Dupont working for improved disease control strategies in oilseed rape

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Oilseed rape is the most important source of vegetable oil in temperate zones and the third most important oil-producing crop in the world after soya and palm. With its increasing popularity, come new agronomic challenges, as growers seek higher yields, oil contents and profits. In this context, the threat from a wide range of diseases is very significant.
The aim of this paper was to present studies on picoxystrobin efficacy on *Sclerotinia sclerotiorum,* to discuss the effect of picoxystrobin mixtures with insecticides on honeybees and to present DuPont activities for more precise, better targeted disease control in oilseed rape.

Picoxystrobin effect on *Sclerotinia sclerotiorum* control and yield effect of winter oilseed rape.

Picoxystrobin is a well known QoI fungicide used in cereals and has recently gained further interest in other crops due to its unique attributes. Picoxystrobin is currently registered for *Sclerotinia* control in oilseed rape in the United Kingdom and is under evaluation in other countries worldwide on a broad range of crops and diseases.
The field studies with picoxystrobin in oilseed rape were conducted according to the EPPO Guideline “Efficacy evaluation of fungicides. Root, stem, foliar and pod diseases of rape.” (EPPO Standard PP 1/78(3). 2002) in France, Germany, United Kigndom, Poland and Czech Republik in 2008 and 2009. The following fungicides were tested: Acanto® (picoxystrobin 250 g/L SC), Amistar® (azoxystrobin 250 g/L SC), Cantus® (boscalid 500 g/kg WG) and Proline® (prothioconazole 250 g/L EC). Fungicides were applied to oilseed rape at BBCH 60 to 65.
Results in Table 1 demonstrate an excellent activity of picoxystrobin on *Sclerotinia sclerotiorum* in oilseed rape in over 30 field trials across Europe. Although the differences between picoxystrobin and standard products were not statistically significant the trend for superior disease control and yield responses from picoxystrobin over standard fungicides was clearly visible.

In addition, an *in vitro* study on *Sclerotinia sclerotiorum* was undertaken at DuPont European R&D Center in Nambsheim, France to confirm the tendency observed in the field. The intrinsic activity of picoxystrobin has been evaluated using 5 isolates collected in 2006 and 2007 from the commercial fields or untreated plots from the field trials in the main oilseed rape producing European countries (UK, Germany, France, Poland). Also one reference isolate - 282, sensitive to the majority of OSR fungicides, originating from SRPV in Reims, France was tested to confirm assay consistency.
The following doses of commercial products containing picoxystrobin (250 g of picoxystrobin/L, SC) and azoxystrobin (250 g of azoxystrobin/L, SC) were used in this test: 0; 0.0019; 0.0156; 0.125;1 and 8 ppm. Each isolate was replicated twice. The effective dose required to inhibit growth of the fungus by 50 % (EC 50) in mg/L was determined for each isolate by using a linear regression analysis of the percentage of inhibition of growth figures against the Log10 of the concentration of the fungicide tested. The test was repeated twice. In Table 2 the EC50 values with confidence intervals for picoxystrobin and azoxystrobin obtained for each isolate are given.

Table 1. *Sclerotinia sclerotiorum* control and oilseed rape yield as influenced by Acanto® 250 SC applications.

Treatment	Disease control (%)	Yield (% untreated)	Treatment	Disease control (%)	Yield (% untreated)	Treatment	Disease control (%)	Yield (% untreated)
	n = 26	n = 18		n = 8	n = 8		n = 7	n = 7
Acanto® 150gai/ha	66.0	110.5	Acanto® 150gai/ha	56.2	107.8	Acanto® 150gai/ha	76.2	105.9
Acanto® 200gai/ha	73.4	110.6	Acanto® 200gai/ha	67.7	108.2	Acanto® 200gai/ha	80.7	108.0
Acanto® 250gai/ha	77.5	112.5	Acanto® 250gai/ha	73.8	111.9	Acanto® 250gai/ha	87.8	111.4
Amistar® 250gai/ha	70.9	110.2	Cantus® 250gai/ha	71.2	110.8	Proline® 175gai/ha	87.9	110.5
Untreated	20.7*	100.0	Untreated	16.1*	100.0	Untreated	10.9*	100.0

*The average level of disease in the untreated check (% pest severity)

Table 2. EC50 and MIC values of 6 isolates of *Sclerotinia sclerotiorum* to picoxystrobin (250 g/L SC) and azoxystrobin (250 g/L SC) in a 24-well cell culture plate on YBA medium.

Isolates	Picoxystrobin				Azoxystrobin				Azoxystrobin/ Picoxystrobin	
	EC_{50}	EC_{50} lower	EC_{50} upper	MIC	EC_{50}	EC_{50} lower	EC_{50} upper	MIC	EC_{50}	MIC
07GER01	0.0146	0.0145	0.0146	0.125	0.0985	0.0649	0.1468	1	6.75	8.00
07PL14	0.0156	0.0156	0.0156	0.125	0.1109	0.1098	0.1119	1	7.11	8.00
07FR05	0.0399	0.0321	0.0496	8	0.3536	0.3504	0.3568	8	8.85	1.00
07FR04	0.0251	0.0211	0.0300	1	0.2655	0.1184	0.7255	8	10.57	8.00
06UK01	0.0187	0.0184	0.0189	0.125	0.1811	0.1651	0.1990	8	9.71	64.00
282	0.0140	0.0139	0.0141	0.125	0.0958	0.0580	0.1582	8	6.83	64.00
Average (log-based) Anova, P<0.05	0.0198			0.35	0.1616			4	8.17	11.31

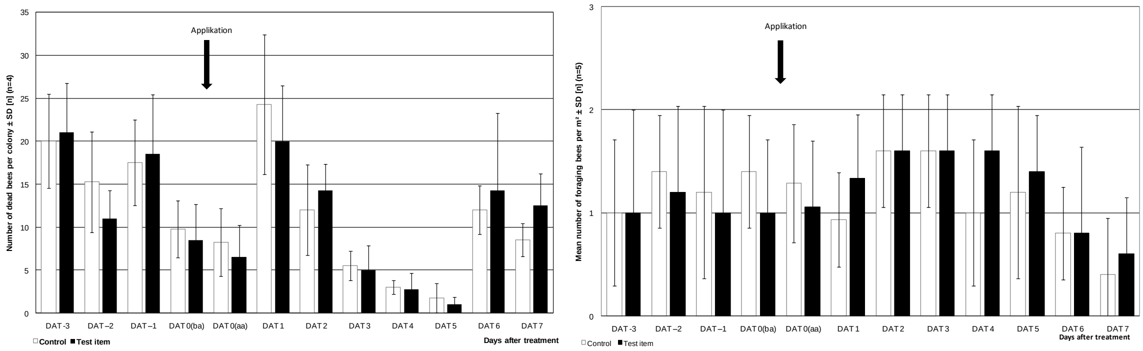
The geometrical mean EC50-value for picoxystrobin was about 8-fold lower and statistically different than that for azoxystrobin ($p<0.05$) indicating significantly higher potency of picoxystrobin for *Sclerotinia sclerotiorum* control. Each EC50 for picoxystrobin was statistically lower than that obtained for the same isolate with azoxystrobin (the confidence intervals did not overlap). Also the geometrical mean MIC-value for picoxystrobin was significantly lower (about 11-fold) and statistically different than that for azoxystrobin ($p<0.05$).
These data confirm the tendency observed in the field trials on *Sclerotinia sclerotiorum* on oilseed rape, where picoxystrobin applied at 250g a.i./ha was on average more efficient (77.5% control) than azoxystrobin applied at the same dose (70.9% control).

Effects of picoxystrobin mixtures with insecticides on honeybees under field conditions after application during bee-flight in witer oilseed rape.

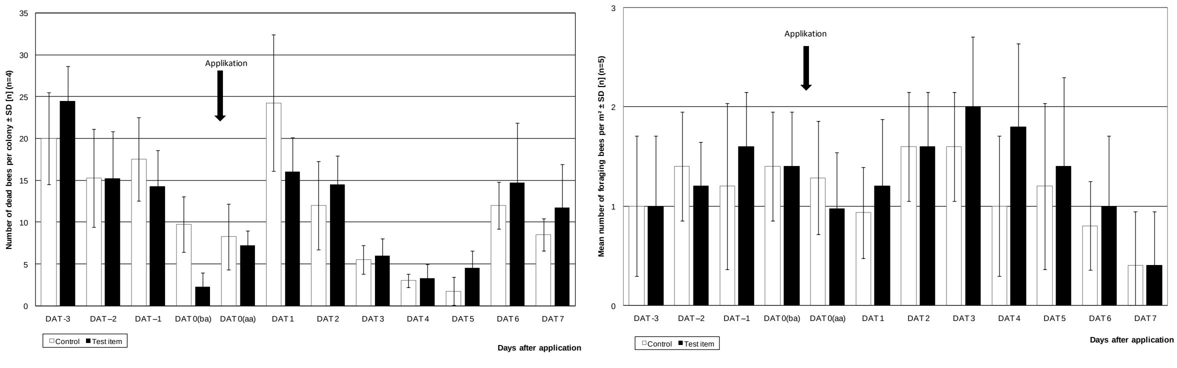
Picoxystrobin has a favourable ecotox profile including low risk for honeybees (proven in laboratory studies) and also for other beneficial arthropods. However, as fungicides may be mixed with insecticides honeybee field studies were conducted under typical commercial conditions to investigate potential effects of picoxystrobin mixtures with Biscaya® (thiacloprid) and Karate Zeon® (lambda-cyhalothrin) considering spray applications during daily bee-flight in flowering winter oilseed rape.

Honeybee (*Apis mellifera* L.) field studies were conducted according to the EPPO Guideline "Side-effects on honeybees" (EPPO Standard PP 1/170(3). 2001). The honeybees (4 colonies each) were exposed for 7 days to untreated control fields and fields treated at BBCH stage 65 during the day while the honeybees were actively foraging with the test item containing Picoxystrobin 250 g/L SC plus Biscaya® or Picoxystrobin 250 g/L SC plus Karate Zeon®. The application rates of the test items were 250 g picoxystrobin/ha plus 72 g thiacloprid/ha or 250 g picoxystrobin/ha plus 7.5 g lambda-cyhalothrin /ha. Mortality, behaviour and foraging activity of the honeybees were assessed daily 3 days before spray application and up to 7 days after treatment. Data concerning honeybee colony strength and honeybee brood development, including brood nest size and mean comb area covered with eggs, larvae, capped cells, honey and pollen were assessed once before treatment and in weekly intervals up to 4 weeks after treatment.
The application of the Picoxystrobin 250 g/L SC plus Biscaya® at a rate of 250 g picoxystrobin/ha plus 72 g thiacloprid/ha or Picoxystrobin 250 g/L SC plus Karate Zeon® at a rate of 250 g picoxystrobin/ha plus 7.5 g lambda-cyhalothrin/ha under field conditions to flowering winter oilseed rape *Brassica napus* L. during daily bee flight caused no effects on honeybee mortality, foraging activity, behaviour, colony weight, colony strength and brood development.
Exemplarily results on mortality (mean number of dead honeybees per colony, left figures) and on foraging activity (mean number of foraging honeybees per m² winter oilseed rate, right figures) for sites tested in North-Eastern Germany are given in the following figures.

Effects of Picoxystrobin 250 g/L SC plus Karate Zeon® at 250 g picoxystrobin/ha plus 7.5 g lambda-cyhalothrin/haon honeybee mortality (left) and foraging activity (right)
(White bars = control. Black bars = picoxystrobin plus lambda-cyhalothrin)



Picoxystrobin 250 g/L SC plus Biscaya® at 250 g picoxystrobin/ha plus 72 g thiacloprid/ha on honeybee mortality (left) and foraging activity (right)
(White bars = control. Black bars = picoxystrobin plus thiacloprid)



The System for Forecasting Disease Epidemics (SPEC) – for more cost-effective, better targeted, and more environmentally acceptable pest control.

DuPont, in co-operation with independent experts, strongly support research aimed at the improved understanding of the epidemiology, forecasting and risk assessment from oilseed rape diseases. The System for Forecasting Disease Epidemics (SPEC), developed in Poland together with Institute of Plant Genetics, Polish Academy of Sciences (PAN), in Poznan, with the aim of improving stem canker management through optimization of fungicide treatments, is an example of such co-operation. This disease causes important economic losses in Poland, on average 10-15% per year, up to 60% in some regions and seasons. SPEC is the largest monitoring system of its kind in Europe and the third largest in the world. Starting from autumn 2004 a network of 10 volumetric spore samplers located in different geographical locations has been constantly operating. At present the monitoring of airborne ascospores concerns mainly *Leptosphaeria maculans* and *L. biglobosa,* responsible for stem canker epidemics. The release of fungal spores is done under natural conditions, using oilseed rape stubble from the previous season, separately collected in each region. The sampling is monitored using a Burkard or Lanzoni 7-day recording spore traps. The samplers suck 10 L of air min^{-1}, and fungal spores (as well as pollen grains) are deposited on a vaseline-coated cellophane tape mounted on a rotating drum, placed in each sampler. Once a week the tape is taken off the drum and cut longitudinally to two pieces, as well as across to seven fragments, corresponding to seven days of the week. The spores of *L. maculans* and *L. biglobosa* are counted on each half of the tape and recorded as daily ascospore concentration per m^3 of air. The other half piece of the tape is studied using PCR-based molecular methods. The results are presented in the form of communications for the farmers and they are immediately passed to the registered end-users, using the website and SMS text messages. Two different websites are dedicated to the users of the SPEC system: the educational website (www.spec.edu.pl) as well as the commercial website (www.dupont.pl). These websites offer complex information service and they are visited by more than 10 thousand users per year. At present, the SPEC system is helping to undertake decisions in oilseed rape protection against stem canker, but studies of the ascospore concentration of *Pyrenopeziza brassicae* responsible for light leaf spot, were also done and there are close perspectives of monitoring *Sclerotinia sclerotiorum, Botrytis cinerea* and *Alternaria* spp.