Testing the efficacy of single applications of five insecticides against Scaphoideus titanus on common grapevines

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Testing the efficacy of single applications of five insecticides against *Scaphoideus titanus* on common grapevines

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The appearance of the Flavescence Dorée phytoplasma and the possibility of its rapid spread by its vector, the American Grapevine Leafhopper (*Scaphoideus titanus* Ball) (AGVL), represent a serious threat to viticulture production in Slovenia and elsewhere in Europe. Insecticide treatment represents one of major means of reducing the abundance of AGVL in wine-growing areas. In the period from 2010 to 2011, five insecticides against AGVL were tested, and the efficacy of the substances was calculated according to the number of nymphs (at 2, 7, 14 and 21 d after application), which were counted on the grapevine leaves. In both years, the highest cumulative efficacy (96–97% or 0.01 nymphs/leaf) was confirmed for thiamethoxam, whereas chlorpyrifos-methyl showed a very high cumulative efficacy (87–89% or 0.01–0.02 nymphs/leaf). In 2010, indoxacarb exhibited only a 43% (0.09 nymphs/leaf) cumulative efficacy; however, its substitute in 2011, chlorpyrifos, was more effective (73% or 0.05 nymphs/leaf). A single application of pyrethrin demonstrated only 45–61% (0.05–0.09 nymphs/leaf) efficacy. Based on the results of our research, we recommend the use of thiamethoxam and chlorpyrifos-methyl against AGVL.

**Keywords:** American grapevine leafhopper; common grapevine; grapevine yellows; flavescence dorée; *Scaphoideus titanus*; insecticides; *Vitis vinifera*

1. Introduction

Grapevine yellows is a pathological syndrome affecting grapevines. It is caused by phytoplasmas and is spread by insects and other arthropods (Battle et al. 2000; Weintrub and Beanland 2006; Bertaccini and Duduk 2009). The phytoplasmas occur in many grape-growing areas in different parts of the world and are increasingly significant (Beanland et al. 2006). The most important types of grapevine yellows appearing in European vineyards are Bois Noir (BN) and Flavescence Dorée (FD), both of which cause economic damage and diminish the quality and quantity of crops (Boudon-Padieu and Maixner 2007). The symptoms of different grapevine yellows are similar, yet there are essential differences in the epidemiology of the individual species of grapevine yellows, depending on the different bionomics and life cycle of their vectors (Boudon-Padieu 2005; Gallet et al. 2011a, 2011b; Kessler et al. 2011). FD is considerably more harmful than BN (Bressan et al. 2006) and can be transmitted very rapidly via vectors throughout vineyards across large areas (Bressan et al. 2006; Belli et al. 2010).

The most important vector of FD is the American Grapevine Leafhopper (*Scaphoideus titanus* Ball, Hemiptera, Cicadellidae) (AGVL) (Marzachi et al. 2001; Bressan et al. 2006). Though originating from North America, *S. titanus* was introduced into France (Decante and van Helden 2006) and then spread to Spain and Portugal, through Italy and then to the Balkans (Battle et al. 1997; Angelini et al. 2001; Boudon-Padieu and Maixner 2007; Krnajic et al. 2007; Papura et al. 2009; Delè et al. 2011). Owing to its habit of feeding on phloem, *S. titanus* can transmit phytoplasmas from infected plants to healthy ones. The AGVL is primarily associated with grapevines. Although several decades ago, it was still believed that grapevines were the only host of this leafhopper (Vidano 1966), the results of recent studies show that *S. titanus* can be live on other plants: Old Man’s Beard (*Clematis vitalba* L.) (Angelini et al. 2004), Creeping Buttercup (*Ranunculus repens* L.) and White Clover (*Trifolium repens* L.) (Jermini 2011). This harmful pest, which develops one generation per year, hibernates in the egg stage (Rigamonti et al. 2011), most often on two-year-old wood (Bertin et al. 2007) and three-year-old wood, and less frequently on one-year-old grapevine wood (Bressan et al. 2005). *Scaphoideus titanus* hatches from eggs in the second half of May and has five developmental stages. FD is most frequently transmitted by adult leafhoppers, though, occasionally, third instar nymphs also are vectors (Seljak 1993). FD can spread very quickly when both infected plants and

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a large population of the leafhopper occur at the same location (Bertin et al. 2007).

Because no effective natural enemy is known to date, AGVL populations should be suppressed using insecticides during the host plants’ growth period (Malausa et al. 2003; Nusillard et al. 2003; Boudon-Padieu and Maixner 2007). There are no environmentally acceptable manners of suppression. Maintaining a low population of this vector is a key means of preventing the spread of FD, as has been confirmed by the preliminary results of our monitoring of leafhopper populations in vineyards treated with insecticides: AGVL population densities have been significantly reduced in vineyards treated with insecticides, predominantly chlorpyrifos-ethyl and chlorpyrifos-methyl (Seljak 2008). Nevertheless, it is important to consider that insecticide use for suppressing AGVL in vineyards depends on the manner in which the grapes are produced and also on the size of the leafhopper population.

Early insecticide applications against the juvenile stages of the insect leave no significant residues in the wine (Bosio et al. 2003). Thus, the aim of our two-year research study was to evaluate the efficacy of the early application of certain insecticides for suppressing AGVL in vineyards.

2. Materials and methods

2.1. Experimental site

The experiment was performed in the years 2010 and 2011 in a 10-year-old vineyard growing the grape variety “Cabernet Sauvignon” (Vitis vinifera L.) from the Vipavska dolina (the village Lože pri Vipavski kraj, 45°50’ N, 13°56’ E, 139 m.a.s.l.) winegrowing district, a sub-Mediterranean region of Slovenia. The variety was grafted onto rootstock SO4 (Vitis berlandieri × Vitis riparia), and the Double Guyot training system was implemented. The vines were planted on marl soil, with distances of 0.9 m between the vines in a row and 2.4 m interlinear, for a total of 4630 vines per hectare. Between rows, the vineyard has permanent sod (uncultivated) as a mixture of different grasses (Poaceae) and clovers (Fabaceae) from 3 years of age. The vineyards were fertilised only with manure (10 t per hectare) and mineral fertilisers PK 20-20 (225 kg per hectare; manufactured by Adriatica S.p.A., Loreo, Italy) prior to deep ploughing.

2.2. Spraying programmes

Five different spraying procedures were performed against the AGVL in 2010 (Table 1) and six in 2011 (Table 2). In our research all tested products were applied only once in both growth periods of the grapevines. However, the insecticides were not applied completely in accordance with the measures for the prevention and suppression of FD on plants and for managing FD in Slovenia, namely that only two tested products (chlorpyrifos-methyl and chlorpyrifos) are recommended for single applications. In contrast, thiamethoxam can be used up to three times, indoxacarb twice and pyrethrin once to twice within the grapevine growing period. In 2010, the plants were sprayed nine times against grape powdery mildew (Erysiphe necator Schwein.) and grape downy mildew (Plasmopara viticola [Berk. & M.A. Curtis] Berl. & De
Toni); in 2011, the same two pathogens were controlled with seven applications. The fungicides used against grape powdery mildew were sulphur (six times in 2010, five times in 2011), quinoxyfen (once in 2010, twice in 2011), tebuconazol (once in 2010) and tebuconazol + trifloxystrobin (once in 2010). The grape downy mildew was controlled with cyazofamid (once in 2010), mancozeb (twice in 2010, once in 2011), metalaxyl (twice in both years) and folpet (four times in 2010 and 2011).

2.3. Treatments

Only one application of the tested insecticides was performed in each year. In 2010, the application was performed on 23 June between 05:30 h and 09:00 h; at that time, the average air temperature was 16.2°C, and the relative humidity of the air was 74.3%. The grapevines were in the phenological growth stage BBCH 73 (groat-sized berries; the bunches begin to hang). In 2011, the insecticides were applied on 13 June, when the phenological growth stage (Anon. 2001) of the grapevines was BBCH 69 (end of flowering) (Table 3). At the time of spraying, as was done as in former year, the average air temperature was 17.4°C and the relative humidity of the air was 70.1%.

AGVL individuals were first (L1) and second (L2) instars at the time of application in both years. The trial was set in a 0.44-ha vineyard; the plot size in 2010 was 620 m², whereas it was 744 m² in 2011. The experiment was designed in three blocks (plots): in 2010, each plot involved 5 treatments (4 insecticides tested and untreated plants) (Table 1); in 2011, each of the 3 plots had 6 treatments (single applications of 4 insecticides, in which chlorpyrifos-methyl was tested at 2 concentrations, plus untreated plants) (Table 2). All of the treatments within the plots were repeated once, and the area of the single treatment within each plot was 31 m². No other insecticide products were used during the trial. The entire trial was performed in compliance with good plant protection practices and with the EPPO standards (EPPO standards 2004). In both years, the spraying was performed using an air-assisted axial sprayer Zupan ZM 350 (manufacturer and supplier: Zupan d.o.o., Malečnik, Slovenia). The amount of water per hectare was 446 L, the pressure was 10.0 bar, and the speed was 5.11 km/h. We used brown ATR 80 Albuz nozzles.

Several control observations of AGVL were performed prior to the insecticide application to establish the number of hatching nymphs and to determine the exact time for the insecticide application. In 2010, these observations were performed twice: on 10 June when we found 22 nymphs on 100 control leaves, and on 15 June when we found 54 nymphs on 100 control leaves. In 2011, the first control was performed on 1 June (42 nymphs on 100 control leaves), the second was performed on 6 June when we found 46 nymphs on 100 control leaves. In both years, the population of AGVL was estimated to be high, and the damage threshold for the nymphs was exceeded (Lessio et al. 2011a). In both years, the application of insecticides was performed when third (L3) instar nymphs were first found, at approximately one week after the second control observation.

2.4. Estimation of spraying procedure efficacy

Four assessments (counts) of the leafhopper nymphs were performed in both years during the vegetative growth of the vines. The dates of assessments in 2010 and 2011 were 2, 7, 14 and 21 days after the day of insecticide application. In 2010, the counting was conducted on the 25 June, 30 June, 7 July and 14 July; in 2011, the assessments were performed on 15 June, 20 June, 27 June and 4 July. For the efficacy calculation, a modified Garbay et al. (2001) method for grapevine leaves was used. We counted the living leafhopper nymphs on 99 leaves in each plot (33 leaves were collected from the upper, 33 leaves from the middle, and 33 leaves from the lower parts of the plants). The leaves were sampled from 10 randomly selected plants in the centre of each plot. We also determined the developmental stage of the leafhopper nymphs found on the leaves.

2.5. Data analysis

An analysis of variance (ANOVA) was conducted to establish the differences among the average number of

<table>
<thead>
<tr>
<th>Year</th>
<th>Date of nymph counting</th>
<th>BBCH growth stage</th>
<th>Developmental stage of nymphs</th>
</tr>
</thead>
<tbody>
<tr>
<td>2010</td>
<td>25 June</td>
<td>73</td>
<td>L1 and L2; first L3 individuals</td>
</tr>
<tr>
<td></td>
<td>30 June</td>
<td>73–75</td>
<td>L1 and L2; first L3 individuals</td>
</tr>
<tr>
<td></td>
<td>7 July</td>
<td>75–77</td>
<td>L1, L2, L3, and L4; first L5 individuals</td>
</tr>
<tr>
<td></td>
<td>14 July</td>
<td>77</td>
<td>First adults</td>
</tr>
<tr>
<td>2011</td>
<td>15 June</td>
<td>69</td>
<td>L1 and L2; first L3 individuals</td>
</tr>
<tr>
<td></td>
<td>20 June</td>
<td>73</td>
<td>L1, L2, and L3; first L4 individuals</td>
</tr>
<tr>
<td></td>
<td>27 June</td>
<td>75</td>
<td>L1, L2, L3, and L4; first L5 individuals</td>
</tr>
<tr>
<td></td>
<td>4 July</td>
<td>77</td>
<td>L2, L3, L4, and L5; first L1 individuals</td>
</tr>
</tbody>
</table>
nymphs in the different treatments. Before the analysis, each variable was tested for homogeneity of the treatment variances. If the variances were non-homogeneous, the data were log-transformed (Y) before conducting the ANOVA. A Student–Newman–Keuls multiple range test was used to separate the mean differences among the studied parameters in all of the treatments. All of the analyses were performed using the Statgraphics Centurion XVI program (Statgraphics Centurion 2009). The efficacy of the insecticides was calculated using Abbott’s formula (Abbott 1925), and the results are expressed in percentages.

3. Results

3.1. Efficacy of insecticides in 2010

The general, statistical analysis confirmed the significant influence of the insecticide (F = 74.01; df = 4; P < 0.0001) and the date of counting the nymphs (F = 10.22; df = 3; P < 0.0001) on the average number of nymphs per leaf. In contrast, the experimental block (F = 6.10; df = 3; P = 0.0004), the height of leaves (F = 1.49; df = 32; P = 0.0581) and the selected plants did not have any influence (F = 1.34; df = 32; P = 0.5609). The results for the cumulative effect of all four assessments showed that the significantly lowest average number of nymphs per leaf (0.01 ± 0.01) was on the plants treated with thiamethoxam, whereas the significantly highest average number of nymphs per leaf (0.15 ± 0.01) was established on the untreated plants. With regard to efficacy, thiamethoxam, was followed by chlorpyrifos-methyl (0.02 ± 0.00), pyrethrin (0.05 ± 0.01) and indoxacarb (0.09 ± 0.01) (Figure 1). A significantly higher average number of nymphs per leaf (0.09 ± 0.01) was established in the first assessment on the 25 June, whereas such differences were not established in the remaining three assessments (30 June and the 7 July, 0.05 ± 0.01; 15 July, 0.06 ± 0.01). No significant differences were found for the average number of nymphs per leaf between the three heights at which we sampled the leaves: 0.07 ± 0.01 nymphs/leaf in the lower third of leaf biomass and 0.06 ± 0.01 nymphs/leaf in the middle and topmost thirds.

The individual statistical analysis for all four dates of assessment confirmed a significant (F = 74.01; df = 4; P < 0.0001) influence of the insecticide on the number of nymphs on the leaves. All four of the assessments showed the significantly lowest average number of nymphs per leaf on the plants treated with thiamethoxam (Figure 1). Two days after insecticide application, we found an average of 0.02 ± 0.01 nymphs per leaf, but in the following three assessments, the nymphs were no longer present. A rapid effect of decreasing the pest population was also observed with chlorpyrifos-methyl: the efficacy (0.01 ± 0.01) at the first assessment did not differ significantly from thiamethoxam, with similar results between the preparations being confirmed at the third assessment. Among the four insecticides tested, the significantly weakest initial effect was observed with indoxacarb (0.16 ± 0.02); no significant differences were found between this indoxacarb preparation and pyrethrin at the third (0.06 ± 0.01) and the fourth assessments (0.05 ± 0.01).

At all four assessments, thiamethoxam attained more than 90% efficacy, achieving a cumulative efficacy of more than 97% (Table 4). At the first three assessments, chlorpyrifos-methyl showed an efficacy above 90%; this insecticide achieved a cumulative efficacy exceeding 88%, though its efficacy dropped below 70% in the last assessment. In the first days after its application, pyrethrin demonstrated almost 72% efficacy, an efficacy that oscillated between 45% and 64% in the following three assessments. The latter value was approximately 4% higher than the cumulative efficacy of this insecticide. Indoxacarb showed the weakest efficacy against the AGVL, not exceeding 62% at individual assessments, with approximately only 43% cumulative efficacy.

**Figure 1.** Mean number of *Scaphoideus titanus* nymphs per grapevine leaf subjected to five different treatments in 2010. The mean values followed by the same letter do not differ (P > 0.05) according to the Student–Newman–Keuls multiple range test. The bars represent the standard error of the mean number of nymphs per leaf.
3.2. Efficacy of insecticides in 2011

The general statistical analysis confirmed the significant influence of insecticide \((F = 109.49; \text{df} = 5; P < 0.0001)\) and leaf height \((F = 1.35, \text{df} = 32; P = 0.0478)\) on the average number of nymphs per leaf; in contrast, the date of counting the nymphs \((F = 1.16, \text{df} = 3; P < 0.0001)\), plants selected for sampling \((F = 33.49; \text{df} = 2; P < 0.0001)\), and the experimental block \((F = 2.75, \text{df} = 3; P = 0.0612)\) did not show an influence. The results of all four assessments showed the significantly lowest average number of nymphs per leaf \((0.01 \pm 0.00)\) on the plants treated with thiamethoxam and 0.16\% chlorpyrifos-methyl; on the untreated plants, this number \((0.17 \pm 0.01)\) was, as expected, the lowest. Of the two most efficient preparations, chlorpyrifos-methyl at 0.1\% resulted in a significantly higher number \((0.02 \pm 0.00)\) of nymphs per leaf; and a significantly higher average number of nymphs per leaf \((0.09 \pm 0.01)\) resulted from pyrethrin application. Chlorpyrifos was significantly more effective because we found an average of \(0.05 \pm 0.01\) nymphs/leaf on the grape leaves treated with this preparation (Figure 2).

Between the dates of assessment, we did find significant differences in the average number of nymphs per leaf, counting an average of \(0.06 \pm 0.01\) nymphs per leaf on 15 June, 27 June and 4 July; on 20 June, this number was \(0.05 \pm 0.00\). Significant differences in the average number of nymphs per leaf were established between the three heights at which the leaves were sampled: the lowest third of leaf biomass exhibited the significantly highest number \((0.08 \pm 0.01)\), whereas the topmost third had significantly the lowest \((0.03 \pm 0.00)\) number of nymphs per leaf. Each sampled leaf in the middle third showed an average of \(0.06 \pm 0.00\) nymphs.

The individual statistical analysis at all four assessment dates confirmed the significant \((F = 109.49; \text{df} = 5; P < 0.0001)\) influence of the insecticide on the number of nymphs on the leaves. At the first two assessments, no significant differences between the average number of nymphs per leaf were found for the most efficient insecticides, thiamethoxam \((0.01 \pm 0.01\) for both assessments) and chlorpyrifos-methyl at both concentrations \((0.02 \pm 0.01\) and \(0.01 \pm 0.00\) at the lower or \(0.01 \pm 0.00\) and \(0.01 \pm 0.01\) at the higher concentration). Conversely, at the fourth assessment, the significantly lowest average number of nymphs per leaf \((0.00 \pm 0.00)\) was found with thiamethoxam (Figure 2). At all four assessments, pyrethrin had the significantly weakest effect, and, in the first assessment, we did not confirm significant differences between its effect \((0.07 \pm 0.01)\) and the effect of chlorpyrifos.

### Table 4. Efficacy of single applications of four insecticides tested against *Scaphoideus titanus* nymphs in 2010 using Abbott's formula.

<table>
<thead>
<tr>
<th>Insecticide</th>
<th>Dose</th>
<th>25 June</th>
<th>30 June</th>
<th>7 July</th>
<th>14 July</th>
<th>Average</th>
</tr>
</thead>
<tbody>
<tr>
<td>Actara 25 WG</td>
<td>200 g/ha</td>
<td>90.48</td>
<td>100.00</td>
<td>100.00</td>
<td>100.00</td>
<td>97.62</td>
</tr>
<tr>
<td>Reldan 22 EC</td>
<td>1000 mL/ha</td>
<td>95.24</td>
<td>100.00</td>
<td>90.91</td>
<td>69.23</td>
<td>88.85</td>
</tr>
<tr>
<td>Kenyatox verde</td>
<td>800 mL/ha</td>
<td>71.43</td>
<td>64.28</td>
<td>45.45</td>
<td>61.54</td>
<td>60.68</td>
</tr>
<tr>
<td>Steward</td>
<td>125 mL/ha</td>
<td>23.81</td>
<td>42.86</td>
<td>45.45</td>
<td>61.54</td>
<td>43.42</td>
</tr>
<tr>
<td>Untreated</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
</tbody>
</table>

![Figure 2](image_url)

Figure 2. Mean number of *Scaphoideus titanus* nymphs per grapevine leaf subjected to five different treatments in 2011. The mean values followed by the same letter do not differ \((P > 0.05)\) according to the Student–Newman–Keuls multiple range test. The bars represent the standard error of the mean number of nymphs per leaf.
(0.06 ± 0.01). The effects of chlorpyrifos 25 CS in the third (0.05 ± 0.01) and the fourth assessments (0.05 ± 0.01) did not significantly differ from those of 0.1% chlorpyrifos-methyl (0.03 ± 0.01 for both assessments); at the second assessment (0.03 ± 0.01), it also did not differ from thiamethoxam and 0.16% chlorpyrifos-methyl (both at 0.01 ± 0.01).

Thiamethoxam at all four assessments attained more than 94% efficacy, with more than 96% cumulative efficacy (Table 5). Chlorpyrifos-methyl at a 0.16% concentration at the first three assessments exceeded 94% efficacy, and this insecticide cumulatively exceeded 92% efficacy, even though its efficacy dropped below 82% at the last assessment. The effect of the same preparation at a 0.1% concentration was slightly weaker, as its efficacy already dropped below 83% at the third assessment; cumulatively, the preparation reached 87% efficacy. The efficacy of chlorpyrifos at the four assessments oscillated between 68% and 81%; cumulatively, the preparation produced 73% mortality of the nymphs. In 2011, pyrethrin showed the weakest insecticidal effect, though its initial efficacy (64%) halved (31%) in less than three weeks; cumulatively, pyrethrin did not exceed 45% efficacy.

### Table 5. Efficacy of single applications of four insecticides tested against *Scaphoideus titanus* nymphs in 2011 using Abbott’s formula.

<table>
<thead>
<tr>
<th>Insecticide</th>
<th>Dose</th>
<th>15 June</th>
<th>20 June</th>
<th>27 June</th>
<th>4 July</th>
<th>Average</th>
</tr>
</thead>
<tbody>
<tr>
<td>Actara 25 WG</td>
<td>200 g/ha</td>
<td>96.15</td>
<td>97.06</td>
<td>94.29</td>
<td>98.36</td>
<td>96.47</td>
</tr>
<tr>
<td>Reldan 22 EC</td>
<td>1600 mL/ha</td>
<td>97.44</td>
<td>94.12</td>
<td>95.71</td>
<td>81.97</td>
<td>92.31</td>
</tr>
<tr>
<td>Reldan 22 EC</td>
<td>1000 mL/ha</td>
<td>91.03</td>
<td>95.59</td>
<td>82.86</td>
<td>78.69</td>
<td>87.04</td>
</tr>
<tr>
<td>Pyrinex 25 CS</td>
<td>1000 mL/ha</td>
<td>71.79</td>
<td>80.88</td>
<td>71.43</td>
<td>68.85</td>
<td>73.24</td>
</tr>
<tr>
<td>Kenyatox verde</td>
<td>800 mL/ha</td>
<td>64.10</td>
<td>45.59</td>
<td>38.89</td>
<td>31.15</td>
<td>44.93</td>
</tr>
<tr>
<td>Untreated</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
</tbody>
</table>

In both years, Actara 25 WG had the best effect: in the first year, the efficacy reached 100% at three of the four assessments, whereas its efficacy was not less than 94% at all of the assessments in the second year. Actara 25 WG also had the highest cumulative efficacy at 96–97%. The good effect of this preparation is due to the systemic active substance thiamethoxam, which metabolises slowly in plants and remains for longer periods (Roberts and Hutson 1999; Perello and Dal Bello 2011).

Satisfactory effect against the leafhopper nymphs was also exhibited by the insecticide Reldan 22 EC, which had higher than 90% efficacy in the first three assessments in both years. Such a high efficacy was maintained for 2 weeks, whereas the efficacy of this preparation diminished at the last assessment: it dropped to 69% in the first year and to 79–82% in the second year. Our results accord with those of Del Carlo et al. (2002) who reported more than five-fold decrease in chlorpyrifos-methyl content in vine leaves at 3 weeks after application. Cumulatively, Reldan 22 EC had 87–92% efficacy, and the higher percentage of efficacy was reached at the higher (0.16%) concentration, which was tested only in the second year of the study. The good initial effect of this preparation with the active substance chlorpyrifos-methyl, which acts as an inhibitor of acetylcholinesterase (Roberts and Hutson 1999), was expected. Indeed, Del Carlo et al. (2002) reported an essentially unchanged chlorpyrifos-methyl concentration in vine leaves at 6 days after treatment. Furthermore, the results of a related study also showed a good effect of organic phosphate insecticides on the nymphs of *Scaphoideus titanus* (Bosio et al. 2004). The results of our research confirm lesser persistence of chlorpyrifos-methyl when compared with thiamethoxam. Therefore, we suggest this compound be used in the AVGL treatment schedule during the first week of July, a time when this stage BBCH 73 on the 25 June 2010, we counted an average of 21 nymphs on 100 grapevine leaves; while on 15 June 2011 (BBCH 69) we found 20 nymphs. These results confirm the presence of extensive populations of the leafhopper in the experimental vineyard in both years, exceeding the damage threshold of the nymphs, which is 0.02 nymphs/5 leaves/plant according to Lessio et al. (2011a).

In 2011, pyrethrin showed a high initial efficacy of 64%, which dropped to 69% in the first year and to 79–82% in the second year. Our results accord with those of Del Carlo et al. (2002) who reported more than five-fold decrease in chlorpyrifos-methyl content in vine leaves at 3 weeks after application. Cumulatively, Reldan 22 EC had 87–92% efficacy, and the higher percentage of efficacy was reached at the higher (0.16%) concentration, which was tested only in the second year of the study. The good initial effect of this preparation with the active substance chlorpyrifos-methyl, which acts as an inhibitor of acetylcholinesterase (Roberts and Hutson 1999), was expected. Indeed, Del Carlo et al. (2002) reported an essentially unchanged chlorpyrifos-methyl concentration in vine leaves at 6 days after treatment. Furthermore, the results of a related study also showed a good effect of organic phosphate insecticides on the nymphs of *Scaphoideus titanus* (Bosio et al. 2004). The results of our research confirm lesser persistence of chlorpyrifos-methyl when compared with thiamethoxam. Therefore, we suggest this compound be used in the AVGL treatment schedule during the first week of July, a time when this
preparation can also suppress the grape berry moth (Clysia ambiguella Hübner) and European grapevine moth (Lobesia botrana [Denis & Schiffermüller]), which are two of the most directly harmful insect species in Slovenian vineyards (Matis and Miklavc 2003) and others throughout Europe (Moshos 2006; Sharon et al. 2009).

The efficacy of the insecticide Kenyatox Verde, which is licensed for use in organic viticulture production, was relatively low in both years. The active ingredient (pyrethrin) of mentioned product was obtained from Dalmatian chrysanthemum (Chrysanthemum cinerariifolium [Trev.] Bocc.) (Lessio and Alma 2006; Lessio et al 2011a). In the first year of our study, its cumulative efficacy was 61% and only 45% in the second year. Due to the chemical properties of pyrethrin, which include chemical degradation and rapid photo-oxidation in sunlight (Roberts and Hutson 1999), we recommend that this preparation be applied in the evening and that the procedure be repeated several times (Lessio et al 2011b) when nymphs appear. Under these circumstances, we can expect satisfactory results with this insecticide, as previously reported by Caruso and Mazio (2004).

The efficacy of the insecticide Steward in the first year of the research did not exceed 62% at any of the assessments, whereas the cumulative result showed little more than 43% efficacy. The efficacy of the preparation was, thus, the weakest among the tested preparations, and we did not include it in 2011. The results of our research cannot confirm the results of Ioriatti et al. (2006) who established that indoxacarb was effective on young Pandemis heparea (Denis and Schiffermüller) caterpillars, as it was effective on older nymphs in our research. However, we can confirm the results by Bosio et al. (2004) who reported an insufficient effect of indoxacarb on AGVL.

In the second year of the study, we tested the insecticide Pyrinex 25 CS (chlorpyrifos as the active substance) rather than Steward. Pyrinex 25 CS was slightly weaker in comparison to Actara 25 WG and Reldan 22 EC: its efficacy at the individual assessments oscillated between 71% and 81%, with a cumulative efficacy of 73%. It is known that the quantity of the active substance chlorpyrifos applied to leaves can be quickly reduced, primarily due to evaporation (Roberts and Hutson 1999; Leistra et al. 2006), which could have slightly diminished its efficacy in our experiment.

Recent work shows that the AGVLs is probably not the only insect that can spread FD, as it was established that the European Lantern Fly (Dictyophara europaea [L.]) can transmit FD from an infected old man’s beard plant to a grapevine (Filippin et al. 2009). Molecular analyses also identified strains of FD in Japanese Leafhopper (Oriusitus ishidae [Matsumura]), the importance of which in the epidemiology of grapevine yellows is completely unknown to date (Mehle et al. 2011). The results of systematic research show that, in Slovenia, FD is an endemic phytoplasma which became noxious to grapevines only after the AGVL had colonised in the area.

We can conclude that the phytoplasma diseases of grapevines are expanding and, as such, require close control and study (Boudon-Padieu 2005). FD is a very important disease in viticulture (Margaria and Palma-no 2011a, 2011b) and the most dangerous in the group of grapevine yellows (Boudon-Padieu 2005). The risk of vineyards becoming infected with FD is also closely connected to the likelihood of transmission by vectors (Boudon-Padieu 2005). Thus, one of the key measures for preventing the spread of FD is the suppression of AGVL using insecticides. The results of our research show the highest efficacy with the neonicotinoid thiamethoxam and the organic phosphate insecticide chlorpyrifos-methyl (Lessio et al. 2011b). Thus, we recommend both insecticides for use in conventional and in integrated viticulture production.

Further investigations should be performed to test the hypothesis that managing Scaphoideus titanus with insecticides reduces the spread of FD. Indeed, these data are crucial to make appropriate recommendations about the use of the most effective insecticides against the pest.

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