other supporting reports about direct and indirect involvement of the DAG motif in the N-terminal part of the virus coat protein in non-persistent virus transmission (Gal-On *et al.*, 1992; Baulcombe *et al.*, 1993)

Although most lines of evidences support the bridge hypothesis the exact mechanism behind non-persistent virus transmission is still unknown.

5.2.3. Aphid species transmitting potato viruses

There are three main aphid species which colonize potato crops in the UK. Colonizing aphids cause two kinds of damage to potato, direct damage which happen because of sucking plant sap and depleting plant nutrients. This kind of damage is normally controlled by insecticides. The second kind of damage is indirect by transmitting plant viruses, and this kind of damage is more difficult to control with insecticides.

M. persicae is considered to be the most important aphid which infests potato crops. Their importance is as a vector as it has the ability to transmit the majority of plant viruses efficiently. PLRV is mainly transmitted by *M. persicae* but five other aphid species can make a contribution in some circumstances. On the other hand, potyviruses (PVY, PVA, and PVV), and the other aphid transmissible potato viruses (PVM, AMY and CMV are vectored by at least 40 aphid species (see Table 1). In contrast to PLRV transmission, PVY and the other potyviruses are transmitted in the field mainly by non-colonizing aphid species although *M. persicae* is a more efficient vector in the laboratory. It is believed that the most important non-colonizing aphids are cereal aphids which migrate from cereal crops to potato crops as their flight activity is correlated with the outbreak of the PVY and PVA epidemics in potato crops (Sigvald, 2007; Pickup, 2008). Species of aphid that are considered vectors is constantly expanding and Verbeek *et al.*, (2010) recently reported additional aphid species which are capable of transmitting PVY in the Netherlands.

TABLE 1 APHID SPECIES WHICH ARE REPORTED TO TRANSMIT PVY IN POTATO CROPS AROUND THE WORLD

Aphid species/common name	Reported by	Virus isolate	Transmission efficiency*	Other potato viruses transmission
Acyrthosiphon pisum	Van Hoof, 1980	PVY ^N	14%	PLRV, PVS
A. primulae	Ragsdale et al, 2001	PVY ^N	15%	NA
Aphis citricola	Raccah et al, 1985	PVY in pepper	6.2%	CMV
Aphis craccivora	Ferreres et al, 1993	PVY in pepper	4%	NA
Aphis fabae	Van Hoof, 1980	PVY ^N , PVY ^O	24%	PLRV
Aphis glycines Soybean aphid	Davis <i>et al</i> , 2005; Davis and Radcliffe, 2008 (USA)	PVY ^O , PVY ^N , PVP ^{NTN}	14% to 75% depends on strain	CMV, AMV, PLRV
A. gossypi	Raccah et al, 1985	PVY ^O	31%	PLRV, PVA
Aphis nasturtii	Sigvald, 1984	PVY ⁰	7.1%	PLRV, PVA,PVS
	Van Hoof, 1980	PVY ^N	9%	
Aphis pomi	Harrington and Gibson, 1989	PVY ^O	2%	
Aphis spp.	Harrington <i>et al</i> , 1986	PVY ^O , PVY ^N	6%	CMV
Aphis sambuci	Harrington and Gibson, 1989	PVY ⁰	4.3%	NA
	De Bokx and Piron, 1990	PVY ^N	12%	NA
Aulacorthum solani	Van Hoof, 1980	PVY ^N , PVY ^O	5%	PLRV
	Piron , 1986	PVY ^N	12.5%	PVA
Brachycaudus helechrysi	Harrington <i>et al</i> , 1986	PVY ⁰ PVY ^N	7.2% PVY ⁰ 0.9% PVY ^N	NA
	Harrington and Gibson, 1989	PVY ^N	5.9 %	
Brachycaudus spp	Piron, 1986	PVY ^N	14.7%	NA
B. cardui B. amygdalinus B. rumexicolens	Perez, 1995	PVY pepper	NA	
Capitophorous hippophoes	Van Hoof, 1980	PVY ^N	3%	NA
Capitophorus eleagni	Halbert et al, 2003	PVY ^O	2%	NA
Capitophorus spp	Perez 1995	PVY pepper	NA	
Cavariella aegopodii	Piron , 1986	PVY ^N	0.4%	NA
Cavanella aegopouli	Harrington and Gibson, 1989	PVY	0.2%	INA.
Cavariella pastinaca	Salazar 1996	PVY ^N	NA	NA
Cryptomyzus ballotae	Harrington et al. 1989	PVY	100%	NA
Cryptomyzus galeopsidis	Piron , 1986	PVY ^N	17.4%	NA
Cryptomyzus ribis	Piron, 1986	PVY ^N	15.4%	
Diuraphis noxia	Perez, 1995	PVY-Pepper	4-7%	
Dysaphis spp	Harrington and Gibson, 1989	PVY ⁰	1.8%	
Drepanosiphum platanoidis	Powell <i>et al</i> , 1992	PVY ^N	0.6%	
Hyadaphis foeniculi	Piron, 1986	PVY ^N	14.7%	
Hyalopterus pruni	Piron, 1986	PVY ^N	13.9%	
Hyperomyzus lactucae	Piron, 1986	PVY ^N	17.4%	
Macrosiphum euphorbiae	Van Hoof, 1980	PVY ^N	29%	PLRV
Metopolophium dirhodum	Van Hoof, 1980	PVY ^N	3%	
Metopolophium albidum	Van Hoof, 1980	PVY ^N	11%	
Metopolophium festucae	Harrington et al. 1986	PVY ⁰	0.5%	
Myzaphis rosarum	Harrington et al. 1986	PVY ^O	10%	
Neomyzus <i>circumflexus</i>	Salazar 1996	PVY ⁰ , PVY ^N	NA	PLRV
Myzus ascalonicus	Verbeek et al., 2010	PVY ^N , PVY ^{NTN} , PVY ^N W	REFs = 0- 0.01	PLRV
Myzus cerasi	Harrington and Gibson, 1989	PVY ^{O,} PVY ^N	3.2%	
Myzus certus	Van Hoof, 1980	PVY ^N	71%	

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Aphid species/common name	Reported by	Virus isolate	Transmission efficiency*	Other potato viruses transmission
Myzus ligustri	Harrington and Gibson, 1989	PVY ⁰	50%	
Myzus myosotidis	Harrington et al.1986	PVY ⁰	100%	
M. persicae nicotianae	Kanavaki et al. 2006	PVY ^N	15.3%	PLRV
	Halbert et al. 1995	PVY	NA	
N4	Van Hoof, 1980	PVY ^N	50%	PLRV, PVA, PVS
Myzus persicae	Harrington and Gibson, 1989	PVY ^O , PVY ^N	8.4%	
	Piron, 1986	PVY ^N	71.1%	
Phorodon humuli	Van Hoof, 1980	PVY ^N	35%	PLRV
Rhopalosiphum insertum	Van Hoof, 1980	PVY ^N	50%	
Rhopalosiphum maidis	Helbert et al, 2003	PVY ⁰	1.5%	
· · ·	Kostiw, 1979	PVY ⁰	2.7 %	
	Van Hoof, 1980	PVY ^N	2%	
Rhopalosiphum padi	Piron 1986	PVY ^N	11.5%	
	Harrington and Gibson, 1989	PVY ⁰	2.4%	
Rhopalosiphum pseudobrassicae	Ragsdale et al. 2001			
Shizaphis graminum	Perez, 1995	PVY pepper	NA	
	Harrington and Gibson, 1989	PVY ⁰	0.1%	
Sitobion avenae	Piron, 1986	PVY ^N	1.8%	
Sitobion fragariae	Harrington and Gibson, 1989	PVY ⁰	0.5%	
Situbion naganae	Piron, 1986	PVY ^N	10.1%	
Sitobion graminum	Verbeek et al., 2010	PVY ^{NTN} , PVY ^N W	REFs = 0- 0.05	
Staphylea tulipaellus	Salazar 1996	PVY ^N	NA	PLRV
Therioaphis trifolii Therioaphis sp	Perez 1995	PVY pepper	NA	
	Harrington and Gibson, 1989	PVY ⁰	0.5%	
Uroleucon spp.	Piron, 1986	PVY ^N	8.3%	

* the values (%) are derived from different methods of assessing transmission and are not directly comparable. Different clones of the same aphid species may give different results.

6. MINERAL OILS

6.1. Introduction

Essential plant oils have been used in the control of harmful organisms for a long time commencing in ancient societies such as Greece, Egypt, India, Persia and China. Agricultural oils are different in origin, as they are derived from industrial processes including by products made during distillation of crude oil to produce petroleum. Petroleum (mineral) oils are a mixture of aromatic, naphthenic, and paraffinic structures. In agriculture they were classified first according to the time of application for example: on dormant plants as winter oils which can be used to control overwintering insects, or as summer season oils which are safe for application during the growing seasons. The winter oils are heavier (longer chained) products. At present, Petroleum oils are available in many commercial brand names such as: horticulture oils, spray oils, white mineral oils, stylet oils, superior oil, volvic oil, etc.

6.2. Mineral oils and potato crops

The earliest report about oil inhibition of non-persistent viruses was from Bradley *et al.* (1962); they reported that paraffin wax oil inhibited PVY transmission. Further investigations were carried out after that to investigate the effect of oil on transmission of other non-persistent viruses on different crops (Loebenstein *et al*, 1964, 1966, 1970; Vanderveken and Semal, 1966; Vanderveken, 1968, Bhargava and Paul Khurana, 1969; Peters and Lebbink, 1973; Walkey and Dance ,1979; Ferro *et al.*, 1980; Asjes and Blom-Barnhoorn ,2001).

Many review papers have been published on mineral oils (Vanderveken, 1977; Simons and Zitter 1980; Loebenstein and Raccah, 1980; Raccah *et al.*, 1980; Simons, 1982; Sharma and Varma, 1982; Raccah, 1986) all of which reach the conclusion that mineral oils do protect crops from non-persistent viruses. However, to achieve this requires frequent application because of their contact mode of action and plant phytotoxicity and yield loss occur with increasing oil concentrations. Most of the literature indicates that mineral oils suppress transmission of non-persistent and semi-persistent viruses, but have no effect on persistent viruses. However, Zitter and Everett (1979) reported that Florida tomato yellow virus (TYV), which is a persistent virus sharing many characteristics with PLRV luteovirus, can be controlled by weekly applications of mineral oil.

6.3. Characteristics of mineral oils used for inhibition of virus transmission

Mineral oil activity is dependent on their physio-chemical properties including saturation, viscosity and distillation (De Wijs *et al*, 1979). De Wijs *et al*. (1980) reported that paraffinic mineral oils are the most active mineral oils in inhibiting non-persistent virus transmission, and the best oils are those with a viscosity range of between 66-150 SUS (Saybolt Universal Seconds). Similarly, oils with a viscosity range of between 60-120 SUS, were reported to be more efficient in preventing virus spread (Simon and Zitter, 1980).

The characteristics of the best mineral oils for inhibition of virus transmission were reported by De Wijs *et al.* (1980) to be as follow:

- Viscosity Gravity Constant (VGC) of 0.790-0.819
- Viscosity between 66-150 SUS
- Boiling range between 370-420 °C
- Mean molecular weight of 340-380 Daltons
- Unsulphonated residue (USR) of 95-100, indicating the absence of aromatic structures, which are inactive and known to be phytotoxic
- Paraffin-pour point should be below 0 °C which indicates that n-paraffins do not form a very important part of the oil because they are not active
- Low content of naphthenic structures, as they seem to be inert and increase oil viscosity

In addition to these characteristics, emulsifiers have a significant effect on oil performance in inhibition of virus transmission. Performance of oils is normally enhanced by increasing the amount of emulsifiers to a critical point (0.75-1.25%). Volume, after which increasing emulsifiers will decrease efficiency for unknown reasons (Simons and Zitter, 1980).

6.4. Conditions for applying mineral oil

In addition to having the above characteristics, effectiveness of mineral oil is related to spray conditions. Boiteau and Singh (1981) found that oil efficacy in decreasing PVY transmission was largely affected by oil concentration, and somehow to the delivery rate; however, spray pressure has no significant effect. In contrast, Simon and Zitter (1980) reported that oil efficiency was improved by increasing the delivery rates and the spray pressure. Zitter and Simon and (1980) recommended the following procedures during oil application to prevent PVY spread in pepper crops in Florida: 1-2 % oil concentration, 200-400 psi spray pressure, addition of emulsifier to oil formulation (0.75-1.25%), and using spray nozzles to achieve small droplet size. Good coverage is essential when using mineral oil to control virus spread. The top surface of the plant may be exposed more often to probing by winged non-colonising aphids which alight but do not move to the lower side of plant leaves (Bradley, 1963). This is normally achieved by weekly application or twice-weekly application for rapidly growing plants. Coverage will be poor if mineral oils are sprayed on a wet surface and it has been recommended that other chemicals are applied one day after oil application (Simon and Zitter, 1980). Regarding persistence, it was reported that oils normally persist for 10-14 days post application (Simon et al., 1977), and a small amount of rain (< 25 mm daily) will not remove oil film from oil treated leaves (Simon and Zitter, 1980). However, Boiteau and Wood (1982) investigated the persistence of different mineral oil formulations on potato leaves, and the effect of natural and artificial rainfall on oil efficiency. They concluded that there was no significant effect of the timing and amount of rainfall on the residual amount of mineral oil.

Oil persistence is also affected by temperature, Simon *et al.*, (1977) found that the efficacy of Sunoco 7E oil lasted longer at 16°C compared with 24°C and 32°C. This can be explained in two ways: 1. Growing conditions for the plant are better at higher temperatures, and thus the leaves will expand more, decreasing the oil coverage on the plant. 2. The virus inhibitory characteristics of mineral oils may be damaged or lost when oils are subjected to higher temperatures.

Sunlight has a negative effect on oil spray persistence on the plant surface. For example, photo degradation of mineral oils was reported after exposure to ultraviolet light (Hodgkinson *et al*, 1999). It has been reported that oil applications are carried out

during the night in France to reduce the water requirements during the spraying process and avoid the effects of strong sunlight which may scorch the plants (E. Anderson pers comm.).

Concentration and application frequency of oil sprays are controversial issues. However, the general tendency is that oil concentration should be in the range of 1-3%, depending on the oil type. Concentrations below 1% are reported to be less effective, whereas concentrations above 3% are toxic to the plant and significantly decrease crop yield (Groove *et al*, 2008; 2009). Zitter and Simon and (1980) reported that adding solvents to mineral oil decreases the oil's viscosity and reduced the effectiveness.

The application frequency should provide continuous coverage to the new plant growth. In some reports a few applications (3-8) during the growing season were enough to offer reasonable protection (Shands, 1977; Martin-Lopez *et al*, 2006), while in others more frequent applications at weekly intervals were required (Boiteau *et al.*, 2008; Groves *et al.*, 2008; French typical programme, Denufbourgh, 2009, personal communication with Eric Anderson – see details in section below 'reports of mineral oil application'). Simon *et al* (1977) reported that oils are less effective on young leaves compared with mature leaves. However, this discrepancy in PVY transmission on young and mature leaves should not be directed to oil effect, because viruses and their vectors prefer young plant growth. Also the rapidly developing crop canopy produced leaves that are untreated with oil.

6.5. Mode of action of mineral oil in inhibiting virus transmission

Since Bradley (1962) discovered their action on virus transmission, considerable work has been carried out to investigate the mode of action of oils in inhibiting virus transmission and many hypotheses have been proposed (Bradley *et al*, 1963, Vanderveken, 1977; Simons *et al.*, 1977; Qiu and Pirone, 1989; Powell, 1992; Wang and Pirone, 1996, Powell 2005). However, even now, the exact mode of action of mineral oils in inhibition of non-persistent virus transmission is still uncertain.

6.5.1. Early observations

Early investigations into the mode of action of oil in preventing virus transmission confirmed that the inhibition mechanism is unrelated to the plant host, virus particle shape, and aphid vector species (Vanderveken, 1977). The following paragraphs outline hypotheses for the mechanism of oil in inhibiting virus transmission:

1. Asphyxia is the term which has been used to describe the oil's physical mode of action in killing insects and mites. This is suffocation as a result of physical blockage of the insect's respiratory system (spiracle openings) by the oily film. This direct mode of action is probably correct for insect control, but the effect of oil on virus transmission is unlikely to be related to this mode of action. This is supported by the fact that mineral oils can reduce the transmission of non-persistent and semi-persistent viruses, but are less effective in preventing the transmission of persistent viruses. Nonetheless, Martin-Lopez *et al.* (2006) found that both mineral and vegetable oils achieved considerable mortality on aphid populations after 72 hours, with mortality ranging between 70-83% depending on the oil type. Yankova *et al.* (2009) also found

that the mineral oil Akarzin at 0.4%, and another two plant oils (Turpentine 1%, Eucalyptus 1%) gave over 80% efficiency in controlling *M. persicae*.

2. Bradley (1963) demonstrated that PVY transmission inhibition occurred when aphids accessed oil before acquisition or between acquisition and inoculation. This means that oil decrease both acquisition and inoculation of PVY. He found that PVY transmission decreased significantly even after a brief contact between the aphid labium and oil treated leaves. In order to explain his finding Bradley (1963) hypothesized that inhibition of PVY transmission after treatment with oil could result from either removal of virus particles from their attachment sites on the aphid's stylet, or by enhancing this attachment so that it became irreversible. He added that aphids may acquire oil while they are acquiring virus, and the oil is then inoculated into the plant with the virus. The oil may then prevent virus multiplication by interfering with the host cells in some way.

3. An early proposal was that mineral oils may inhibit non-persistent virus transmission by interfering with the physiology of the plants they are applied to or by modifying the metabolism of plant cells. However, it was possible to eliminate this hypothesis as inhibition of transmission occurs when only the insect is exposed to the oil – i.e. without treating the virus source or the recipient plant with oil (Vanderveken, 1977).

4. Another hypothesis was reported by Loebenstein *et al.* (1964). They suggested that acquisition and transmission are not affected by oil. However, the infection process itself is terminated post inoculation due to the oil action. This hypothesis was supported by Peters and Lebbink (1975) working on CMV (cauliflower mosaic virus); by using an aphid free system they confirmed that mechanical virus transmission was impeded when the virus inoculum was mixed with oil or when oil was applied after manual inoculation. Moreover, Peters and Lebbink, reported that CMV infectivity was restored after separating the oil from the virus particles by centrifugation. There are several lines of evidence which suggest that oil affects both parts of the transmission is inhibited when oil comes into contact with either the aphid's stylet or the virus particles before acquisition or during any of the transmission processes (Bradley, 1963; Russell, 1970)

5. Mechanical surface adherence is a hypothesis to explain the transmission of styletborne viruses proposed by Van der Want (1954) that suggests that the variation in the surface structure of stylets is behind the adsorption and elution of virus particles. Kassanis and Govier (1971a,b) found that aphid transmission of PVY^C and PAMV (non- aphid transmissible viruses) occurs only when aphids have initially fed on a plant infected with PVY^O or on a plant co infected with either PVY^C or PAMV and PVY^O. In order to understand their observations Kassanis and Govier (1971a,b) used Van der Want (1954) hypothesis to explain that PVY^C and PAMV were not adsorbed directly to the stylet, but PVY^O modified the charge or the surface structure of the stylet to allow PYY^C and PAMV to be adsorbed onto the stylet or to the adsorbed PVY^O particles. Similarly, Vanderveken (1977) stated that oils could modify the surface structure or charge of the stylets and inhibit adsorption or elution of virus particles.

6. Vanderveken (1977) concluded that two hypotheses should be considered to explain how oil inhibits non-persistent virus transmission: 1 A modification effect which may affect the stylet charge and prevent virions from adsorption onto or elution from

the stylet. 2. Impedance of charge exchange between aphid stylet, virus particles, and plant cells caused by the insulating properties of oils.

6.5.2. Recent investigations

The number of studies that deal with the mode of action of mineral oil in preventing virus transmission has declined in recent years. However, those that have been done were focused on two major subjects. The first is the interference of oil on aphid feeding behaviour. This was made possible after Tjallingii (1978) developed monitoring of aphid probing behaviour using the electrical penetration graph (*EPG*) *technique*. The second direction in recent investigations was in support of the assumption that oil may interfere with the chemistry of virus retention on aphid stylet receptors.

6.5.3. Mineral oil and aphid feeding behaviour

Simons *et al.* (1977) found that *M. persicae* required a longer time to initiate probing on oil treated plants, and sap ingestion was also decreased. It was suggested this delay decreased the chance of viruses being passed on to an uninfected plant. Interference of the sensory structures of the aphid labium or stylet was also reported to be responsible for the change in the probing behaviour induced by oils on treated plant surface (Harris, 1977). However, there are conflicting reports that suggest that mineral oils have no effect on aphid feeding behaviour (Bradley *et al*, 1963; Vanderveken, 1968; Peters and Lebbink, 1975).

EPG of aphids feeding on oil treated plants revealed that their probing behaviour was affected and that the presence of oil delays the initiation of aphid stylet penetration (Powell, 1992) or facilitates probing (Ameline *et al.*, 2009). However, both previous reports concluded that there was no correlation between the observed feeding behaviours and virus transmission inhibition which may suggest other reasons for virus transmission inhibition by oils. Powell (1992) suggested that a delay in stylet penetration was not responsible for inhibition of virus transmission by oil application. He suggested instead that other factors were responsible, in line with those suggested by Qiu and Pirone (1989) (see next section). Recently, Ameline *et al.*, (2009) reported that oil affects the feeding behaviour of *M. euphorbiae*. However, this modification in feeding behaviour was not enough to explain the seven-day protection from non-persistent virus transmission provided by oils. Like Powell (1992) they concluded that mechanisms in addition to interference with aphid probing behaviour underlay the mode of action of oil in inhibiting virus transmission.

6.5.4. Mineral oil and virus retention

In agreement with Bradley's early observations, Qiu and Pirone (1989) were able to confirm that oil decrease virus acquisition by aphids through artificial membranes as effectively as from plant source. They concluded that oil decreased tobacco etch virus (TEV) transmission when *M. persicae* were exposed to oil before or after acquiring Hc-Pro, virus particles or a mixture of both.

Powell (1992) mentioned that oil can inhibit virus transmission by affecting both the retention and inoculation processes. He speculates that after being exposed to oil it is normally carried on aphid mouthparts but gradually released during successive penetrations. This explanation is in agreement with the observations by Bradley *et al.* (1963) that virus transmission was inhibited if aphids were exposed to oil before,

during or after acquisition, and the fact that transmission inhibition is increased if both the virus source and the target plants are treated with oil. The early observations about the effect of mixing mineral oil with plant infectious sap on the mechanical transmission of virus (Lobenstein *et al.*, 1964) also provides strong support for Powell's hypothesis. More recently Wang and Pirone (1996) reported the possible involvement of oil in damaging the virus helper component protein (HC-Pro)'s interaction with aphid receptors in the stylet. They labelled TEV particles with radioactive isotope, and found that the ability of aphids to transmit these labelled virions was significantly affected when aphids probed on oil treated membrane or plant leaf before virus acquisition. Their data suggest that mineral oil inhibited virus retention as there was a lack of labelled virus particles in the stylet compared with the control. This correlation between transmission efficiency and retention of labelled virus particles in the stylet, led the above researchers to speculate that a hydrophobic layer of oil is retained in the food canal after probing oil treated membrane, which may affect virus retention in the food canal.

Powell 2005 determined that inoculation is associated with the first intracellular activity (subphase II-1) following maxillary puncture of an epidermal cell is associated with active injection of saliva directly into the cytoplasm. When aphids acquire transmissible combinations of HC and potyvirus, virions are retained within the maxillary stylets (Berger & Pirone, 1986) where they show HC-mediated adherence to the cuticular lining of the food canal (Ammar *et al.*, 1994; Wang *et al.*, 1996). Uptake of virions occurs when the maxillary stylet tips puncture the plasma membrane of an epidermal cell (Lopez-Abella *et al.*, 1988; Powell, 1991).

An alternative hypothesis is that virions are released by salivation. This is possible because, although the salivary canal remains distinctly separate from the food canal for almost the entire length of the aphid stylet bundle, the two canals converge 2-4 µm from the tips (Forbes, 1969). At the point of convergence, the maxillary stylets form an enclosed common duct (Kimmins, 1986) where mixing of food and salivary canal contents may occur. Ingested virions adhering to the cuticular lining of the common duct may therefore be flushed out during saliva secretion into the plant, providing an alternative 'ingestion-salivation' hypothesis for transmission (Martin et al., 1997). Uptake of virions occurs when the maxillary stylet tips puncture the plasma membrane of an epidermal cell. The third intracellular activity (II-3) is associated with efficient uptake of non-persistently transmitted viruses and therefore represents active ingestion of cytosolic fluid by the aphid. The common duct represents the likely functional retention site and, although many virions flow past this position during ingestion, there may be no credible mechanism by which they can be inoculated. This new information is consistent with the hypothesis that oil works on the retention of virions in the common duct.

Recently, Wrobel (2007) tried to investigate the effect of mineral oil on successive infectious probes that an aphid performs. In agreement with the retention impairment hypothesis he found that efficiency of the oil in decreasing the number of successive infections was enhanced when both virus source and test plant were treated with oil. He suggested that mineral oil may act by inactivating virus particles on the aphid stylet while being acquired from an oil treated source.

Although many hypotheses have been proposed, the exact mode of action of oil in preventing non-persistent virus transmission remains uncertain. However, the balance of current evidence suggests that the process controlling attachment and release of virus particles from their receptors on aphid mouthparts is disrupted by oil (Qiu and Pirone 1989; Wang and Pirone, 1996, Powell *et al.*, 1998). This is supported by the

fact that oil inhibits transmission of the non-persistent and semi-persistent viruses but not persistent viruses. Further investigation should be done in order to understand the virus-vector relationship more clearly.

6.5.5. Reports of successful mineral oil applications in decreasing virus spread in potato crops

Initial laboratory work by Bradley (1963) indicated that application of 0.03 mg of paraffinic oil per 10 cm² of plant leaf surface was enough to give 60% reduction in PVY transmission. Investigations into the effectiveness of oils in disrupting virus transmission were extended to field conditions. In some reports, field experiments were performed in parallel to those conducted in the laboratory. In other reports, laboratory experiments were performed first, followed by field investigations. The majority of findings suggest that measurement of mineral oil effectiveness decreased considerably under field conditions (Lobenstein *et al.*, 1970, Shands, 1977). Zitter and Ozaki, (1978) attributed this difference to the wrong choice of oil-emulsifier, improper oil concentration, and poor coverage of oil on the plant.

The literature about mineral oil use for controlling virus spread in potato crops is summarised in Table 2 and can be found in the following papers:

In Canada, Bradley *et al.* (1966) found that six applications of mineral oil during a season on a weekly basis was enough to suppress PVY spread in potato crops. Two oil spray programmes were used: In the first programme, oil was initially sprayed at 0.5 gal/acre (4.675 l/ha), then the amount of oil was doubled in the second, third, and fourth applications to 1 (9.35), 2 (18.7), and 4 (37.4) gal/acre (l/ha) respectively. Another two applications were performed using 4 gal/acre (37.4 l/ha). This programme reduced virus transmission by 56-88%. In the second programme, the first two oil sprays were applied at 2 gal/acre (18.7 l/Ha), then the remaining four applications at 4 gal/acre (37.4 l/ha). This decreased virus transmission by 71-88%. Phytotoxicity was not of great importance as the yield was not affected although occasional burning on petioles and leaflets was observed.

Boiteau and Singh (1982) also tested the effects of the following eight commercial mineral oils on PVY transmission in potato fields: JMS Stylet oil, Triton B-1956 Co-op Surfactant Conc. Pfizer XA Kornoil Agr. Adjuvant Green Cross Booster Plus Kornoil Conc. Adjuvant, Atplus 300F

They found that these oil formulations gave comparable results of between 35-64% reductions in PVY spread when applied to potato. The best reduction of PVY spread was 64% which was consistent after applying oil at 1.5-3% emulsion in water without causing any phototoxic effect on the plant. Application of oil at lower concentration (1%) gave less reliable results than application at 1.5%. The reason for the comparable results achieved by the eight oil formulations was believed to be because they had the same viscosity range (60-120 SUS).

In Oraono, Maine, USA, Shands (1977) found that 6 weekly application of paraffin oil, N.F (Atrooil QS-1540A, Esso Imperial Oil, Ltd) at 2.5 gal/acre (23.4 l/ ha) of on potato crop decreased the PVY infection by 69-75%. In another year, the same author found that 5 applications of mineral oil achieved 64%, 55% decrease in PVY transmission when applied at 1.25 and 2.5 gal/acre (11.7 and 23.4 l/ha), respectively. No phototoxic effect or significant decrease in yield after oil application was reported.

In Maine, USA mineral oils were used on a seed potato crop to control PVY spread as reported by Simons and Zitter (1980) from a personal communication with D.F. Hammond. The conditions described were that 0.75% JMS stylet oil decrease PVY incidence by 3.36 % when applied at weekly intervals. No phytotoxicity was reported at this low concentration, and the loss in yield due to oil application did not exceed 3%

In England, considerable work has been done by Gibson and co-workers (Gibson and Cayley (1984); Gibson and Rice (1986); Gibson *et al.* (1988)) to investigate the effect of oils on non-persistent transmission in potato fields. However, their work focused on the effect of mixing oil with pyrethroids which will be discussed in another section of the review (section 6.5.8).

In Ireland Bell (1980) investigated the effect of mineral oils on PVY^N transmission in potato fields of cv. Record. He reported that 8 applications at weekly intervals of 3% Bayol 52 decreased virus spread by 59% when compared with unsprayed controls. 14 applications at weekly intervals during the season achieved 62% reduction. Application of a Captafol fungicide after the 6th week spray of mineral oil resulted in severe phytotoxicity and significant yield loss.

In Poland, Turska (1980) found that spray with 0.3% mineral oil emulsion in water decreased virus incidence by 50%. They found that the type of oil affected the yield loss, but no difference in phytotoxicity was observed with five potato cultivars.

Wrobel (2005) investigated the effect of presprouting, haulm destruction and mineral oils on PVY spread in potato crops. He reported that presprouting of seed potato has no effect in decreasing PVY spread. However, mineral oil application decreased incidence in progeny tubers by 50%. A combination of mechanical and chemical methods was more effective than a single method. PVY incidence was greatly decreased when mineral oil was used in combination with haulm destruction. Using this approach, the infection rate was decreased by 68-79% in cv Mila, and by 80-84% in *cv*. Balbina. He recommended that a combination of early mechanical and chemical vine killing with weekly application of mineral oil provided optimal protection in seed potatoes of susceptible PVY cultivars.

Wrobel and Arbanowicz (2005) investigated the effect of several adjuvants on potato phytotoxicity. They found that oils induced no phytotoxic symptoms when applied at 1% once a week for 10 weeks. However, with increasing oil concentration phytotoxicity was observed. This was least with the oil Sunspray 850 EC, and no phytotoxicity was observed with rape oil.

Wrobel (2006) conducted a three-year field experiment to investigate the effect of the mineral oil Sunspray 850 EC on PVY, PVM and PLRV transmission. He found that regular oil treatment decreased the aphid population by 60%, and the percentage of PVY, and PVM in resultant tubers was considerably decreased. The effect was

particularly significant with PVY spread in the susceptible cultivar Mila. Thus he recommended applying mineral oil to highly susceptible PVY cultivars.

Wrobel (2007) found that the ability of *M. persicae* to transmit PVY to successive potato plants in greenhouse conditions was significantly affected after oil application. This author found that the strongest effect was achieved when Sunspray 850 EC mineral oil was applied to both virus source and test plant.

In France, many commercial white mineral oil products are available and they are effective at decreasing non-persistent virus spread. The products include:

FINAVESTAN (Total Fina Elf) VAZYL-Y (CCL) ACTIPRON PLUS (CEREXAGRI) OIL ANTI VIRUS Y (JPB, GERMIPHYT (Germicopa) OLEOPHOLINE (JP Industrie)

The recommended rate for applying mineral oil is at 10-15 l/ha dissolved in 250-450 l of water (3 - 4% oil), depending on the susceptibility of potato variety to potyvirus, primary infection level and the vector pressure. It is important to apply oil at 8 l/ha, 8 l/ha, and 10 l/ha after 30%, 70%, and 100% of plant emergence, respectively. After these initial sprays a programme of 12-15 l/ha at weekly intervals is continued until tuber maturity. Finally, two application of 12, and 10 litre/ha are applied when the plants start to senesce. This information came from an FNPPPT Booklet. (FNPPPT = Fédération Nationale des Producteurs de Plants de Pomme de Terre).

From a personal communication with Denufbourgh (2009) the typical use of mineral oils is 120 I. It is used in conjunction with other insecticidal products on potato in France is as follows:

- 1. E30 5 I oil in 150 I water (3%)
- 2. E50 7 I oil in 210 I water (3%)
- 3. E100 10 I oil in 300 I water (3%)
- 4. 75 ml Hallmark Zeon +10 l mineral oil
- 5. 10 I mineral oil
- 6. 0.3 kg plenum + 10 l mineral oil
- 7. 10 I mineral oil
- 8. 1.5 | Dovetail + 10 | mineral oil
- 9. 10 I mineral oil
- 10. Teppeki + 10 liters mineral oil
- 11.10 I mineral oil
- 12.75 ml Hallmark Zeon +10 l mineral oil
- 13.10 I mineral oil

In the Netherlands all the commercial oils are 99% identical, they differ in the emulsifier that comprises the remaining 1%. The following are the characteristics of currently used mineral oil (Cropspray 11E, De Sangosse Ltd) in the Nederlands according to Foster, 2009, (personal communication with Eric Anderson)

Viscosity cSt at 40°C AS	19.9 min to 22.0 max		
Unsulphonated residue	92% min		
Gravity API at 60 F	ASTM D287	31.0 min	
Density at 15°C		0.86	
Pour point		-15C	
Boiling point		368 - 382⁰C	

Cropspray 11E is a high-grade adjuvant oil containing 99 % of highly refined paraffinic oil. It is safe to apply as often as required to a maximum of 2.5% although it is normally used at 1%.

Aphid monitoring and haulm destruction are the most important methods to control potyviruses in Dutch potato crops. However, farmers are using up to 8 applications per season of 10 l/ha of paraffinic mineral oils totaling 80 l to suppress the transmission of new recombinant aggressive strains of PVY.

Schepers *et al.* (1977) reported that mineral oil spray on potato cultivars Bintje, Eersteling, and Eigenheimer which were growing adjacent to PVY^N infected potato crops, was effective in considerably decreasing PVY^N spread. This occurred when oil was applied from emergence at fortnightly, weekly and twice weekly intervals at a rate of 1000L/ha of 2.5% oil emulsion. However, oil caused leaf necrosis later in the season, and yield was decreased by 8.5, 10, and 22% depending on the frequency of sprays.

Peters (1977) reported that oils can decrease virus spread when applied to potato crops. However, a negative effect of oil on tuber weight and burning leaves prevented more widespread application of oil usage in seed potato crops.

De Bokx and Cuperus (1978) reported that oil spray can decrease PVY spread, but oil also decreases potato yield and masks disease symptoms by severe leaf injury.

Schepers and Bus (1978) found that application of 7.5-25 L/ha of mineral oil on potato cv. Bintje, starting at emergence and repeated at weekly intervals, achieved a 60-80% decrease in PVYN spread, and the yield decreased by only 2-8%. However, yield loss was increased to 20-30 % when Maneb and Tin (Sn) were used to control *Phytophthora infestans*, and the resultant damage prevented identification of virus-infected plants. They recommended not using copper compounds, but Maneb and Chlorothalonil could be used provided that oil was not mixed with them. They also reported that oil spray is expensive and may be justified on basic seed crops only.

Schepers and Bus (1979) reported that the mineral oil Schering 11E restricted PVY^N transmission considerably. However, oil spray decreased yields by 2-11% regardless of concentration and time of applications.

In Germany, Wenzel and Foschum (1973) reported that 3% oil applied weekly lead to a 84% decrease in virus spread, relative to untreated control.

In Switzerland, Munster and Carnu (1978) reported that mineral oil was effective in decreasing PVY spread by up to 92% and they were investigating the effect of decreasing the concentration from 3% to 1% and the frequency of application from 6 times to 4 times during the season.

Carnu and Gehriger (1981) then reported that mineral oil application at 10 day intervals was effective in achieving 50% decrease in PVY incidence. They also reported that the mineral oil Sandoz was approved for use in seed potato production for preventing PVY spread. Application of oil at 10 days interval achieved a 50% reduction in virus infection under high pressure infection conditions. Yield was only decreased by 5-10%.

In Serbia (formerly Yugoslavia), Milosevic (1996) compared the efficiency of weekly application of 3% white oil (80% mineral oil), and 40-day application of 0.1 % Metasystox (Demeton-s-methyl), on PVY, and PLRV spread in a potato crop. The efficiency of mineral oil in suppressing PVY spread was 47% compared to the control. On the other hand, Metasystox had no effect on PVY spread but decreased PLRV spread by 69.2%

In Italy, D' Amato *et al.* (1981) investigated the effect of the mineral oil (Omlocin) in combination with Sumicidin (fenvalerate) on virus transmission in a seed potato crop *cv.* Jaerla. They found that potato yields were 22.7 t/ha when oil was applied alone compared with 28.33 t/ha when both compounds were applied. In the mixed programme the percentage of virus infected tubers decreased to 9.64% compared with 24.03% in the untreated control.

In Cyprus, Ioannou and Lordanou (1987) investigated the effect of some insecticides and oil formulations on seed potato crops under high artificial pressure of PVY inoculum and natural population of aphid vectors.

They reported that 1-3 % foliar oil spray reduced PVY incidence by 70-95% without greatly reducing the aphid population. They attributed the effect of the oil to transmission inhibition rather than symptom suppression as the degree of protection was related to the concentration and frequency of application. 3% oil spray at weekly intervals gave the best result (90-95% control) and 1% at weekly intervals achieved 70-75% control. They did not notice a difference between the use of two mineral-oil formulations (Albolineum and Luxan Oil). However, weekly application with 3% of Luxan Oil caused severe leaf scorching, while Albolineum was non-toxic in all the concentrations and application frequencies used.

In contrast, soil treatment with the systemic granular insecticides aldicarb or thiofanox and, to a lesser extent, foliar sprays with the specific aphicides ethiofencarb or pirimicarb gave substantial control of aphids but failed to significantly reduce the spread of PVY.

They concluded that 4-5 weekly sprays starting from 80% emergence, with 1-3% of Albolineum oil emulsions, is effective in controlling PVY incidence under conditions of high virus pressure in seed potato crops.

6.5.6. Reports of successful mineral oil applications in decreasing potato virus spread in crops other than potato

In addition to their effectiveness in limiting PVY spread on potato crops, use of mineral oils in decreasing PVY spread in other crops has been documented. Loebestein *et al,* (1970), decreased CMV and PVY spread in pepper nurseries by applying oil emulsions when the aphid population was high.

In Florida, USA, it has been reported that mineral oils (Stylet-Oil) were successfully used to control virus spread in pepper, cucumbers, and squash (Zitter and Ozaki, 1978). They reported that JMS stylet oil decreased virus spread by 3-8 fold in the experimental plots and offered complete protection under field conditions. In laboratory conditions Simons and Zitter (1980) found that 0.75% JMS stylet oil was effective in decreasing PVY transmission on pepper by a maximum of 50% with no phytotoxic effect.

Laboratory experiments in England, on tobacco seedlings has confirmed that Bayol 52Esso petroleum Plc, was effective in decreasing PVY transmission by 23.5% compared to the control. SC811, another mineral oil, achieved 23.8% decrease in PVY transmission (Gibson and Rice, 1986). Marco, (1993) reported that the light mineral oil (Virol) offered a 60% decrease in PVY and CMV spread in a pepper crop in Israel. Powell *et al.*, (1998) reported that the 1% spray of the mineral oil Sunoco 7E significantly reduced PVY transmission to tobacco plants. They found that transmission efficiency decreased to 2.5% compared to 36.7% for untreated control. In Hungary, mineral oils decreased PVY transmission on pepper by 73% (Suranyi, 1999).

Recently, Margaritopoulos *et al.*, (2009) used the light mineral oil (Bio-lid 80 E, W, SIPCAM Hellas) on tobacco plants in Greece to decrease PVY spread. They found that mineral oil offered a 44.5% reduction in virus transmission rate compared to an untreated control when applied on the test plant, and 40% reduction in PVY transmission when applied to the virus source plant.

6.5.7. Current investigations of mineral oil inhibition of virus spread in potato crops

There are several currently running projects investigating the effects of commercially available mineral oils on PVY transmission. In ongoing experiments Groves *et al.*, (2008), at the University of Wisconson, USA, are investigating the effect of some commercial oil formulations on PVY transmission. Initial results indicate that Aphoil, JMS Stylet oil, and QRD-416 offered comparable virus reduction (about 30%) on potato when applied 1-2 times a week.

In another ongoing project in **Manitoba**, **Canada** by McLaren *et al.* (Agriculture and Agri-Food Canada), the effect of commercial mineral oils on the transmission of PVY^O and PVY^{N: O} isolates are being investigated. They reported that Superior 70 oil decreased PVY transmission by 33%. In earlier work they also found that the mineral oil Aphoil (Agsco.Inc, Grand Forks, USA) controlled PVY spread in seed potato.

Regarding phytotoxicity and crop yield, McLaren *et al.*, (2008) reported that in 2007 they noticed some phytotoxic effect of oil spray on cv. Russet Burbank, but there was

no impact on the yield as both control and test crops produced 15.0 tons/acre (37 tonnes/Ha). Further studies are planned which will investigate the oil application rates on potato yield on different cultivars.

6.5.8. Combination of oils with insecticides and other crop practices

The effect of mineral oils in disrupting PVY transmission described in the previous sections can be enhanced by mixing with insecticides, or by incorporating crop borders.

Bradley (1963) argued that virus sources for non-persistent viruses could be targeted by mineral oil, as was also suggested by Simon (1960). The latter paper described decreased PVY transmission in pepper by applying insecticides on the potential virus sources outside the field. Bradley (1963) suggested that the effectiveness of insecticides may be enhanced if they were applied with oils, also to target vectors which had encountered virus sources outside the field.

Hammond (1979) investigated the effect of 0.75% oil emulsion on the mosaic diseases caused by PVY, and PVA in potato seed plots in **Maine, USA.** He reported that the mean control of virus transmission achieved was 24.8% and 8.3% when oil was applied at weekly or fortnightly intervals respectively. Incorporating oil with insecticides increased the percentage of control to 74.5%. Raccah *et al.*, (1983) were the first to report the enhanced effects between mineral oils and pyrethroids in controlling non-persistent viruses. These authors found that mixing the pyrethroid fenprithrin (vivithrin) with the mineral oil virol produced enhanced control of CMV in the laboratory.

In **England**, Gibson and Cayley (1984) reported after that application of a mixture of the mineral oil (Sunoco 7E) and the pyrethroid insecticide, cypermethrin, provided better inhibition of virus transmission than the application of each component separately. In addition to a decrease in PVY incidence they also found that colonizing aphid density was reduced compared to the control. Electrostatic spraying of a mixture of cypermethrin and paraffin oil enhanced the deposit of chemical on the plant and decreased the spray volume required. Efficiency of the two mineral oils, Bayol 52, and SC811 in decreasing PVY spread on tobacco in the laboratory were greatly enhanced when mixed with low doses of WL85871 (Fastac, 10% ec. an enriched form of the pyrethroid cypermethrin (Gibson and Rice, 1986)

Similarly, Bell (1989) reported that 54% reduction in PVY transmission occurred in a potato crop in **England**, when potato was sprayed in a mixture of mineral oil SC811 and cypermethrin. Gibson and Cayley (1984) found that the knock-down effect of pyrethroids was not increased by mixing with oil. However, they reported that increased toxicity is the most likely reason of enhancing the effect of oils by mixing them with pyrethroids.

This kind of enhanced effect of mineral oils in improving insecticides was reported for insecticides in chemical groups other than pyrethroids. For example, in **Italy**, a new oil formulation called Biolid, characterized by a low content of aromatic hydrocarbons, was reported to be effective in decreasing PVY and CMV spread in tobacco crops. It was applied at 5 day intervals for 50 days starting from transplanting. However on days 29 and 52 post transplant a mixture of imidacloprid at 51 /ha and 90ml/ha

respectively was required for optimal control (lovieno *et al.*, 2005). More recently in **Spain**, Martin-Lopez *et al.* (2006) reported that application of mineral oil or rapeseed oil at 10 ml/litre (1% oil) in combination with a low dose of imidacloprid (1/5 the recommended concentration) decreased PVY transmission in potato fields. They found that the mineral oil /imidacloprid combination gave a 60% reduction in PVY transmission compared with a 40% decrease after using the rapeseed oil /imidacloprid mixture. Enhanced contact effect of insecticides after mixing with oil is not always consistent. This phenomenon has been reported after mixing pyrethroides and one of the new neonicotinoides (Biscaya), which has an oil dispersion formulation with 1% oil (E. Anderson unpublished). Mixing of other insecticides such as endosulfan, aldicarb, and pirimicarb with mineral oil does not enhance its effects.

Border crops are a field practice normally used in some countries, particularly the USA, to offer protection of the main crop from invading insects and diseases. Border plants, such as cereals, can be crops in their own right (Difonzo *et al.*, 1996; Hooks and Fereres (2000); Fereres (2006); Olson *et al.* (2005). However, examples of the combined effect of oil application and border crops will be discussed here.

In the **USA**, Radcliffe (2006) reported that insecticide applications in planted field borders, combined with oil applications in the field, are effective in controlling non-persistent viruses like PVY and this practice can preserve natural enemies and decrease the cost of insecticides.

In **Canada**, Boiteau *et al.* (2009) applied Superior oil (Loveland Products, Inc, 1%) at weekly intervals for 7-15 weeks on the borders of a potato field. The border itself was composed of potato cultivar Kennebec. This selective application of mineral oil provided a high rate of protection from PVY infection within the main crop. However this protection varied considerably from one year to the next. For example, the virus inhibition rates were 47-59% in 2004, 57-63% in 2005, and 79-97% in 2006.

6.5.9. Limitations of mineral oils in controlling virus spread on potato crops

6.5.9.1. Persistence in field conditions

In field conditions the effectiveness of oil at reducing virus transmission is less than that achieved in the laboratory. Weather and rainfall in particular, lead to the rapid degradation or removal of the oil from plant surface. Plant growth habit and persistence of oil is another reason for degradation of the effects of oil in the field. Plant viruses multiply more effectively in the young leaves of infected plants which are also the more favoured sites for aphid vectors to feed on. These areas develop and the surface area expands rapidly, leaving the leaf without sufficient oil coverage until the next application. In order to continually cover these areas an intensive application of oil is required (Simon et al. 1977). A second problem occurs as there is a correlation between increased oil application and yield reduction. The evidence on how frequent application of mineral oils will affect the economy of the crop is complicated. This will be a combination of cost of continuous applications and any reduction in yield. Bradley (1963) reported that mineral oil can effectively persist on plant leaves in the laboratory for up to two months, suggesting that the oil may last. However, in the literature contradictory information exists about the most effective number of applications. The overall trend though is that protection is improved by increasing the number of application (Boiteau and Singh, 1982).

6.5.9.2. Phytotoxicity and crop yield

Plant phytotoxicity resulting from applying high-doses of oil (generally more than 3%) is the most important restriction preventing oil being widely used. It is well established that oil performance increases with the increased concentration of oils applied. However, plant yield is adversely affected as at higher concentrations the oil becomes toxic to the plant (Simons and Zitter, 1980; Webb an Linda, 1993; Boiteau and Singh (1982); Ioannou and Lordanou (1987; Peters (1977); Wrobel and Arbanowicz (2005); Martin-Lopez et al. (2006) reported that there was no yield reduction or phytotoxicity observed on potato crops when sprayed 8 times with 1% of the mineral oil, Sunspray Ultrafine.

6.5.9.3. Incompatibility with other chemicals

In addition to the phytotoxic effect of mineral oils when applied at high concentrations, Simons and Zitter (1980) reported that incompatibility occurs between the JMS stylet oil and some fungicides including: sulphur, chlorothalonil (Bravo), and dichlone (Phygon). Similarly, Boiteau and Singh (1982) suggested fungicide application should be carried out at least 24 hours after oil application, as they found that under some circumstances application of fungicides directly after oil resulted in significant increases in phytotoxicity. They stated that "foliar phytotoxicity may hinder field readings by potato inspectors". This was reported for the fungicides Du-ter, Bravo, Difolatan, polyram, and Dithane. In France the use of mineral oil in conjunction with the fungicide fluazinam has also been reported as causing crop stunting by shortening of stem internodes (from a personal communication with Olivier Denufbourgh). Symptoms on oil treated potato that could be misidentified as virus and mislead field inspectors was also reported by De Bokx and Cuperus (1978). Another example was found in Cyprus by Ioannou and Lordanou (1987) who noticed a difference between the use of two mineral-oil formulations (Albolineum and Luxan Oil) where weekly application with 3% of Luxan Oil caused severe leaf scorching.

6.5.9.4. Environmental impact

Environmental impact is something to consider when evaluating oil usage, as paraffinic oil does not degrade readily in the soil and the biodegradable vegetable oil is less effective in reducing potyvirus transmission (Martin-Lopez *et al.*, 2006).

Another potential environmental impact of applying mineral oil to the potato crop is that oil could kill insects indiscriminately, and thus kill beneficial insects. This is not desirable for integrated pest management programmes (IPM). However, McLaren *et al.*, (2008) reported that oils are less harmful to natural enemies because of their relatively low toxicity compared with other pesticides. It should be noted that the use of natural enemies in seed potato crops is not an effective means of preventing virus spread.

Oil name /author	Virus/ crop Country	Dose /frequency	% Disruption of virus transmission	Phyto-toxicity or yield loss	
Oil from B43 wax (Bradley, 1962)	PVY/tobacco Canada	leaves were coated with oil	60%	N/A	
Paraffin oil, viscosity 125-135 Fisher Scientific, Montreal (Bradley, 1963)	PVY/tobacco Canada	0.13 mg/10 cm ²	> 60%	NA	
(Imperial Oil, Ltd) Bradley, 1966	PVY/potato Canada	6 applications At weekly intervals, at 0.5, 1, 2, 4, 4, and 4 gal/acre	56-88%	Occasional burning on petioles and leaflets No yield loss	
(Imperial Oil, Ltd) (Bradley, 1966)	PVY/potato Canada	6 applications At weekly intervals, the first two applications at 2 gal/acre, then the last four at 4 gal/acre	71-88%	Occasional burning on petioles and leaflets 6 application caused 15% yield loss	
Paraffin oil Atrooil QS-1540A (Esso Imperial Oil, Ltd) (Shands, 1977)	PVY/Potato Oraono, Maine, USA	1% 2.5 gal/acre 6 weekly applications	69-75%	No damage	
Refined mineral oil (Paraffin oil, N.F + 11.1% Triton-x172 ^{®4)} (Shands, 1977)	PVY/Potato Oraono, Maine, USA	1% at 1.25 gal/acre 5 weekly application	64%	No damage	
Sunoco 7E oil (Simon <i>et al</i> , 1977)	PVY/ Pepper Florida, USA	7500 PPM	PVY % after 6 days was 45% at 32°C 27.5% at 24°C 12.5% at 16°C	NA	
	PVY/PVA Seed potato Maine, USA	0.75% weekly	24.8%	NA	
Mineral oil Hammond, 1979		0.75% fortnightly	8.3%	NA	
		Accompanied by insecticides	75.4%	NA	
Sunoco 7E oil (Simons and Zitter, 1980)	PVY/Pepper Florida, USA	0.75%	Between 20-45%	No damage	
Sunoco 11E oil (Simons and Zitter, 1980)	PVY/Pepper Florida, USA	0.75%	Between 20-50 %	No damage	
		Weekly oil 0.75%	3.36%		
JMS stylet oil (Simons and Zitter, 1980)	PVY/ seed Potato	Fortnightly oil 0.75%	0.31%	No phytotoxicity	
Personal communication with D.F. Hammond	Maine, USA	Aldicarb at planting + Weekly oil at 0.75%	-0.52%	Yield loss 3%	
Eight commercial oil formulations: JMS Stylet oil, Triton B-1956 Co-op Surfactant Conc. Pfizer XA, Kornoil Agr. Adjuvant, Green Cross Booster Plus Kornoil Conc. Adjuvant Atplus 300F (Boiteau and Singh, 1982)	PVY/Potato New Brunswick, Canada	1.5-3%	35-64%	After 3% oil + When fungicides applied before 24 hours gap (could cause virus symptoms masking)	

Oil name /author	Virus/ crop Country	Dose /frequency	% Disruption of virus transmission	Phyto-toxicity or yield loss
Bayol 52Esso petroleum Plc + WL85871 (Fastac, 10% ec.) (Enriched form of Cypermethrin) (Gibson and Rice, 1986)	PVY ^O /PVY ^N potato/tobacco Laboratory England	1% oil 0.0001% WL85871	both 49% Bayol 23.5% WL85871 22.6%	NA
SC811 (97.5% e.c.) Chiltern Farm Chemical Ltd, + WL85871 (Fastac, 10% ec.) (Enriched form of Cypermethrin) (Gibson and Rice, 1986)	PVY ⁰ /PVY ^N potato/tobacco Laboratory England	1% oil 0.0001% WL85871	both 53% SC811 23.8% WL85871 11.88%	NA
SC811(Chiltern Farm Chemical Ltd,) + Cypermethrin 50 g a.i/ha (Bell, 1989)	PVY ^N /Potato England/UK	1.5% 9 l/ha 10 days intervals from emergence	34% alone 54% mixed	No
JMS stylet oil™ (Qiu and Pirone, 1989)	TEV/Tobacco Kentucky, USA	0.75%	54% inoculation 64% Acquisition Acquisition	NA
SC811 (Chiltern Farm Chemical Ltd,) (Gibson <i>et al</i> , 1989)	PVY ^O /PVY ^N /tobacco Laboratory England	0.1- 0.2 %	diminished for 30 minutes, and decreased significantly after that	NA
Suneco 7E (Powell, 1992)	PVY/ Tobacco England	1% suspension in water	25% acquisition 11.6% inoculation	NA
Mineral oil (Virol) 80% medium light oil+ 20% water emulsifiers + Reflective whitewash (Yalbin) (Marco, 1986, 1993)	PVY-Pepper CMV Israel	1% oil 10% Whitewash	Oil 37% Both 65%	Whitewash caused slight damage More harmful after mixing with oils
JMS stylet oil (JMS Flower Farms, Vero Beach, FL) Wang and Pirone (1996)	TEV/ tobacco Kentucky, USA	0.75% vol/vol emulsion in water	Significant decrease in retention	NA
Mineral oil Sunoco 7E (Powell <i>et al</i> ,1998)	PVY Tobacco England UK	1%	34.2%	NA
Biolid + imidacloprid (lovieno <i>et al.</i> , 2005)	PVY, CMV tobacco Italy	Oil Every 5 days during the 50 days after transplanting imidacloprid Two times 29, 52 days from transplanting At 5,000 ml/ha, and 90 ml/ha respectively	24.65 PVY 24% CMV	No
Sunspray Ultrafine 85% w/v Agrichem,UK paraffin oïl + imidacloprid (Martin-Lopez <i>et al.</i> , 2006)	PVY/Potato Spain	Oil 10ml/liter emulsion in water 8 times during the season imidacloprid Before sowing 350g/L 40ML/100 KG	60%	Yes After7.5-10 ml/L

Oil name /author	Virus/ crop Country	Dose /frequency	% Disruption of virus transmission	Phyto-toxicity or yield loss
Sunspray 850 EC mineral oil 98.8 % oil+ 1.2% emulsifier (Wroble, 2007)	PVY Potato in greenhouse Poland	3.75%	Four successive transmission compared to seven in the control	N/A
Sunspray 850 EC mineral oil 98.8 % oil+ 1.2% emulsifier (Wroble, 2007)	PVM /Potato in greenhouse Poland	3.75%	Two successive transmission compared to seven in the control	N/A
Aphoil (Groves <i>et al.,</i> 2008)	PVY/potato Field USA University of Wisconsin	2-4 % 2 times a week	31.9%	No significant effect
JMS Stylet oil (Groves <i>et al.,</i> 2008)	PVY/potato USA University of Wisconsin	0.75-1.5 % 1-2 times a week	29.9%	No significant effect
QRD-416 (Groves <i>et al.,</i> 2008)	PVY/potato University of Wisconsin USA	1.% 1-2 times a week	27.2%	No significant effect
Superio oil, Loveland Products, inc + crop border (Boiteau et al., 2009)	PVY/Potato Canada	1% weekly for 7-15 weeks on crop borders only	47-59% 2004 57-63% 2005 79-975 2006	N/A
Mineral oil Dc Tron (Olubayo <i>et al</i> ., 2008)	PVY, PLRV/ potato National potato Research Centre NPRC Kenya PVY ⁰	5 I a.i/ha Once a month for three months on potato in stores	Significant	NA
Superior 70 oil McLaren <i>et al.</i> , 2008 Ongoing project	PVY ⁰ PVY ^{N:0} Manitoba Canada	NA	33%	Yes But no effect on yield
Mineral oil French FNPPPT Booklet 2009	Potyviruses/pot ato France	10-15 litre/ha 15 application during season	Effective	NA
Bio-lid 80 E,W, SIPCAM Hellas (Margaritopoulos <i>et al.</i> , 2009)	PVY/tobacco Greece	Weekly application at 5g a.i./l	44.5% reduction in inoculation 40% reduction in acquisition	N/A

7. INSECTICIDES

7.1. Introduction

Chemical control of potato viruses started by chance before the exact vectors responsible for virus transmission were discovered. In Broadbent's review (1957) named 'Insecticidal control of plant viruses' he described early attempts to control plant viruses chemically. According to that review, in 1931 a mixture of lead arsenate, nicotine sulphate, and Bordeaux mixture was effective in decreasing the incidence of PLRV. Nicotine was not effective in controlling potato viruses but it did succeed in increasing potato yield because of preventing direct insect damage. The failure of nicotine to prevent potato virus spread occurred when nicotine was sprayed, dusted or fumigated. Dusting or spraying with the natural plant product derris (rotenone) also failed to control PLRV spread.

A second stage in controlling plant viruses started with the introduction of the contact persistent insecticides. DDT and parathion belonged to this new type of insecticide and they created a revolution in the field of pest control. DDT quickly replaced all the previous insecticides to control potato pests as concluded from Hill's (1948) paper which reviewed 108 publications on this subject within a three year period. Broadbent (1957) concluded that most of the effort on the potato crop was directed at controlling persistent viruses and PLRV in particular. The general conclusion was that persistent viruses can be controlled using contact insecticides if the infection was within the field. However, protection from PLRV failed when infected aphids arrived from outwith the field. Despite the consistent results in controlling persistent viruses by contact insecticides, there was variation in results reported about non-persistent viruses between different researchers. For example, in one report, Parathion failed to protect potato from PVY infection even if sprayed at weekly intervals (Broadbent, 1957). On the other hand, in a second report, moderate inhibition of PVY transmission was achieved by using DDT, Endrin, and parathion (Broadbent *et al.*, 1956)

In a third report, PVY and PVA spread were not decreased by DDT and parathion when applied at 2- and 4-day intervals respectively Broadbent (1957).

This conflict between results could be explained as a result of experimental and environmental variability. This includes weather conditions, vector pressure, virus source, experimental design, and the statistical methods used to analyse the results.

A third stage in controlling plant viruses commenced with new products and changes in the way that insecticides could be delivered to the target. In the 1960s two new chemical groups: organophosphates and carbamates were introduced at the same time as new insecticide products that could gain entry into the insect through the plant. Schradan and Demeton (organophosphates) were the first of these insecticides used to control potato viruses. The characteristics of these newly developed insecticides included: systemic and longer activity and they were effective in controlling the persistent viruses such as PLRV. However, progress in producing new insecticides had less impact on the incidence of non-persistent viruses. This is because the majority of these new insecticides still did not act quickly enough to stop aphids from probing and thus control viruses transmitted in this manner (Broadbent, 1957; Woodford *et al.*, 1983).

Mineral oils were the first chemicals which provided reasonably consistent results in controlling non-persistent viruses in crops. These remained the best method until

the discovery of the **synthetic pyrethroids** in the 1970s, and their effects in suppressing non-persistent viruses when applied alone or in combination with mineral oils (Gibson *et al.* 1982; 1983; Gibson, 1983; Gibson and Campbell, 1986; Gibson and Rice, 1986; Gibson and Cayley, 1984). This new class of insecticides was used extensively after the 1980s to control PVY spread. However, there are some considerations regarding their most effective use for example, their repellent effect may increase virus spread especially when applied at sub-lethal concentration (Robert *et al,* 2000). Pyrethroids are also more economically effective when mixed with mineral oil as their effectiveness is considerably enhanced (Gibson and Rice, 1989).

Currently, additional new insecticides belonging to different chemical groups have been introduced and their effect on virus transmission is still being fully investigated. This includes imidacloprid, thiacloprid, *and* acetamiprid which belong to a new chemical group called neonicotinoids. Pymetrozine and Flonicamid are additional new insecticides belonging to the pyridine and pyridinecarboxamide groups respectively.

7.2. Synthetic insecticides

7.2.1. Background

Direct damage of insect attack on crops can be controlled using insecticides to keep the insect population below damaging threshold. However, the indirect damage caused by vectoring plant viruses is more difficult to prevent. This is because the aphid vectors can come from a variety of sources both winged and wingless and from within and without the crop (Mathew, 1991).

Insecticides have been used extensively to control vector-born plant viruses, and the epidemic spread of persistent viruses like PLRV is normally controlled by chemical application of newly developed systemic insecticides. Unlike the conventional contact insecticides, systemic insecticides provide consistent and long lasting protection from persistent viruses through their transport via the plant phloem system.

Non-persistent viruses on the other hand are also transmitted by phloem feeding insects, but they multiply in the epidermis of the plant. In this case the vectors acquire the virus by briefly puncturing cells *en route* to the phloem or while assessing whether the plant is a suitable host. In these situations the insects may not probe deeply enough to reach the phloem system, avoiding large doses of systemic insecticides. Moreover, the accumulation of non-persistent virus in the epidermis of the plant tissue make the transmission feasible by non-colonizing aphid species, whereas the transmission of phloem restricted viruses is limited to a limited number of colonizing aphid species. Indeed, more than 40 aphid species have been reported to transmit PVY in potato crops, but only a few of them are potato colonizers (see Table 1). Interestingly, the non-colonizing aphids are believed to be responsible for spreading early infection despite being very poor vectors in laboratory conditions. For example, in the Netherlands Van Hoof (1977) found that 100% infection with PVY happened before *M. persicae* migrate, which indicated that other vectors are responsible for that and in particular R. padi. In Sweden Sigvald (2007) reported that non-colonizing aphid species are far more important in PVY transmission compared with *M. persicae*, and data from suction traps indicated that R. padi is the most common. In Scotland, SAC information notes suggest that aphid management should start early to prevent the non-colonizers aphids from transmitting viruses to seed potato crops and in particular *R. padi* which are caught in traps from the first sampling date.

Collectively, in order to control non persistent viruses, a very fast acting pesticide is required, or aphid's probing behaviour should be prevented. Irwin (2000) reported that insecticides which decrease the aphid's probing behaviour are the best candidates to control non-persistent viruses.

Unfortunately, the older classes of contact pesticides act slowly. Persistent organophosphorus (OP) and carbamates and their systemic products were of limited use because of the virus accumulation in the plant epidermis and the non-colonizing behaviour of the majority of the aphid vectors of non-persistent viruses. Consequently, most of the attempts to decrease transmission of non-persistent failed when using synthetic insecticides. The only exception was pyrethroids which is one of the most important classes of new pesticides which act quickly. But, their fast mode of action was generally insufficient to prevent or decrease transmission of potyviruses alone.

Laboratory studies suggest that the antifeeding effects of the new insecticides belonging to the neonicotinoid and pyridine groups could have a future in controlling non-persistent virus transmission. However more investigation is required before validating such products in virus control programmes on potato.

A summary of all the literature referring to insecticide use in preventing potyvirus spread can be found in Tables 3 and 4.

7.2.2. Organophosphate and Carbamate insecticides and their effectiveness in controlling non-persistent virus spread in potato

Despite the great success of organophosphate and carbamate insecticides in controlling PLRV spread in potato crops, there are many reports of failures in controlling PVY in potato crops (Broadbent *et al*, 1956; Burt *et al*, 1960, 1964; Gibson *et al*, 1982; Bell, 1989). However, there are some reports where limited but inconsistent success in suppressing viruses transmitted non-persistently on potato crops have been reported.

7.2.2.1. Methamidophos

Methamidophos is an organophosphorous insecticide, used extensively for controlling vectoring of PLRV in potato crops with great success. However, it has recently been reported that methamidaophos efficiency is very limited when compared with neonicotinoid insecticides (Mowry, 2005).

Van Toor *et al*, (2009) reported that both calendar and targeted methamidophos treatments in potato crops in New Zealand were effective in decreasing aphid populations to negligible level. However, they concluded there was no impact of this insecticide on virus incidence in potato tubers, but this may have indicated either that the insecticide is ineffective or there was a high level of virus inoculum in the seed potatoes used for their experiment.

Combined effect of insecticide application on crop edges were investigated by Carroll *et al.* (2004), they tried to estimate the effect of targeting field borders only with methamidophos to control *M. persicae* population. Their results indicate that *M. persicae*, which is the main vector for PLRV, was controlled by the application of spray only to field borders. However, they used final seed certification as a measure of success. They achieved a 15 times reduction in the cost of pesticide application using this approach.

7.2.2.2. Pirimicarb

In the UK, the only carbamate insecticide, which is still recommended for use on potato crops is pirimicarb. However, *M. persicae* resistant to this insecticide are reported to be widespread (Fenton *et al.*, 2010), which will limit its future use. In addition, pirimicarb was reported to have no effect in suppressing the spread of the non-persistent viruses on potato, and the beneficial characteristics of pyrethroid insecticides in suppressing non-persistent virus spread appears to be diminished when these insecticides are used in combination with pirimicarb (Eric Anderson, personal communication).

7.2.2.3. Other organophosphates and carbamates

Application of insecticides in soil to control plant virus spread in potato crops has been investigated. Burt *et al* (1960) found that Rogor and Thimet applied at planting were effective in decreasing PLRV spread, but only slightly decreased PVY spread.

Virus transmission inside potato seed stores has been reported in Kenya where the organophosphate dimethoate was effective in decreasing the spread of PLRV and PVY (Olubayo *et al.*, 2008).

7.2.3. Pyrethroid insecticides and their effectiveness in controlling non-persistent virus spread in potato.

Pyrethroids are different from the other two groups of insecticides in terms of mode of action, and efficiency of non-persistent virus-vector control. They are fast acting insecticides (knockdown effect). While pyrethroids decrease aphid transmission of non-persistently transmitted viruses alone, when combined with mineral oil the performance is enhanced considerably (Gibson *et al.*, 1982). This combined effect of pyrethroids with oils will be discussed under another section in this review. While pyrethroids were the only class of insecticides to show inhibition of non-persistent virus transmission, their repellent effect is sometimes believed to cause more virus spread by increasing aphid flights and probing activities (Gibson and Rice., 1989)

7.2.3.1. Deltamethrin

Deltamethrin is one of the most studied and widely used pyrethroids to control aphid spread of viruses on potato crops. Highwood (1980) demonstrated that pyrethroids are effective in decreasing virus spread in the field. Gibson *et al*, (1982) investigated this phenomenon in the laboratory, and reported that PVY spread can be controlled after application of deltamethrin. They attributed this effect to the incapacitation of the aphid vector after exposure to the insecticide which may lead to infectivity being lost. Alternatively, the repellent effect of the compound could prevent acquisition and/or inoculation. In another study by the same group, Rice *et al.* (1983) found that deltamethrin was still effective in controlling PVY transmission by pyrethroid- resistant clones of *M. persicae* on potato crops in England.

7.2.3.2. Cypermethrin

It has been reported that the pyrethroid, cypermethrin, decreased non-persistent virus spread in potato crops possibly by acting on the aphid vector in a similar way to deltamethrin. Gibson and Rice (1986) reported that the enriched form of cypermethrin (WL85871) was effective in diminishing PVY acquisition by 23.8% compared with the untreated control.

In an attempt to investigate the effects of insecticides on aphid probing behaviour and the ability to transmit PVY, Collar *et al.* (1997) used the EPG technique (Tjallingii, 1978). They found that both probing behaviour and transmission efficiency were affected when cypermethrin was applied and aphids allowed 10 minutes to feed (acquisition access period, AAP). Collar et al. (1997) treated a virus source with cypermethrin and investigated the consequences for aphid behaviour and PVY transmission. They found that aphids were paralysed after 2.5 min exposure to infected treated source, thus virus inoculation was prevented when AAP was more than 2.5 min. In addition, stylet penetration was shorter and less frequent on insecticide treated leaves, and this observation agrees with the other reports about aphid probing behaviour related to insecticides treatment (Atiri *et al.*,1987; Lowery and Boiteau, 1988). Cypermethrin's performance in decreasing virus spread like other pyrethroids is enhanced by mixing with oil (Gibson *et al.*, 1984; Bell, 1989).

7.2.3.3. Lambda-cyhalothrin

Lambda-cyhalothrin is the most recently developed pyrethroid. It is marketed in different countries around the world under different commercial names. It is the most widely used contact fast acting insecticide on potato crops. Recently it has been used as a tank mix with systemic neonicotinoid insecticides, and prior to this as a formulated mixture with primicarb.

Lambda-cyhalothrin is widely recommended for use in virus control programmes in the UK, USA, and New Zealand. There are some studies that are currently investigating the repellent effect of this insecticide on the actual levels of PVY transmission and preliminary results support the assumption that this disrupts transmission. However, further investigations are required before making a final conclusion.

Lambda-cyhalothrin was reported to be effective in limiting potyviruses spread in brassica crops (Bedford *et al*, 1998).

7.2.3.4. Other Pyrethroids

Bifenthrin was reported to decrease the spread of PVY and PLRV in potato stores (Olubayo *et al.*, 2008).

7.2.4. Insecticides belonging to new classes and their effectiveness in controlling non-persistent virus spread in potato

The neonicotinoid insecticides were introduced recently to pest management programmes. This group includes some of the most promising insecticides for controlling vectors of plant viruses. Imidacloprid, thiacloprid, thiamethoxam, and acetamiprid are currently being evaluated for their effect on disrupting virus transmission. Pyridine azomethine is another new class of insecticides, with pymetrozine as the compound used in formulations.

7.2.4.1. Imidacloprid

Imidacloprid belongs to a new class of insecticides called chloronicotinyl nitroguanidines or neonicotinoids. Insecticides belonging to this group target the nicotinic acetylcholine receptor. It is a systemic insecticide and acts through ingestion and contact. Its use has expanded rapidly due to its low human toxicicity. Imidacloprid is the first insecticide developed from this group, and the most studied in respect of virus transmission. It has been reported to be effective in decreasing transmission of many, mainly persistent, plant viruses vectored by different species of insect vector. On potato crops imidacloprid is effective in controlling the persistently transmitted PLRV (Boiteau and Singh, 1999; Mowry, 2005). There are also some reports about antifeeding effects of low concentrations of imidacloprid on aphid behaviour (Nauen, 1995). However; there are only a few reports about the effectiveness of neonicotinoids in suppressing PVY spread. Imidacloprid was reported to be ineffective in reducing PVY spread in potato crops in Canada when applied in soil at planting followed by two foliar treatments after mid July (Boiteau and Singh, 1999). Alyokhin et al. (2002) decreased the imidacloprid rate applied to potato crop by applying insecticide as foliage instead of furrow application. They found that the later application gave better control of the aphid itself; however, the PVY transmission was not decreased by such treatment. On the other hand, when they compared in furrow treatment to foliage treatment, they found that the latter decreased virus spread significantly, although the foliar application was applied at a lower rate. Efficacy of imidacloprid was greatly enhanced when mixed with mineral oil (Martin-Lopez *et al.*, 2006). They reported that mixing imidacloprid with oil might enhance the contact action and the persistence of insecticide as happens with the toxicity of pyrethroids when mixed with mineral oils. The mixture diminished the aphid population after 16 hours of application and the combination of mineral oil and imidacloprid achieved a 59.3% decrease in PVY spread compared with the imidacloprid only.

Van Toor *et al.* (2009), reported that seed treatment with imidacloprid alone or with a combination of fortnightly sprays of either methamidophos or lambda cyhalothrin were not effective in decreasing PVY infection, although the levels of virus in their controls were very low (0.4%) and they were not sure of the levels of virus in their seed. Collar *et al.* (1997) investigated the probing behaviour of *M. persicae* after a 10 min exposure to insecticide-treated pepper leaf. His results indicated that imidacloprid did not have a significant affect on aphid probing behaviour or on the transmission efficiency. In addition, there is some evidence of negative interacting effects of imidacloprid with other insecticides on PVY transmission. For example, PVY spread was 2.2% enhanced after application of cypermethrin on a potato crop that emerged from imidacloprid-treated seed (Martin-Lopez et al, 2006).

7.2.4.2. Thiacloprid

Thiacloprid is the second most common neonicotinoid which has shown promise for decreasing non-persistent virus transmission. Biscaya, the forumulation with thiacloprid as the active ingredient, was reported to be effective in decreasing the spread of PVY, PVA, and PLRV when tank mixed with Hallmark Zeon at the first and the third spray in virus control programmes (Farmers Guardian, 2009). A spray programme of Hallmark Zeon (lambda-cyhalothrin) + pymetrozine (Plenum) alternating with Hallmark Zeon+ Aphox (Pirimicarb) for the first six sprays then Hallmark Zeon+ Aphox (Pirimicarb) for two sprays resulted in 5.5% PVY incidence in daughter tubers compared to 8% in the untreated control. However, PVY incidence in daughter tubers was decreased to 0.5 % when Biscaya was substituted at the first and the third spray in the above spray programme (Farmers Guardian, 2009).

7.2.4.3. Thiamethoxam

Thiamethoxam (Actara) is another new neonicotinoid insecticide commercialized recently. Actara can be applied in soil or as a foliar spray and Cruiser is another marketed formulation, but for oilseed rape seed treatment only (Maienfisch *et al.*, 2001). In a recent investigation by Daniels *et al.*, (2009), it was reported that Thiamethoxam affects the feeding behaviour of the cereal aphid *R. padi* when applied at sub-lethal dose on wheat crops. Starved aphids subjected to a sub-lethal dose of this insecticide showed feeding impairment that is associated with decrease in body water content, development, and fecundity. They speculated that this pesticide may have antifeeding effects when applied at low doses, and they recommended further studies to investigate the effect of sub-lethal doses on virus transmission.

7.2.4.4. Other neonicotinoids

Acetamiprid (InSyst) is one of the new neonicotinoid insecticides introduced recently and incorporated in vector control programmes in the UK. Clothianidin is another new neonicotinoid which interferes with the aphid feeding behaviour and decreases PVY transmission (Nolte *et al*, 2009)

7.2.5. New non-neonicotinoid chemicals

7.2.5.1. Pymetrozine

Pymetrozine belongs to a new class of insecticides (pyridine azomethine) and has been available since the 1990s. It has antifeeding properties, and kills insect by starvation, but it mode of action is still unclear. Pymetrozine has been widely used in potato crops to decrease PLRV spread, and it has some effects on decreasing PVY transmission in laboratory conditions. Harrewijn and Piron (1994) found that pymetrozine affected PVY acquisition but not transmission. Similar results were recently reported by Davis *et al.* (2009).

By employing the EPG technique, Harrewijn (1997), found that at high does pymetrozine prevented the aphids from inserting their stylets, while at lower doses the aphids recovered and reinserted their stylets. In addition to decreasing PVY spread, pymetrozine was reported to suppress transmission of other viruses by its antifeeding properties. This includes viruses transmitted persistently by aphids on potato (Harrewijin and Piron, 1994; Mowry, 2005) or persistently and semi-persistently by the same type of vector or other vectors on other crops (Castle *et al.*, 2009; Bedford *et al.*, 1998).

However, Van Toor *et al.* (2009) reported that pymetrozine did not significantly decrease PVY or even PLRV incidence in harvested potato tubers in New Zealand. Their results were similar whether they followed calendar spray programmes or when applied after the number of caught aphids in the traps reached certain thresholds. This conflict between the finding of the previous authors and the many successful examples led them to speculate that virus contamination of seed potato used in their experiments may be high. Indeed their results indicate that their untreated control potatoes sometimes had more virus than their treated controls. This observation correlates well with Robert *et al* (2000) who reported that initial virus inoculum is a substantial factor in determining the transmission of aphid-born viruses spread.

In addition, pymetrozine is reported to be effective in decreasing PVY spread on crops other than potato. Margaritopoulos *et al.*, (2009) reported that pymetrozine, which has been used since 1990 in Greece, is effective in decreasing PVY spread in tobacco crop. They found that pymetrozine can decrease both acquisition and transmission compared with the control and the protection that is offered by these pesticides is comparable with that of light mineral oil. Pymetrozine was also effective in decreasing semi-persistent viruses (Bedford *et al*, 1998), and the persistent PLRV (Mowry *et al.*, 2005).

7.2.5.2. Flonicamid

Flonicamid is a novel, selective insecticide belonging to the pyridinecarboxamide group used for control of homopterous pests. It was recently commercialized worldwide although it is not available for farmers in some countries yet (Van Toor *et al.*, 2009). In the UK, it is incorporated into aphid and virus control programmes on potato crops. The compound has a strong feeding inhibition effect and kills aphids by starvation (Morita *et al*, 2007). The effects of this new insecticide on virus transmission have yet to be revealed. However, it seems that its antifeeding properties are similar to pymetrozine and by analogy it may be useful in decreasing non-persistent virus spread in potato.

7.2.5.3. Spirotetramat

Spirotetramat (Movento) is a new insecticides belonging to the tetronic acid group, and is reported to decrease PVY transmission in American potato fields Nolte *et al*, (2009).

7.2.6. Other chemical compounds which affect non-persistent virus spread on potato crops

In addition to the synthetic insecticides and mineral oils, there some synthetic chemical which showed some influence of virus transmission in potato crops.

Bradley and Ganong (1957) found that treating tobacco with thouracil or trichothecin at 500ppm, and 10ppm respectively, inhibited aphid from transmitting PVY to treated plants. However, these two chemical caused damage to plant at these concentrations. in further studies Bradley found that trichothecin induced more than 50% reduction in virus transmission when applied two days before or four hours after aphid transmission. Bradley and MacKinnon (1958) found that trichothecin prevented systemic infection by many viruses including PVY, however, it had no effect on PLRV. The repellent effect of the dodecanoic acid and polygodial on the transmission of some plant viruses by aphids was reported by Gibson *et al.*, (1982). Polygodial decreased the acquisition of PVY; however, dodecanoic acid increased virus acquisition. Powell *et al.* (1996) also investigated the repellent effects of polygodial on stylet penetration behaviour and non-persistent transmission of plant viruses by aphids and concluded that it had not effect on stylet penetration and subsequent transmission of PVY or CMV by aphids.

Aphid alarm pheromones were reported to affect aphid settling behaviour and their capacity to transmit plant viruses (Dawson et al, 1982). Gibson *et al.* (1984) also tested the effects of some derivatives of the aphid alarm pheromones in inhibition PVY transmission. They found that (E)-b-farnesene and another saturated ester of acetylene dicarboxylic acid decreased virus transmission.

7.3. Current programmes recommended for control of virus spread on potato crops around the world

7.3.1. UK

In the UK, all the current virus control programmes on potato crops were designed to control PLRV spread, and there are no specific programmes to control non-persistent virus transmission (Eric Anderson pers comm). Parker *et al* (2006) reported that the insecticide use of the potato industry in the UK is largely dominated *by* the carbamate insecticide pirimicarb and pymetrozine which belongs to the pyridine azomethine group. In addition, pyrethroid insecticides are used, alone and in a formulated mixture with pirimicarb. Chemical programmes to control virus spread in potato crops are quite similar between different parts of the UK. However, it has been separated to the following:

In England, PVY, and PVA are an increasing problem. Fera currently runs the Potato Council aphid monitoring and forecasting programme using about 100 yellow water traps located in the main potato production areas. Information about aphid flights is presented on this website: <u>http://aphmon.fera.defra.gov.uk</u>. Information at the website is updated twice a day, and it can provide up-to date information for seed potato producers about aphid flights. In addition, farmers can sign up to receive weekly information, and email alerts when the number of aphids requires chemical spray in a certain region.

In addition, to water traps, Rothamsted Research and Scientific Advice for Scottish Agriculture (SASA) are running a network forecasting system based on 16 suction traps in different sites of the UK.

In England the most recent recommended procedure is to use tank mixes to avoid insecticides resistant *M. persicae*. The main component of the mixture should be rapid knock-down pyrethroid insecticides, and the second component one of the insecticides: pirimicarb, thiacloprid, thiamethoxam, pymetrozine, flonicamid, or acetamiprid. It is recommended to apply one of the active systemic insecticides after using pirimicarb as resistance has been reported. However, no more than two applications of the new neonicotinoids (thiacloprid, thiamethoxam, or acetamiprid) should be applied.

In Scotland, SASA monitors four suction traps in different locations in Scotland. They are used for aphid and virus forecasting (Pickup and Brewer, 1994) YWTs are also used by growers at a local level. SASA research indicates that there is a correlation between PVY spread and the number of non-colonizing aphid species and in particular *R. padi, M. dirhodum*, and *B. helechrysi*. Virus control programmes normally start after aphid being caught in the traps (source: information notes, SAC).

In Scotland, SAC's most recent advice recommends that tank mixtures of two distinct chemical groups are used in order to avoid insecticide resistant *M. persicae*. Therefore, lambda-cyhalothrin is recommended for use with one of the systemic insecticides, pirimicarb, pymetrozine, thiacloprid, flonicamid, and acetamiprid. The last four insecticides are used to combat insecticide resistance as there are no local reports of resistance to these insecticides.

The time of application in seed potato fields should start at emergence where PVA susceptible cultivars are being grown (Desiree, Estima, Hermes, Golden Wonder, Marfona, Kerr's Pink, Red Pontiac, and Russet Burbank). For the first three treatments it is advised to use formulated mixtures of the Lambda-cyhalothrin with Pirimicarb (Dovetail, Claytin Groove and Mortice). After potato aphids appear tank mixtures of lambda-cyhalothrin with the already mentioned systemic insecticides are recommended.

Treatment should be repeated every 7-10 days, and the systemic insecticide component of the mixture should be alternated to eliminate any chance of resistance development.

On Scottish ware potatoes, insecticide sprays are not recommended unless aphid population are large. When a treatment is required it is recommended to use any of the insecticides used on seed potato (source: SAC website, Information notes).

In Northern Ireland, PVY is also the most widespread potato virus. In order to control PVY spread in seed crops on susceptible Irish potato varieties such as Premiere, Maris Piper, Cultra, and Kerr's Pink, it is recommended to start chemical spray with insecticides after 80% emergence of the crop, then application should be repeated fortnightly until August or three weeks before desiccation. To avoid insecticide resistant *M. persicae* it is recommended to tank mix a pyrethroid such as Dovetail or Hallmark Zeon with one of the translaminar products with one of the systemic insecticides (pirimicarb, thiacloprid, thiamethoxam, pymetrozine, and flonicamid). However, thiamethoxam and pymetrozine should be avoided during the flowering period due to their harmful effect on bees.

On ware potato, aphid transmission of potato viruses is less important. It is recommended to monitor aphid population on weekly basis and to start spraying when aphid population reaches five aphids per leaf (source: Aphid control in potatoes, Irish Department of Agriculture and Rural Development, 2008).

7.3.2. USA

Nolte *et al.*, (2009) reported that among the commonly used insecticides on potato crops in Idaho, there are some which decrease PVY spread. This includes clothianidin (Belay), imidacloprid (Admire Pro), imidacloprid (Provado), pymetrozine (Fulfill), and spirotetramat (Movento). Insecticides are grouped according to their mode of actions, and farmers are recommended to avoid using insecticides from the same group in succession to avoid resistance problems.

7.3.3. France

To control virus spread on susceptible potato cultivars in France 12 insecticide applications are recommended in conjunction with mineral oils. Insecticide sprays start after 80 % emergence (3 leaves) at weekly intervals, and stop when the plants start to senesce. It is recommended that the first and the last two sprays contain esfenvalerate or lambda-cyhalothrin. During the second, third, seventh, and eighth, it is recommended to use one of these three combinations lambda-cyhalothrin + carbamate, esfenvalerate + fenitrothion, or chlorpyriphosethyl+ cypermethrin. Insecticides classified as antifeedant (pymetrozine, flonicamid) are recommended during the third, fourth, fifth, and sixth applications.

7.3.4. Spain

Martin-Lopez *et al.* (2006) recommended that treatment of seed potatoes with imidacloprid followed by mineral oil application during a season is effective in decreasing PVY incidence to a controllable level.

7.4. Current projects which investigate the chemical control of non-resident viruses by insecticides in potato crops

A project entitled "Evaluation of the chemical management, inoculum sources and aphid vectors for potato virus Y" is currently running in Wapato, Washington by the United States Department of Agriculture. It is investigating the effect of different insecticide treatments on the transmission of PVY by *M. persicae* and *R. padi.* There are three related projects: 1. Improved Diagnostics and Management Strategies for Potato Viruses. 2. Strategies to Manage Potato Virus Y and Eradicate the Tuber Necrotic Variants Recently Introduced into the U.S. 3.Management of Nematodes and Virus Diseases Affecting Potato and Grain Crops.

Research is also ongoing in Idaho, to investigate which of the currently used insecticides in potato crops are effective in achieving control of aphids and limit PVY spread in the field (Nolte *et al.,* 2009). In Colorado, USA, another more comprehensive project called "Improved potato yield and quality through disease suppression and optimum certified seed potato production" is currently ongoing. This project involves using chemical insecticide control to suppress PLRV, PVY, and PVS transmission.

In the UK, Syngenta is currently investigating the repellent effect of lambda-cyhalothrin in decreasing PVY transmission on potato crops (Tait, Farmers Weekly, 2009).

7.5. Limitations of synthetic insecticides in controlling nonpersistent viruses on potato crops

7.5.1. Effectiveness of synthetic insecticides in controlling nonpersistent viruses

As discussed above, neither the older conventional contact insecticides nor systemic organophosphorus (OP) and carbamate insecticides were effective in controlling viruses transmitted in a non-circulative manner. There are a lot of early reports which showed PVY control failure in potato crops (Broadbent *et al.*, 1956; Burt *et al.*, 1960; 1964; Webley and Stone, 1972; Reagan *et al.*, 1979).

Pyrethroids are the only class of insecticides which showed some degree of virus control. However, their effect alone was not sufficient to completely eliminate viral spread and thus they should be applied with mineral oil to enhance their efficacy.

The effect of newer classes of insecticides on non-persistent virus transmission is promising. However, further studies will continue to be helpful in evaluating their efficacy in the field in order to refine aphid control programmes on potato crop.

Perring *et al*, (1999) reported that insecticides are normally less efficient in controlling the spread of infection of plant viruses from point sources within a field in general and non-persistent viruses in particular. Targeting non-colonizing aphids outside the field by applying insecticides on the plant harbouring the vectors or external virus is more

important to control the seasonal spread of virus. But application of the insecticides outside the target field is less feasible.

Van Toor *et al.* (2009) reported that five chemical applications were not significantly effective in reducing the tuber infection with PLRV or PVY or increased potato yield in New Zealand. These chemical applications were methamidophose applied fortnightly as a foliage spray to plant emerged from imidacloprid treated and untreated tubers, lambda cyhalothrin, when aphids exceed the threshold (10 per/150) on potato plants emerged form imidacloprid treated or untreated tubers, pymetrozine, when aphids exceed the threshold (10 per/150) on potato plants emerged from imidacloprid treated or untreated tubers, pymetrozine, when aphids exceed the threshold (10 per/150) on potato plants emerged from imidacloprid free tubers. However, they recommended that seed treatment of imidacloprid, and foliar spray of lambda-cyhalothrin or pymetrozine when alate aphids exceeds 10 per/150 potato leaves is effective in keeping aphid population and virus damage under threshold.

In conclusion, the reported reliability of insecticides in controlling non-persistent virus spread in potato field was mixed (see reviews by Broadbent, 1957; Perring *et al.*, 1999; Robert *et al*, 2000; Irwin *et al*, 2000, Radcliffe and Ragsdale, 2002). The reason for that is possibly the very short acquisition or inoculation required for aphid vector to transmit such viruses (Perring *et al.*, 1999), which enable aphids to acquire or inoculate virus before many insecticides can fully act (Gibson and Rice, 1989). This does highlight a major difference between field and laboratory studies. In the laboratory an experimenter will transfer the aphids, speeding up the process, whereas in the field the aphids feed on an infected plant and then must survive long enough to feed on a second uninfected plant.

7.5.2. Insecticide resistance

Sustainability in insecticides is an important objective when designing virus control programmes. This is because of the capability of virus vectors to develop resistance to certain groups of pesticides when applied frequently for a long time and the harmful effect of the insecticides on the natural enemies and the environment in general. There are limited biochemical targets in the insect to be attacked by insecticides. For example, Harrington *et al.* (1989) noticed that repeated application of cypermethrin with mineral oil caused increases of insecticide resistant *M. persicae* in treated potato fields. Unfortunately, many insecticides share a similar mode of action as they act on the nerve systems. In order to decrease resistance development in aphid population to insecticides, application of insecticides belongs to different groups should be alternated. This problem can be overcome by using an integrated programme.

7.5.3. Incompatibility between insecticides used for virus control

Suppression of the spread of the non-persistent viruses by insecticides is not always achievable and can even be increased by their application due to the increase in vector activity and their destructive effects on natural enemies (Gibson and Rice, 1989).

Insecticides which are effective in controlling persistent viruses are not necessarily effective for non-persistent viruses. This phenomenon is of great importance when designing chemical control programmes on potato to avoid unintended virus dissemination in potato fields. It was reported that at a high level of virus infection in small scale potato production, application of parathion or shradan increased virus spread. It is possible that the sprayed plants were stressed and attracted more aphids. In some circumstances DDT and parathion increased the incidence of PVY and PVA in potato fields (Broadbent, 1957). It was suggested that the insecticide application prolonged the life of the plant by decreasing the insect damage. However, aphids can carry infections from neighbouring infected unsprayed potatoes which senesce before those which have been sprayed with insecticides (Broadbent, 1956)

Incompatibility of mineral oils with insecticides has also been observed. From a personal communication with D.F. Hammond (Maine, USA), Simons and Zitter (1980) reported that JMS stylet oil applications slightly increased PVY spread when the insecticide aldicarb (Temik) was applied in soil at planting. In another report by Zitter and Simon (1980) they mentioned that although aldicarb is effective in controlling PLRV in potato fields, it caused an increase in PVY incidence compared with untreated plots.

Shanks *et al.* (1965) reported that PVY transmission by viruliferous aphids was higher when the recipient tobacco plants were sprayed with parathion. This enhanced effect became even greater when both virus source and target plants were sprayed with the insecticide.

Martin-Lopez *et al.* (2006) found that a control programme starting with treating potato seeds before sowing with imidacloprid, and subsequently spraying with cypermethrin 8 times during a season lead to a 2.2% increase in PVY incidence in potato crops in Spain.

7.5.4. Environmental damage

One of the most important considerations on the use of synthetic insecticides is their potentially harmful effects on bees and natural enemies. In addition, they can have residual effect on crops and pollute the environment. However, the selective nature of the newly synthesized insecticides (neonicotinoids, pymetrozine, and flonicamid), makes them applicable to IPM programmes, as they affect selectively phloem sucking insects and cause minor effect on the natural enemies (Ishaaya, *et al.*, 2007). Natural enemies are not used in the production of high quality seed.

7.6. Examples of papers examining the effects of insecticides on controlling the spread non-persistent viruses on potato crops

			Field/		
Insecticide	Chemical group	Virus/ plant	laboratory	Efficiency	Author
Deltamethrin (Decis, 2.5% e.c.: Procida, France)	Pyrethroids	PVY/Tobacco England	Greenhouse	Effective	Gibson <i>et al</i> , 1982
RU-15525 (Kadethrin,10% e.c. Wellcome Research Laboratories, UK)	Pyrethroids	PVY/Tobacco England	Greenhouse	Effective	Gibson <i>et al</i> , 1982
Deltamethrin (Decis, 2.5% e.c.: Procida, France)	Pyrethroids	PVY/Tobacco England	Laboratory	Effective	Rice <i>et a</i> l. 1983
WL85871 (enriched form of Cypermethrin)	Pyrethroids	PVY/Tobacco England	Laboratory	Effective	Gibson and Rice,1986
Imidacloprid	Neonicotinoids	PVY/potato Canada	Field	Not effective	Boiteau and Singh, 1999
Imidacloprid	Neonicotinoids	PVY/potato Canada	Field	Effective	Alyokhin <i>et al</i> , 2002
Methamidophos (Monitor)	Organophosphates	PVY/Potato USA	Field Borders only	Effective	Carrol <i>et al</i> ., 2003
Bifenthrine	Pyrethroids	PLRV and PVY/ potato Kenya	Stores	Effective	Olubayo <i>et al</i> ., 2008
Dimethoate	Organophosphates	PLRV and PVY/ potato Kenya		Effective	Olubayo <i>et al</i> ., 2008
lambda-cyhalothrin (Karate Zeon technology)	Pyrethroids	PVY/Potato New Zealand	Field	Not effective	Van Toor <i>et al.</i> 2009
Methamidophose (Tamaron)	Organophosphates	PVY/Potato New Zealand	Field	Not significant	Van Toor <i>et al.</i> 2009
Pymetrozine (Chess WG)	Pyridines	PVY/Potato New Zealand	Field	Not significant	Van Toor <i>et al.</i> 2009
Pymetrozine (Plenum 50 WG, Syngenta)	Pyridines	PVY/tobacco Greece	Laboratory	Effective	Margaritopoul os <i>et al</i> , 2009
Clothianidin (Belay)	Neonicotinoids	PVY/Potato USA	Field	Effective	Nolte <i>et al,</i> 2009
Pymetrozine (Fulfill)	Pyridines	PVY/Potato USA	Field	Effective	Nolte <i>et al,</i> 2009
Imidacloprid (Provado),	Neonicotinoids	PVY/Potato USA	Field	Effective	Nolte <i>et al,</i> 2009
Spirotetramat (Movento ⁾	Tetronic acid	PVY/Potato USA	Field	Effective	Nolte <i>et al,</i> 2009

TABLE 3 COMMON INSECTICIDES FOR CONTROL OF NON-PERSISTENT VIRUS SPREAD IN POTATO CROPS

TABLE 4 COMMON INSECTICIDE MIXTURES FOR CONTROL OF NON-PERSISTENT VIRUS SPREAD IN POTATO CROPS

Insecticide mixture	Chemical group	Virus/ crop	Dose /frequency	Effects	Author
Imidacloprid + Cypermethrin	Neonicotinoid + Pyrethroid	PVY/Potato Spain	Imidacloprid Before sowing 350g/L 40ML/100 KG 8x applications of Cypermethrin	2.2% PVY increase	Martin-Lopez et al. 2006
Imidacloprid (Gaucho) + Iambda-cyhalothrin (Karate Zeon technology)	Neonicotinoid + Pyrethroid	PVY/Potato New Zealand	Seed tubers 134 ml/ha Imidacloprid lambda-cyhalothrin 10 ml/ha after 10 winged aphids/150 leaves	Not significant	Van Toor <i>et al.</i> 2009
Imidacloprid (Gaucho) + Methamidophose (Tamaron)	Neonicotinoid + organophosphate	PVY/Potato New Zealand	Imidacloprid Seed tubers 134 ml/ha 480 ml/ha Methamidophose Every 14 days	Not significant	Van Toor <i>et al.</i> 2009
Mixture A Hallmark Zeon (lambda- cyhalothrin) + pymetrozine (Plenum) Mixture B Hallmark Zeon + Aphox (Pirimicarb)	Pyrethroid + Pyridine Pyrethroid + Carbamate	PVY /potato England	Mixture A alternatively with Mixture B for the first 6 sprays then Mixture B for the last 2 weeks	5.5% PVY incidence in daughter tubers compared to 8% in the untreated control	Cambridge university farm, Farmers Guardian, 2009
Mixture A Hallmark Zeon (lambda- cyhalothrin) + pymetrozine (<i>Plenum</i>) Mixture B Hallmark Zeon + Aphox (Pirimicarb) Treatment C Thiacloprid (Biscaya)	Pyrethroid + Pyridine Pyrethroid + Carbamate	PVY /potato England	Mixture A alternatively with Mixture B for the first 6 sprays then Mixture B for the last 2 weeks *Biscaya was applied at the first and the third spray alone	5.5% PVY incidence in daughter tubers compared to 8% in the untreated control	Cambridge university farm, Farmers Guardian, 2009

8. CONCLUSIONS AND DISCUSSION

8.1. Conclusions - Mineral Oils

The number of insecticides available to control non-persistent viruses may be reduced or restricted when EU regulations 91/414/EEC and 1107/2009 take effect over the coming years. While some active ingredients may remain, mineral oils currently represent an untapped control method to prevent virus spread in UK potato fields. It has been reported that, to be effective in preventing virus spread, mineral oils should be applied at concentrations in the range of 1-3 % and the application should be repeated frequently to cover newly emerging leaves. Concentration values above 3% appear to be accompanied by increasing plant toxicity and it is widely noted that this can include changes in plant appearance. It is recognised that concern over altered appearance is an overriding factor where visual crop inspections form the basis of seed certification, as occurs in the UK, and Boiteau and Singh (1982) specifically mention this in relation to their work in Canada.

Our judgement is that, with the shortage of highly effective control measures, mineral oils could be incorporated into virus control programmes on seed potato crops. In particular, ware or other crops where virus symptoms are a secondary consideration could be used for immediate application of oil. Assessment of effectiveness and inspectability could also occur on these crops. Another avenue to assess the effectiveness of oils would be to use the Estima crops which are post harvest tested as a matter of routine. Further field related studies are required to validate the usage of mineral oils more widely on seed crops as each variety could differ in their response. The input level of virus is also likely to affect the outcome as oils work better when both the virus inoculum and aphid vector pressure are low (Simon and Zitter, 1980; Martin-Lopez *et al.*, 2006). In particular the statutory bodies SASA/Fera must make an informed decision on the impact of oils on their certification schemes and ways to help expedite this decision should be explored.

Should oils receive approval for use on seed crops then the core characteristics of a successful mineral oil are well described and understood. Minor issues about emulsifiers and additives which may enhance oil persistence, oil concentration, and number of applications could be identified. Conflict exists between different studies relating to the optimal oil concentration and application frequency, which are both important issue to consider when incorporating oil into virus control programmes. Experiments performed in different geographical regions and at different times using different oil formulations can draw different conclusions. Thus, the absolute effect of the use of mineral oil in particular regions can be underestimated or overestimated when extrapolating from published work. It is a prerequisite therefore to conduct some evaluation of the effectiveness of oil in the country where it is to be applied. Any enhancement of mineral oils when combined with pyrethroids should also be examined. This would facilitate the sustainable use of pyrethroids within a mineral oil spray programme. This rationalized application of pyrethroids should delay the selection of pyrethroid resistant *M. persicae* on potato. The genetics of resistant population is now well understood (Fenton et al 2010) and would complement this approach. However, it should be noted that the majority of the potyvirus vectors carry no resistance and their control should be a priority until pyrethroid resistant populations of *M. persicae* become a problem. Investigation of synergy between the new classes of insecticides and oil is also required.

Further work is required on the environmental fate of mineral oils after application and the effect of tank mixing oils with fungicides. There is some evidence that blight control could become a problem in a mineral oil regime. An understanding of this is important if the seed potato industry is to take advantage of the potential benefits of the use of oils in a way that is acceptable to the certifying authorities and is also practicable for commercial growers.

8.2. Conclusions - Insecticides

A limited number of insecticides have been reported to partially prevent non-persistent virus transmission in general and on potato crops in particular. The most consistent of these are pyrethroid based. Sustainability in using these insecticides is one of the most important objectives when designing virus control programmes. This is because of the high cost of insecticides and most importantly the capability of virus vectors to develop resistance to certain groups of pesticides when applied frequently and for long time. Not to mention, the undesirable effect of the insecticides on the general insect community and environment. Some reports consider insect natural enemies, but, unlike ware, no high health seed potato production system can rely on naturally occurring predators to prevent virus spread.

Insecticides in the organophosphates (OP) and carbamate groups have been used extensively and, to some extent, randomly by potato farmers to control plant viruses for many years. These two classes of insecticides have succeeded in decreasing PLRV incidence in potato crops to a manageable level. Encouraged by the successes with PLRV, farmers may have thought that they could control PVY spread with the same classes of insecticides. In order to achieve virus control and to decrease resistance developments in aphid vectors, the application of insecticides should be alternated so that different modes of action are used sequentially. It has been suggested that continuous application of ineffective insecticides could initiate a hyperactive response by winged aphids which may lead to increased probing or flight behaviour and as a result enhance virus spread (Simons and Zitter, 1980). However, this comment is only generally relevant to *M. persicae* and PLRV as the overwhelming populations of PVY vectors are not resistant and not on the crop.

Similarly the new classes of insecticides (neonicotinoids and pyridines) work differently and should be applied in rotation with the above classes of insecticides to minimize the chance of developing resistance against their mode of actions. However, it is not clear just how useful these chemicals are likely to be in preventing potyvirus spread.

Developing new insecticides with different mode of actions is another possible way to avoid resistance problem. A product which combines rapid kill and repellency would be the most useful in preventing both PLRV and potyvirus spread. However, this is practically hindered by the limited targets in the insects for the insecticides to act on, and the time and money needed to develop such new insecticides. In order to introduce a new insecticide commercially is a very long and costly process. This normally takes 10-15 years to develop one pesticide, costs *c.a.* 300 million dollars, and the life time of pesticides is relatively short compared with the development time.

In Table 4 the effectiveness of some insecticide rotation schemes were presented. There are some occasions where pesticides rotation was not effective in decreasing virus spread (Van toor *et al.* 2009), and other occasions where, the combined application has a negative effect (Lopez-Martin *et al*, 2006). However, in other reports,

the enhanced effects of different pesticides were proven to be effective in controlling potyvirus spread on potato (E. Anderson 2009, unpublished).

8.3. Discussion

Controlling non-persistent viruses in potato crops using insecticide or mineral oil sprays is difficult. The total numbers and diversity of aphid vectors which can participate in the transmission process, along with the lack of information about the exact mechanism of transmission, are just some of the factors which compound this problem. There is no single highly effective insecticide or mineral oil which completely suppresses vector transmission in the field. Weidemann (1988) concluded that different control methods should be combined to interrupt the PVY^N epidemic cycle, as any single method will have limited effect on its spread. Mature plant resistance can decrease PVY^N spread later in a growing season, thus pre-sprouting potato seeds and early planting combined with early burn down will be effective in decreasing PVY^N incidence in potato crops. In agreement with Weidemann (1988), we conclude that there are many reported strategies for decreasing virus transmission, but none completely prevent it. It is difficult to conclude from the literature which is the best current strategy as contradictory data exist. This conflict can be attributed to the experimental conditions and that they were conducted in different countries and at different times. Even in the same country, many factors affecting virus transmission can change over a 50 year period. For example, it has been reported by Hollings (1955) that harsh weather conditions in Scotland are favourable for seed potato production compared with England and Wales. This is because of the reduction in over-wintering aphids or volunteer potatoes which can initiate infection in the early season. This situation may now be different under climate change and aphids are surviving milder winters in greater numbers and groundskeepers are now more common. Two decades ago it was demonstrated that groundkeepers were already an important source of virus in some areas (Barker, 1984).

However, the impact of climate change on local weather is far more complex and the winters of 2008/2009 and 2009/2010 were two of the coldest for some time with considerable frosts. In addition to changes in weather, there has been a change in the relative importance of virus vectors depending on their population dynamics, and the general composition of the cropping environment. For example, the leaf curling plum aphid (*Brachycaudus helechrysi*) was reported to be the most abundant aphid vector of PVY in England more than 30 years ago (Harrington *et al.*, 1986). Changes in agricultural practices have introduced winter crops of wheat, barley and oilseed rape. As a result, cereal aphids are now coming to the fore as the most important vectors of PVY (Sigvaldt, 2007; Pickup *et al.* 2008).

8.3.1. What can be concluded from the literature and suggested as the best practice to control non-persistent virus spread?

Insecticides are most capable of controlling non-persistent virus transmission caused by colonizing or non colonizing aphids when there are no external virus sources and a low percentage of infection inside the potato field. Low virus incidence can be achieved by growing high quality certified seed and removing groundkeepers with regular inspection and roguing of infected plants during a season. Prior assessment of areas earmarked for seed production should also be considered.

During a season early insecticide applications may act to decrease the transmission by removing colonizing aphids. Complete prevention of colonizing aphids from spreading potyvirus infection by chemical application on the potato crop is unlikely, due to the short time required by the aphid to inoculate the virus into plants. However, colonising aphids are more likely to remain on any potato plants they probe. In contrast, insecticides are much less effective in controlling spread by non-colonizing aphid species. Non colonizing aphids will avoid exposure to insecticides as they will only be present on the crop for a short time and they will avoid systemic insecticides because they do not feed on potato plant phloem.

The following points have been concluded from the available literature:

1. Although oil effects on virus transmission were reported about 50 years ago, its mode of action has still to be elucidated. Moreover, this topic has a very low level of recent interest. In fact, the most recent publication which investigated or handled the mechanism behind oil effects on virus transmission inhibition was reported almost 20 years ago (Wang and Pirone, 1996). The reasons behind such neglect are not clear, but the speculation is that scientists are currently still trying to understand the mechanism of non-circulative virus transmission. Perhaps oil studies will increase if the transmission process is clearly understood.

2. To control non-persistent virus spread many studies recommended oil application alone or combined with other pesticides. However, oil application in preventing virus transmission is still limited and even discouraged in some countries where seed certification programmes are based on visual field inspections. The reason for that is the assumption that oil spray will induce symptoms in potato which can be misleading for inspection and impact on the efficiency of seed certification programmes and there is some grounding to this. Clearly, any phytotoxicity will cause abnormal plant growth and yellowing which has the potential to mask viral symptoms and lose yield. Peters (1987) mentioned that in addition to the high cost of oil application, oil has not became common practice because of the difficulty to rogue infected plants, and the incompatibility of Fentin compounds to control late blight. De Bokx and Cuperus (1978) and Schepers and Bus (1978) also comment on masking of viral symptoms. However, all the reported cases of severe phytotoxicity and yield loss resulted after application of a high rate of oil which is not essential in achieving reasonable protection. Moreover, oil formulations have been improved in the last 2-3 decades towards more effective and less harmful products, while these reported phototoxic or virus masking effects were reported in the 1980s when oil formulations were very limited. Oil usage in the UK should therefore consider the advantages and disadvantages related to oil application for non-persistent virus control. These studies should be conducted on different potato cultivars, and by using different concentrations and oil formulations. Any recommendations should cover optimal types

of mineral oils, oil concentration, timing and number of applications per season, the best method of application, and recommended combination with insecticides and other crop practices.

3. The literature suggests that fast-acting synthetic insecticides such as pyrethroids are also partially effective in reducing non-persistent virus spread. The case for any other active substance group having an impact is less clear.

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