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(Article begins on next page)



# UNIVERSITÀ DEGLI STUDI DI TORINO

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# **Integrated management of *Thrips tabaci* (Thysanoptera: Thripidae) on onion in north-western Italy: basic approaches for supervised control**

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## **Abstract**

**BACKGROUND:** *Thrips tabaci* Lindeman (Thysanoptera: Thripidae) is a major pest on onion, *Allium cepa* L., worldwide. In 2010, research was conducted in a commercial onion field in north-western Italy in order (i) to evaluate the efficacy of different insecticides and of the SAR activator acibenzolar-*S*-methyl, (ii) to correlate thrips infestation levels with bulb size and weight at harvest and (iii) to implement a reliable thrips sampling method. Efficacy of the three active ingredients spinosad, lambda-cyhalothrin and acibenzolar-*S*-methyl on local thrips populations were also evaluated in laboratory bioassays.

**RESULTS:** During field surveys, the highest and the lowest thrips infestations were observed in plots treated with lambda-cyhalothrin and with spinosad and acibenzolar-*S*-methyl respectively. The effectiveness of spinosad was also confirmed in laboratory bioassays. At harvest, bulb size and weight did not significantly differ between treatments. A high correlation with visual inspection made plant beating a suitable sampling method for routine practice, enabling a good estimate of thrips infestation.

**CONCLUSION:** Damage caused by thrips is often not severe enough to warrant the frequent pesticide applications the crops receive in north-western Italy. The use of spinosad and acibenzolar-*S*-methyl is suggested as an alternative to conventional insecticides for the preservation of natural enemies.

## **1 INTRODUCTION**

Onion thrips *Thrips tabaci* Lindeman (Thysanoptera: Thripidae) is one of the most serious pests of onion (*Allium cepa* L.) and other *Allium* spp. in many parts of the world.<sup>1-5</sup> Heavy infestations of *T. tabaci* can sometimes kill young plants,<sup>6</sup> but thrips feeding on leaves can usually reduce onion yield. Both adult and larval stages can be responsible for economic damage by reducing onion bulb size and weight<sup>7</sup> and causing yield losses of up to 50%.<sup>8</sup> In addition, *T. tabaci* is the vector of Iris yellow spot virus (IYSV), genus *Tospovirus* (Bunyaviridae), a severe and widespread disease infecting onion, leek, iris and wild *Allium* species.<sup>9-12</sup> In Italy, IYSV was detected in Veneto,<sup>13</sup> Emilia Romagna<sup>14</sup> and recently also in Piedmont (personal observation). Therefore, tospovirus spread and severity should be carefully monitored, and the actual impact of thrips injuries on onion yield, which has not yet been investigated in Italy, should be assessed.

Thrips chemical control is difficult because of their small size and cryptic habits.<sup>6,7</sup> Nevertheless, many intensive treatments are applied on a routine basis to prevent thrips infestations. In Italy, only

a few insecticides, mostly pyrethroids (e.g. lambda-cyhalothrin), are authorised on onion, resulting in repeated applications of similar active ingredients (AIs). As reported worldwide, a high treatment frequency selects for resistance to the most-used compounds, especially synthetic pyrethroids,<sup>2,3,15 – 18</sup> even though there have been no studies documenting resistance in onion thrips in Italy. Here, spinosad has been recently allowed on onion, with a maximum of two consecutive applications or three total applications per year. This product is well known to be one of the most effective insecticides against *T. tabaci*.<sup>11,15,19</sup> Moreover, spinosad is a reduced-risk insecticide for many useful arthropods, as required for the conservation of thrips predators.<sup>20–22</sup>

Pesticide impact on human and environmental health begs for alternative pest management approaches, such as the use of straw mulch<sup>11,23,24</sup> or thrips-resistant onion cultivars<sup>25</sup> or the evaluation of new bioinsecticides<sup>4</sup> and of biologically active plant volatiles<sup>1</sup> against *T. tabaci*. Moreover, novel chemical and biological control agents appear to be promising for reducing the use of conventional insecticides; in particular, activators of the natural systemic acquired resistance (SAR) response found in most plant species are worthy of attention. The SAR response can be induced by products such as acibenzolar-*S*-methyl, a structural analogue of salicylic acid, used as an effective alternative to many bactericides and fungicides for the control of several diseases.<sup>23,26,27</sup> The potential value of SAR compounds for the control of tospoviruses and their vector thrips has been studied.<sup>11,23,28</sup>

Independently of the employed chemical, a supervised control based on action thresholds or tolerance levels is one of the major aspects of integrated pest management.<sup>5,22,29</sup> As a consequence, a reliable sampling method that is fast and easily adoptable is needed for correct monitoring of *T. tabaci* on onion crops. With the aim of implementing efficient and environmentally friendly thrips control, research was carried out in a commercial onion field in north-western Italy to compare the efficacy of different pest management strategies. Thus, the effectiveness of some insecticides registered for use on onion and of the SAR activator acibenzolar-*S*-methyl was evaluated on thrips infestations: (1) in the field during the growing season; (2) in laboratory bioassays; (3) in relation to the crop yield and quality at harvest. Moreover, for correct monitoring of thrips populations, three sampling methods were tested in order to assess the most feasible routine practice for growers.

## 2 MATERIALS AND METHODS

### 2.1 Field experiments

Field experiments were conducted in a commercial onion field of about 8 ha located in Castellazzo Bormida (province of Alessandria, Piedmont, 44° 50' 45" N, 8° 34' 41" E, 105 m a.s.l.) in 2010. The experimental site was flanked on all sides by at least 2 m of insecticide-free onions within the grower's field. Experimental plots (10.5 m<sup>2</sup> each) consisted of six 7 m long onion rows, and rows were spaced every 0.21 m. Plots were separated within a row by 1 m and spaced every 1.5 m. Onions of the golden onion cultivar 'Derek' were seeded on 12 April, and never irrigated throughout the season; diseases and weeds were controlled using pesticides recommended for onion production that do not affect the *T. tabaci* population.

The trial was arranged in a randomised complete block design with four replicates for each of six treatments (24 plots). The six treatments consisted of: one untreated control (T1); one routine treatment, which included the same insecticides as those adopted in the commercial field (T2); two treatments based on repeated pyrethroid applications without or with mineral oil (T3 and T4); one treatment based on both spinosad and pyrethroid applications (T5); one treatment based on

application of the plant activator acibenzolar-*S*-methyl (T6) (Table 1). The tested active ingredients (AIs) and formulations were: acibenzolar-*S*-methyl 500 g AI kg<sup>-1</sup> (Bion<sup>®</sup> 50 WG; Syngenta Crop Protection, Milan, Italy); chlorpyrifos methyl 200 g AI L<sup>-1</sup> + cypermethrin 20 g AI L<sup>-1</sup> (Daskor<sup>®</sup> EC; Dow AgroSciences, Milan, Italy); lambda-cyhalothrin 15 g AI L<sup>-1</sup> (Karate<sup>®</sup> Zeon 1.5; Syngenta Crop Protection, Milan, Italy); mineral oil 800 g AI L<sup>-1</sup> (Biolid E<sup>®</sup>; SIPCAM SpA, Milan, Italy); spinosad 120 g AI L<sup>-1</sup> (Success<sup>®</sup> SC; Dow AgroSciences, Milan, Italy). Chemicals were used at the recommended application doses (label) for field crops using 600 L solution ha<sup>-1</sup> of onion crop; the rates and timings of applications are listed in Table 1.

Starting in May, onion plots were surveyed weekly for the presence of *T. tabaci*. At the first occurrence of onion thrips infestation on the crop on 1 June (pre-S, i.e. the sampling before chemical applications), four insecticide applications were sprayed on 6 June, 21 June, 1 July and 12 July. Thrips sampling (S1, S2, S3 and S4, i.e. sampling after the first, second, third and fourth applications respectively) was carried out 1 week after each cluster of sprays on 13 June (S1), 28 June (S2), 8 July (S3) and 19 July (S4). The number of green leaves per plant was approximately 2–4 in pre-S (prebulbing stage), 4–6 in S1 (early bulbing), 6–8 in S2 (late bulbing), 8–10 in S3 (late bulbing-sizing) and 10 or more in S4 (sizing). Thrips adults and larvae were counted by visual inspection of five plants randomly selected at three points of each plot (15 plants plot<sup>-1</sup>).

As an alternative to the visual inspection, two other sampling methods were tested. The first method consisted of the beating onto a plastic tray (350 × 250 mm) of three plants randomly selected at three points of each plot (9 plants plot<sup>-1</sup>); thrips adults and larvae were counted, collected with a mouth aspirator and transferred to the laboratory. The second method consisted of collection of two intact plants randomly chosen at three points of each plot (6 plants plot<sup>-1</sup>); plants were individually capped with a plastic bag and gently pulled with a hand spade, sealed in the bag and labelled. In the laboratory, each plant was dissected and examined leaf by leaf to detect adults and larvae. Adult thrips collected by both sampling methods were then observed under a stereomicroscope at 100× magnification and identified to the species level according to Mound *et al.*<sup>30</sup>

During field surveys, onion plants were carefully checked for IYSV symptoms. At harvest, on 10 September, 15 plants randomly selected at three points of each plot (45 plants plot<sup>-1</sup>) were collected (180 plants treatment<sup>-1</sup>), labelled and allowed to dry for a 3 week period under a shelter. Afterwards, bulbs were individually weighed on a precision balance and checked for diameter and height with a sliding caliper.

## 2.2 Laboratory bioassays

*Thrips tabaci* adults and larvae collected by beating in untreated plots on 8 and 19 July (S3 and S4) were used in the laboratory bioassays. Thrips were temporarily transferred to 1000 mL gauze covered glass jars containing fresh organic green bean pods (*Phaseolus vulgaris* L.) as a food source and oviposition site, and corrugated cardboard on the bottom provided pupation sites. Jars were stored at 25 °C and 60% RH under a 16 : 8 h light : dark cycle. The toxicity of the plant activator acibenzolar-*S*-methyl and of the two insecticides lambda-cyhalothrin and spinosad on *T. tabaci* was evaluated by two methods: a leaf-dip bioassay and a vial bioassay. The active ingredients were used in the same formulations and doses as those adopted in the field experiment in order to obtain baseline data on susceptibility. Acibenzolar-*S*-methyl was tested in the vial bioassay to evaluate a potential side effect on thrips even in the absence of the plant.

### 2.2.1 Leaf-dip bioassay

In the leaf-dip bioassay, discs of 35 mm diameter were cut from the white section of insecticide-untreated leek leaves; they were then dipped in the chemical solution or water only for the untreated control for 10 s, and dried on tissue paper in a fume hood for 2 h. When dried, discs were placed one each in Plexiglas cages,<sup>31</sup> on damp filter paper to maintain the turgidity of the leaf. Ten specimens of *T. tabaci* (including adults and larvae) were collected from rearing jars and transferred onto the treated discs by gently tapping the inverted pooter. Cages were then closed with a fine gauze (200 × 200 mesh inch<sup>-1</sup>) to allow ventilation and stored at 25 °C under a 16 : 8 h light : dark cycle. Five cages were used for each of the four treatments: acibenzolar-*S*-methyl, lambda-cyhalothrin, spinosad and untreated control. The numbers of living and dead individuals on each disc were recorded after 24 and 48 h using a stereomicroscope (thrips were considered dead if they failed to move when gently touched with the tip of a paintbrush). The leaf-dip bioassay was repeated twice.

### 2.2.2 Vial bioassay

The vial bioassay method is an adaptation of the thrips insecticides bioassay system (TIBS),<sup>16</sup> carried out in the laboratory rather than directly in the field. In addition, thrips were put into a plastic 1.5 mL microcentrifuge tube (instead of 0.5 mL) previously treated with the product being tested. In the tube cap, a small well contained a 10% sugar–water solution with blue food colorant (E133; Eurofood SpA, Milan, Italy). The solution was sealed into the well with a small piece of stretched parafilm through which thrips could feed on the 10% sugar solution. The food colorant was added to the solution to facilitate determining whether the parafilm membrane had been broken and the solution contaminated the tube when the assay was read. The tube, but not the cap, was treated with the product by filling the tube to its top with the chemical solution or water only for the untreated control. After 4 h, the chemical solution (or water) was poured out and the tube was allowed to dry overnight. Ten thrips (including adults and larvae) were collected from rearing jars and transferred into the treated tubes (by gently tapping a paintbrush on the rim of the tube). Five replicates were used for each of the four treatments: acibenzolar-*S*-methyl, lambda-cyhalothrin, spinosad and untreated control. Thrips mortality was assessed after 24 and 48 h with the help of a stereomicroscope. The vial bioassay was replicated twice.

## 2.3 Statistical analysis

For the field data, the mean numbers of adults, larvae and total thrips per plant were log-transformed to achieve homogeneity of variance (Levene) and normality (Shapiro–Wilk). To check the correlations between adult, larval and total thrips populations, data were analysed by the parametric Pearson correlation. Mean numbers of total thrips were then analysed by univariate analysis of variance (ANOVA) for randomised blocks (treatments and blocks were the factors). When there were significant differences between treatments ( $P < 0.05$ ), means were compared using the Tukey *post hoc* test at  $P < 0.05$ .

Correlations between mean numbers of thrips per plant collected by visual inspection and by plant beating or plant dissection were analysed by the non-parametric Spearman's rank correlation test; sampling dates were used as pseudoreplications.

Data on harvested onions were analysed by ANOVA after tests of homogeneity of variance (Levene) and normality (Shapiro–Wilk).

Percentage survival data obtained in the laboratory bioassays were transformed to arcsine square root values before being analysed; the non-parametric Kruskal–Wallis test was chosen because data were inhomogeneous; means were compared using the Mann–Whitney *U*-test at  $P < 0.05$ .

All statistical analyses were performed using the software SPSS v.17.0 (SPSS, Chicago, IL).

## 3 RESULTS

### 3.1 Field experiments

#### 3.1.1 Treatment efficacy against onion thrips

All phytophagous adult thrips collected on onions were *T. tabaci*, with populations composed of both adults and larval stages. By visual inspection, overall larval stages were 74% of the total sampled thrips ( $n = 83\ 256$ ). Percentages of larvae were similar in all treatments; however, these percentages were variable throughout the growing season, with 74, 76, 79, 52 and 85% of specimens in total sampled on 1 June, 13 June, 28 June, 8 July and 19 July respectively. In all treatments, the total thrips (adults plus larvae) population trends very much reflected both adult and larval populations taken separately (Pearson correlation coefficients 0.96–0.99;  $P < 0.001$ ;  $n = 30$ ) (data not shown). For this reason, data on the total thrips population were chosen to compare the efficacy of the tested treatments.

The mean numbers of onion thrips collected in the plots of each treatment, on five sampling dates, are reported in Table 2. On 1 June, before the first chemical application, mean numbers ranged between 0.7 and 1.4 thrips plant<sup>-1</sup> without any significant differences between the treatments (ANOVA:  $df = 5, 63$ ;  $F = 0.7$ ;  $P = 0.6$ ;  $n = 12$ ). Then, thrips populations started to increase and reached maximum infestation levels on 8 July in treatments T3 and T4, and on 19 July in the other treatments. The maximum mean numbers ranged from 86.9 thrips plant<sup>-1</sup> (T5) to 196.8 thrips plant<sup>-1</sup> (T6) (Table 2).

The numbers of onion thrips per plant were significantly higher in the treatments of lambda-cyhalothrin without or with mineral oil (T3 and T4) than in the untreated control (T1) after the second chemical application on 28 June (ANOVA:  $df = 5, 63$ ;  $F = 9.6$ ;  $P < 0.001$ ;  $n = 12$ ) and after the third chemical application on 8 July (ANOVA:  $df = 5, 63$ ;  $F = 35.0$ ;  $P < 0.001$ ;  $n = 12$ ). On this sampling date (S3), thrips infestations were significantly higher in T3 and T4 than in all the other treatments; on the other hand, in T5, where lambda-cyhalothrin was sprayed for the first time after two applications of spinosad (see Table 1), thrips numbers were significantly the lowest, as well as in T6 where acibenzolar-Smethyl was applied. This last treatment showed the lowest thrips numbers also on 13 and 28 June; by contrast, on 19 July, when plants started to wither, the SAR activator showed statistically the highest numbers of *T. tabaci* (ANOVA:  $df = 5, 63$ ;  $F = 3.7$ ;  $P = 0.006$ ;  $n = 12$ ).

In the commercial onion field, no plant showing IYSV symptoms was detected during field sampling over the entire growing season, and at harvest the yield was approximately 5.5 t ha<sup>-1</sup>. Data on weight (g), diameter (mm) and height (mm) of 180 harvested bulbs for each treatment are reported in Table 3. On average, bulbs were 114.3 g in weight, 59.1 mm in diameter and 63.3 mm in height; no significant differences were found between treatments (ANOVA:  $df = 5, 63$ ;  $F = 0.9, 1.2, 1.5$ ;  $P = 0.5, 0.3, 0.2$ ;  $n = 12$ ). Nevertheless, the lowest values of all measures were observed in T3 and

especially in T4, based on lambda-cyhalothrin and on lambda-cyhalothrin + mineral oil respectively (Table 3).

### 3.1.2 Sampling method evaluation

Both plant beating and plant dissection were less accurate methods for thrips sampling than visual inspection, but they showed a good correlation with this method. The linear regressions between the mean numbers of thrips per plant sampled by plant beating and by plant dissection versus visual inspection are reported in Figs 1 and 2 respectively. The relationship with visual inspection was highly significant both for plant beating (Spearman correlation:  $\rho = 0.92\text{--}0.95$ ,  $P < 0.001$ ;  $n = 30$ ) and for plant dissection (Spearman correlation:  $\rho = 0.87\text{--}0.92$ ,  $P < 0.001$ ;  $n = 30$ ) (Figs 1 and 2). Overall larvae were 28 and 77% of thrips totally recorded (adults plus larvae) by plant beating ( $n = 16\,099$ ) and by plant dissection ( $n = 2\,903$ ) respectively, compared with 74% sampled by visual inspection (see above). By plant beating, almost equivalent numbers of adults but very low numbers of larvae were collected, as the slope coefficients in the equations suggest (Fig. 1). By plant dissection, the ratio between adults and larvae was similar to that of visual inspection; on the other hand, thrips densities were highly underestimated above all with high infestation levels (Fig. 2). The value of 20 thrips  $\text{plant}^{-1}$ , sometimes used as a threshold for insecticide application in north-western Italy, corresponds to a mean thrips density of 16.8 adults or 2.0 larvae sampled by plant beating, and of 2.8 adults or 4.3 larvae sampled by plant dissection (Figs 1 and 2).

## 3.2 Laboratory bioassays

The percentages of adults and larvae alive were significantly different between the treatments, in the leaf-dip bioassay both after 24 h (Kruskal–Wallis:  $df = 3$ ;  $\chi^2 = 24.1, 27.5$ ;  $P < 0.001$ ;  $n = 10$ ) and after 48 h (Kruskal–Wallis:  $df = 3$ ;  $\chi^2 = 24.1, 26.2$ ;  $P < 0.001$ ;  $n = 10$ ) (Fig. 3), and in the vial bioassay both after 24 h (Kruskal–Wallis:  $df = 3$ ;  $\chi^2 = 34.9, 35.6$ ;  $P < 0.001$ ;  $n = 10$ ) and after 48 h (Kruskal–Wallis:  $df = 3$ ;  $\chi^2 = 33.0, 31.9$ ;  $P < 0.001$ ;  $n = 10$ ) (Fig. 4). In the untreated control, percentages of mortality after 24 h in both bioassays never reached 10%. Spinosad was the most effective active ingredient, as only 3.1% of adults and 2.3% of larvae were alive after 24 h, and no thrips survived after 48 h in the leaf-dip method, whereas there were no thrips alive after 24 h in the vial method (Figs 3 and 4). Lambda-cyhalothrin showed contrasting results in the two bioassays. When leaves were treated (leaf-dip method), after 24 h the number of live adults was 85.3%, not significantly different from the untreated control, whereas the number of live larvae was statistically higher than with spinosad treatment but lower than the control (Fig. 3a). After 48 h, both live adults and larvae were significantly higher than after spinosad and lower than the control (Fig. 3b). Nevertheless, in the vials treated with lambda-cyhalothrin, after 24 h only 4.0% of adults survived, statistically the same as spinosad, while 37.8% of larvae were still alive (Fig. 4a). After 48 h, none of the adults survived and only 4.1% of larvae were alive; both percentages were not significantly different from those obtained with spinosad (Fig. 4b). With acibenzolar-*S*-methyl treatment, the percentage of live adults was statistically the same as the control in both bioassays after 24 h, and only in the vial method after 48 h, whereas it was statistically lower than the control in the leaf-dip method after 48 h. On the other hand, percentages of live larvae were significantly lower than the control in both bioassays after 24 and 48 h (Figs 3 and 4).

#### 4 DISCUSSION AND CONCLUSIONS

Total thrips population trends very much reflected both adult and larval population trends throughout the growing season in all tested treatments. Adults are highly vagile and they can move rapidly between treated plots; therefore, only larval stages are sometimes considered to evaluate insecticide efficacy.<sup>5</sup> Anyway, in the case reported here, because of the high correlation between larvae, adult and total population trends, the presence of adults was found to reflect the efficacy of the treatments in the same way as the presence of larvae.

Independently of the treatments, *T. tabaci* populations increased, starting from the end of June and the beginning of July, as observed in Spain<sup>32</sup> and in New York,<sup>24</sup> owing to favourable climatic conditions (i.e. high temperatures and low humidity). At the end of the growing season, the maximum infestation levels were very high, from 87 to 197 thrips plant<sup>-1</sup>, similar to observations in other countries.<sup>3,24,32</sup>

During the field experiments, lambda-cyhalothrin applications were followed by the highest infestation levels, especially starting from the second consecutive repetition of this AI. Although there have been no studies documenting resistance of local thrips populations to pyrethroids in Piedmont or elsewhere in Italy, resistance of onion thrips to pyrethroids (including lambda-cyhalothrin) has been reported worldwide.<sup>2,3,15-18</sup> Moreover, insecticide resistance can rapidly increase over the growing season as a consequence of selection pressure.<sup>16</sup> The reiteration of lambda-cyhalothrin is a practice usually adopted by most of the onion growers in Piedmont; the present results confirmed the failure of such a control strategy against thrips infestations, as already observed in other countries.<sup>2,3,16,17</sup>

In the laboratory, lambda-cyhalothrin showed variable toxicity in the two bioassay methods. When the AI was applied on a vegetal portion (leaf-dip bioassays), the insecticide efficacy was quite low; in fact, for adults after 24 h of exposure, it was statistically the same as in the untreated control, corroborating the results obtained in the field experiments. On the other hand, in the vial bioassays, the efficacy of lambda-cyhalothrin was much higher, and after 48 h of exposure it was statistically comparable with that of spinosad. In this method, thrips are confined in a very small space and they are forcibly exposed to the AI more than when they are in ventilated cages where only the vegetal portion is treated. Hence, this situation may intensify insecticide toxicity and cause higher thrips mortality in relation to the leaf-dip method. A similar behavior was observed for abamectin and chlorpyrifos;<sup>19</sup> their efficacy in TIBS was higher than in the leaf-dip method owing to the bioassay substrate and formulation of the insecticides. However, other authors have demonstrated thrips resistance to lambda-cyhalothrin using TIBS<sup>15</sup> or both leaf-dip assays and TIBS.<sup>33</sup> Further investigations using TIBS with *T. tabaci* directly collected on onion foliage and at different concentrations of lambda-cyhalothrin are needed to determine the concentration–mortality responses of onion thrips populations in this geographic area.

The effectiveness of spinosad against *T. tabaci* was confirmed, even considering the restriction of use to two consecutive applications or three total applications per year. The long persistence and residual activity of this insecticide against different pests under field conditions are generally known.<sup>34,35</sup> Therefore, the low number of thrips per plants recorded in T5 (two spinosad applications followed by two lambda-cyhalothrin applications) on 8 July, after the first lambda-cyhalothrin application, was most likely due to the residual activity of the previous spinosad application (21 June). Spinosad efficacy was supported under laboratory conditions; no thrips survived after 48 h exposure

to the AI at field concentrations in both the leaf-dip and vial bioassays. Spinosad proved to be the most insect toxic among the tested products, and its toxicity was analogous in both tested methods, as has already been observed.<sup>19</sup>

The potential value of SAR compounds for tospovirus and vector thrips control has been shown.<sup>11,23,27,28</sup> In the present field experiments, the statistically lowest values of thrips infestations were observed in treatment with acibenzolar-*S*-methyl during the growing season, except at the last sampling date. This product activates the natural resistance system in the plant, so its action is strictly correlated with plant phenology: when plants started to wither (starting from the second decade of July), they were probably less prompt in reacting to any physiological stimulus. In the laboratory bioassays, when applied on a portion of plant (the leaf-dip method), acibenzolar-*S*-methyl showed an effect, even if minor, on thrips adults only after 48 h, while a side effect on larval stages was recorded as soon as after 24 h and even in the absence of plant material (the vial method). Although its potential direct insecticide effect on thrips should be further assessed, the use of this SAR activator in controlling *T.tabaci* appears promising in order to reduce the use of conventional insecticides. Nevertheless, in Italy, acibenzolar-*S*-methyl is not currently allowed on onion crops, and its optimum rate and timing should be carefully investigated before recommending its use.

Unexpectedly during the whole growing season, the untreated control never showed the highest infestation levels. The potential or increased abundance of natural enemies was not surveyed, but their preservation in the untreated plots resulting in pest suppression can be assumed.<sup>22,32</sup> The same beneficial effect due to the conservation of thrips predators can be assumed in the plots treated with spinosad, which is a reduced-risk insecticide for useful arthropods, as well as in the plots treated with acibenzolar- *S*-methyl.

In spite of high thrips infestation levels recorded, especially on the two last sampling dates, the commercial farm did not experience yield losses attributable to thrips injuries. However, consistent with Kendall and Capinera,<sup>7</sup> thrips damage results in significant yield reductions during the midseason, when bulb diameter and weight increase and when leaf initiation decreases, and not at the end of the growing season, corresponding to further bulb growth and collapse of the leaf bundle. The total onion yield, around 5.5 t ha<sup>-1</sup>, was consistent with the local average yield for the cultivar ‘Derek’ (5.4 t ha<sup>-1</sup>); moreover, the mean value of bulb weight (114.3 g) was higher than the mean regional value (106.5 g) (data from regional variety trials performed by local technical assistants). Hence, the damage caused by thrips, at least on this onion variety in north-western Italy, may not be severe enough to warrant the frequent pesticide applications the crop receives. In addition, the total onion yield was not significantly affected by the different tested treatments, even if the lowest values of bulb weight, diameter and height were observed in the two treatments based on the reiteration of lambda-cyhalothrin, in which thrips populations were significantly higher from the end of June. Moreover, onion yield was not significantly affected by the four applications of acibenzolar-*S*-methyl, consistent with Cole,<sup>36</sup> whereas a phytotoxic effect, causing a bulb yield reduction of up to 27%, was observed with ten weekly acibenzolar-*S*-methyl applications.<sup>26</sup>

Supervised control of onion thrips based on accurate plant monitoring proved to be very effective on leek in this geographical area.<sup>22</sup> From this perspective, finding a suitable and simple sampling method for correct monitoring of *T.tabaci* on onion crops is crucial. In the present experiments, visual inspection gave the most accurate data on thrips populations; contrary to expectation, overall plant dissection produced only 38% of thrips counted by direct visual inspection in the field. This result

was probably biased by different factors such as the time elapsed between plant field collection and laboratory dissection, the moisture in the bag, the difficulty of detecting dead thrips inside the bag and thrips escape from plants above all with high population levels (see Fig. 2). Visual inspection takes a very long time to be applied, especially with high infestations. Both plant beating and plant dissection showed a strong relationship with visual inspection, so the regression equations can be used in practice to adjust the number of thrips per plant recorded with the two proposed methods. This is particularly useful in estimating the real number of larvae by beating plants because, with this method, larvae are only partly captured, most of them remaining among the leaves at the base of the plant.<sup>37</sup> Nonetheless, plant beating is the most suitable sampling method for preserving commercial crops and a more feasible routine practice for growers than counting insects leaf by leaf, directly in the field or after plant dissection. The number of onion plants to be inspected for a correct population estimate is also very important: five plants each in ten different areas of a field for a total of 50 plants per field is ideal,<sup>38</sup> even fewer plants can be examined when they are selected by stratified random sampling.<sup>29</sup>

In Piedmont, the current action threshold suggested on onion against *T. tabaci* is fixed at around 20 thrips plant<sup>-1</sup>. The action thresholds recorded in the literature range from 0.1 to 2 thrips plant<sup>-1</sup> in New Zealand,<sup>29</sup> and from 4 to 30 thrips plant<sup>-1</sup>, depending on onion plant growth stage, in Canada,<sup>8</sup> the United States<sup>5,38,39</sup> and Honduras.<sup>40</sup> Lower action thresholds are often recommended for thrips-susceptible varieties and during the bulbing stage, whereas higher action thresholds are recommended for moderately tolerant varieties (such as ‘Derek’), young plants or plants near maturity. Based on the present study, if the action threshold of 20 thrips plant<sup>-1</sup>, corresponding to around 17 adults or two larvae sampled by plant beating, was applied, the first cluster of insecticide sprays would be delayed until late June or early July. On the other hand, for the growth stage following the bulbing, when plants are near maturity, this action threshold should be most likely to be augmented. This approach would allow for fewer targeted interventions with selective insecticides, enabling the conservation and spontaneous crop colonisation of natural enemies.

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## Tables

**Table 1.** Active ingredients, rates and numbers and timings of applications of the products sprayed in the experimental plots in the onion field during the growing season.

Treatment	Active ingredient	Rate (L ha <sup>-1</sup> )	No. <sup>a</sup>	Timing
T1 <sup>b</sup>	-	-	-	-
T2 <sup>c</sup>	Lambda-cyhalothrin+mineral oil	1.3+1.25	1	6 June
	Chlorpyrifos methyl+cypermethrin	1.5	1	1 July
	Spinosad+mineral oil	0.8+1.25	1	12 July
T3	Lambda-cyhalothrin	1.3	4	6 June; 21 June; 1 July; 12 July
T4	Lambda-cyhalothrin+mineral oil	1.3+1.25	4	6 June; 21 June; 1 July; 12 July
T5	Spinosad	0.8	2	6 June; 21 June;
	Lambda-cyhalothrin	1.3	2	1 July; 12 July
T6	Acibenzolar-S-methyl	0.2	4	6 June; 21 June; 1 July; 12 July

<sup>a</sup>Number of applications.

<sup>b</sup>Untreated control.

<sup>c</sup>Thrips control strategy adopted by the grower.

**Table 2.** Mean numbers ( $\pm$  SE) of *Thrips tabaci* per plant sampled by visual inspection in the six tested treatments during field surveys<sup>a</sup>.

Treatment	Pre-S <sup>b</sup> : 01-June	S1 <sup>b</sup> : 13-June	S2 <sup>b</sup> : 28-June	S3 <sup>b</sup> : 08-July	S4 <sup>b</sup> : 19-July
T1	0.97 $\pm$ 0.34	2.27 $\pm$ 0.46	10.88 $\pm$ 2.65 bc	57.77 $\pm$ 20.53 b	168.05 $\pm$ 29.22 ab
T2	1.40 $\pm$ 0.42	2.07 $\pm$ 0.44	16.65 $\pm$ 2.04 ab	42.60 $\pm$ 8.73 b	127.07 $\pm$ 42.34 b
T3	1.08 $\pm$ 0.73	2.45 $\pm$ 0.74	23.68 $\pm$ 4.57 a	152.87 $\pm$ 13.83 a	146.18 $\pm$ 26.25 ab
T4	0.60 $\pm$ 0.26	1.75 $\pm$ 0.54	24.48 $\pm$ 4.41 a	165.22 $\pm$ 13.59 a	114.65 $\pm$ 23.64 ab
T5	1.17 $\pm$ 0.55	3.48 $\pm$ 1.01	10.25 $\pm$ 1.66 bc	4.95 $\pm$ 1.12 c	86.87 $\pm$ 18.61 b
T6	0.72 $\pm$ 0.18	2.87 $\pm$ 0.44	8.23 $\pm$ 1.38 c	9.57 $\pm$ 1.77 c	196.82 $\pm$ 29.97 a
<i>P</i>	0.598	0.440	<0.001	<0.001	0.006
<i>F</i> <sub>5,63</sub>	0.738	0.975	9.645	35.029	3.664
SED <sup>c</sup>	0.202	0.251	0.194	0.337	0.267

<sup>a</sup> Statistical analyses were performed on log-transformed data which are not shown. Within a column, means followed by different letters are significantly different ( $P < 0.05$ , Tukey's test following ANOVA). ANOVA results ( $P$  and  $F$  values,  $df = 5, 63$ ;  $n = 12$ ) are reported.

<sup>b</sup> Pre-S represents the sampling before chemical applications; S1, S2, S3, and S4 represent sampling after the first, second, third and fourth applications, respectively.

<sup>c</sup> Standard Errors of the difference values.

**Table 3.** Mean values ( $\pm$  SE) of weight, diameter and height of bulbs collected in the experimental plots for the six tested treatments at harvest<sup>a</sup>.

Treatment	Weight (g)	Diameter (mm)	Height (mm)
T1	116.35 $\pm$ 4.84	59.02 $\pm$ 0.92	63.25 $\pm$ 0.64
T2	114.21 $\pm$ 4.29	61.58 $\pm$ 2.70	64.20 $\pm$ 0.92
T3	111.79 $\pm$ 4.34	58.45 $\pm$ 0.82	63.08 $\pm$ 0.92
T4	109.52 $\pm$ 2.49	57.94 $\pm$ 0.41	61.76 $\pm$ 0.65
T5	117.99 $\pm$ 3.10	58.93 $\pm$ 0.58	63.46 $\pm$ 1.00
T6	115.70 $\pm$ 3.60	58.78 $\pm$ 0.63	63.96 $\pm$ 0.75
<i>P</i>	0.493	0.339	0.200
<i>F</i> <sub>5,63</sub>	0.891	1.160	1.508
SED <sup>b</sup>	4.684	1.666	0.989

<sup>a</sup> There were no significant differences in bulb weight, diameter or height between treatments; ANOVA results (*P* and *F* values, *df* = 5, 63; *n* = 12) are reported.

<sup>b</sup> Standard errors of the difference values.

## Figure captions

**Figure 1.** Relationship between mean numbers of adults, larvae and total thrips collected per plant by beating and by visual inspection. Population densities range from 0.1–250 thrips plant<sup>-1</sup> (A) and 0.1–30 thrips plant<sup>-1</sup> (B). Lines represent the best fit line; regression equations and linear regression coefficients ( $R^2$ ) are reported in the figure label. The Spearman coefficient (rho) and two-way significance values (sig) are also given. The arrow indicates the value of 20 thrips plant<sup>-1</sup> recommended as a threshold for insecticide application in north-western Italy.

**Figure 2.** Relationship between mean numbers of adults, larvae and total thrips collected per plant by plant dissection and by visual inspection. Population densities range from 0.1–250 thrips plant<sup>-1</sup> (A) and 0.1–30 thrips plant<sup>-1</sup> (B). Lines represent the best fit line; regression equations and linear regression coefficients ( $R^2$ ) are reported in the figure label. The Spearman coefficient (rho) and two-way significance values (sig) are also given. The arrow indicates the value of 20 thrips plant<sup>-1</sup> recommended as a threshold for insecticide application in north-western Italy.

**Figure 3.** Mean ( $\pm$  SE) survival percentages of adults, larvae and total *Thrips tabaci* 24 h (A) and 48 h (B) after exposure to onion leaf discs treated with the tested products at field concentrations (leaf-dip bioassays). Bars within adult, larva and total thrips treatments labelled with different letters (small, capital, and Greek letters, for adults, larvae and total thrips respectively) are significantly different ( $P < 0.05$ , Mann-Whitney following Kruskal-Wallis,  $df = 3$ ).

**Figure 4.** Mean ( $\pm$  SE) survival percentages of adults, larvae and total *Thrips tabaci* 24 h (A) and 48 h (B) after exposure to vials treated with the tested products at field concentrations. Bars within adult, larva and total thrips treatments labelled with different letters (small, capital, and Greek letters, for adults, larvae and total thrips respectively) are significantly different ( $P < 0.05$ , Mann-Whitney following Kruskal-Wallis,  $df = 3$ ).

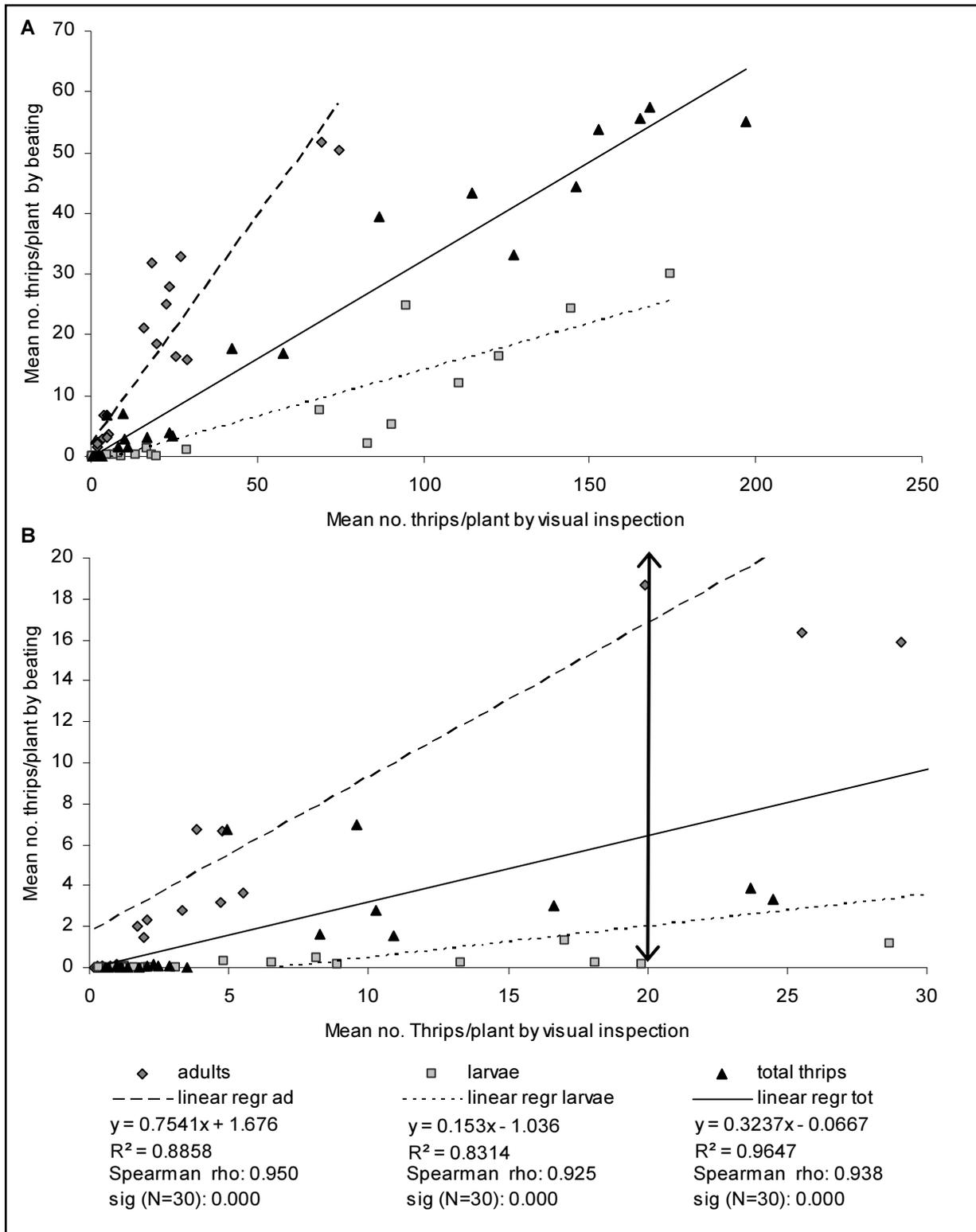


Figure 1

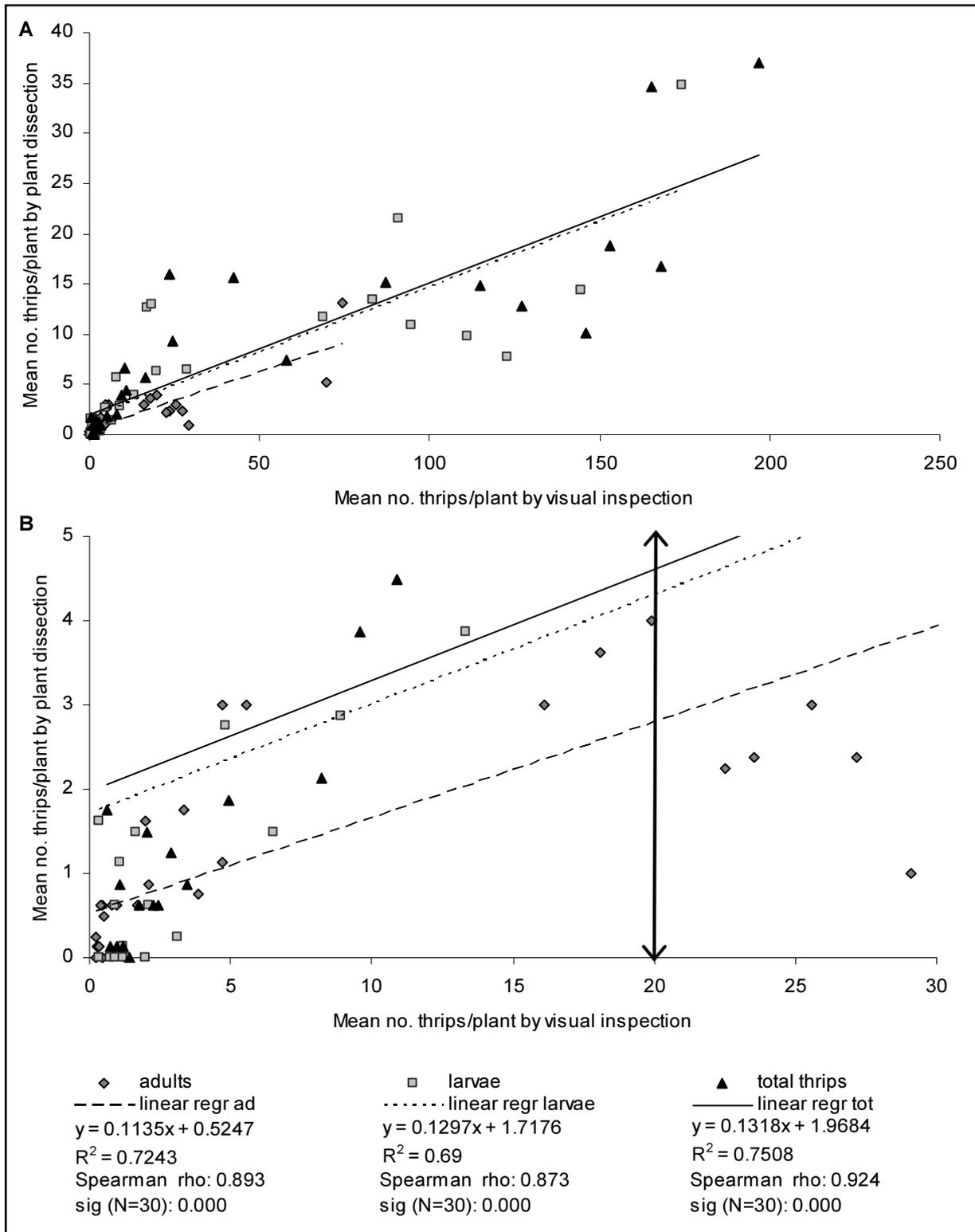
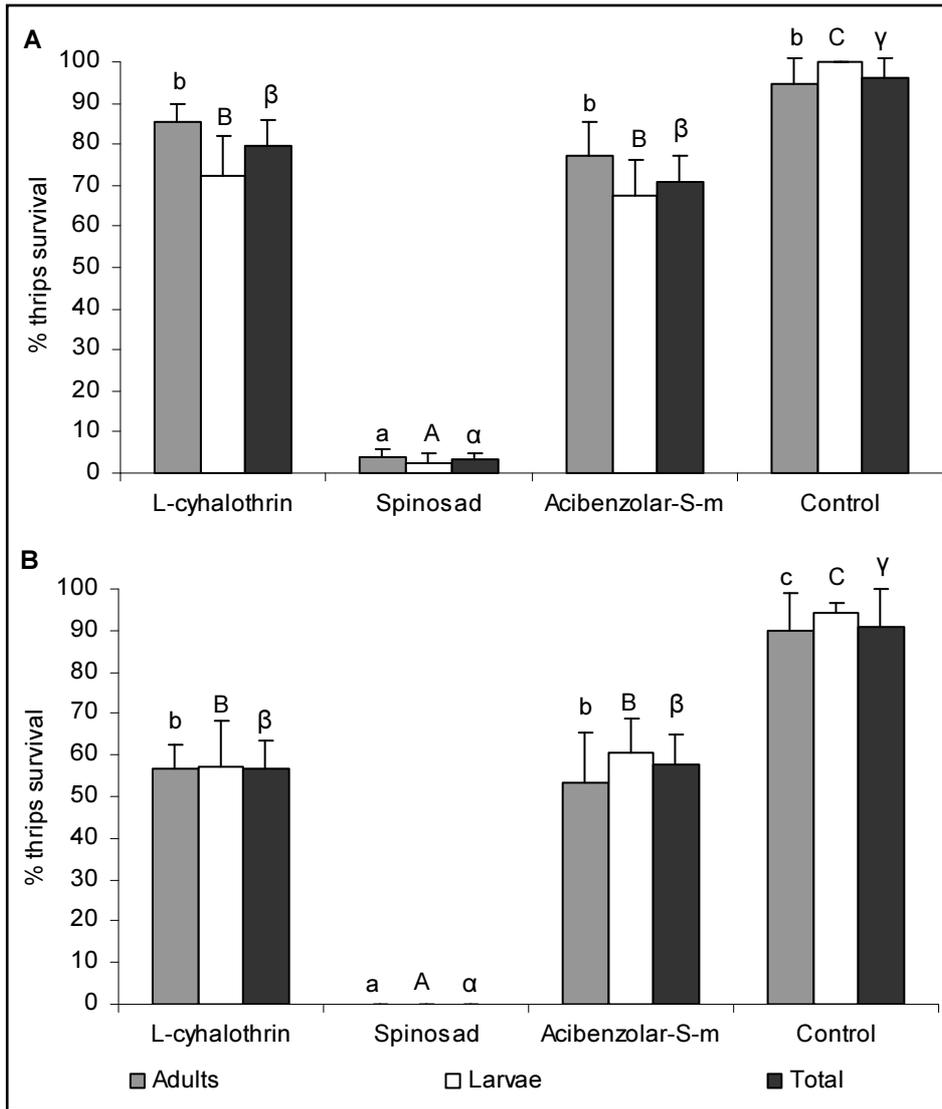


Figure 2



**Figure 3**

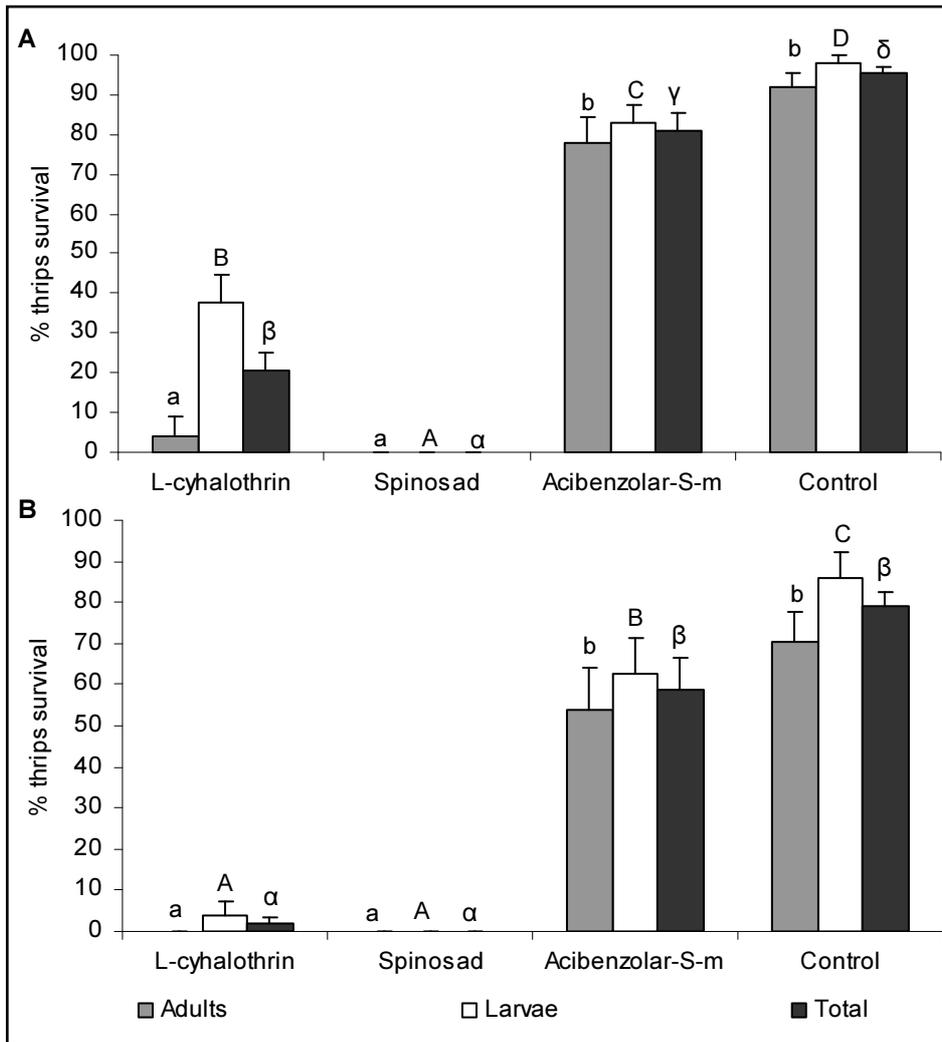


Figure 4