Could it be possible to disturb the insects' behaviour and to reduce damages of pollen beetle and stem weevil?

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Introduction

Concerns about the adverse impacts of pesticides on the environment and their inevitable negative side-effects on non-target organisms have been growing since the 1960's. As a consequence, regulatory bodies take into account the environmental effects of pesticides applications, leading to increased restrictions in their use or to their banning (van der Werf, 1996). For winter oilseed rape (*Brassica napus* L.) in France, insecticides constitute the largest component of the Treatment Frequency Index (TFI). Among insects, Pollen beetle (*Meligethes aeneus* Fabricus) (Coleoptera:Nitidulidae) and stem weevil (*Ceuthorhynchus napi* Gyl) are among the most damaging insect pest of WOSR throughout severe bud and stem injuries (Alford et al., 2003). The pollen beetle adults feed on pollen from flowers and buds, and females lay their eggs on buds suitable for oviposition (2-3 mm long) (Nilsson, 1994). Serious yield losses, over 70 %, can result from pollen beetle attacks due to bud abortion (Nilsson, 1987; Ekbom & Borg, 1996). The main actual way to control pollen beetle and stem weevils is the intensive use of pesticides treatments, but the emergence of metabolic resistance to pyrethroïds has made beetle attacks more severe. Designing new chemical-free crop management, which could reduce pest injuries, damage- and yield losses is a challenge for this crop. Since each cultural practice is likely to modify the interaction with the pest, it could possible to design cropping systems to minimize crop loss induced by pest population with a limited use of chemical control (Valantin-Morison et al., 2007, Mediene et al., 2010). However, there is a lack of knowledge on pest-crop-natural enemies interactions. Host quality is considered as a key factor in host's selection and various farming practices could modify host quality. So N fertilization, sowing density and different cultivar and/or species may play a significant role in attractiveness of the crop through "push-pull" actions, through its influence on emissions of crucifer-specific olfactory cues involved in host plant location (Valantin-Morison et al. 2007; Evans and Allen-Williams, 1994; Smart and Blight, 2000; Staley et al. 2010, Cook et al., 2007). Furthermore nitrogen application and cultivar and/or species may have an important effect on compensation mechanisms. The aim of this study is to analyse, quantify and understand the effect of cultivar and nitrogen management on the occurrence of stem weevil and pollen beetle and on their damage on the crop, in order to design "insecticides-free" strategies.

Materials and methods

Experimental design

Several experiments were carried out in 2006, 2007 and 2009, 2010 in the north of France (Versailles (Ile de France) 48°80'N, 2°13' E and Coudres (Normandie) 48°86'N, 1°24). The experimental plots were laid in a three factorial split-plot design with three or two repeated blocks, depending on the year. Cultivar was the treatment randomly assigned to each sub-plot of nitrogen levels and insecticides treatments. For each experiment, from 2 to 5 cultivars and mixing cultivar, composed of 4 different cultivars, were sown at normal density (40 plants/m^2). Three nitrogen regimes were applied: N1: 50% of nitrogen rate is applied early (BBCH phenological Gowth Stage) (GS) N°33 and GS50; N2 : 100% of nitrogen rate is applied early on growth stage between GS33 and GS50 and N3 : 100% of nitrogen rate is applied late in the season on growth stage between GS50 and GS57.

Pests' abundance and damage

Crop attacks by insects were estimated either through a number of insects on a sample of plants and the occurrence of the damage. Pollen beetle population surveys were carried out during the bud development stage between GS 50 and GS 65 every years, while stem weevils attacks were recorded in 2007, 2010 and 2011. During this period, pollen beetles were sampled using a D-Vac suction sampler every week on 10 plants per micro-plot, randomly selected. As it was difficult to trap stem weevil, we counted the number of stem weevil holes per plant after during the stem elongation, 1 week after the first emergences of stem weevils detected in the Moericke traps, (between GS53 to GS60). At the end of flowering (for damage of stem weevil the first year) and in early summer at GS 80 (for pollen beetle damage and stem weevil damage the second and third year) we randomly selected 10 plants per 1 m^2 micro-plot to assess yield components (number of racemes and pods), the proportion of plants with distorted stem and the pollen beetle damage. By counting the number of podless stalks and the number of pods for each plant we estimated the blooming flower abortion linked to the incidence of blossom pollen beetle damage (Nilsson, 1994).

Periode of flowering	*Early flowering*		*Late flowering*		*Mixing cultivars*
Location of field trials / genetic types	*Hybrids*	*Line cultivars*	*Hybrids*	*Line cultivars*	
Grignon (78) 2006-07				Campala, Grizzly	
Versailles (78) 2007-08		Ecrin	Exagone	Campala, Grizzly	×
Versailles (78) 2009-10	ES Hydromel	Olphi	Exagone	Alesi, Grizzly	×
Coudres (27) 2009-10		Alpaga	Mendel	Kadore	
Versailles (78) 2010-11	ES Hydromel	Oplhi	Exagone	Grizzly	×
Coudres (27) 2010-11	ES Neptune	Alpaga	Mendel	Kadore	×

Table 1 : Characteristics of cultivars experienced in the several experiment trials.

Plant growth

For each micro-plot, whole plants from a micro-plot of 1 m^2 were sampled during the bud emergence (BBCH- Growth Stage (GS) 53), in the middle of flowering (GS65) and at pod filling stage in early summer (GS80). The plants were counted and the roots separated from the aerial parts after washing. For each micro-plot sample, dry above ground biomass (following 48 h of drying at 80°C) were determined. The first year and in 2011, when the numbers of stem weevil holes were counted (from GS53 to GS60), the height and the diameter of the plants were measured. At the pod filling stage (GS80), the number of branches and the height of the plant were measured on a 10 randomly chosen plants per micro-plot. Glucosinolates analyses were only realised at one stage in 2007, and were done using HPLC methods.

Statistical analysis

Data were explicitly tested for normality and homogeneity of variance using the Shapiro-Wilk statistic and Levene's tests, respectively. Normal-theory statistical analysis was used on continuous variables such as dry biomass by applying standard analysis of variance procedures. Proportions, such as proportion of plants attacked by rape stem weevil, were analysed after an Arcsin√ transformation to stabilise the variance among groups. Non-parametric test, such as Kruskal Wallis, has been used when variables were not normally distributed.

Results and discussion

Occurrence of pollen beetle

The number of pollen beetles per plants exhibited very large variations among the three years from zéro to 10 pollen beetles/plant. Therefore, in some statistical analysis the data of the three years has not been pooled. In Normandie, the attacks were very low the two successive years (2010 and this year 2011) and therefore, we did not considered the data collected in Normandie to analyse the effect of crop management on pollen beetles.

In this field study, the effect of cultivar was significant every year despite a very low incidence of pollen beetle in 2008 and 2010. This effect could partially be due to the early flowering nature of the cultivars and may be to their more attractive volatile profile. Cultivars which flower earlier were more heavily infested by pollen beetles/plants than cultivars which flower late, which is in accordance with the effect of turnip rape trap effect already observed either in laboratories and in semi-field studies (Cook et al., 2006 and 2007). This suggests that pollen beetles were more attracted to old plant. It is confirmed by the significant effect of bud stage on pollen beetle abundance. Effects of fertilization on pollen beetle abundance has also been observed, but only the first year (spring 2007): the plants which received nitrogen applications earlier were significantly higher infested by pollen beetles. The very low incidence of pollen beetles in 2008 and 2009 might explain the absence of effect of nitrogen regime during these years. The attractiveness by the insects on plants seems to be also related to crop biomass, height and stem diameter, which confirmed observations of Valantin-Morison et al., 2007. Moreover pollen beetles incidences were significantly different among cultivar with the same earliness of flowering, suggesting the effect of volatiles profile. When glucosinolates have been measured, the cultivars which exhibited higher isothiocyanates concentrations, exhibited also a higher number of pollen beetles/plant, which has never been observed on field scale.

Damage of stem weevils

Early flowering cultivars (line or hybrids) exhibited significant differences in phenological stage growth during stem elongation and inflorescence emergence (data not shown). Similarly as for pollen beetles, the plants of early flowering cultivars were more heavily attacked by stem weevils and exhibited a higher number of holes. This suggest that the differences in growth stage of cultivars during stem elongation might be responsible for the significant effect of cultivars in the number of holes per plants (figure 1). The effect of cultivars were also observed regarding to the proportion of distorted plants, despite it was not statistically significant. Additionally, stem weevil incidence was significantly different among cultivar with the same earliness of flowering (in spring 2007), suggesting the effect of volatiles profile. Moreover, important effects of fertilization on stem weevil damage were also recorded (figure 1A). In spring 2007, plants, which received nitrogen supplies earlier, were more attacked and conversely in spring 2010 whatever the cultivar (except Olphi), the plants, which received late applications of nitrogen, were more attacked by stem weevils. This contradictory effect has to be analysed with the synchronization or a-synchronisation of nitrogen application, rate growth of the plants and stem weevils emergence.

Despite these contradictory and disperse results, this brings new challenges for crop management, through “pest tolerance” cultivar and relevant nitrogen application, in accordance with insects-plant synchronization.

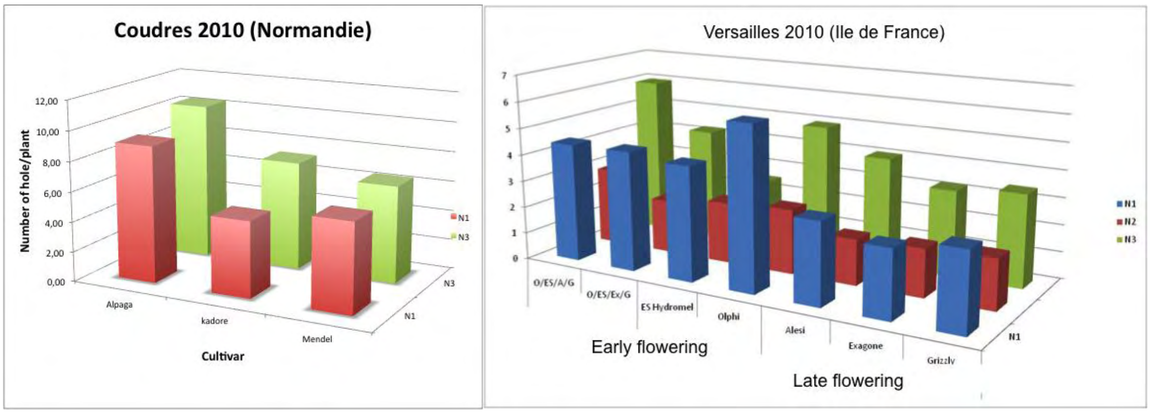

Figure 1: Effect of Nitrogen regimes and cultivars in Stem weevils holes in 2010 in Coudres (A) and on Versailles (B).

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