

## Control of air-borne field spread of tulip breaking virus, lily symptomless virus and lily virus X in lilies by mineral oils, synthetic pyrethroids, and a nematicide in the Netherlands

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### Abstract

The inhibitory effect on the spread of viruses in lilies viz., tulip breaking virus (TBV; non-persistently aphid-borne, potyvirus,) lily symptomless virus (LSV; non-persistently aliphid-borne, carlavirus), and lily virus X (LVX; potexvirus of unknown etiology), was studied of brands of mineral oil (Luxan oil H and Duphar-7E oil) and synthetic pyrethroid insecticides (l-cyhalothrin and deltamethrin), and a nematicide (aldicarb) in crops in which virus-infected plants were present as virus sources. The spread of TBV and LSV were controlled by sprays of mineral oil and insecticide, while that of LSV was also limited by the soil-applied nematicide. The spread of LVX was reduced by the insecticides and, not effectively by the mineral-oil spraying, by which data the mode of transmission may be presumed to be by an insect in the persistent or semi-persistent manner.

Mixtures of mineral oil and pyrethroid were more effective in the reduction of spread of TBV and LSV than either components tested alone. The mineral oil was the most effective component in the mixtures in which pyrethroid added a slight extra effect. The addition of pyrethroid did not mask either the lower efficacy of the oil brand Duphar-7E oil, or the diminished inhibitory effect of low dosages of oil. The normal rate of mineral oil gave similar control to that of a mixture of mineral oil at half rate plus the pyrethroid at full dosage. Low rates of oil, or even synthetic pyrethroids alone may be used on cultivars which suffer of the loss of bulb weight by the use of normal or decreased rates of oil. Weekly sprays were more effective than fortnightly sprays. The rate of control by the weekly sprays ranged between 90 and 95% for Luxan oil H at half dosage plus the full rate of pyrethroid. Weekly sprayed synthetic pyrethroids alone onto the virus sources and the plants to be infected gave 60-70% control. The weight ratios tended to be slightly reduced if the half dosage of the efficient Luxan oil H was used. Factors which affect the control of the air-borne field spread of viruses by mineral oils and synthetic pyrethroid insecticides in lilies are discussed.

*Additional keywords:* Luxan oil H, Duphar-7E oil, deltamethrin, cyhalothrin, aldicarb.

### Introduction

The inhibitory effect of mineral oil on the non-persistent transmission of tulip breaking virus (TBV; potyvirus, aphid-borne) has been exploited by routine spraying of lilies since the mid 1960s (Asjes, 1976; Asjes, 1984). Similarly the effect on the non-persistent lily symptomless virus (LSV; carlavirus, aphid-borne) became evident when

virus-tested lilies were produced from the mid 1970s onwards (Asjes, 1976). In the routine of spraying of lilies<sup>1</sup> we became aware of the vulnerability of certain cultivars to reduction of bulb weight due to the sprays of mineral oil. The Asiatic hybrids were fairly tolerant (Asjes, 1984), whereas cultivars of the Oriental hybrids showed substantial reductions in bulb yields (C.H.M. Hendriks, N.A. Hof and B.J. Kok, see Asjes, 1989b). Therefore, the use of synthetic pyrethroids which proved effective in the control of stylet-borne virus spread (Asjes, 1981; Gibson et al., 1982; Rice et al., 1983), although less efficiently than mineral oils (Asjes, 1981) was considered. The combined use of mineral oils and synthetic pyrethroids tested on potatoes (Gibson and Cayley, 1984; Perrin and Gibson, 1985; Gibson and Rice, 1986; Bell, 1989) indicated diminished amounts of oil to be used for control. Maybe that would minimize the reduction of bulb weight without loss of efficacy of control.

In this paper data will be presented on the reduction of spread of tulip breaking virus, lily symptomless virus, and lily virus X in lilies by different oil brands and synthetic pyrethroids applied in different rates and frequency of spraying, and on the use of a nematocide with a systemic effect in plants as insecticide.

## Material and methods

*Plant material and experimental lay-out.* In 1987 9-10 cm circumference bulbs of lily cv. Enchantment were retrieved from a certified lot which contained about 0.5% TBV, 2% LSV, and 0% LVX. In 1988 another lot of cv. Enchantment contained about 0.5% TBV, 1% LSV, and 0% LVX. Bulbs of cv. Connecticut King contained 0% of all viruses, and the lot of cv. StarGazer 100% LSV, 0% TBV, and 5.5% LVX. These percentages were not subtracted from the infection rates obtained in the field, except for cv. Star Gazer.

In 1987 each plot consisted of four rows planted in the 1 m-wide beds in the direction of the flanking paths. In each of the two central rows of the plots 20 bulbs of the certified material were planted plus ten virus-infected bulbs of each cv. Enchantment and Sun Ray in the centre of the rows. Both of the two outer rows of a plot contained 30 bulbs of the certified lot. This made a total of 100 bulbs of certified material per plot. The ten virus-infected bulbs of cv. Enchantment contained LSV and TBV, while those of cv. Sun Ray contained either no virus, or LSV, or LVX, or LSV + LVX. So altogether the inoculation pressure in the plots was about 4% TBV, 12% LSV, and 3% LVX.

In one of the 1988 experiments (Exp. 1) a similar lay out of the plots was applied. The infected material was of cv. Enchantment with TBV and LSV, and of cv. Montreux with LSV and LVX. The overall infection pressure in the plots was about 8% TBV, 9% LSV, and 5% LVX. In a second experiment of 1988 (Exp. 2) five pairs of two 1 m-wide rows with ten bulbs per row were either planted per plot with certified material of cv. Enchantment, or cv. Connecticut King, or cv. Star Gazer, while each pair of rows was flanked by two rows of cv. Montreux infected with LSV and LVX. So each plot was subject to an infection pressure of about 20% LSV and 40% LVX. The rows lay

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<sup>1</sup> Total acreages of lilies increased considerably in The Netherlands since the early 1960s, e.g., in 1960: 100 ha, 1970: 223 ha, 1975: 593 ha, 1980: 1119 ha, 1985: 1686 ha, 1989: 2571 ha, and 1990: 2419 ha.

perpendicularly to the direction of the paths.

In 1987 the bulbs were planted in early April, and in 1988 in early April in the first experiment and in late April in the second one. In both years each plot was flanked by a transverse fallow row and a row of dahlias, which crop is not susceptible to TBV, LSV, and LVX. Dahlias were also planted on beds of half width (0.5 m) alongside the beds with the plots. In 1988 in Experiment 2 the intermediate beds were of full width (1 m). The dahlias were full-grown in July and then a little taller than the lilies. The trial beds mostly comprised 12 plots both in 1987 and in 1988 (Exp. 1). Six plots lay per bed in experiment 2. All beds were flanked by paths of 50 cm width. In both years treatments were arranged in three randomized blocks.

*Chemicals.* The brands of the mineral oil were 'Luxan oil H' (93% oil; Luxan BV) and 'Duphar-7E oil' (99% oil; Philips-Duphar BV), of which the characteristics were earlier reported (Asjes, 1984). The amounts at full rate (12.5 and 15 l ha<sup>-1</sup>, respectively) were emulsified in volumes of water equivalent to 400 l ha<sup>-1</sup>, as were the half and quarter dosages of the oils. The synthetic pyrethroids were in PP 321 (5% 1-cyhalothrin e.c.) and in Decis (2.5% deltamethrin e.c.) both as liquid formulation at full rates of 15 g ha<sup>-1</sup> and 10 g ha<sup>-1</sup>, respectively. The full, half and quarter dosages were all sprayed in 400 l water per hectare. The nematocide with a systemic insecticidal effect in Temik 10 G (10% aldicarb e.c.) was slightly incorporated into the soil at a rate of 30 kg ha<sup>-1</sup> in mid-May and twice at about six week intervals afterwards. Spraying of fungicides, e.g., maneb/zineb, to control *Botrytis* infection, was performed independently of the applications of mineral oils or pyrethroids.

*Application of sprays.* Emulsions prepared within half an hour before use were sprayed with a propane knapsack sprayer (Birchmeijer Helico nozzles 1,2) at a pressure of 400 kPa. Spraying was started in May when the lily plants were about 5-10 cm tall growing in an open stand with bare soil in between. The plants grew about 5 cm per week until July, when flower buds were picked. The last spray applications were made early in September for cvs. Enchantment and Connecticut King, and late in September for cv. Star Gazer.

Both the healthy and virus-infected plants were sprayed. Spraying was done during calm weather to minimize spray drift and it was performed during the day when it was cloudy, or later in the afternoon in case of sunny and dry weather at mid-day.

*Assessment of bulb weight and virus infection.* The bulbs of cvs. Enchantment and Connecticut King were harvested in October, and those of cv. Star Gazer in November. The bulbs were stored at 0-1 °C till January. The bulbs were weighed after appropriate cleaning. The mean weight ratios were calculated as (weight of *n* treated bulbs): (weight of *n* control bulbs) × 100. The weight of the bulbs was not considered to be affected by the effect of current-season infections as determined in earlier experiments (unpublished results). The presence of TBV, LSV, and LVX was assessed by Double Antibody Sandwich ELISA. The virus percentages in Tables 2 and 3 were calculated on the basis of single infections per plant of either two viruses, viz., LSV and TBV, or LVX. This means that the multiple infections shown in Table 1 were calculated to be single plants infected either by one of the two or three viruses. The data of LVX summarized in Table 4 comprise the infection of plants in many plots as is shown by the figures between brackets.

Table 1. Single and multiple infections by tulip breaking virus, lily symptomless virus, and lily virus X in untreated plots of virus-control experiments in lily cv. Enchantment in 1987 and 1988.

Virus infection	Percentage in	
	1987 ( <i>n</i> = 1155)	1988 ( <i>n</i> = 1144)
Lily symptomless virus	29.3	19.8
Lily virus X	0	4.5
Tulip breaking virus	5.6	9.7
Lily symptomless virus + lily virus X	5.5	2.5
Lily symptomless virus + tulip breaking virus	5.3	3.7
Lily virus X + tulip breaking virus	0.1	1.0
Lily symptomless virus + lily virus X + tulip breaking virus	0.4	1.0
Total	46.2	42.2

## Results

*Complexity of infection.* The results of the virus spread into single or multiple infections in cv. Enchantment of untreated plots in 1987 and 1988 are shown in Table 1. The data indicate that LSV spread most. The rates of multiple infection were fairly high in these untreated plots. These rates became progressively lower as the spread of viruses was inhibited in the sprayed plots.

*Effect of mineral oil and synthetic pyrethroid.* The results on the effect of mineral oils and synthetic pyrethroid insecticides in 1987 are shown in Table 2. The data indicate that the Duphar-7E oil generally was less effective than the Luxan oil H. The full dosages of cyhalothrin and deltamethrin without oil limited the virus spread very considerably. The combined mineral oil/pyrethroid sprays generally were more effective in the control than either component alone. Combined sprays with Luxan oil H tended to decrease the bulb yields slightly more than those of Duphar-7E oil. The addition of pyrethroids, especially cyhalothrin mostly added to the control provided by the mineral oil, especially Luxan oil H. The addition of pyrethroids did not compensate for the less effectiveness of the Duphar-7E oil.

The results of the mineral-oil and insecticide spraying in 1988 (Exp. 1) are shown in Table 3. The addition of pyrethroids to the mineral oil showed a slight increase in the efficiency of reduction of virus spread. The prevention of virus spread was best in mixtures of pyrethroids with mineral oil at half dosage. Both the pyrethroids and the mineral oil alone reduced the virus spread very considerably. The weekly sprays gave more control than the fortnightly application. Weekly sprays containing half dosages of Luxan oil H slightly decreased the bulb yields.

*Spread and control of lily virus X and lily symptomless virus.* The results on the spread and control of LVX, and LSV and LVX are shown in Table 4 and 5, respectively. The synthetic pyrethroids were most effective in the control of LVX and substantially

Table 2. Effect of weekly mineral-oil sprays without and with synthetic pyrethroid insecticide on the spread of lily symptomless virus and tulip breaking virus added together in lily cv. Enchantment in 1987.

Treatment	Dosage	% virus	% spread reduction				Weight ratio			
			mineral oil	mineral oil + cyhalothrin	mineral oil + delta-methrin	mineral oil	mineral oil + cyhalothrin	mineral oil	mineral oil + cyhalothrin	mineral oil + delta-methrin
Untreated				46.2			0		100	
Luxan oil H	0.25	14.9		6.3	16.9	68	86	98	101	103
Luxan oil H	0.5	11.1		3.9	6.4	76	91	95	94	101
Luxan oil H	1	5.7		4.4	3.5	88	90	95	86	93
Duphar-7E oil	0.25	24.1		18.5	20.8	48	60	105	104	102
Duphar-7E oil	0.5	17.2		7.6	21.6	63	84	101	101	99
Duphar-7E oil	1	5.0		10.1	13.3	89	78	95	99	93
Cyhalothrin	1	<i>without mineral oil</i>		14.7			68		97	
Deltamethrin	1			16.4			64		101	

Table 3. Effect of sprays of mineral oil and synthetic pyrethroid insecticides in different dosages and spray frequency (7 and 14 days) on the spread of lily symptomless virus and tulip breaking added together in lily cv. Enchantment in 1988 (Exp. 1).

Treatment	Dosage of mineral oil + insecticide	% virus		% spread reduction		Weight ratio	
		7	14	7	14	7	14
Untreated	0 + 0	37.7		0		100	
Luxan oil H	0.25 + 0	6.1	12.0	84	68	96	99
Luxan oil H + cyhalothrin	0.25 + 0.5	10.2	9.4	73	75	99	102
Luxan oil H + cyhalothrin	0.25 + 1	5.2	8.1	86	78	99	104
Luxan oil H + deltamethrin	0.25 + 0.5	4.5	12.2	88	68	99	99
Luxan oil H + deltamethrin	0.25 + 1	3.4	7.0	91	81	103	104
Luxan oil H	0.5 + 0	4.4	11.8	88	69	95	101
Luxan oil H + cyhalothrin	0.5 + 0.5	2.7	2.7	93	93	94	97
Luxan oil H + cyhalothrin	0.5 + 1	2.0	5.0	95	87	94	99
Luxan oil H + deltamethrin	0.5 + 0.5	1.7	3.1	96	92	95	96
Luxan oil H + deltamethrin	0.5 + 1	1.7	10.1	95	73	96	102
Cyhalothrin	0 + 1	11.4	15.6	70	58	99	101
Deltamethrin	0 + 1	14.4	13.7	62	64	100	102

Table 4. Effect of weekly mineral-oil and synthetic pyrethroid insecticide sprays on the spread of lily virus X in lily cv. Enchantment in 1987 and 1988.

Treatment	% virus			
	1987		1988	
Untreated	3.1 <sup>1</sup>	(11) <sup>2</sup>	9.1	(12)
Luxan oil H <sup>3</sup>	3.9	( 9)	3.3	(12)
Luxan oil H + cyhalothrin	0.2	( 9)	0.1	(24)
Luxan oil H + deltamethrin	0.9	( 9)	0.3	(24)
Cyhalothrin	0	( 3)	0.9	( 6)
Deltamethrin	0.7	( 3)	0.5	( 6)

<sup>1</sup> In one plot about 50% of the plants became infected by LVX. This excessive datum has been left out in the calculation of the average infection rate. Otherwise this rate would have been 7.1%

<sup>2</sup> The figure between brackets concerns the number of plots of which the average virus rate was calculated.

<sup>3</sup> Irrespective of the rate of mineral oil or synthetic pyrethroid insecticide.

effective for LSV. Aldicarb was substantially effective for both LVX and LSV. The mineral oil effective in the prevention of LSV spread was not likewise at all in the reduction of LVX spread.

Table 5. Effect of mineral oil, synthetic pyrethroid, and nematicide on the spread of lily virus X (LVX) and lily symptomless virus (LSV) in lily cvs. Connecticut King, Enchantment, and Star Gazer in 1988 (Exp. 2).

Treatment	Connecticut King				Enchantment				Star Gazer	
	% virus		% spread reduction		% virus		% spread reduction		% virus	% spread reduction
	LVX	LSV	LVX	LSV	LVX	LSV	LVX	LSV	LVX	LVX
Untreated	30.9	20.7	0	0	26.0	12.1	0	0	38.6	0
Cyhalothrin	1.7	5.0	94	76	2.6	4.9	90	60	1.3	97
Luxan oil H + cyhalothrin	0.7	2.7	98	87	2.4	2.4	91	80	1.2	97
Aldicarb	8.7	6.5	72	70	10.0	4.0	62	61	8.4	78

## Discussion

The reduction of the spread of the non-persistently transmitted viruses like lily symptomless virus and tulip breaking virus by mineral oils and synthetic pyrethroids confirmed earlier results (Asjes, 1981; 1984; 1985). The potexvirus character of the LVX (Stone 1980; Memelink et al., 1990) implicated that the spread was not assumed to be affected by insecticide sprays. However, the data showed otherwise. The transmission of lily virus X being persistent or semi-persistent was based on the data that the mineral oil only slightly at the most reduced the LVX spread, whereas pyrethroids and Aldicarb were highly effective (Tables 4 and 5). The inhibition of LVX spread by insecticide, e.g., synthetic pyrethroids and Aldicarb, and an experimental insecticide CKB 1638 (unpublished results; Fritsche et al., 1984), was not to be expected for potexviruses. The vector species is not known yet as aphids, e.g., *Myzus persicae*, *Macrosiphum euphorbiae*, and *Dysaphis tulipae*, were unable to transmit the virus from single infections as well as from those additionally infected with LSV and TBV (A.F.L.M. Derks, personal communication). The transmission of LVX by the mechanical inoculation of bulb scales to simulate conditions of injury during the handling of the bulbs after harvest, or during the scaling of the bulbs for propagation purposes failed to be effective at all, whereas LSV was transmitted at low rate (Asjes and Segers, 1987).

The effect of mixtures of mineral oil and synthetic pyrethroid insecticide has been earlier tested, however, with only one oil brand at a time (Raccach et al., 1983; Gibson and Cayley, 1984; Perrin and Gibson, 1985; Gibson and Rice, 1986; Bell, 1989; Raccach et al., 1989). In this study in which two oils were tested, the additional inhibitory effect of pyrethroids on the virus spread did not compensate for the difference in efficiency of the two oil brands tested (Table 2; Asjes, 1984). The effect of pyrethroids additional to the mineral oil generally was much less than that induced by the mineral oil alone (Table 3).

The range of control was 90-95% by the weekly spray applications of the most effective oil brand at half dosage in a mixture with synthetic pyrethroid at full rate. The inhibitory efficacy of the pyrethroids of 60-70% (Tables 2 and 3) by spraying both the

virus sources and the plants to be infected doubled the efficiency compared to spraying only the plants to be inoculated (25-35%; Asjes, 1985). The slightly better efficacy of cyhalothrin compared to deltamethrin in 1987, which was the reverse in the weekly sprays in 1988, can not be explained. The addition of permethrin, cypermethrin or fenvalerate in Luxan oil H at half rate were similarly effective as the pyrethroids tested in 1987 (unpublished results).

Aldicarb used as soil disinfectant to control the root lesion nematode *Pratylenchus penetrans* and the free-living root nematode *Rotylenchus robustus* in lilies proved to be widely effective by its inhibition of LVX spread, but its insecticidal impact as systemic agent was also effective in the limitation of the spread of the non-persistent LSV. The effect was the more so unexpected as resident aphids were not ever observed on the lilies. A similar effect on the non-persistent beet mosaic virus in sugar beet has recently been reported (Hurej and Peters, 1989). The effect of aldicarb and similar compounds may be a subject for further investigation.

Oil sprays has been used routinely for control of non-persistently aphid-borne viruses lilies up to the late eighties. In the last years this routine has gradually changed to the use of mineral oil plus synthetic pyrethroids. Further progress in the culture of virus-tested bulbs of the many cultivars produced in the seventies and eighties, and subsequently that of highly qualified planting material has been made possible by the efficient use of mineral oil and/or synthetic pyrethroids. The growers may foster this use if they will also consider the impact of the effects which may counteract the efficiency of the reduction of virus spread, e.g., the rate of virus sources in or adjacent to crops, interval of sprays, and efficacy of oil brands as shown by the results above. Moreover, the vulnerability of some cultivars (C.H.M. Hendriks, N.A. Hof and B.J. Kok, see Asjes, 1989b) to loss of bulb yields due to the use of mineral oil will enhance the use of pyrethroids. The seasonal susceptibility of lilies needs further investigation to make the spraying more efficient. The planting density should be high to enhance further low rates of virus spread (Asjes, 1976). The growth of weeds may inexplicably interfere with the efficacy of the sprayings. The daily and seasonal weather conditions may also affect the efficiency of the sprays, and this topic requires further study.

Progressively, scientists and growers have to evaluate these factors in order to spray as little as possible to minimise the burden to the environment without a substantial loss in the control of viruses in lilies (Asjes, 1989a; 1989b; 1989c). The resistance of some cultivars to some viruses (Asjes, 1989a) but unfortunately not to all will necessitate the need to spray mineral oil and the liquid formulations of synthetic pyrethroids for the time being and in the near future unless the genetic transformation of lilies is successful in the short term to incorporate resistance against these viruses (S.A. Langeveld, A.F.L.M. Derks, personal communication).

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