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Migration of *Scaphoideus titanus* Ball from the surrounding vineyards into the nursery

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ABSTRACT

A one-year study of the mobility of leafhopper *Scaphoideus titanus* Ball (ST), vector of Flavescence dorée phytoplasma, was conducted in 2009 and included three Slovenian nurseries in the Primorska winegrowing region, as well as two Italian and one Swiss nursery, and was aiming at evaluating the risk of spreading ST with vine propagating material. The presence of ST was monitored by the means of yellow sticky traps that were placed at three distances in the nurseries and the nearby vineyards on three parallel lines (replicates). The traps were equidistantly placed from the nurseries' and vineyards' borders in order to determine the possible distance gradient. The monitoring started in the latest nymphal stages (middle of June) and terminated in the first decade of October. Insecticide treatments with deltamethrin (Decis 2.5 EC) were applied in nurseries, according to the instructions for use and recommendations for suppression of ST. On all Slovenian plots in the vineyards, the population level of ST was high (1796 adults in average / vineyard or 18 adults per trap / week), which indicated a great migration potential of the vector towards the nurseries. Catches were very low (54 adults in average / nursery or 0.4 adults / trap / week) on two nurseries' plots (Lože and Slap), although high numbers of ST adults were caught in the adjacent untreated vineyards. On the Poreče nursery plot, insecticide treatments were not sufficient to avoid high numbers of ST catches (in total 481 adults or 3 adults / trap / week). This could be related to the sub-optimal timing of the first insecticide treatment and/or migration of adults from two nearby untreated vineyards located at a distance of 70 and 150 m. The distance between the nurseries and vineyards did not affect ST catches in the nurseries.

Key words: *Scaphopideus titanus*, Flavescence Dorée, migration, nursery

IZVLEČEK

MIGRACIJA AMERIŠKEGA ŠKRŽATKA (*Scaphoideus titanus* Ball) IZ BLIŽNJIH VINOGRADOV V TRSNICO

Z namenom preučitve nevarnosti širjenja prenašalca zlate trsne rumenice z razmnoževalnim materialom vinske trte smo v letu 2009 izvedli raziskavo mobilnosti ameriškega škržatka (*Scaphoideus titanus* Ball) (ST) v treh trsnicah v vinorodni deželi Primorska. V manjšem obsegu je bil poskus izveden tudi pri partnerskih institucijah v Italiji in Švici. Zastopanost in nalet ST smo ugotavljali z rumenimi lepljivimi ploščami, ki so bile postavljene na treh različnih razdaljah od roba trsnice in vinogradov v 3 vzporednih ponovitvah. S postavitvijo lepljivih plošč v enakomernih razmakih od roba trsnice in vinogradov v globino smo želeli ugotoviti morebitni gradient naleta. Z monitoringom naleta smo začeli, ko je bil škodljivec v 5. stadiju ličinke (sredi junija) in ga zaključili v prvi dekadi oktobra. Tretiranje z deltametrinom (Decis 2.5 EC) je bilo izvedeno v trsnici, skladno z navodili za uporabo insekticida in s priporočili za zatiranje ST. Populacija ST v vinogradih je bila na vseh treh lokacija v Sloveniji visoka (v povprečju 1796 odraslih osebkov / vinograd oziroma 18 osebkov / ploščo / teden), kar je predstavljalo velik migracijski potencial prenašalca proti trsnicam. Kljub visoki populaciji ST v bližnjih vinogradih je bilo število ujetih ST v trsnicah na lokaciji Slap in Lože nizko (v povprečju 54 osebkov / trsnico oziroma 0,4 osebka / ploščo / teden). Število ujetih odraslih osebkov na lokaciji Poreče je bilo kljub tretiranju z insekticidom visoko (skupno 481 osebkov ali 3 osebki / ploščo / teden). Vzrok za to gre iskati v neustreznem času prvega tretiranja z insekticidom ali v dodatnem naletu ST iz bližnjih vinogradov, ki so bili oddaljeni 70 in 150 m. Razdalja med trsnico in vinogradom ni vplivala na število ujetih ST v trsnici.

Ključne besede: ameriški škržatek, zlata trsna rumenica, migracija, trsnica

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1 INTRODUCTION

The efficacy of insecticide treatments against *Scaphoideus titanus* Ball (ST) (Hemiptera, Cicadellidae), the vector of Flavescence Dorée phytoplasma (FD), in nurseries depends on the term of treatment regarding the vector's development stage, on the proportion of its mortality, insecticide residues on plants and on the migration of adults from the surrounding vineyards. The previous pesticide registration against ST considered only the situation in spacious vineyards, but did not make any allowances for small-sized nurseries (Boudon-Padieu, 2003). This aspect has to be considered also in adopting a suitable insecticide treatment program. Also the distances between vineyards and nurseries and the mobility range of ST are crucial for determining the risk of spread of ST with the vine grafted plants.

ST, a Nearctic leafhopper, was introduced into Europe from the North America before 1958. This univoltine species overwinters in the egg stage. Eggs are deposited into the bark of two or more years old vine branches (Vidano, 1964), rarely also in one-year branches. Nymphs start emerging in the second half of May and adults occur in mid-summer (in the Slovenian climate, at the end of June). They feed exclusively on *Vitis* spp. and cause indirect damages by transmitting FD from infected vines. Although insecticide treatment against FD vectors is mandatory to prevent the progression of the disease, new contaminations may occur with the movement of infested planting material or ST migrations (Boudon-Padeau et al, 2005). The movement of ST is rather limited to the vine's canopy and is not very likely to fly for more than 2.50 m in high (Lessio F., Alma A., 2004). Seasonal and daily fly of ST is partly influenced by increased temperatures and humidity (Lessio F., Alma A., 2004).

In order to determine the risk of FD vector-spreading with the planting material, a one-year study of the mobility of ST was conducted in 2009 in three nurseries in the Slovenian Primorska winegrowing region. The occurrence of ST was monitored using yellow sticky traps. They were placed at three distances in the nurseries and the nearby vineyards in three parallel lines (replicates). The traps were equidistantly placed from the nurseries' and vineyards' borders in order to determine the possible distance gradient. The main part of this study was carried out in Slovenia, where ST is widely spread in all winegrowing regions and the population rates in some vineyards were still fairly large. A small-scale experiment was also conducted in Italy and Switzerland.

The objective of the work was to evaluate the effectiveness of insecticide treatments against ST adults in nurseries in order to reduce the risk of immigration of adults from the nearby vineyards.

2 MATERIALS AND METHODS

2.1 Slovenian trials

In Slovenia, the monitoring was carried out in 2009 at three locations in the Primorska winegrowing region, winegrowing district of Vipava valley (Slap - 45° 50' 73,55" N, 13° 56' 31,57" E, 99 m altitude; Lože - 45° 49' 52,80" N, 13° 56' 43,68" E, 109 m altitude; Poreče – 45° 50' 57,72" N, 13° 56' 20,30" E, 95 m altitude) with four evaluating plots. Distances between the vineyards and nurseries were different, because the major part of vineyards were grown on hilly sides, while nurseries were situated along the river Vipava. Distances between nurseries and vineyards included in experimental plots are shown in the Table 1.

Table 1: Sizes of experimental plots and distances between them

Location (size)	Vineyard nr. 1	Vineyard nr. 2
1. Slap		
Nursery (0.8 ha)	126 m	188 m
Vineyard nr. 1 (0.65 ha)	/	
Vineyard nr. 2 (2.0 ha)		/
2. Poreče		
Nursery (4.5 ha)	150 m	70 m
Vineyard nr. 1 (0.5 ha)	/	
Vineyard nr. 2 (1.1 ha)		/
3. Lože		
Nursery nr. 1 (2.5 ha)	18 m	
Nursery nr. 2 (0.6 ha)		18 m
Vineyard nr. 1 (2.2 ha)	/	
Vineyard nr. 2 (0.43 ha)		/

Except for the second location where a tree-barrier was grown between the nursery and vineyards, no other spatial obstacles were noticed. Yellow sticky traps (Unichem, 17 x 24 cm) were placed on the plots on 17 June. The traps were placed on the nurseries' and vineyards' edges and in at least three parallel lines (replications). They were placed on the inner side of borders, considering the distance of 10 m between traps. Replications were designated in order to cover the whole area of the vector's mobility; distances between them were 20 m.

In vineyards, planted with varieties Merlot, Sauvignon and Cabernet sauvignon, traps were attached with a wire inside the leaf canopy between 1.3 and 1.8 m altitude, depended on the growing type. In nurseries, traps were fixed on 0.5 m long iron stakes. The traps were checked weekly and replaced every three weeks. The monitoring was finished on 8 October. In nurseries, two insecticide treatments against ST were applied. Application data are given in Table 2.

Table 2: Data of insecticide treatments in nurseries in Slovenian experiment in 2009

Plot	First treatment	Second treatment
Slap	13. 7.	6. 8.
Poreče	9. 7.	31. 7.
Lože	20. 7.	7. 8.

For all treatments, the insecticide deltametrin (Decis 2.5 EC) in dosage 0.5 L of the product per hectare was used.

Data analysis

Data were analyzed using nested analysis of variance, where time of sampling were nested within trap distance from border, distance were nested within a plot use (nursery and vineyard), and plot use were nested in location. With this analysis the amount of variability contributed by each factors were estimated. Analysis was performed using STATGRAPHIC 5.0 statistical software.

A small-scale experiment was conducted also in Switzerland (one plot) and Italy (two plots). In order to facilitate trial activities and compare results between the three countries, a common protocol had been established before starting the activities.

2.2 Italian trials

The experiment performed by the Italian partner (IVC) was done on two plots (Pordenone - 46° 2' 48" N, 12° 52' 23" E, 70 m altitude and Verona - 45° 33' 19" N; 10° 46' 42" E; 200 m altitude). The trials started on 16 June 2009 in Verona and on 30 June 2009 in Pordenone, and were finished on 1 October. Insecticide treatments were applied in nurseries and in vineyards. The period of application as well as the number of treatments was different for the locations, as presented here below:

Pordenone
Vineyard - four treatments (18 April – deltametrin, 23 May indoxacarb -, 26 June and 21 July chlorpyrifos ethyl). Nursery - three treatments (deltametrin, chlorpyrifos ethyl).

Verona
Vineyard – one treatment (chlorpyrifos methyl – 14 July). Nursery - seven treatments (20 May - fenazaquin, 29 May - spinosad, 10 June - chlorpyrifos ethyl, 25 June - spinosad, 10 July, 30 July and 20 August - chlorpyrifos ethyl)

2.3 The Swiss trial

A small-scale experiment was carried out on one plot (Anieres – 6° 13' 23,5" E, 46° 16' 24,9" N, 412 m altitude). Traps were set on 3 July 2009 and the trial finished on 5 October. Two insecticide treatments against the leafhopper were applied only in the nursery: on 13 July and 27 July. The insecticide used was lambda-cyhalotrine (Kararate with Zeon Technology (9.43% 100 g/l) in dosage 1.2 l/ha.

3 RESULTS

3.1 Slovenian trials

On all three Slovenian plots, the population level of ST in vineyards was very high, which indicates a high immigration potential of the vector towards the nurseries. It should also be stressed that all Slovenian experimental locations are exposed to frequent and strong NO-SW winds that may influence the movement direction and distance of ST.

Table 3: Results from nested ANOVA analyses of ST catches

Source	Sum of squares	Df	Mean square	F		Variance component	% variance component
Location	6640.6	2	3320.30	0.12		0.00	0.00
Manner of land use	82043.1	3	27347.70	23.34	**	116.86	28.75
Trap distance from border	16403.7	14	1171.69	1.90	*	8.64	2.13
Time of sampling	172895.0	280	617.48	3.34	**	96.14	23.65
Error	194083.0	1050	184.84			184.84	45.47

Significance of F values indicated as follows: *$p < 0.05$; **$p < 0.01$

The results of nested ANOVA indicate that manner of land use and time of sampling contributes significantly ($p < 0.01$, F statistic) to the total variance of number of ST. Manner of land use contribute 28.75% of the total variation, and time of sampling contributed 23.65%. The large variance component for the error (45.47%) indicates large variation among replicates within time of sampling. Trap distance from border is significant at $p < 0.05$, but not at $p < 0.01$ and contribute only 2.13% of the total variation.

Slap location

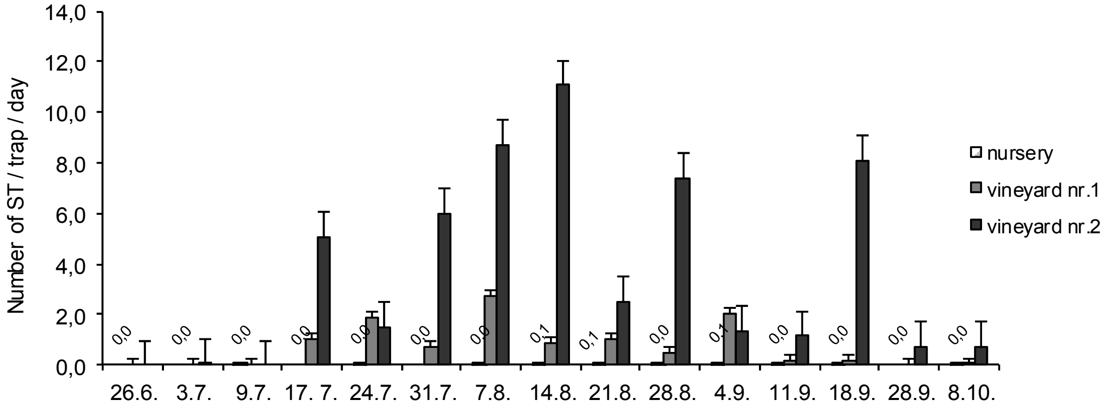

Figure 1: Number of captured adults of ST per trap and day in location Slap

At the first location, the number of captured ST in the nurseries was very low (34 adults in total, 0.0 adults / trap / day) in contrast to the captures in the two vineyards in the neighbourhood, where the average number of adults per trap per day was 0.7 for the vineyard No. 1 and 3.6 for the vineyard No. 2. In this case, the high population rate of ST in vineyards didn't imply the population of ST in the nursery; the reason might be fairly long distances between the nursery and vineyards (126 m and 188 m). Also the position of traps regarding the distance from the nursery edge didn't show differences. Insecticide treatments didn't show reduction of population level, especially the second application of 5. August.

Poreče locatioin

The trial conducted on the second plot revealed a high number of ST captures (481 adults in total or 0.4 adults / trap / day in average) in the nursery, with a pick at the end of July (49% of all captures in the nursery). Since the nursery was treated two times with an insecticide, the high number of ST captures could be explained either by the movement of ST from the neighbourhood untreated vineyards (Vineyard nr. 2) or by insufficient efficacy of the first treatments. After the second treatment (31. July), the population rate in the nursery dropped considerably, but still relatively numerous captures afterwards suggest that one or two additional treatments against ST would be necessary.

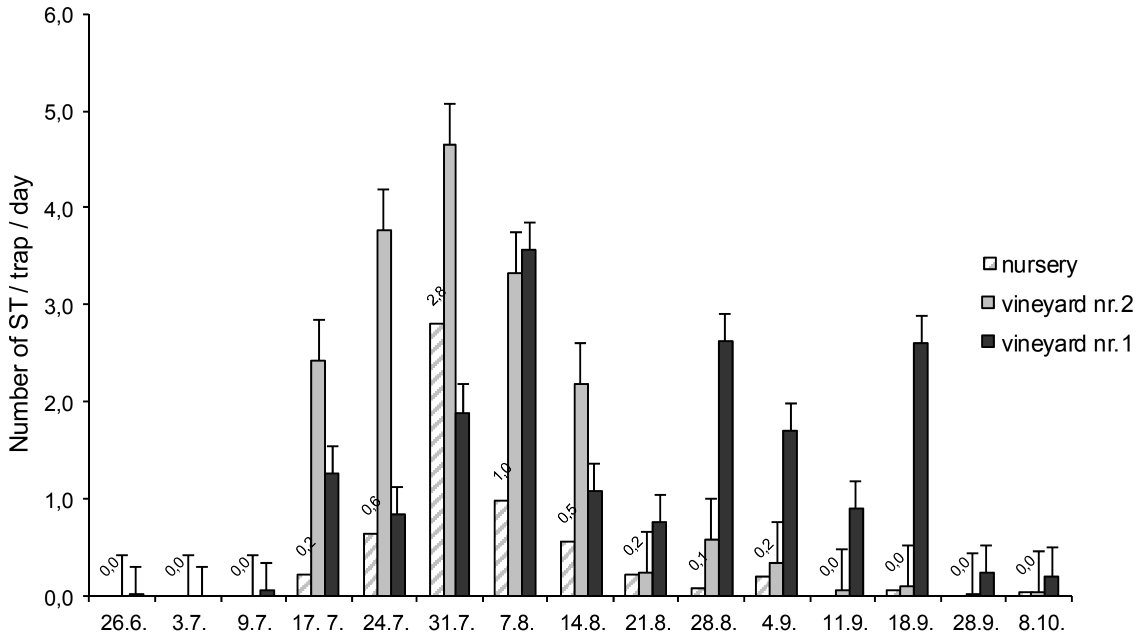

Figure 2: Number of captured adults of ST per trap and day in location Poreče

The highest number of adults was captured on the traps placed 20 meters from the nursery border (182 adults in total or 0.6 adults / trap / day in average). On traps placed on the border line, the mean capture was 0.4 adults / trap /day. 45% of all adults captured at the end of July were found 20 meters from the border line.

Lože location

At this location, two different plots were evaluated because of the different position of the vineyards. In contrast to the previous locations, in this case the distance between the nursery and vineyard was quite short (18 m) so that the possibilities for the movement of ST from the adjacent vineyard to the nursery were much more reliable.

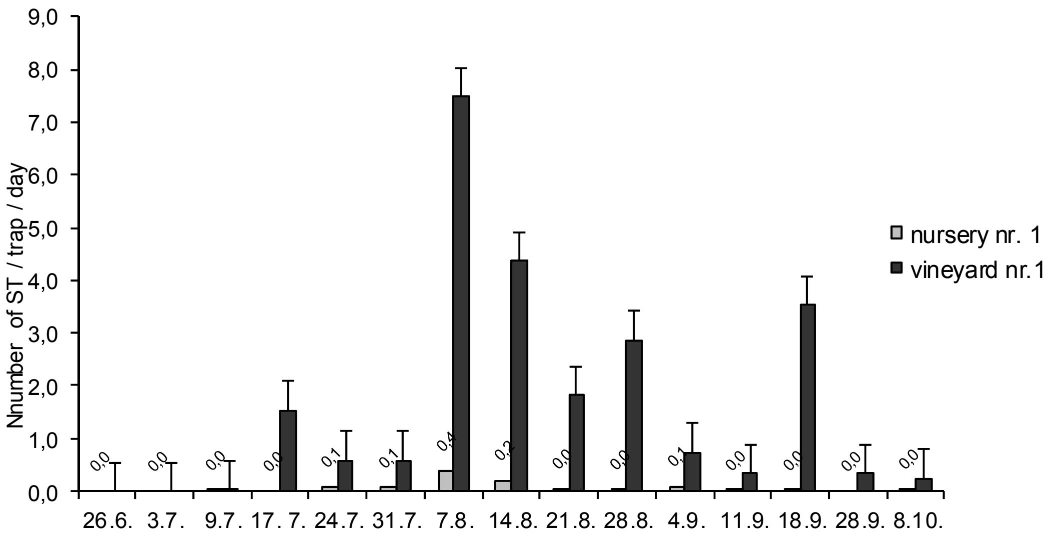

Figure 3: Number of captured adults of ST per trap and day in Lože nursery nr.1

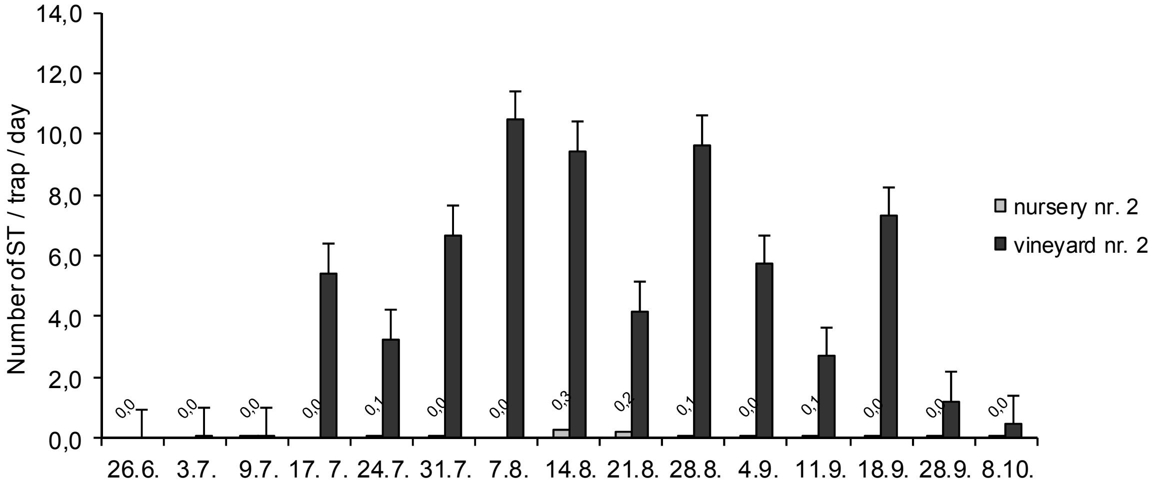

Figure 4: Number of captured adults of ST per trap and day in Lože nursery nr.2

Data gathered show a low number of ST captures in the nursery plot (64 adults in the nursery nr. 1 and 63 adults in the nursery nr. 2 in total; 0.1 adults / trap / day in average), in spite of a very high immigration potential from the vineyards in the nearest neighbourhood. It seems likely that in this case, insecticide treatments kept the ST population in the nursery at a relatively low level. Nevertheless, an additional treatment in mid-August would be necessary. Considerable differences in the population levels between vineyard No. 1 (1.6 adults / trap / day) and vineyard No. 2 (4.4 adults / trap / day) could be explained by irrigating technology that was applied in the vineyard No. 2.

Gradient distribution data revealed the same situation on the plot No. 1 as in other nurseries (the highest number of adults was captured 10 m and 20 m from the border). The gradient distribution for the nursery No. 2 shows just the opposite situation, where the highest number of adults was captured at the border line.

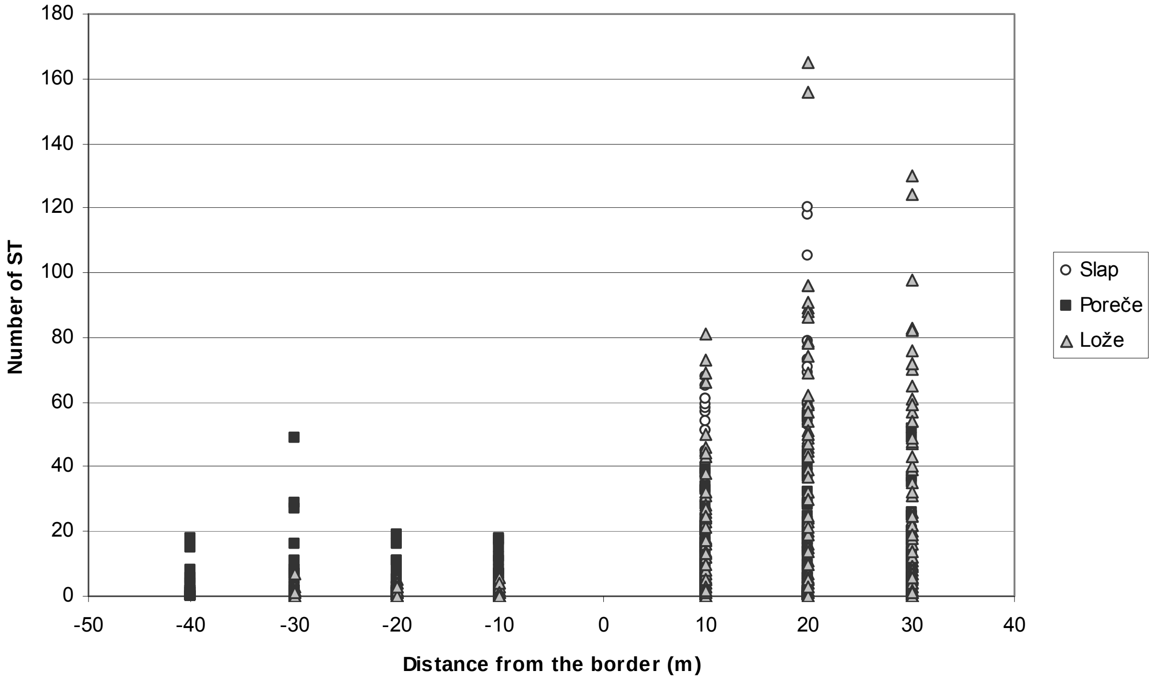

Figure 5: Scatterplot for the number of ST versus the distance of traps from the border

The number of ST catches dropped rigorously at traps posted 30 m from the border in vineyards towards the nursery. As it is shown in Figure 5, the highest number of ST was captured 30 m away from the nursery border.

3.2 Italian trials

In trials done by the Italian partner, no relevant data on mobility was obtained because only 1 ST male adult was captured 30 meters from the border of the vineyard. The situation was more rigorous at the location of Verona, where no catches were detected. The absence of ST catches in nurseries is the consequence of numerous insecticide treatments applied both in vineyards (up to four) and nurseries (up to seven).

3.3 Swiss trials

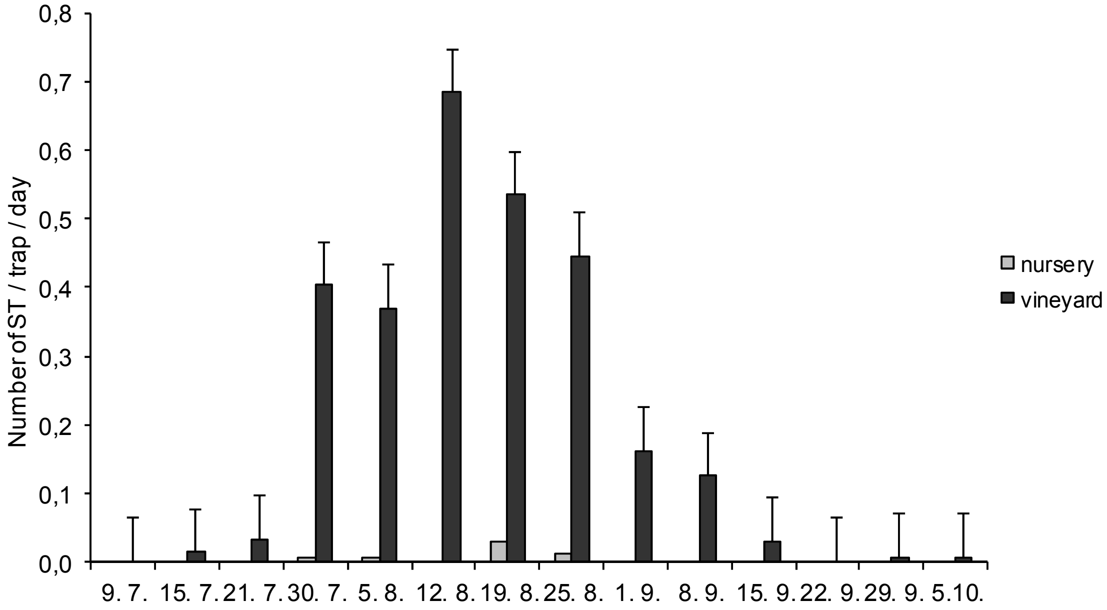

Figure 6: Number of captured adults of ST per trap and day in location Anieres

In Switzerland, two insecticide applications had an important impact on adults' catches in the nursery (98% less catches compared to the vineyard). However, a slight peak of catches could be observed in August, three weeks after the last insecticide application, suggesting immigration from the vineyard. In the nursery, catches on border traps facing the vineyard represented 25% of all captured adults and the highest number of adults was observed on the trap located 70 m from the border (37.5% of catches). In the vineyard, the numbers of captured adults on border traps facing the nursery were the lowest, while they were the highest 10 meters from the border (16% of all catches). However, from the traps-distance-distribution point of view, adult catches did not show a significant gradient.

Sex-ratio of the captured adults was determined in the nursery in order to check the hypothesis if females were more mobile than males. Among the total of 8 catches in the nursery, only 1 adult was identified as a female.
A side differentiation of trap-catches was also done in the nursery to determine if the mobility of ST could be greater than in the vineyard. The results showed that only 2 adults were captured at the vineyard side.

4 DISCUSSION

Immigration risk of ST from nearby vineyards and the efficacy of insecticide treatments in nurseries were monitored. On two Slovenian locations, catches were fairly low in the nursery plots although high numbers of ST adults were caught in the adjacent untreated vineyards. On the third plot (Poreče), insecticide treatments were not sufficient to avoid important catches in the nursery. This could be related to the sub-optimal timing of the first insecticide treatment and/or immigration of adults from two nearby untreated vineyards located at a distance of 70 and 150 m. Population rate in vineyards showed a small influence on the population of ST in nurseries. That could be explained by the preference of ST to remain in vineyards, where leaf canopy provides for easier feeding and reproduction of the vector. Nevertheless, vineyards present the source of ST infection for nurseries.

Insecticide treatments reduced sufficiently the ST population in nurseries in almost all trials except on one Slovenian plot. This case suggests that additional insecticide treatments are necessary if the immigration pressure of ST from adjacent vineyards is high (Boudon-Padieu E., 2005). With four to seven insecticide applications in Italian trials, Italians managed to completely suppress the population of ST in nurseries and vineyards and eliminated the risk of vectors' immigration. Swiss trials showed that males seem to be more mobile than females and that they are trapped in higher numbers in nurseries. If this behaviour is confirmed, the risk of egg-laying on propagation material could be considered as very low.

The variance components analysis shows that the location of the nursery doesn't contribute to the variability of the ST number. About 28% of the variability came from different uses of the plot (nursery vs. vineyard) and 23% from the time of capture of ST. Only 2% of the variability came from the position of catches. Regarding the distance between the vineyards and the nursery, we can conclude that the population of ST in nurseries doesn't depend on this factor but rather on the season and the use of the plot where the technology (insecticide treatment) is also included. Irrespective of the vineyard being 18 m or 126 m far from a nursery, it could be the source of infestation.

As all Slovenian experimental plots were subject to continuous strong wind we conclude that abiotic factors can also enhance the movement range of ST adults towards nurseries.

Main conclusions

- Distance between the nursery and nearby vineyard lower than about 200 m didn't influence the mobility of ST from the vineyard to the nursery.
- Applied suppression technology against ST, particularly proper timing of insecticide treatments, affected the population level of ST in the nursery.
- The population level of ST adults in vineyards did not seem to have a direct effect on the mobility range.
- A decreasing flight gradient of ST from vineyards to nurseries could not be established at distances monitored in these trials.

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