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Pollen beetle (*Meligethes aeneus* Fab) susceptibility to synthetic pyretroids – pilot study in Estonia

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Abstract. Pollen beetle (*Meligethes aeneus* Fab) is a common pest of oilseed rape. The development of resistance to pyrethroids in *M. aeneus* is widespread in northern Europe. In Estonia, the majority of farmers apply insecticides against pollen beetles without considering the threshold levels of the pest. This creates good preconditions for development of resistance in *M. aeneus*. To date the susceptibility tests of pollen beetles have not been conducted in Estonia and it is unknown whether the local pest population has any resistance to pyrethroids. Therefore a pilot study was conducted to ascertain the state of resistance inthe Estonian pollen beetle population. Approximately 200 adult beetles were collected from two intensively managed spring oilseed rape fields in Jõgeva and Tartu County. The susceptibility test was conducted according to IRAC test method No 11, with lamda-cyhalothrin as a standard for testing the synthetic pyrethroid resistance. In 20% lamda-cyhalothrin solution the majority of insects were affected and at the normal field rate (100%) all studied insects were affected which demonstrates that they were not resistant. These results suggest that the pollen beetles are susceptible to synthetic pyrethroids in these monitored regions in Estonia. However, additional studies are necessary to make conclusions about the extent of resistance or susceptibility to pyrethroids in the Estonian beetle population.

Key words: pesticide resistance, lamda-cyhalothrin, susceptibility test

INTRODUCTION

Throughout Europe, pest management on oilseed rape still relies heavily on synthetic pesticides in which the main active components are pyrethroids. Most often insecticides are applied routinely and prophylactically, without considering the pest incidence or the threshold values of the pest population (Williams, 2004). This has led to over-use of chemicals, which might reduce the economic competitiveness of the crop and threaten biological diversity. Due to the awakened public concern about the adverse impact of insecticides on the environment, the number of products for controlling oilseed rape pests has been greatly reduced and for many years only the use of pyrethroids has been permitted (Thieme et al., 2008). Therefore, the most serious current concern is the recent widespread development of resistance to pyrethroids in the pollen beetle *Meligethes aeneus* (Fab.), the most important pest of oilseed rape plants in northern Europe. The first cases of resistance were identified in populations of pollen beetles in Poland in the late 1970s (Lakocy, 1977). Since then, no insecticide

resistance in pollen beetles was reported, either in Poland or other European countries, until the late 1990s (Hansen, 2008). Since then the resistance has spread extensively and has now been reported from France, Denmark, Poland, Germany, Sweden and the UK (Ballanger et al., 2003; 2007; Hansen, 2003; 2007; Heimbach et al., 2006; Nilsson & Ahman, 2006; Wegorek & Zamoyska, 2007; Glattkowski et al., 2008). Due to the significance of the problem, an *ad hoc* EPPO (intergovernmental organization responsible for European cooperation in plant health) workshop was held in 2007 to discuss the severity of the problem and to find alternative options for controlling the pollen beetle populations (Zlof, 2007).

In Estonia, the growing area of oilseed rape has increased 60-fold over the last 15 years (Statistics Estonia, 2009). As expected, the amount of insecticides sold has also increased, and is five times greater today compared to six years ago (Plant Production Inspectorate, 2009). The majority of Estonian farmers apply insecticides against pollen beetles according to the technology scheme of oilseed rape cultivation (1–2 times during green bud and/or flowering growth stage of plants), which does not take into account the pest threshold levels and is based on plant growth stages. These circumstances create good preconditions for the development of resistance in *M. aeneus* against pyrethroids. Since susceptibility tests of pollen beetles have not been conducted in Estonia before, it is not known whether the local pest population has already developed resistance or not. Therefore we carried out a pilot study to ascertain and record the state of resistance in the pollen beetle.

MATERIAL AND METHODS

Thieme et al. (2008) showed that pollen beetles were very active flyers between oilseed rape fields. This makes it difficult to use only field experiments to identify the development of resistance because of the possibility of re-colonization. Thus we conducted a laboratory experiment to test the susceptibility of pollen beetles, following the protocol of the IRAC test method No 11 (Insecticide Resistance Action Committee, 2009). Beetles were collected from two intensively managed spring oilseed rape fields on June 29 and July 8, 2009, in Jõgeva and Tartu County, respectively. Approximately 200 adult beetles were collected at different locations across the field. Beetles were placed into an aerated plastic jar, containing a dry paper towel and some oilseed rape leaves and inflorescences as a food source at the bottom. The insects were shipped to the laboratory within an hour, released into a ventilated cage and left to recover overnight.

The test was carried out with lamda-cyhalothrin as a standard for testing the susceptibility to synthetic pyrethroids. Dilutions of 100% and 20% of field application rate were made in acetone. The experiment was carried out in five replications. Glass vials (26 ml volume) were filled with 1.5 ml of 100% or 20% solution of lamda-cyhalothrin or with only acetone as a control. Vials were rotated at room temperature until the acetone had completely evaporated and there was no liquid detected on the glass. After that, 15 or 10 beetles (June 29 and July 8 respectively) were placed into every vial, capped and stored at room temperature ($20 \pm 2°C$), avoiding exposure to direct sunlight. The number of affected beetles was scored 24 hours later by emptying the vials onto the centre of a white paper with a 15 cm circle drawn in the middle. A

bright light was directed into the circle to stimulate beetles' movement out of the circle; the beetles, which could not exit the circle before a period of one minute, were considered severely affected. Affected and live beetles were counted and the percentage of affected insects was calculated. Differences between treatments were calculated using nonparametric Kruskal-Wallis ANOVA multiple comparison test. Results were evaluated using the 'susceptibility rating scheme' which consists of five categories: highly susceptible (1), susceptible (2), moderately resistant (3), resistant (4) and highly resistant (5). In total 376 adults of *M. aeneus* were analyzed.

RESULTS AND DISCUSSION

The experiment for resistance monitoring is considered successful if less than 20% of beetles in the control are affected. In Jõgeva County only 3% of the insects in the control were affected and therefore the test results are reliable. In the 20% solution the majority of insects (88%) were affected and, at normal field rate, all the studied insects were affected (Fig. 1A). The analysis confirmed a significant difference in the survival rates of the insects between treatments (Kruskal-Wallis: $H_{(2,\ N=225)}= 187.97$; *P<0.0001*). These results classify to the second level in the susceptibility rating scheme and therefore these beetles were determined as susceptible. Although Jõgeva has been one of the most intensive oilseed rape growing regions in Estonia for many years, our results suggest that the pollen beetles have not yet become resistant to synthetic pyrethroids there. Nevertheless, the farmers started to use high insecticide levels only recently, so it is possible that the insects are still susceptible to them. Recently pollen beetles have been observed on freshly sprayed fields in Jõgeva (unpublished data); despite that fact, our study implies that these beetles might have originated from neighbouring fields and arrived after spraying, as shown before (Thieme et al., 2008).

The results of our experiment with pollen beetles from Tartu County cannot be considered as reliable since 28% of the beetles in the control were affected (Fig. 1B). Although insects treated with 100% and 20% field rate were 100% affected, and the beetles could be evaluated as highly susceptible, it is not possible to say if the survival was only influenced by the insecticide or the insects were already affected by some other factors prior to the experiment.

CONCLUSIONS

Despite the increase of oilseed rape-growing areas and intensive insecticide use during the last few years in Estonia, the pollen beetles tested in this study were highly susceptible to pyrethroids. However, additional studies in different regions of the country are necessary in the coming years to determine whether, and to what extent, the pollen beetle population in Estonia could be resistant to pyrethroids.

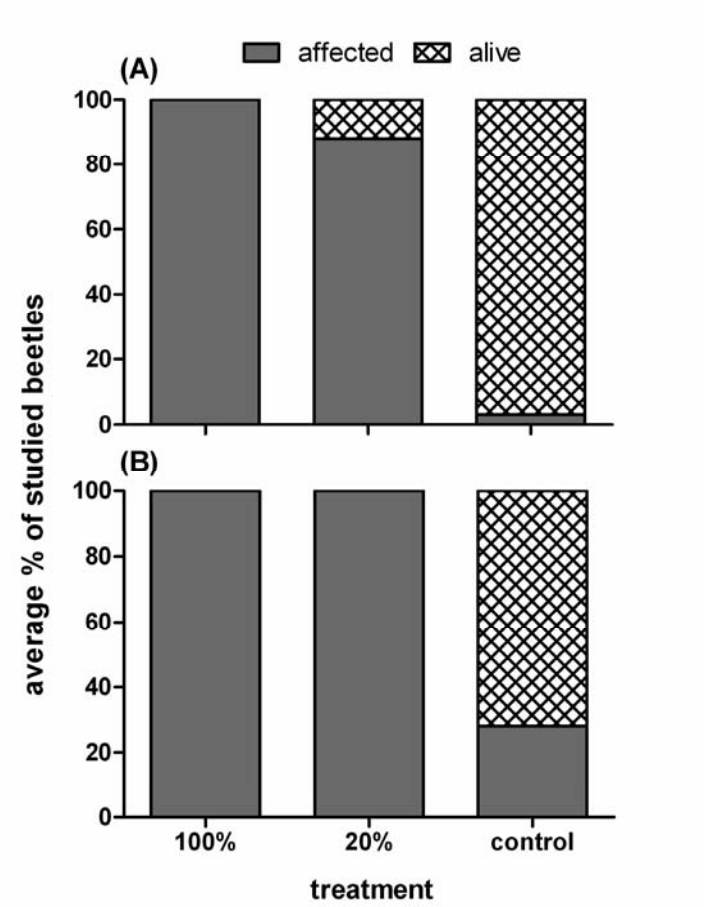

Figure 1. Mean percentage of affected and live *Meligethes aeneus* adults after treatment with pyrethroid (lamda-cyhalothrin) solution (100% and 20% of field application rate and acetone as control) from Jõgeva (A) and Tartu County (B) in 2009.

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