



## **A large field trial to assess the short-term and long-term effects of several 4 insecticides used to control the pollen beetle on parasitic hymenoptera in oilseed rape**

**Jean-Pierre Jansen, Gilles San Martin Y Gomez**

*Plant Protection and Ecotoxicology unit, Life Sciences Department, Walloon Agricultural Research Centre, Gembloux, Belgium*

**Abstract:** A large scale field trial was performed in spring 2015 to assess the effects of several insecticides used to control the pollen beetle on parasitic hymenoptera in oilseed rape. The tested products were Avaunt 150EC, Steward 30WG (both containing indoxacarb), Plenum (pymetrozine), Mavrik 2F (tau-fluvalinate), Biscaya (thiacloprid) and Cyren 4E (Chlorpyrifos-ethyl). The insecticides were applied at their commercial rates soon before flowering on large strips of oilseed rape. A strip was left untreated as control. Insects were sampled with the help of beating methods and sweep net sampling from DBT1 (five day before treatment) to DAT43. The direct effects of the products were assessed on adult pollen beetle (target pest) and adult parasitic hymenoptera (Tersilochinae and Pteromalidae). In the context of IPM, long-term effects were assessed on pollen beetle larvae to determine their parasitism rate and estimate the balance of parasitic hymenoptera/pollen beetle that could be extrapolated for the next seasons.

All products were effective to control adult pollen beetle population at least three days after application. Avaunt 150EC, Steward 30WG and Cyren 4E were the most effective products with significant reduction of pollen beetle populations 7 days after treatment and for a longer period than the other products.

Significant reductions were observed for Biscaya and Cyren 4E for adults of the Tersilochinae subfamily, the main subfamily parasitizing pollen beetle larvae, with 65% and 62% less insects compared to the control, respectively. No effects were observed with the other insecticides. No insecticides had significant effects on adult Pteromalidae, the main parasitic hymenoptera attacking weevil larvae. The other families were only collected in few numbers. The parasitism of the pollen beetle larvae were significantly reduced by Biscaya and Cyren 4E, compare to control and all the other insecticides tested. The balance between the parasitic wasp and the adult pollen beetle for the next seasons, extrapolated from the analysis of the larval parasitism gave similar results, with Biscaya and Cyren 4E leading to a reduction of the balance in favour of the pollen beetle, with 70% and 62% less parasitic hymenoptera expected for the next year, respectively. The other insecticides did not impact negatively this balance and were considered as least as neutral for the parasitic wasps.

In the context of IPM, Avaunt 150EC, Steward 30WG, Plenum and Mavrik F demonstrated efficacy in term of adult pollen beetle control and did not present a negative impact on the balance parasitic wasp/pollen beetle that could be obtained at the next generation. Therefore, the use of these insecticides can be recommended in oilseed rape to control pollen beetle populations in IPM, with an absence of negative short-term and long-term effects.

**Key words:** oilseed rape, pollen beetle, parasitoid, Tersilochinae, Pteromalidae, insecticide

## Introduction

The pollen beetle has become one of the most important pest problems in oilseed rape in Western Europe during these last 20 years. The repeated applications of insecticides in spring on this crop led to the apparition of population resistant to most of the products (Thieme *et al.*, 2010; Slater *et al.*, 2011). As the insecticide solutions appear more and more as a dead end, other control methods, including natural control by beneficial arthropods, are now considered. Specific hymenoptera belonging to the Tersilochinae subfamily (Hym.; Ichneumonidae) were identified as the key pollen beetle natural enemy (Ulber *et al.*, 2006; 2010 a), with larval parasitism rates across Europe in the range of 25-50% in unsprayed crops, or even higher in some cases. However, beneficial insects alone could be not sufficient for an effective control in all cases and insecticides still need to be used, at least from times to times. In this context, products that could be compatible with beneficial arthropods are requested in order to maintain the potential of natural control for the forthcoming years.

Several insecticides still in use in oilseed rape were already assessed on pollen beetle parasites. Most of the products were highly toxic for the adults, with reduction of the adult populations regularly higher than 50% compared to the untreated control. These products had also a negative impact on pollen beetle parasitism (Halden, 2004; Ulber *et al.*, 2010 b). Compared to other pyrethroids insecticides, tau-fluvalinate (Mavrik 2F) appeared as the less toxic product, but the effects cannot be considered as negligible in all studies.

In 2013 and 2014, large field trials were performed in Belgium to assess the effects of different products (Jansen and San Martin, 2014 a; 2014 b). The main results were that insecticides as pymetrozine, tau-fluvalinate and phosmet had no detrimental effects on pollen beetle larval parasitism, leading to comparable levels of parasitoids' production for the next years than the untreated control, while thiacloprid and chlorpyrifos-ethyl drastically reduced these populations and furthermore reduced the potential of natural control for the next seasons. The aim of the field trial performed in 2015 and presented here was to repeat these experiments and include new products containing indoxacarb, in order to determine their ecotox profile compared to products already assessed.

## Material and methods

### *Test products*

The test was implemented in a commercial oilseed rape field cropped under the current agricultural practices for the area. The field was located at Onhaye (Ferme de Lenne, 50°14'20"N, 4°51'40"E). The trials (about 5,0ha with the borders) were implemented in a 18.0 ha commercial oilseed rape field. No insecticides were applied before and during the tests, except the test products. The part of the field not used for the test was treated with Biscaya (Thiacloprid) two days before the application of test products. The whole field was treated with a fungicide during flowering (Cantus 0.5 kg/ha, boscalid 50% WG). The field site was divided into 7 strips of 30 m × 200 m that received one of the 6 insecticide treatments or was left untreated (control). Each strip was divided into four plots of 30 m × 50 m, each plot being considered as a replicate. All samplings and visual counts were done in the 20 m × 40 m central part of each plot. The field was nearly completely surrounded by woods.

The tested insecticides were Plenum (pymetrozine, 500 g/l, WG), applied at 150 g/ha, Biscaya (thiacloprid, 240 g/l, OD) applied at 300 ml/ha, Mavrik 2F (tau-fluvalinate, 240 g/l, EW) applied at 200 ml/ha, Avaunt 150EC (indoxacarb, 150 g/l, EC) applied at 170 ml/ha, Steward 30WG (indoxacarb 300 g/kg) applied at 85 g/ha and Cyren 4E (chlorpyrifos-ethyl,

480 g/l EC) applied at 375 ml/ha. The doses corresponded to the commercial recommended rates. The insecticides were applied on the 22<sup>th</sup> of April at the end of the bud stage (BBCH 55-59), on basis of a volume of 200 l spray mixture/ha.

### ***Sampling techniques***

Different sampling techniques were used during the experiments, according to the main target insects followed and the climatic conditions: beatings (plastic trays and funnels) and sweep net methods. For more details on the sampling techniques, please consult previous publications (Jansen & San Martin, 2014 a; 2014 b). The first samples were taken 1 day before treatment, then 1 and 3 days after treatment (DAT) and weekly from DAT7 to DAT43.

### ***Presentation of the results***

The short term effects that concerned the field where the products were applied and the longer-term effects that concerned the pest/beneficial balance for the following years were both taken into consideration.

The short-term effects were assessed on adults of pollen beetle (target pest) and adults of parasitic hymenoptera, identified at the subfamily level for Tersilochinae (mainly pollen beetle larval parasitoid) and family level for Pteromalidae (mainly seed weevil larval parasitoid).

The long-term effects were assessed on pollen beetle larvae populations and pollen beetle larvae parasitism. These two results were used to estimate the balance between adult parasitoid and adult pollen beetle that could be produced by the treated area for the next growing season, according to the insects that survived to the insecticide treatments. These balances were calculated as follow for each plot:

$$R = (L_p/L_{np}) * 100$$

R = balance; L<sub>p</sub> = pollen beetle larvae parasitized sampled in the corresponding plot; L<sub>np</sub> = pollen beetle larvae unparasitized sampled in the corresponding plot

A R value = 100 is indicating that the corresponding plot will potentially produce the next year the same number of adult parasitoids as adult pollen beetle, if all larvae gave an adult for the next season. It corresponds also to a level of parasitism of 50%. There are different factors that will affect the success of unparasitized and parasitized larvae development in the soil after the season (tillage, climatic and pedologic conditions), but it is considered that these factors are not related to the insecticides used and will equally affect all groups, treated or not. Thus, the estimation of these ratios could be a basis for a comparison of long-term adverse effects of different products tested on the same populations and untreated control. It can also be considered as a simple modification of the expression of the percentage parasitism data.

### ***Statistical analysis***

The results were analysed with the help of R software. One-way Anova tests were performed at 0.05 level. Pair-wise comparisons were performed using Tukey (multiple comparisons) and Dunnet-test (comparison to the control for efficacy results) at 0.05 level. Several data were log transformed to normalise the distribution before analysis.

## Results

### *Adults of pollen beetle*

The numbers of adult pollen beetles sampled at the different dates are given in Figure 1. All treatments were effective one and three days after product application, with a reduction of pollen beetle population higher than 90%. After one week, the plants start to be in flowering and the pollen beetles were redistributed in all plots, leading to a migration of the population from the control to the other plots. After these dates, the field was fully flowering and the pollen beetle no longer considered as a pest.

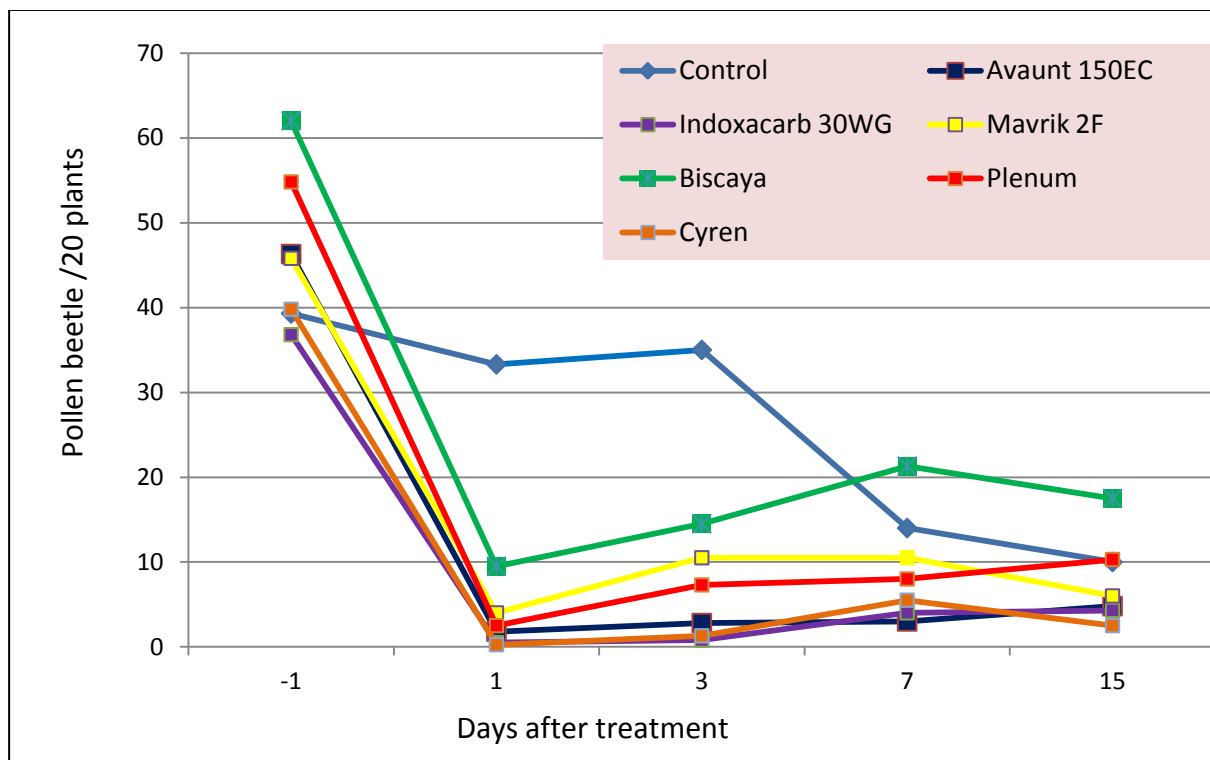


Figure 1. Populations of adult of pollen beetles sampled before and after application of the insecticides. Number of pollen beetle for 20 plants.

### *Parasitic hymenoptera adults*

The effects of insecticides on numbers of adult parasitoids sampled during the experiments are listed in Figure 2 (Tersilochinae) and Figure 3 (Pteromalidae), the two main groups of hymenoptera found. Specimens of other families were sampled from time to time (less than one per plot, in a mean) and were not considered. The results of all samples, from DAT1 to DAT43 were pooled.

A mean of 17.0 adults of Tersilochinae were collected in the control plots. No insecticides were different from the control and the only difference was observed between two treatments, the one with the most abundant population (Avaunt 150EC, 19.8 adults and the less abundant (Cyren 4E, 11.0 adults).

No differences between treatments and control were observed for the number of adults of Pteromalidae. These results were similar to those obtained the previous years (Jansen & San Martin, 2014 a; 2014 b).

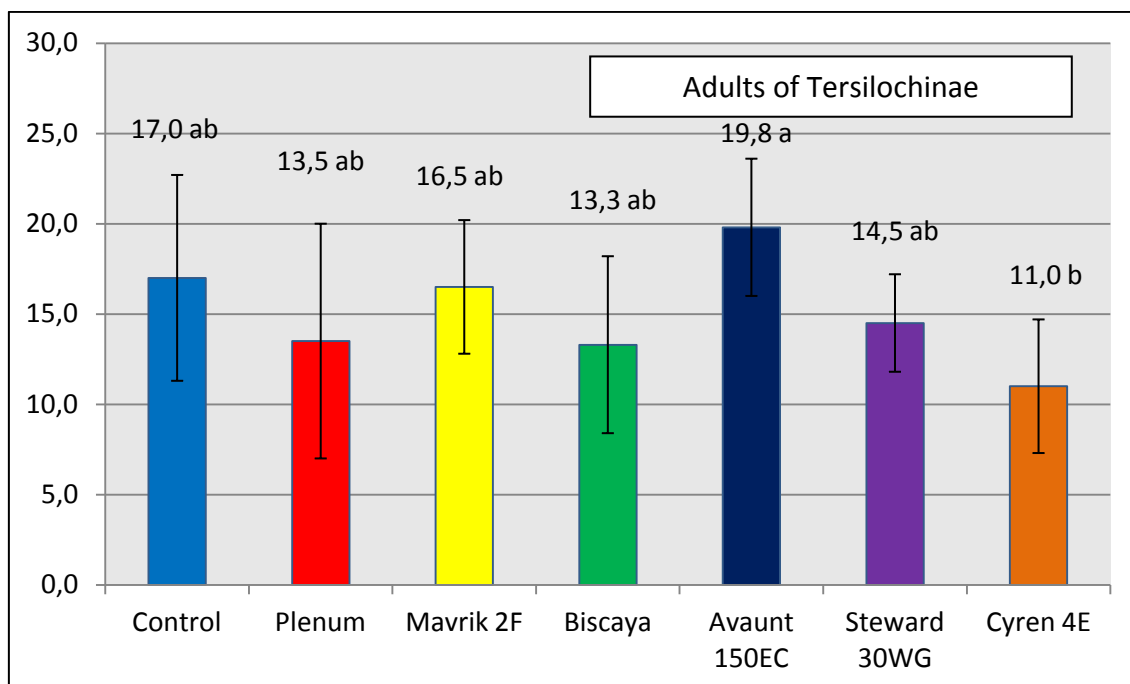


Figure 2. Number of adults of Tersilochinae sampled in each treatment from DAT1 to DAT43. GLM, One-way Anova ( $p = 0.05$  level) followed by pair-wise Tukey test for population (results followed by the same letter are not statistically different) and Dunett test for efficacy (a = not different to control, b = different to control).

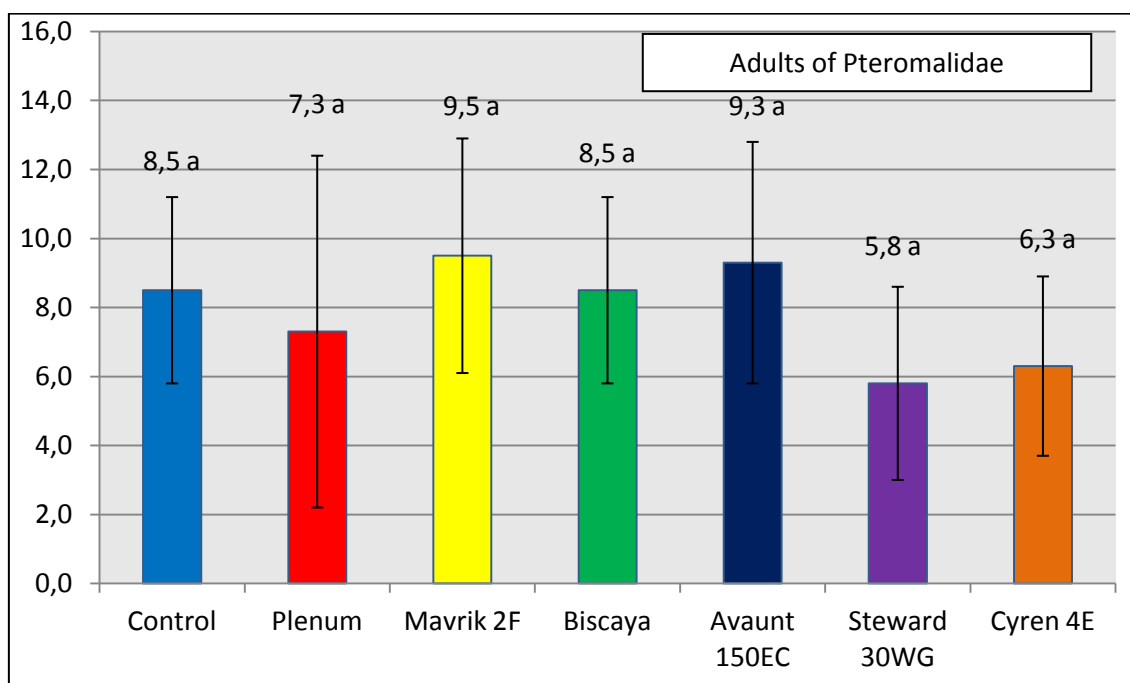


Figure 3. Number of adults of Pteromalidae sampled in each treatment from DAT1 to DAT43. GLM, One-way Anova ( $p = 0.05$  level) followed by pair-wise Tukey test for population (results followed by the same letter are not statistically different) and Dunett test for efficacy (a = not different to control, b = different to control).

### ***Long term effects – Pollen beetle larvae and parasitic hymenoptera larvae***

The parasitism rates of larvae of pollen beetle are given in Figure 4. It was low in the control, with only 3.48% of the larvae parasitized, while the mean parasitism rates generally observed in Belgium is about 10%. Despite this low level, significant differences were observed with two products, Biscaya and Cyren 4E. The other ones, including the two indoxacarb formulations that were tested for the first time, were not different from the control and also significantly different from the two most toxic products. For the products that were previously assessed with the same methodology in 2013 and 2014, the results were similar, with the absence of effects of Plenum and Mavrik 2F and significant effects of Biscaya and Chlorpyrifos based products.

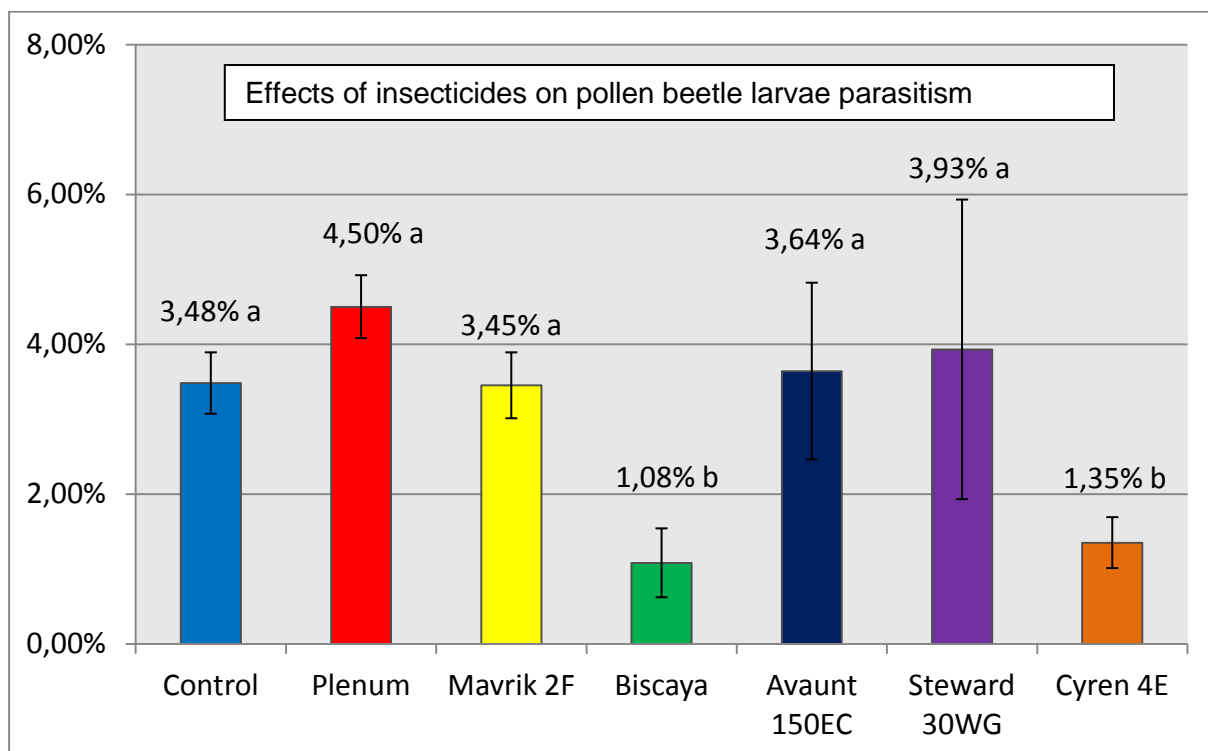


Figure 4. Effects of insecticides on pollen beetle larval parasitism. Percentages of pollen beetle larvae parasitized collected between DAT1 and DAT43. One-way Anova ( $p = 0.05$  level) followed by pair-wise Tukey test (results followed by the same letter are not statistically different).

When the parasitism rates are used to estimate the potential impact of the insecticides for the next generation (Figure 5), comparing the number of adult of parasitoids that will be produced by cohort of 100 pollen beetle, the main conclusions as for larval parasitism can be established from the results, with an important reduction of the ratio parasite/pollen beetle with Biscaya and Cyren 4E and no effects for the other products. For products already assessed in 2013 and 2014, the conclusions were similar.

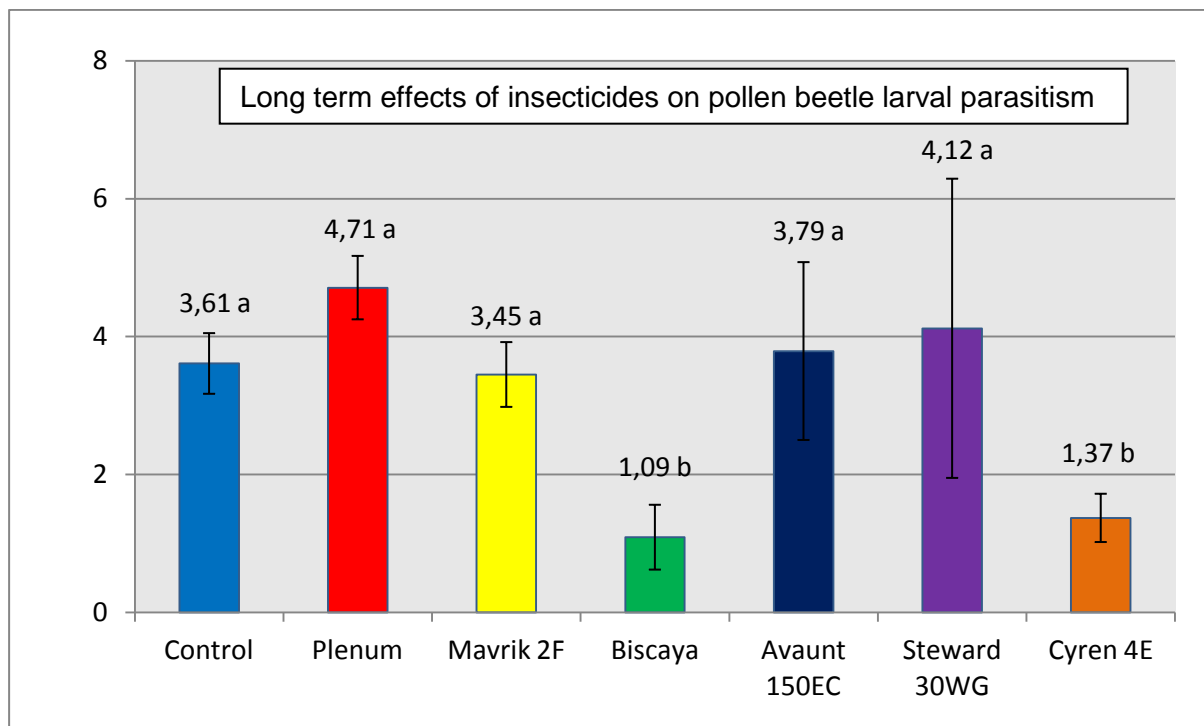


Figure 5. Long term effects of insecticides on pollen beetle larval parasitism. Assessment of potential number of adult parasitic wasps that could be obtained at the next generation for 100 adults of pollen beetle. One-way Anova ( $p = 0.05$  level) followed by pair-wise Tukey test (results followed by the same letter are not statistically different).

## Discussion

The products assessed during this study have shown different effects on pest and beneficial populations. In term of adult pollen beetle control, all treatments showed efficacy on the pest directly after the application and can be considered as effective for pollen beetle control, at least soon before flowering.

The effects of the products on adult parasitic wasps were significant for two products, Biscaya and Cyren 4E, with reduction of 70 and 60% of the populations. Plenum, Mavrik 2F, Avaunt 150EC and Steward 30WG were not different from the control. The same conclusions were found for the long-term effects. For Biscaya, Cyren 4E, Plenum and Mavrik 2F, the results were comparable to those obtained in 2013 and 2014, with the same methods, but on another field site and other pest and beneficial populations, climatic conditions, levels of parasitism, date of treatment and agricultural conditions.

When all results obtained during the 2013-2015 period were pooled (Table 1), two categories of products appeared: those that are both effective to control pollen beetle adult before flowering and had potentially no long term impact on parasitic wasps that attack this pest: Plenum, Steward 30WG, Avaunt 150EC, Mavrik 2F and Phosmet, and those that have both short term effects on adult parasitic wasps and long term effects on the balance parasitic wasps/pollen beetle for the next generations. Some of these products were tested three successive years, other ones two years (Phosmet) or only one year (Indoxacarb based products) but with two different formulations. According to the similarity of results obtained with different formulation and between years despite sometimes huge difference in term of

insect populations, timing of application of products and climatic and agricultural conditions, a relative high confidence can be given to the methodology followed and to the main conclusions.

Table 1. Evaluation and classification of the products used in the years 2013-2015.

<b>Product (active substance)</b>		<b>2013</b>	<b>2014</b>	<b>2015</b>
Plenum	R (%)	32.50%	2.20%	-30.50%
(pymetrozine)	<b>IOBC class</b>	<b>2</b>	<b>1</b>	<b>1</b>
Mavrik 2F	R (%)	38.40%	14.20%	4.40%
(tau-fluvalinate)	<b>IOBC class</b>	<b>2</b>	<b>1</b>	<b>1</b>
Steward 30WG	R (%)	-	-	-5.00%
(Indoxacarb)	<b>IOBC class</b>	-	-	<b>1</b>
Avaunt 150EC	R (%)	-	-	-14.10%
(Indoxacarb)	<b>IOBC class</b>	-	-	<b>1</b>
Boravi WG	R (%)	-1.10%	2.40%	-
(Phosmet)	<b>IOBC class</b>	<b>1</b>	<b>1</b>	-
Biscaya	R (%)	78.30%	100.00%	69.80%
(Thiacloprid)	<b>IOBC class</b>	<b>4</b>	<b>4</b>	<b>3</b>
Pyrinex / Cyren 4E	R (%)	60.30%	57.10%	62.00%
(Chlorpyrifos-ethyl)	<b>IOBC class</b>	<b>3</b>	<b>3</b>	<b>3</b>

On basis of this sum of experiments and results, a positive list, containing Plenum, Indoxacarb based products, Phosmet and Mavrik 2F can be recommend for the farmer to be used to control the pollen beetle before flowering, because they are both effective and have no long term impact on parasitic hymenoptera, the main pollen beetle natural enemies in Belgium. On the other hand, despite a good short term efficacy on the pest, Biscaya and Chlorpyrifos based products are not recommended because of their important long term impact on parasitic hymenoptera populations.

It must also be noted that most of the results obtained on Tersilochinae in oilseed rape followed the same trends that results obtained with the same products on other parasitoid species in the laboratory, on inert surfaces or on plants (Van de Veire & Tirry, 2003; Medina *et al.*, 2008; Jansen, 2010).



## References

- Halden, P. 2004: Influence of Tau-Fluvalinate on parasitoid abundance and parasitisation rate of pollen beetles in spring oilseed rape (in Swedish). Master Science Thesis, Dept. Crop Science SLU, Alnarp, Sweden.
- Jansen, J.-P. 2010: Beneficial arthropods and pesticides: building selectivity lists for IPM. IOBC-WPRS Bulletin 55: 23-47.
- Jansen, J.-P., Defrance, T. & Warnier, A. M.: 2011: Side effects of flonicamide and pymetrozine on five aphid natural enemy species. BioControl 56: 759-770.
- Jansen, J.-P. & San Martin, G. 2014 a: A large field trial to assess the short-term and long-term effects of 5 insecticides used to control the pollen beetle on parasitic hymenoptera in oilseed rape. IOBC-WPRS Bulletin 103: 9-16.
- Jansen, J.-P. & San Martin, G. 2014 b: A large field trial to assess the short-term and long-term effects of 4 insecticides used to control the pollen beetle on parasitic hymenoptera in oilseed rape. IOBC-WPRS Bulletin 104: 67-74.
- Medina, P., Morales, J. J., Gonzalez-Nunez, M. & Vinuela, E. 2008: Is the use of some selected insecticides compatible with two noctuid endoparasitoids: *Hyposoter didymator* and *Chelonus inanitus* ? IOBC-WPRS Bulletin 35: 51-58.
- Slater, R., Ellis, S., Genay, J.-P., Heimbach, U., Huart, G., Sarazin, M., Longhurst, C., Müller, A., Nauen, R., Rison, J.-L. & Robin, F. 2011: Pyrethroid resistance monitoring in European populations of pollen beetle (*Meligethes* spp.): a coordinated approach through the Insecticide Resistance Action Committee (IRAC). Pest Man. Science 67: 633-638.
- Thieme, T., Heimbach, U., Müller, A. 2010: Chemical Control of Insect Pests and Insecticide Resistance in Oilseed Rape. In: Biocontrol-Based Integrated Management of Oilseed Rape Pests (ed. Williams, I. H.): 313-335. Springer, The Netherlands.
- Ulber, B., Williams, I. H., Klukowski, Z., Luik, A. & Nilsson, C. 2010 a: Parasitoids of oilseed rape pests in Europe: Key species for conservation biocontrol. In: Biocontrol-Based Integrated Management of Oilseed Rape Pests (ed. Williams, I. H.): 45-76. Springer, The Netherlands.
- Ulber, B., Klukowski, Z. & Williams, I. H. 2010 b: Impact of insecticides on parasitoids of oilseed rape pests. In: Biocontrol-Based Integrated Management of Oilseed Rape Pests (ed. Williams, I. H.): 337-355. Springer, The Netherlands.
- Van de Veire, M. & Tirry, L. 2003: Side effects of pesticides on four species of beneficials used in IPM in glasshouse vegetable crops: “worst case” laboratory tests. IOBC-WPRS Bulletin 26(5): 41-50.