Findings of insecticide monitoring in Nordic and Baltics

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Key findings

Pollen beetles:

- λ-cyhalothrin resistance is widely spread throughout the region. Efficacy of λ-cyhalothrin was similar in Scandinavia over time, while a decrease in susceptibility was observed in the Baltic countries
- τ-fluvalinate remains effective. However, a drop in susceptibility was observed in 2018
- Acetamiprid remains effective in Scandinavia, but decreased susceptibility was observed in Lithuania

Clover head weevils:

- Control issues have been reported. λ-cyhalothrin resistance seems widely spread throughout Denmark. Most of the tested samples proved resistant in different degrees to λ-cyhalothrin.

Clover seed weevils:

- λ-cyhalothrin resistance has been observed in Denmark. Some of the tested samples proved resistant in different degrees to λ-cyhalothrin.

Introduction

This report aims to give a short and concise overview of the data obtained by members of the NorBaRAG insecticide subgroup from 2016-2021 and described the overall trends observed for resistance monitoring over a five-year period for pollen beetles and three-year period for clover weevils.

All members of NorBaRAG are encouraged to contribute to the collection of data for future reports by sending an overview of data to the insecticide subgroup chair. A range of actives were tested according to appropriate IRAC methods.

Method

A total of three active substances with two different modes of action were tested using standard IRAC susceptibility test methods. The tested actives were λ-cyhalothrin, τ-fluvalinate (synthetic pyrethroids) and acetamiprid (neonicotinoid).

The standard IRAC susceptibility test is performed as an Adult Vial Test (AVT), where the effect on insects by several dose rates are tested. Glass vials are coated with the relevant active by adding the dissolved active to the vials and letting the solvent evaporate under rotation of vials. Glass vials coated with acetone functions as control treatment.

Figure 1: Example of adult vial test

The target insects (normally 10 adults per vial) are placed in glass vials coated with the relevant insecticide active for 24 hours, after which number of affected and alive beetles are noted and %affected beetles is calculated. In some cases, the insects are moved to control vials for recovery. To ensure movement, beetles should be exposed to light during the experiment.

Active	Mode of action (IRAC)	IRAC Method used	Amount of active tested (ng/cm^2)
λ-cyhalothrin	Pyrethroid (3A)	Method 011	75
τ-fluvalinate	Pyrethroid (3A)	Method 011	480
Acetamiprid	Neonicotinoid (4A)	Method 021	400

Methods 011 and Method 021 are developed for testing pollen beetles. For more information on specific IRAC methods, please see https://www.irac-online.org/methods/.

The monitoring results for pollen beetles are divided into three parts, presenting data for each active substance individually. The monitoring results for clover head weevils and clover seed weevils are combined in a separate section.

Brassicogethes aeneus

Brassicogethes aeneus (also known as *Meligethes aeneus*) is an important pest of oilseed rape particularly in Western Europe. The larvae are up to 3 mm long and white with brown sclerotised plates. Eggs are laid in the flower buds of the host plant. The larvae develop within the flowers. Oviposition damage to the buds of oilseed rape can cause the flowers to drop off. Both adults and larvae feed on the pollen and nectar in the flowers.

Resistance to pyrethroids has been reported in this species since 1999, especially against λ-cyhalothrin being the most monitored pyrethroid. Initial reports suggest that pyrethroid resistance first occurred in North-East France and over the following years has spread to other countries in Europe. Investigations into the mechanism of resistance has primarily identified enhanced metabolism by P450 monooxygenases as the prime mechanism of resistance to pyrethroids. More recently, a target site mutation has been identified in some λ-cyhalothrin resistant populations from Denmark and Sweden[1].

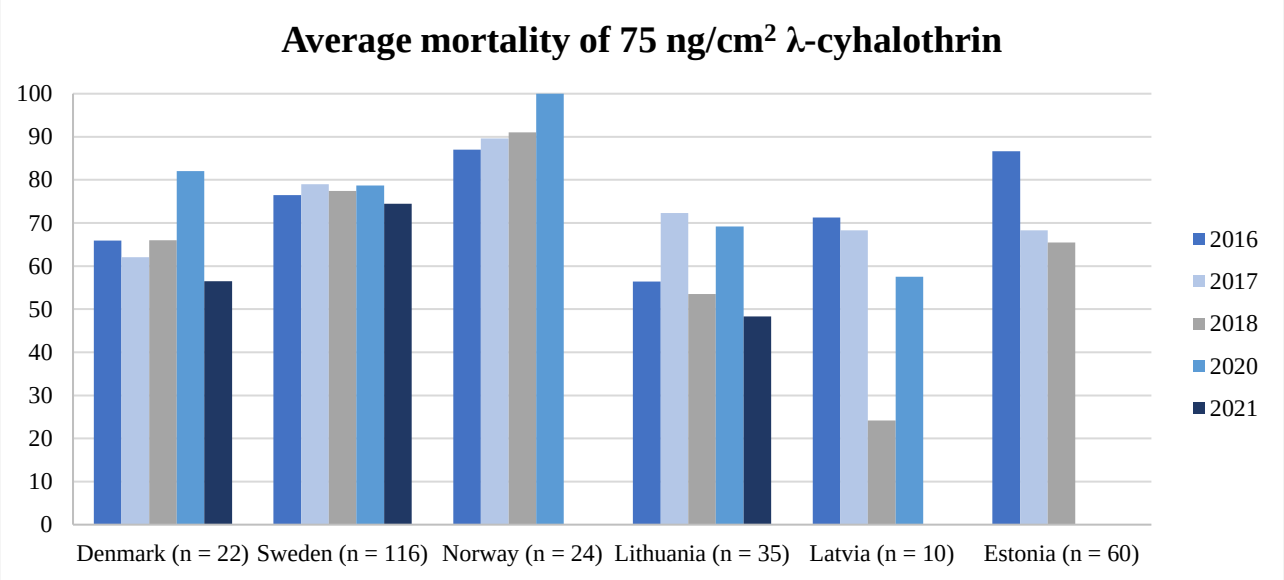
λ-cyhalothrin

IRAC Method 011 tests the effect of synthetic pyrethroids, including λ-cyhalothrin, on pollen beetles in a laboratory setting. It is widely used for monitoring of sensitivity of *B. aeneus* throughout Europe. Beetles are

[1] Kaiser, C., Jensen, KM.V., Nauen, R. et al. J Pest Sci (2018) 91: 447. https://doi.org/10.1007/s10340-017-0856-x

placed in glass vials coated with λ-cyhalothrin for 24 hours, after which number of affected and alive beetles are noted and %affected beetles is calculated. Glass vials coated with acetone functions as control treatment.

The presented data represent the efficacy of 100% field rate (7.5 g/ha λ-cyhalothrin). Data was obtained from Latvia, Estonia, Lithuania, Finland, Denmark, Norway and Sweden. No data from Finland is included as data was limited. A total of 270 population samples were collected and tested in the lab against λ-cyhalothrin. Samples where the control mortality was above 20% were not included.

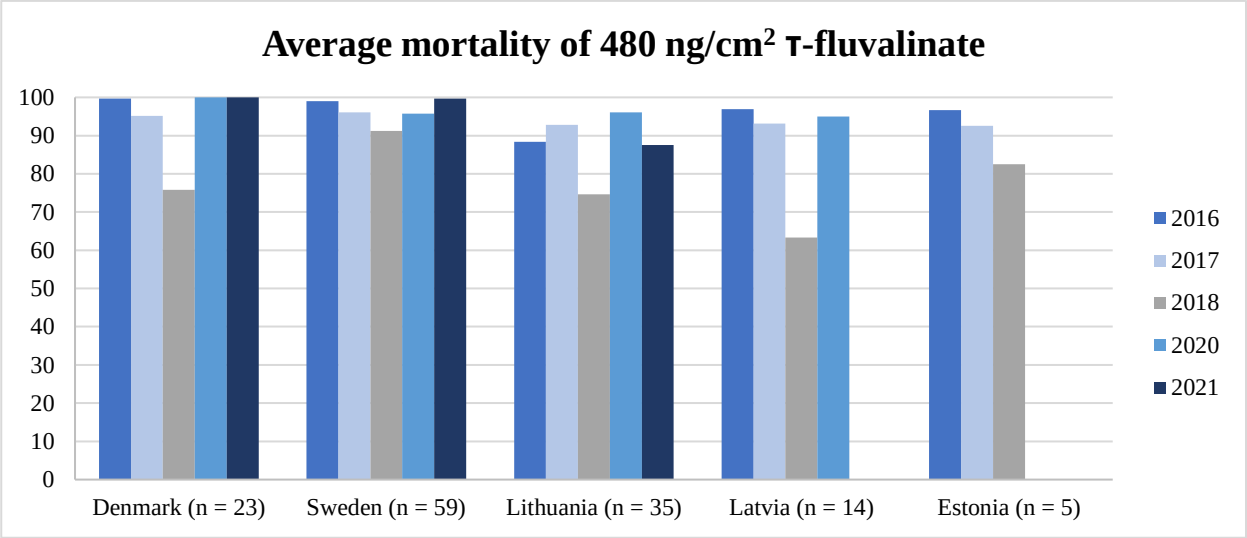


The average mortality of beetles exposed to 75 ng/cm^2 λ-cyhalothrin remained at a similar level in Denmark, Sweden and Norway over a five-year period. However, a decrease in susceptibility was observed in 2021 for Denmark. In Lithuania, susceptibility varies greatly between years, possibly due to the relative low number of samples tested each year (3-10 samples per year). For Latvia and Estonia, mortality by λ-cyhalothrin on pollen beetles seems to decrease over time, with a significant drop in Latvia in 2018 which were countered in 2020. But no new 2021 data was included for Estonia and Latvia. Most of the pollen beetle population samples were resistant to λ-cyhalothrin to various degree, indicating a possible issue with controlling pollen beetles in the field.

τ-fluvalinate

IRAC Method 011 tests the effect of synthetic pyrethroids on pollen beetles in a laboratory setting. The standard of Method 011 is λ-cyhalothrin, however, τ-fluvalinate can also be tested with this method as long as differences in inherent potency is considered. Like for λ-cyhalothrin, beetles are placed in glass vials coated with τ-fluvalinate for 24 hours, after which number of affected and alive beetles are noted and %affected beetles is calculated. Glass vials coated with acetone functions as control treatment.

The presented data represent the efficacy of 100% field rate (480 ng/ha τ-fluvalinate). Data was obtained in 2016-2018 and 2020-2021 from Latvia, Estonia, Lithuania, Denmark and Sweden. No data from Finland was available. Furthermore, only 2018 and 2020 data exists for Norway so they are not included here. A total of 136 population samples were collected and tested in the lab against τ-fluvalinate. Samples where the control mortality was above 20% were not included.



The average mortality of beetles exposed to 480 ng/cm^2 τ-fluvalinate was above 88% in all countries in 2016 and 2017. However, the 2018 data suggests a decrease on susceptibility of pollen beetles against τ-fluvalinate. This was the case for both the Nordic and the Baltic countries. The shift in susceptibility was most expressed in Denmark, Lithuania and Latvia. On the other hand, data from 2020 and 2021 suggest susceptibility of pollen beetles as mortality of 480 ng/cm^2 τ-fluvalinate was above 95% in all countries where trials were conducted.

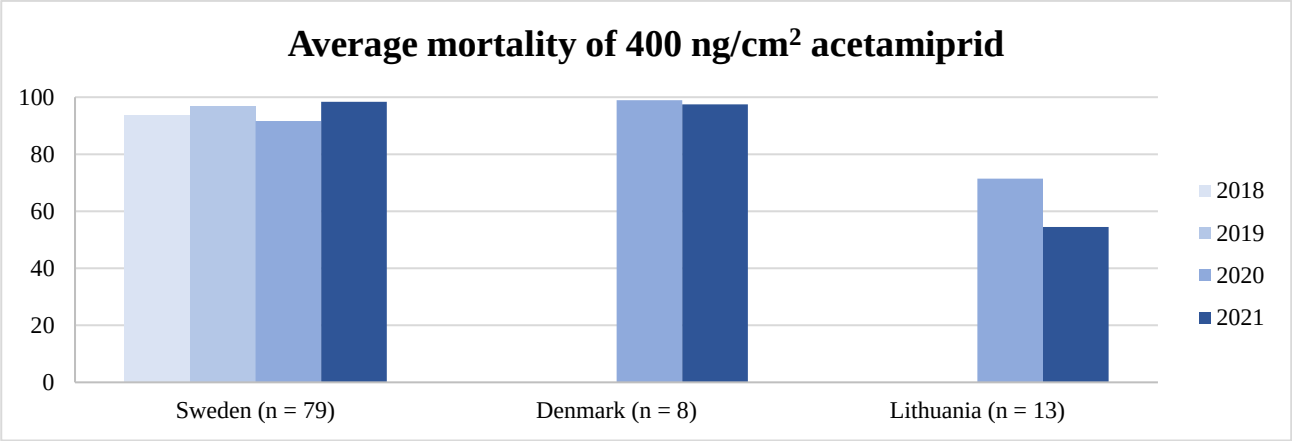
Conclusion on pyrethroids

Both λ-cyhalothrin and τ-fluvalinate function as sodium channel modulators, stimulating repetitive nerve discharges, leading to death through paralysis. Results obtained in the laboratory setting using the adult vial test is not directly referable to field performance of the product but can give indications of possible control issues. Based on 2016-2021 trials, control issues for λ-cyhalothrin might already be present in the field as data show a large proportion of resistant population samples. The efficacy of 100% field rate of τ-fluvalinate was higher than that of λ-cyhalothrin, but for both actives, susceptibility seems to decrease from 2016-2018 primarily in the Baltic region. However, in 2020 increased susceptibility of both actives were observed.

Acetamiprid

Method 021 tests the effect of neonicotinoids on pollen beetles in a laboratory setting. The standard neonicotinoid used is thiacloprid, however, acetamiprid can also be tested with this method if differences in inherent potency is considered. Both acetamiprid and thiacloprid belong to the group of chloropyridinyl neonicotinoids, so it is sensible to also use Method 021 for the monitoring of acetamiprid. Please note that thiacloprid is no longer approved as the grace period ended in 2021. Therefore, acetamiprid is now used as the representative for neonicotinoids in monitoring efforts. Beetles are placed in glass vials coated with acetamiprid for 24 hours, after which number of affected and alive beetles are noted and %affected beetles is calculated. Glass vials coated with acetone functions as control treatment.

The presented data represent the efficacy of 100% field rate (40 g/ha acetamiprid). Data is presented for Sweden (2018-2021), Denmark and Lithuania (2020 and 2021), where a total of 100 population samples were collected and tested in the lab against acetamiprid. Samples where the control mortality was above 20% were not included.



The average mortality of beetles exposed to 400 ng/cm^2 acetamiprid was above 90% in all four years in the Swedish and Danish samples. The data suggest that currently there is no issues in control of pollen beetles, at least in Sweden and Denmark. However, the 13 Lithuanian samples showed a reduced susceptibility of pollen beetles against acetamiprid to a similar degree as observed for thiacloprid before the ban of thiacloprid. More data is required to get the full overview of possible control issues of pollen beetles for the Baltic region.

Conclusion on neonicotinoids

Acetamiprid function as postsynaptic nicotinic acetyl choline receptor modulators, stimulating repetitive nerve discharges, leading to death through paralysis. Results obtained in the laboratory setting using the adult vial test is not directly referable to field performance of the product but can give indications of possible control issues. A total of 100 population samples from Sweden, Denmark and Lithuania were collected and tested against acetamiprid. Based on these data, there is no issues in acetamiprid control of pollen beetles currently in Scandinavia, but issues are already present in the limited number of samples from Lithuania. More data from these and the other countries in the Northern Regulatory Zone is required to give the full overview of the resistance status in the NorBaRAG countries.

White clover weevils

White clover (*Trifolium repens*) is an important component in grassland mixtures and as a green manure crop in Denmark. The White clover seed weevil (*Protapion fulvipes*) and the Clover head weevil (*Hypera meles*) are important pests in clover production. Adults and larvae feed externally on the leaf, stem and pollen, which can cause severe seed yield losses.

λ-cyhalothrin

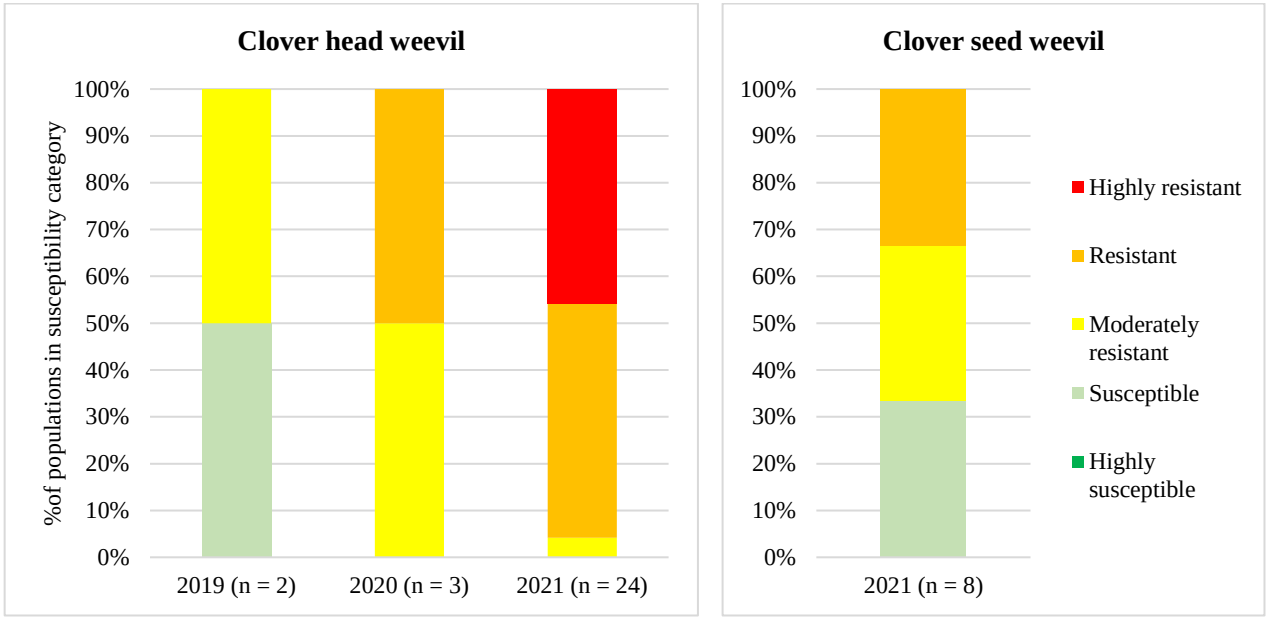
IRAC Method 011 is designed for testing λ-cyhalothrin on pollen beetles in a laboratory setting. The recommended field dose for λ-cyhalothrin for clover weevils is the same as for pollen beetles, therefore IRAC Method 011 can also be used for characterization of λ-cyhalothrin susceptibility.

Weevils are placed in glass vials coated with λ-cyhalothrin for 24 hours, after which number of affected and alive weevils are noted and %affected weevils is calculated. Glass vials coated with acetone functions as control treatment. Sample populations are classified into 5 susceptibility levels, ranging from highly susceptible to highly resistant. This classification will be used in this report. Classification of susceptibility levels depends on the effect of 15 ng/cm^3 and 75 ng/cm^3 (20% and 100% field rate) on weevil samples.

Dose rate (ng/cm^3)	Affected	Classification
75	100%	Highly susceptible
15	100%	
75	100%	Susceptible
15	<100%	
75	90-100%	Moderately resistant
75	50-90%	Resistant
75	<50%	Highly resistant

Data was obtained from Denmark over a three-year period. A total of 29 clover head weevil population samples and eight clover seed weevil population samples were collected and tested in the lab against λ-cyhalothrin.

λ-cyhalothrin susceptibility classification



Of the 29 clover head weevil population samples tested in 2019-2021, a few populations proved susceptible to λ-cyhalothrin. Most of the population samples were resistant to λ-cyhalothrin to different degrees, indicating a possible issue with controlling clover head weevils in the field. In fact, control issues have already been reported. Clover head weevils are much larger than pollen beetles but treated with a similar dose rate. A calibration of vial-test is needed to be more exact in determining λ-cyhalothrin susceptibility for clover head weevils.

Eight clover seed weevil population samples were tested in 2021. There was an almost even distribution of susceptible. Moderately resistant and resistant samples. Of the eight samples, none were highly susceptible nor highly resistance. The initial data could indicate potential resistance issues also for the clover seed weevil. Hopefully, more data will be available in coming years to give a better picture of the efficacy of λ-cyhalothrin against clover weevils.

Overall conclusion

The susceptibility of pollen beetles over a five-year period against a range of insecticides was assessed. A total of three active substances with two different modes of action were tested using standard IRAC susceptibility test methods developed for pollen beetles.

More than 270 population samples of pollen beetles were tested in 2016-2020 at 100% field rate. Results show that resistance against λ-cyhalothrin is widely spread throughout the region. Efficacy of λ-cyhalothrin was maintained in the Nordic countries but decreased in the Baltic countries. For τ-fluvalinate, acceptable efficacy is maintained. From April 2020, the neonicotinoid thiacloprid is no longer approved. Acetamiprid is now used as the reference component in monitoring of neonicotinoid resistance. Monitoring data on acetamiprid was tested in Sweden, Denmark and Lithuania in 2018-2021. For Scandinavia, data showed average mortalities above 90% in all years, while decreased susceptibility was observed in Lithuania.

A total of 29 population samples of clover head weevil and eight of clover seed weevil were tested according to IRAC Method 011 and classified according to susceptibility against λ-cyhalothrin. For clover head weevils, the control issues reported by farmers was supported by data from 2019-2021, that showed decreased susceptibility of the clover head weevil, with no susceptible samples in 2021. For the clover seed weevil, no control issues have been reported yet, but the data presented here, indicate that resistance may be emerging. More data in weevil susceptible would be beneficial in coming years to get a better understanding of λ-cyhalothrin resistance status.

Acknowledgements

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