



# PURE

# Pesticide Use-and-risk Reduction in European farming systems with Integrated Pest Management

Grant agreement number: FP7-265865

Collaborative Project SEVENTH FRAMEWORK PROGRAMME

# D3.2

# Updated list of tested IPM solutions which further improve the sustainability of maize based systems

**Due date of deliverable:** M 39

Actual submission date: M 39

**Start date of the project**: March 1<sup>st</sup>, 2011

Duration: 48 months

Workpackage concerned: WP3

Concerned workpackage leader: Dr. Maurizio Sattin

**Organisation name of lead contractor: UDCAS** 

Project co-funded by the European Commission within the Seventh Framework Programme (2007 - 2013)				
Dissemination Level				
PU Public	PU			
PP Restricted to other programme participants (including the Commission Services)				
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<b>CO</b> Confidential, only for members of the consortium (including the Commission Services)				



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# Glossary

# Definitions used in this study

European Network for Durable Exploitation of crop protection strategies
Pesticide Use-and-risk Reduction in European farming systems with Integrated
Pest Management
Work Package 3
Maize-Based Cropping System
Integrated Pest Management
European Corn Borer, Ostrinia nubilalis
Mediterranean Corn Borer, Sesamia nonagioides
Western Corn Rootworm, Diabrotica virgifera virgifera LeConte



# 1. General introduction

This is the second deliverable of the WP3 providing an updated list of IPM solutions by reporting 1) the 2012 and 2013 results from advanced and innovative IPM strategies for maize tested on-station in France and of the 2011-2013 from the on-station trial in The Netherlands studying the effect of various tillage regimes with chemical or mechanical weed control in maize (Task 3.3a), 2) the updated results from IPM tools tested in on-farm experiments during the first 2 years of PURE and the new IPM tools tested in 2013 together with their costbenefit analysis (Task 3.3b), 4) the ex-post assessment of the new IPM tools tested on-farm in 2013 (Task 3.4), all in comparison to the conventional approach. Results of the two long-term on-station experiments in Italy and Hungary (Task 3.2) did not involve all crops of the rotations tested each year and these results will be reported in the last deliverable after analysing their effect during 2014 maize that is present in all systems tested.

#### Geographical areas covered and partners involved

Three regions from European maize cultivation areas (in Italy/France, Germany/The Netherlands, and Hungary/Slovenia) were selected that represent the range of various geographic, climatic and cultivation types as follows:

- Southern conditions (Po Valley in Northern Italy and southern France); average characteristics are medium-heavy soils, relatively mild winters and warm-hot summers, water availability (medium-high rainfall or irrigation), high yield potential. Weeds, soil born insects, ECB and WCR (and MCB in southern France). Partners involved: CNR Italy, ACTA/Arvalis France, IAS France.
- Eastern conditions (eastern Hungary and eastern Slovenia), with continental climate, medium-low rainfall during maize growing season and generally no irrigation available, medium yield potential. Key-pests: ECB and WCR for Slovenia. Partners involved: UDCAS - Hungary and KIS - Slovenia.
- Central conditions (southern Germany and The Netherlands), mild-warm summers and medium-high rainfall, medium-low yield potential (in terms of grain). Key pests: weeds (and ECB in Germany). Partners involved: JKI - Germany and Stichting DLO – The Netherlands.



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# **2.** Task **3.3a**: On-station experimentation in France (comparison of cropping systems with different crop protection strategies)

ARVALIS – INSTITUT DU VEGETAL (Agrosite de PAU – MONTARDON)

## Introduction

In this project, several European countries implemented experimental platforms of cropping systems based on maize: Italy, Hungary and France. Italy and Hungary are studying a 3-year rotation, with an experiment over 4 years (2011-2014) whereas France is working on 2-year rotations during 3 years (2012-2014).

In France, the experiment is located in Sendets (64) and managed by the Pau Montardon Station of Arvalis Institut du Végétal.

In this experiment, we are interested in evaluating the pest management and productivity of the different cropping systems. The 2012 and 2013 experimental results are presented in this report.

# Materials & methods

#### **Experimental layout**

The « system » experiment follows the project protocol concerning the on-station experiments which can be found in the previous deliverable (D3.1). The experiment includes 8 plots of  $300 \text{ m}^2$  replicated 3 times. Only one factor is tested: cropping systems with 5 different treatments. A cropping system is characterized by all the decisions taken in relation to a field, including established crops, their successions and crop management (i.e. fertiliser, pesticides, mechanical weeding).

Five cropping systems are studied:

- Treatment 1 (T1): maize monoculture , conventional system
- ✤ Treatment 2 (T2): maize monoculture, advanced system
- Treatments 3 and 6 (T3, T6) : winter wheat-maize rotation, advanced system
- Treatments 4 and 7 (T4, T7): soybean-maize rotation, advanced system
- Treatments 5 and 8 (T5, T8): soybean-maize rotation, innovative system



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Three rotations are studied:

- $\rightarrow$  maize monoculture
- $\rightarrow$  maize-soybean rotation
- $\rightarrow$  maize-winter wheat rotation

Maize rotation with another crop such as winter wheat or soybean could have a beneficial effect on the maize present every second year, because cropping interventions are different and disrupt the lifecycle of pests, which causes a decrease in weed and pest populations. The pesticides applied also differ depending on crops present each year, and switching active ingredients could limit the development of weed or pest resistance.

Three types of pest management are studied:

- $\rightarrow$  Conventional
- $\rightarrow$  Advanced (IPM1)
- $\rightarrow$  Innovative (IPM2)

The conventional system is the most commonly used cropping system in the area. It includes a deep tillage before sowing, seed treatments, herbicides (pre- and post-emergence) and insecticides (if necessary) with an integrated programme. In this experiment, the conventional system is continuous maize.

The advanced system is based on the conventional system, but with less dependence on pesticides and integrated with agronomic methods like hoeing. The advanced system is tested in continuous maize (not PURE), in maize-soybean rotation (PURE system) and in maize-winter wheat rotation (not PURE).

The innovative system reduces the use of herbicides and insecticides (by reducing the number of applications and the quantity of applied products) more than in the advanced system. There is no post-emergence herbicide and no seed dressing. This innovative system is only tested in the maize-soybean rotation.

The experimental design allows that each crop in a rotation is present every year (i.e. maize present all years) and this allows the year effect to be ruled out (i.e. climatic replication), an independent factor that could affect experimental results. Figure 1 shows the cropping succession in each system during these 3 years: 2012 to 2014, with the 5 PURE systems and the other 3.



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	T1	T2	Т3	<b>T</b> 4	T5	<b>T6</b>	<b>T</b> 7	<b>T</b> 8
2012	Maize	Maize	Maize	Maize	Maize	Cereals	Soybean	Soybean
	conv	IPM1	IPM1	IPM1	IPM2	IPM1	IPM1	IPM2
2013	Maize	Maize	Wheat	Soybean	Soybean	Maize	Maize	Maize
	conv	IPM1	IPM1	IPM1	IPM2	IPM1	IPM1	IPM2
2014	Maize	Maize	Maize	Maize	Maize	Wheat	Soybean	Soybean
	conv	IPM1	IPM1	IPM1	IPM2	IPM1	IPM1	IPM2
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Fig. 1 Diagram of the cropping succession during the experimentation.

Complementary systems, not required by the PURE project, have been added. This resolves the PURE project problem, by reducing the year effect and testing a continuous maize rotation, currently used in this area, as an advanced system. The PURE project treatments are encircled in orange in Figure 1.

The systems included in the PURE project are:

- treatment T1, conventional system, continuous maize rotation
- treatments T4 and T7, advanced system, maize-soybean rotation
- treatments T5 and T8, innovative system, maize-soybean rotation

The systems added to the PURE experimentation are:

- treatment T2, advanced system, continuous maize rotation
- treatments T3 and T6, advanced system, maize-winter wheat rotation

#### **Observation protocols**

During this experiment, several observations have been made on cropping systems and their impacts on pests. Crop development has also been monitored.

Observation protocols have therefore been created. When no date information is given, this means protocols were the same in 2012 and 2013.



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Aerial pests:

ECB (Ostrinia nubilalis)

Monitoring by a pheromone trap, checked twice a week. Traps were set on 25<sup>th</sup> April, and ran until 26<sup>th</sup> September (*traps followed from 11/05 to 24/09 in 2012*).

MCB (Sesamia nonagrioides)

Monitoring by a pheromone trap, checked twice a week. Traps were set on 25<sup>th</sup> April, and ran until 26<sup>th</sup> September (*traps followed from 11/05 to 24/09 in 2012*).

Helicoverpa armigera

Monitoring by a pheromone trap, checked twice a week. Traps were set on 25<sup>th</sup> April, and ran until 26<sup>th</sup> September (*traps followed from 11/05 to 24/09 in 2012*). *The two following aerial pests were only monitored by pheromone traps in 2013:* 

➤ Agrotis segetum

Monitoring by two pheromone traps in 2013. Traps were set on 25<sup>th</sup> April, and checked twice a week until 26<sup>th</sup> September.

➤ Agrotis ipsilon

Monitoring by two pheromone traps in 2013. Traps were set on 25<sup>th</sup> April, and checked twice a week until 26<sup>th</sup> September.

#### Soil pests:

➢ <u>Wireworms</u>

Monitoring adults by:

- Barber traps: 2 traps per plot. From 26<sup>th</sup> April (when maize had begun to emerge) to 23<sup>rd</sup> October, checking once a week (*traps followed from 14/05 to 04/07 in 2012, 8 weeks*).
- Pheromone traps: one for *A. sordidus*, the other one for *A. lineatus*. Checking twice a week, from 26<sup>th</sup> April to 23<sup>rd</sup> October 2013 (harvest). *No pheromone traps in 2012*.
   Monitoring larvae by:
- Bait traps (Kirfman, adapted by Chabert & Blot): Set on 17<sup>th</sup> and 18<sup>th</sup> July, and removed on 31<sup>st</sup> July. Funnels added on 5<sup>th</sup> August, checking on 19<sup>th</sup> August. *In 2012: 3 traps per plot, set on 29<sup>th</sup> May, 31<sup>st</sup> May or 1<sup>st</sup> June to 13<sup>th</sup> June, 14<sup>th</sup> June or 15<sup>th</sup> June and checking 2 weeks later.*

In 2012: 4 lines of 10m length per plot, checked three times during maize development.

Garden Symphilan (Scutigerella immaculata):

Bait potatoes traps, 12 traps in the experiment, from 11<sup>th</sup> to 14<sup>th</sup> May, checking after 3 days.



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<u>Wireworms, Garden Symphilan, slugs (and Black cutworm in 2012): Damage assessment</u> In 2013: Crop plant inspections: two 20-meter rows per plot; checked 6 times between mid-May and mid-July (16/05, 03/06, 12/06, 21/06, 02/07 and 12/07)

In 2012: 4\* 10-meter rows per plot, checked 3 times (at 2-4 leaf stage and 6-7 leaf stage).

Weeds:

Grasses and broadleaves

<u>Weed density assessment</u> (total density and specific densities): 4 observations of 6 areas (40cm X 78cm) per plot: end of June, beginning of July, end of August and at harvest. *In 2012: 4 observations of 2 areas (40cm X 76cm) before harvest.* 

Weed biomass assessment: Removal of weeds before harvest (done in 2012 and 2013).

Yields:

- Quantitative measurements
  - Maize biomass assessment on 11.4m<sup>2</sup> (3 rows x 5m) before harvest. Measure of moisture content and dry matter
  - Grain yield assessment on 159.6m<sup>2</sup>, with 14% moisture content at harvest
  - Soybean total yield assessment on plot area

In 2013 only:

- Soybean total biomass assessment on plot area
- Wheat total yield assessment on plot area, measurement of moisture content, thousand kernel weight, number of kernels per cob, specific weight.
- Qualitative measurements

Mycotoxin analysis on 2-3 kg grain sample at harvest.

#### **Crop management practices**

#### Crop management in Maize

According to the protocol and the observations on the experimental field, 3 crop management systems have been elaborated for maize, independently of cropping rotations:

- conventional system (representative of what is commonly done in the area),
- advanced (=IPM1) system (a more environmentally-friendly system, with hoeing and localized spraying)
- innovative (IPM2) system (more environmentally-friendly system than the advanced one: there is no post-emergence herbicide, no seed dressing and localized spraying)



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#### Table 1 Crop management strategies in maize

**Legend:** Differences between crop management practices are shown **in red** (conventional, IPM1 and IPM2). the 2012 operation dates are **in purple** 

**Note:** Doses have been calculated for one hectare depending on the kind of spraying (overall, row or band spraying)

Maize	Conventional	IPM1 advanced	IPM2 innovative
Ploughing Disking	Tillage (25cm) 09/03/13 (13/03/12) cultivator (2 runs on a row) : 14/04/13 (2/05/12)	Tillage (25cm) 09/03/13 (13/03/12) cultivator (2 runs on a row) : 14/04/13 (2/05/12)	Tillage (25cm) 09/03/13 (13/03/12) cultivator (2 runs on a row) : 14/04/13 (2/05/12)
starter fertilizer (NH <sub>4</sub> ) <sub>3</sub> PO <sub>4</sub>	150 kg/ha (P : 40 U/ha N : 20U) 15/04/13 ( <i>03/05/12</i> )	150 kg/ha (P : 40 U/ha N : 20U) 15/04/13 (03/05/12)	150 kg/ha (P : 40 U/ha N : 20U) 15/04/13 ( <i>03/05/12</i> )
Sowing	82000 gr/ha. DKC5783 (in 2012 : Antalya) 15/04/13 (03/05/12)	82000 gr/ha. DKC5783 ( <i>in</i> 2012 : Antalya) 15/04/13 (03/05/12)	82000 gr/ha. DKC5783 (in 2012 : Antalya) 15/04/13 (03/05/12)
Seed dressing Crows	Royal Flo (thirame)	Royal Flo (thirame)	
Seed dressing diseases	Influx XL (Fludioxonil, mefenoxam)	Influx XL (Fludioxonil, mefenoxam)	Influx XL (Fludioxonil, mefenoxam)
Seed dressing Wireworms	Cruiser 350 (thiametoxam)	Cruiser 350 (thiametoxam)	
Slug pellets	Mesurol Pro (mecaptodimethur) 19/04/13 (18/05/12)	Mesurol Pro (mecaptodimethur) 19/04/13 (18/05/12)	Mesurol Pro (mecaptodimethur) 19/04/13 (18/05/12)
Weed control pre- emergence	Trophée (Acetochlore 400g/L) 4L/ha + Lagon (aclonifen 500g/L + Isoxaflutole 75g/L) 0,6L/ha Sprayer with low drift nozzles, between seeding and emergence, 17/04/2013 (04/05/12)	Trophée (Acetochlore 400g/L) 4L/ha + Lagon (aclonifen 500g/L + Isoxaflutole 75g/L) 0,6L/ha Sprayer with low drift nozzles, between seeding and emergence, 17/04/2013 (04/05/12)	Trophée (Acetochlore 400g/L) 4L/ha + Lagon (aclonifen 500g/L + Isoxaflutole 75g/L) 0,2L/ha Local application in seed slot (1/3 of the area) Sprayer with low drift nozzles, between seeding and emergence, 17/04/2013 (04/05/12)
Weed control post-emergence	Callisto ( mesotrione 500g/L) 0,5L/ha + Banvel 4S (Dicamba=salt of dimethylamine 480g/L) 0,4 L/ha. Sprayer with low drift nozzles. 25/05/13 at the 5 leaf stage (05/06/12)	- Banvel 4S (Dicamba=salt of dimethylamine 480g/L) 0,26 L/ha (only done in 2013) Sprayer with low drift nozzles. Band application (2/3 of the area) 25/05/13 at the 5 leaf stage. ( <i>In 2012 : no post emergence</i> <i>herbicide</i> ) Hoeing *2 : 06/06/13 (5 leaf stage) and 25/06/13 (9 leaf stage) (05/06/12 and 22/06/12)	- - - - Hoeing *3 6/06/13 (5 leaf stage), 14/06/13 (6 leaf stage) and 25/06/13 (9 leaf stage) (31/05/12, 15/06/12 and 22/06/12)
N Fertilization urea	200 U/ha, row application 2/07/13 (08/06/12)	200 U/ha, row application 2/07/13 (08/06/12)	200 U/ha, row application 2/07/13 (08/06/12)
Harvest	23.10.13 (29/10/12)	23.10.13 (29/10/12)	23.10.13 (29/10/12)



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Table 1 shows that the conventional system requires lots of chemical treatments: seed dressing and pre- and post-emergence herbicides; whereas the innovative system does not use seed dressing or post-emergence herbicides, and needs less pre-emergence herbicide. Indeed, weed control is by hoeing. The advanced system is a mix of the 2 other systems. This system has seed dressing and the same quantity of pre-emergence herbicide as the conventional system but no post-emergence herbicide application. Hoeing is used also in this system, but less than in the innovative system (three times).

#### Crop management in Soybean

Soybean	IPM1 advanced	IPM2 innovative
Ploughing Disking	Tillage (25cm) 09/03/13 (13/03/12) cultivator (2 runs on a row) : 14/04/13 (2/05/12)	Tillage (25cm) 09/03/13 (13/03/12) cultivator (2 runs on a row) : 14/04/13 (2/05/12)
<b>K</b> <sub>2</sub> <b>O</b>	120 U/ha 09/03/13 ( <i>13/03/12</i> )	120 U/ha 09/03/13 ( <i>13/03/12</i> )
Sowing	ECUDOR 376 000 gr/ha 15/04/13 (03/05/12)	ECUDOR 376 000 gr/ha 15/04/13 (03/05/12)
Slug pellets	Mesurol Pro (mecaptodimethur) 19/04/13 (18/05/12)	Mesurol Pro (mecaptodimethur) 19/04/13 (18/05/12)
Weed control pre- emergence	Mercantor Gold 1,4L/ha Sprayer with low drift nozzles. 17/04/13 (10/05/12)	Mercantor Gold (S-metolachlor 960g/L), <u>0.47L/ha.</u> 17/04/13 (10/05/12) Row spraying with low drift nozzles (1/3 of the area)
Weed control post- emergence	Pulsar 40. <u>0.66L/ha</u> on 14/05/13. Band application with low drift nozzles (2/3 of the area) Hoeing *3 : 6/06/13, 25/06/13, 10/07/13. ( <i>in 2012 :</i> * Pulsar 40, 0,6L/ha on 05/06/12 and Pulsar 40, 0,6L/ha on 22/06/12. Sprayer with low drift nozzles, band application (2/3 of the area) * no hoeings)	- Hoeing *4 : 6/06/13, 14/06/13, 25/06/13, 10/07/13. (31/05/12, 15/06/12, 22/06/12 and 12/07/12)
Harvest	10/10/13 (17/10/12)	10/10/13 (17/10/12)

 Table 2 Crop management strategies in soybean.

Soybean is not tested in the conventional system; it is only a part of maize rotation in the advanced and innovative systems. Alternatives to chemical methods are favoured, particularly in the innovative system (IPM2).



In the IPM1 advanced strategy, there were some differences between years in terms of weed control. During the 2012 season, there were 2 sprayings of Pulsar (one on 05/06/12, the second on 22/06/12) with 0.6L/ha each time (whereas there is only one application with 1L/ha in 2013). Moreover, there hoeing in 2012. and 3 in 2013. was no Concerning the innovative system IPM2, the only difference is that the first hoeing was one week earlier in 2012.

#### Crop management in wheat

Winter Wheat	IPM1 advanced
Ploughing	cultivator (2 runs on a row) : 29/10/13
Disking	(In 2012 : tillage 25cm on 13/03/12 ; cultivator *2 :
	15/03/12 and 3/05/12)
Sowing	Solveig, 323 000 seeds/ha
	30/10/13 ( <i>3/05/12</i> )
Slug pellets	Mesurol Pro (mecaptodimethur) 3kg/ha, in furrow,
	after emergence 30/10/13 (18/05/12)
Insecticides, seed	Seed dressing : Gaucho 350 (Imidaclopride 350g/L),
dressing	0,200L/qx)
Fertilisation	70 U on 1/02/13
	97 U on 7/03/13
	23 U on 5/05/13
	(In 2012 : No N fertilisation)
Pre-emergence	Archipel 0.25kg/ha, Rafale 0.5L/ha on 20/02/13
weed control	(In 2012 : no pre-emergence weed control)
Post-emergence	In 2013 : no post-emergence weed control
weed control	(In 2012 : Allié (Metsulfuron methyl 20%)
	0,030kg/ha, Sprayer with low drift nozzles, 5/06/12)
Fungicides	Opus 0.5L/ha on 10/04/13 and 21/05/13 and Abacus
	0.8L/ha on 5/05/13
	(In 2012 : no fungicide)
Harvest	30/07/13
	(No harvest in 2012)
Secondary crop	Oat, sown on 23/08/2013

 Table 3 Crop management in wheat.



# Results

#### **Aerial Pests**

#### 2012 Results

To observe aerial pests, pheromone traps were checked twice a week during the entire maize development (11<sup>th</sup> May to 24<sup>th</sup> September 2012). The monitored species are ECB (*Ostrinia nubilalis*), MCB (*Sesamia nonagrioides*) and *Helicoverpa armigera*.

Few MCB were observed in the experimental field, only in May, when the overwintering larvae began to fly. No WCR (*Diabrotica virgifera*) were observed.

A peak was observed for *Helicoverpa armigera* at the beginning of July and a second one at the beginning of September. Few individuals were observed at these two dates. Two generations were revealed. Population decreased in July and there was a new generation at the end of August. There was then a stable number of individuals until harvest.

The ECB have been observed for the first time at the end of June, which corresponds to the 1<sup>st</sup> generation. In conclusion, a small number of insects have been observed on the experimental field. Results are presented in figure 2.



Fig. 2 Evolution of aerial pest numbers in traps.

A very low level of overall damage was observed. Only some plants had *Helicoverpa armigera* attack symptoms. However, some crow damage was observed in the field at the end of May. The IPM2 maize system (T5) had the highest level of damage (average of 15% for

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the three blocks) since no seed dressing was used, whereas Royal Flo seed dressing had been applied in the other plots. The analysis of variance showed that these results are relatively significant (Table 4).

Table 4 Percentages of plants with crow damage by strategy and ANOVA results.						
	Conv. Maize T1	Advanced Maize T2	Advanced Maize T3	Advanced Maize T4	Innovative Maize T5	Average per block
Block 1	O%	O%	O%	O%	O%	0%
Block 2	O%	O%	O%	O%	29%	6%
Block 3	1%	O%	1%	O%	18%	4%
Mean	O%	O%	O%	O%	15%	

#### DETAILED REPORT

Groups	Number of Samples	Sum	Average value	Variance
T1	3.00	1.00	0.33	0.33
T2	3.00	0.00	0.00	0.00
T3	3.00	1.00	0.33	0.33
T4	3.00	0.00	0.00	0.00
T5	3.00	47.00	15.67	214.33

#### ANALYSIS OF VARIANCE

Statistical differences	Sum of squares	Degrees of Freedom	Average of squares	F	Probability	Critical value for F
Between groups	576.93	4.00	144.23	3.35	0.05	3.48
Within groups	430.00	10.00	43.00			
Total	1006.93	14.00				

#### 2013 Results

Aerial pest monitoring took place from 25th April to 26th September using pheromone traps. It focused on ECB, MCB, *Helicoverpa armigera*, *Agrotis ipsilon* and *Agrotis segetum*. Checks were made twice a week and pheromones in the traps were replaced every 3 or 6 weeks depending on species.

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Fig. 3 Number of aerial pests caught in pheromone traps.

Overall, few individuals were found on plots (Fig. 3).

Agrotis ipsilon: catching time was between April and July, when maize was between 6 and 12 leaf stages. A total of 68 individuals were found, most during the laying period (maximum of 13 individuals per check). Nevertheless, no damage caused by this pest was seen on plots.

Agrotis segetum: few individuals were caught at the beginning of May and at the end of June and July, with a maximum of 4 individuals per check. No damage due to this pest was seen.

MCB: only 4 individuals were found, at the end of May and at the end of August. MCB population was low.

Helicoverpa armigera: a total of only 8 individuals were found during the 5 months of monitoring, at the end of June and July.

ECB: few individuals were caught at the end of June, July and August, whereas a high population was seen throughout the experimental period, above all in July and September. Some damage from ECB larvae was recorded in plots in September.



### The research leading to these results has received funding from the European Union Seventh Framework Programme (FP7/ 2007-2013) under the grant agreement n°265865 -PURE

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#### Soil pests

The most common soil pest in maize in the area is wireworm, and particularly *Agriotes sordidus*. To observe them, bait (Kirfman) and Barber traps were used.

#### 2012 Results

#### Bait traps

According to the protocol, bait traps stayed in plots for 15 days, from 29th May to 1st June, when maize was at the 5 leaf stage. 3 bait traps were set per plot. Figure 4 shows the average number of larvae per trap, depending on the treatment.



Fig. 4 Larvae distribution by treatment.

System	Rotation 2012/2013/2014	System
<b>T1</b>	Continuous maize	Conventional
T2	Continuous maize	Advanced (IPM1)
Т3	Maize/Winter Wheat/Maize	Advanced (IPM1)
<b>T4</b>	Maize/Soybean/Maize	Advanced (IPM1)
Т5	Maize/Soybean/Maize	Innovative
		(IPM2)
<b>T6</b>	Cereal/Maize/Winter Wheat	Advanced (IPM1)
<b>T7</b>	Soybean/Maize/Soybean	Advanced
		(IPM1))
<b>T8</b>	Soybean/Maize/Soybean	Innovative
		(IPM2)

In grey, the PURE treatments (not in PURE, T2, T3, T6)

The total number of larvae was very low compared to the rates currently observed in Southwestern France. Wireworms were present in continuous maize in the conventional system with 0.83 larvae per trap (average threshold is 0.5 larvae per trap) and in advanced (IPM1) maize systems (0.78, 0.56 and 0.33 mean number of larvae per trap). The highest number was for the innovative (IPM2) system (0.89 mean larvae per trap) that was expected as these plots

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were not treated with the Cruiser seed dressing. However, according to the statistical analysis, values are not significant so we cannot deduce a real and reliable interpretation of these results.

On the other hand, wireworm larvae were less numerous in soybean and winter wheat plots. It might be that these crops could reduce the wireworm populations for the following maize. This hypothesis was tested in 2013. Figure 5 shows the spatial distribution of wireworms for the 72 bait traps.



Fig. 5 Spatial distribution of wireworm larvae.

#### Barber traps

Barber traps were set to observe adult wireworms. They were set on 24<sup>th</sup> May 2012 (2 per plot) and checked every week for 8 weeks. They were removed on 4<sup>th</sup> July. Figure 6 shows the number of adult wireworms caught during the entire monitoring period by Barber traps.





Fig. 6 Adult wireworm distribution.

Figure 6 shows that a very low number of wireworms were caught in the experimental field (only 7 adults during the entire monitoring period). Adults were only caught in these 4 systems (listed in decreasing order): T8 **soybean**-maize rotation IPM2 system, T5 **maize**-soybean rotation IPM2 system, T7 **soybean**-maize rotation IPM1 system and T6 **wheat**-maize rotation IPM2 system. These results show that the IPM2 system is the most susceptible to wireworms, because they are more numerous. However, these results are not significant because the values are low and so the margins are too small.

Maize stand assessments at the 2-4 and 6-7 leaf stage showed a low rate of wireworm attacks. Only the plot with innovative maize system had 0.4% of attack. So overall, there is no damage in the experimental field caused by wireworms in 2012. This result is very unusual for the area, where wireworms are high level risk pests. However, the innovative (IPM2) system seems to be more susceptible to wireworms and the cause seems to be the absence of Cruiser 350 application.

#### Bait potatoes traps

Bait potatoes traps were set on 11<sup>th</sup> May and removed on 14<sup>th</sup> May. No Garden Symphilan (*Scutigerella immaculata*) was observed. So their presence has not been proved. No damage was observed from other pests (Black cutworm or slugs).

#### 2013 Results

Pheromone traps



Pheromone traps (focused on the two species *Agriotes sordidus* and *Agriotes lineatus*) were set on 25th April and removed on 26th September. Traps were checked twice a week, and a total of 600 individuals were found.

The two species had similar curves, except that *A. sordidus* always had more individuals per check: a total of 450 *A. sordidus* individuals was found, compared to 140 *A. lineatus*, which was expected since *A. sordidus* is the most common wireworm species in south-eastern France (Figure 7). A maximum of 100 wireworms in one check was found in May, when maize was emerging and another peak was observed in June, with 40 individuals. After June, few individuals were caught, never more than 20 individuals per check, and most were *A. sordidus*.

With a high attack level, there should be more than 300 individuals per trap at each check, whereas the peak observed in 2013 corresponded to just 100 individuals, thus with a low risk level in the experimental field.



Fig. 7 Number of wireworms caught in pheromone traps from 06/052013 to 26/09/13.

#### Kirfman's traps:

Kirfman's traps were first set in March 2013 before sowing with 3 traps per plot, and then on 17th and 18th July with 6 traps per plot. In both cases, they were removed after 14 days and larvae were then counted. Figure 8 shows the total number of larvae in each cropping system, for the first catching period in March 2013.

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Fig. 8 Distribution of wireworm larvae in March 2013.

Very few individuals were caught with these traps. No larva was found in the plot treated with the conventional system and only 2 were caught in continuous maize rotation IPM1 system and in soybean-**maize** rotation IPM1 system. Only one larva was found in the maize-**soybean** rotation IPM1 system, in wheat-**maize** rotation IPM1 system and in soybean-**maize** rotation IPM2 system (crop indicated in bold is the one present on the plots in 2013). These values are very low with an average of 0.15 larvae per trap.

Nevertheless, 4 larvae were caught in maize-**soybean** rotation IPM2 system. This tends to show a high level of wireworms in the plot with maize IPM2 as preceding crop. This tendency cannot be confirmed because of the too low number of individuals collected.

All larvae collected were *A. sordidus*, except in soybean-**maize** rotation IPM1 system where *A. lineatus* was also identified (in the  $2^{nd}$  block).

According to the analysis of variance, results are not significantly different, so no reliable interpretations can be made of our data.

Another series of Kirfman's traps was set on 17th and 18th July, with 6 traps per plot. The 144 traps were removed after 14 days, on 31st July. In wheat plots, traps were removed on 29th July because harvesting was done on 30th July. Traps were checked using funnels, set on 5th August and analysed after 14 days. Only a few larvae were collected (24) and Figure 9 gives distribution of larvae for each cropping system. A new analysis method was used: we placed trap contents on funnels. After 15 days, the soil dried and larvae went to the bottom of the funnels, falling into vials that were collected. 14 larvae were collected in these vials, thus 54% of the traps larvae, and the other 46% were still in the soil, and probably dead. Because of their physical aspect, the cause of death seems to be a fungus.

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Fig. 9 Distribution of wireworm larvae for each system in July 2013.

Number of larvae found in continuous maize rotation and in maize IPM1 systems was similar. We can conclude that there is apparently no beneficial effect of rotation. Nevertheless, there was a slightly larger number of larvae in wheat, and in maize which had wheat as preceding crop. A wheat-maize rotation system appears not to be an efficient alternative to control wireworms, compared to continuous maize rotation. We expected a high number of larvae in IPM2 maize systems, because of the absence of insecticide applications. However, this was not the case perhaps because of too late trap periods, as wireworms could have begun their pupations.

IPM2 soybean seemed to have a higher number of larvae compared to other treatments, whereas IPM1 soybean had a low larvae number. These results were not expected, because wireworm larvae seem to be preferentially found in maize, compared to soybean areas. It is possible that a bias was created by the difference in vegetation cover. Indeed, soybean may cover a greater surface than maize, which could increase soil moisture level (higher than in maize soils). Wet conditions are much more attractive to adult wireworms, so many more lay eggs, which leads to a higher number of larvae.

The preceding crop in T5 (maize-soybean rotation IPM2 system) was maize IPM2, which did not receive any insecticide against wireworms. This cropping system was the one with the highest number of wireworm larvae in 2012, so larvae found in 2013 T5 might be the result of the previous year's high larvae rate. Another explanation could be an unequal larvae distribution, caused by « population centres » (small areas where pest density is high). This hypothesis is confirmed by figure 10, which shows the plot heterogeneity regardless of treatments applied. The figure shows that there are areas where wireworms are concentrated: on the right side for blocks 2 and 3, and on the left side of the field for block 1.

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Fig. 10 Distribution of wireworm larvae.

For maize-**soybean** rotation IPM2 system (T5), 9 larvae were found, including 7 dead larvae which seemed to have been killed by fungus (Figure 11). There was only a high proportion of dead larvae in the T5 and only a few dead larvae were found in other treatments (1 in T2 and T3, 2 for T6). We can suppose that in maize-**soybean** rotation IPM2 system, there is a regulation of larvae population, which is different from Cruiser seed dressing. It might be the soybean crop or the absence of Cruiser application that had a beneficial effect on the larvae pathogen development. Another favourable factor could be the increase of soil aeration caused by several hoeings in advanced and innovative systems, which furthers the development of this pathogen.



Fig. 11 Proportion of dead and alive larvae found in each system.

According to the literature, the wireworm population could also be regulated by the soybean crop. Indeed, when soybean makes nodules, it creates isoflavones (which represents 50% of

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the soybean sprouts composition). Isoflavones are phenolic compounds, and contain glycosides. Soybean would excrete a lot of glycosides when a wireworm attack takes place (defensive response). Soybean may therefore have a repulsive action against wireworms.

Focusing on only alive larvae, the highest values were in maize plots, whatever the strategy (conventional, advanced or innovative) and there was always a higher number of larvae in systems with wheat-maize rotations.

Compared to IPM2 soybean system, few larvae were caught in the IPM1 soybean system, and none were dead. There are two explanations: fungi are more numerous or active in IPM1 system, or the repulsive action of soybean against wireworms is more important. This second explanation is confirmed by the absence of larvae found in T7 (maize-soybean IPM1 system). Statistical analysis shows that data are not significantly different.

Figure 12 represents the total number of larvae caught in each treatment for the three monitoring periods (May 2012, April 2013 and July 2013). Results were homogenized on a basis of three traps per plot, thus of 9 traps per treatment. The figure shows the evolution of wireworm larvae presence depending on year and type of system. It can be seen that wireworm populations decreased between 2012 and 2013. This is explained by a « year effect », independent of the experiment. There is a lack of a control sample (number of wireworms in each plot in 2011, before the trial started). It can be noted that soybean IPM2 plots which had the highest larvae number in July 2013 also had the highest values in April 2013 and in May 2012. It is possible that there is a plot bias, but we cannot confirm this because there was no control sample.



Fig. 12 Wireworm larvae evolution per system.



#### Barber traps:

Figure 13 shows the total number of wireworms for 6 traps (2 traps per plot, 3 repetitions) during all the monitoring period. A total of 28 adults were caught, i.e. 0.58 individuals per trap. Wireworm adults were present in all systems, except in wheat/maize rotation IPM1. The most affected plots were those of continuous maize in conventional and IPM1 systems, and then the two IPM2 systems. Hoeing has a repulsive effect on wireworms, and because the only system where there is no hoeing is T1, this causes a bias. Hoeing does not seem to be sufficient to control wireworms because IPM2 maize-soybean systems, where there was no Cruiser but hoeing, had a relatively high level of wireworms. It therefore seems that hoeing and insecticide are both needed, because the IPM1 maize system has fewer adults than the conventional maize system.



Fig. 13 Barber trap catches.

The highest number of adults was caught in June, as we can see very clearly in T1 curve (Figure 14). This is the period when hoeing is done, which means that hoeing is done when adults are most numerous. This regards the hypothesis previously made about hoeing interventions.





Fig. 14 Evolution of wireworm adult catches per treatment using the barber traps.

#### Wireworm damage on plants

Figure 15 shows damage evolution by system. It can be seen a gradual evolution of attacks from 16th May to 12th July. Attacks began in T8 (on maize), where no Cruiser insecticide was applied. On 12th May, maize plants in every scenario began to be affected, but with less than 5% of damage except for T8 which had 12%. The attack peak took place on 2<sup>nd</sup> July. The percentage of attacked plants then decreased because of plant regeneration.



Fig. 15 Wireworm damage evolution.

On 2<sup>nd</sup> July, wireworm damage was highest in T8, in maize/soybean rotation IPM2 system: around 20% of attacked plants (Figure 16). Other systems had lower rates, between 5 and 10% of attacked plants due to the seed dressing (Cruiser insecticide), which was always applied except in IPM2 systems. However, these attack rates are fairly high compared to normal fields treated with Cruiser insecticide. This was due to the rainy weather, which decreased the efficiency of the seed protective effect. There is no significant difference

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between conventional and advanced systems, so there does not seem to be a rotation effect on damage caused by wireworm larvae.



**Fig. 16** Wireworm damage on 2<sup>nd</sup> July 2013.

#### Weeds

#### 2012 Results:

#### Weed density in maize plots:

Figure 17 shows the weed density evolution between May and October in conventional, advanced (IPM1) and innovative (IPM2) systems.





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#### Fig. 17 Evolution of weeds density in maize.

On 31<sup>st</sup> May 2012, the highest weed density was observed in the innovative (IPM2) maize system, followed by the advanced maize system and, with the lowest weed densities, the conventional system. A high increase of weed density was then observed in each system until July. However, this increase was less in the conventional system than in the others. The increase coefficients are almost the same for the advanced and innovative systems. For all systems, the weed density decreased from July to the harvest. At harvest, the advanced maize had the lowest weed density, with 10 weeds/m<sup>2</sup>, followed by the conventional system. In the innovative system, many weeds were between rows and very large.

In the case of innovative (IPM2) system, the use of hoeing is not enough to control weed density of the entire area (particularly next to maize rows). Weeds can grow on rows or close to rows, without being damaged by a technique like hoeing.

The weed species identified are shown in figure 18. The most common weeds were Black Nightshade, Hedge birdweed, Common Chickweed and Fat Hen.

During the development cycle of maize, weeds appear; particularly Black Nightshade which could reach 70 plants/m<sup>2</sup> in the innovative maize system. These plants then disappear, to let Common Chickweed and Cockspur increase in October. Weeds species changed over time. The diagram shows that conventional maize system has a good and regular control of weeds during maize development, which facilitates its growth. The greatest density in the conventional system was less than 40 plants/m<sup>2</sup> with Black Nightshade, Fat Hen and Cockspur as main species.



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The research leading to these results has received funding from the European Union Seventh Framework Programme (FP7/ 2007-2013) under the grant agreement n°265865 -PURE

#### Fig. 18 Diagram of weed species distribution in maize.

However, the highest weed density was found in the innovative system (70 weeds/m<sup>2</sup>). The final density value shows that maize has not been competitive enough against weeds, because mechanical techniques do not give an excellent control of weeds. The high weed density could be harmful to the maize growth and yield. So, combining herbicides and hoeing could be a good solution to control weeds.

The highest weed density was that in the innovative (IPM2) maize system. The conventional system had the lowest weed density values throughout the maize development cycle, except at harvest, whereas the advanced maize had only a small number of Common Chickweed.

#### Weed densities in soybean plots:

The same species were observed in soybean plots (Figure 19). Weed density also had the same evolution, with an increase from May to July, and then a decrease until the harvest (Figure 20). During the first months, advanced (IPM1) soybean system had a higher weed density than innovative (IPM2) system, although the IPM2 system received less herbicide. However, there were only few weeds in the advanced system at harvest, and slightly more in the innovative system. Soybean therefore has a competitive effect against weeds so chemical control is not necessary.



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Fig. 20 Evolution of weed density in soybean.

#### 2013 results

#### Weed density assessment in maize plots:

Figure 21 shows the evolution of all the weeds in systems sown with maize in 2013. There were few weeds on 25th May, except in the advanced maize system which reached around 18weeds/m<sup>2</sup>. The only difference between all systems on this date was the kind of preemergence herbicide treatment (there was no-post emergence weed control, either chemical or mechanical). In T8 innovative system, there was a localized herbicide application in furrow, whereas in other systems application was on the whole plot. Therefore, herbicide row spraying implies an increase in weed density, whereas with overall spraying the plot had very few weeds.



Fig. 21 Evolution of maize weed density by system/treatment.

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After this first assessment, a herbicide treatment was applied on 25th May in conventional and advanced systems. In advanced system plots, there was one hoeing on 6th June, and in innovative systems two hoeings on 6th and 14th June. The second assessment was made on 25th June.

This second assessment showed a low weed density, between 1 and 3 weeds  $/m^2$ . The highest rate was observed in the IPM2 system, which did not receive any post-emergence herbicide treatment, but density differences between systems remained very low.

The last hoeing in advanced and innovative systems was done on 25th July, after monitoring. There was no herbicide treatment on the conventional system. The assessment made on 8th July showed that hoeing treatments were effective to control weeds in the innovative and advanced plots. Weed density had instead increased in the innovative system that was not hoed, which presented 11 weeds/m<sup>2</sup>.

The last assessment was made on 28th August. There had been no weed control between the two last assessments.

Conventional maize system had the lowest weed density (22 weeds/m<sup>2</sup>). Advanced systems, which combined chemical and mechanical weed control, followed with 22 weeds/m<sup>2</sup> (continuous maize and soybean-maize rotation) and 35 weeds/m<sup>2</sup> (wheat-maize rotation). According to these results, rotations do not seem to have a positive effect on weed control for maize based systems. However, it seems that the kind of weed control has an effect on weeds, because maize in the innovative system that had no post-emergence herbicide application, had the highest density with 55 weeds/m<sup>2</sup>. Therefore, hoeing alone does not seem to be sufficient to give a good control of weeds.

Weed identification showed that species proportions differ depending on the system. Conventional systems had more Cockspur, Black Nightshade, birdweeds and chickweeds, whereas advanced and innovative systems had more Black Nightshade but the other species had about the same proportions as in the conventional system.





Fig. 22 Weeds species proportions in maize crops by system.

#### Weed density assessments in soybean plots

As with maize plots, the first assessment took place on 25th May, when only pre-emergence weed control had been done. It was an overall spraying in the advanced system (29 weeds/ $m^2$ ) and a row spraying in the innovative system (62 weeds/ $m^2$ ) (Figure 23). A lot of weeds were therefore present in the innovative system, as was the case for maize.

Before the second assessment on 25th June, a row spraying application was made in the advanced system, and a hoeing on 6th June. There were two hoeings in the innovative system. The graph in Figure 23 shows that weed densities had highly decreased, and that soybean in the innovative system still had a higher rate than in the advanced system.

There was one hoeing in each system. On 8th July, a few weeds were found in each system (less than 3 weeds/ $m^2$ ). Weed control was efficient in this period. The 3rd hoeing was done on 10th July in the advanced system and the 4th in the innovative system.

During the last assessment, soybean in the advanced system had 20 weeds/ $m^2$ , whereas soybean in the innovative system without any post-emergence herbicide had reached 35. There is therefore an effect depending on the type of weed control.





Fig. 23 Evolution of weed densities in soybean plots in the advanced and innovative systems.

According to the following graph, the main weeds identified were Black Nightshade, Cockspur and Fat Hen.



Fig. 24 Densities and proportions of weed species in soybean plots.

#### Weed biomass

Weed biomass was checked on 17th September on all plots. Fresh biomass was weighed and dried in an oven for 24 hours at 105 °C. Results of weed dry matter are shown below.





Fig. 25 Assessment of fresh and dry weed matter.

In maize systems, dry matter rates were similar and varied from 15 to 17%. It can be noted that biomass does not follow the observed density. Indeed, the average fresh matter in conventional system was roughly the same as for innovative systems. Biomass in advanced systems was far less than that found in other systems. There thus does not seem to be a beneficial rotation effect on weed biomass, because advanced systems that include rotations had a higher level of biomass than continuous maize T2 system.

Hoeing therefore seems to slow weed development, contrary to what happened in the conventional system where weed control took place earlier in the maize development cycle. In conventional systems, weeds were less numerous, but more developed.

The presence of an edge effect is possible, because plants have more light when they are close to grass or soybean than when there are only rows of tall maize plants.

It is also probable that the field had a heterogeneous distribution of weeds.

The final hypothesis is that weeds are less numerous but more developed in conventional systems, whereas they are more numerous but less developed in advanced and innovative systems, where late hoeing is practiced.

According to these observations, it seems that combination of chemical action (with efficient weed control), and hoeing (the latest operations slow weed development) is a good solution to control both weed number and development.

Nevertheless, statistical analysis showed that the obtained results are not significant.

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#### **Yields**

#### 2012 Results

The maize yield of each system was assessed using dry matter yield and grain yield. Figure 26 presents the dry matter yield results, obtained after drying the harvested maize. The highest dry matter yield is in the conventional system, which reaches almost 24 tons of dry matter per hectare. Advanced (IPM1) and innovative (IPM2) systems have a lower values (between 21 and 23 tons/ha), so maize plants had a more moderate growth than those in the conventional system. Differences between maize plots in the advanced system (T2, T3 and T4) are only explained by factors beyond the experiment, because T2, T3 and T4 had the same crop management and rotations are not taken into account for this first experimental year.



Fig. 26 Maize dry matter yield.

At harvest, the moisture content of grain maize was 15% and 16% for soybean. Yields are heterogeneous between systems. The chemically protected maize (conventional) has the highest yield with 11.97 t/ha. The advanced system is not very different from the conventional system. The innovative system has lowest yields (7% less than the conventional system). The innovative system is therefore less efficient than other systems in terms of pest and weed control, because the only difference between systems is the crop management. However, the analysis of variance shows that the results are not significant (probability of 0.07).

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Fig. 27 Grain yields PURE 2012 (PURE systems are encircled).

In accordance with the protocol, mycotoxin analyses have been done on maize grains. Results are shown in the following table.

	DON	Fumonisin	Zéaralénon
Conventional	<111 µg/kg	410 µg/kg	< 7,5 µg/kg
IPM1 adavnced	314 µg/kg	434 µg/kg	< 7,5 µg/kg
IPM2 innovative	236 µg/kg	456 µg/kg	< 7,5 µg/kg

Conventional maize grains had less mycotoxin than other systems (DON and Fumonisin). Advanced (IPM1) and innovative (IPM2) systems are more sensitive to pests and further mycotoxin development, which decreased their grain quality. So, there is a higher sanitary risk in IPM1 and IPM2 systems than in the conventional ones. However, all the results were beneath the regulatory threshold for human food.

The wheat was not harvested in 2012 because of lodging (severe winds in May 2012).

## 2013 Results

Despite bad weather conditions in 2013: high rainfall (1700 mm total, compared to the 1050 mm current average) and a cold spring, yields were high (12.6 t/ha on average). The highest



yield was obtained with the advanced system (13.1 t/ha), the lowest one with the innovative system (11.8 t/ha).



Fig. 28 Grain yields for each system.

Level of contamination was low in 2013, DON and FUMO were below the current thresholds and values were similar for the three systems. Zearalenones rates were slightly above the regulatory thresholds (i.e.  $350 \ \mu g/kg$ ) for conventional and innovative systems.

#### Quality assessment

	DON	Fumonisines	Zéaralénone
Conventional	1049 µg/kg	194 µg/kg	387 µg/kg
IPM1	1067 µg/kg	165 µg/kg	$> 607.5 \mu g/kg$
IPM2	837 µg/kg	168 µg/kg	169 µg/kg

Fig. 29 Mycotoxin analysis on grain maize (ELISA Method).

# Conclusion

2012 and 2013 yields were similar for conventional and IPM1 systems. This can be related to the low level of wireworms and the efficiency of weed control. Indeed, the combination of

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hoeing and pre-emergence herbicide application (IPM1 system) seems to be as efficient as the conventional methods.

The experimental field historically had a high level of wireworms, and the surrounded fields are still affected. The very low level of wireworms in plots could be explained by the high frequentation in the field due to numerous measures and observations, and the exceptional climatic conditions (1700 mm cumulated in 2013, whereas the current rainfall average is 1050 mm). These two factors can be considered together or separately.



## **3.** Task **3.3a**: On-station experimentation in the Netherlands (comparison of different tillage and weed management systems)

R.Y. van der Weide, H.F. Huiting & M. Riemens (DLO)

## Introduction

### **Description problem/question**

Herbicides cause problems with respect to the quality of surface and groundwater. European and national regulations demand a reduction of this impact. To achieve this, innovations that result into a reduction of the emission of herbicides and crop protection products in general, and at the same time reduce costs are required.

As a result of the high energy prizes it is becoming more and more economically feasible to reduce the intensity of soil cultivation (ploughing) in agriculture. In the past exploratory research has shown that weed control was the main bottle neck for the introduction of reduced or no till systems and those systems were for that reason not economically feasible. However, recent technological developments and innovations in North America have brought new agricultural systems in which energy use and costs have been reduced considerably.

On top of that, these systems even suppress weed growth and have reduced the dependence of herbicides. At present, these systems are applied in American organic agriculture and further developed in practice. A fundamentally new aspect of these systems is the use of ridge till or no till systems in which crop residues are spread on top of the soil and used as soil coverage instead of being incorporated into soil. In North-America the methods are successfully applied in maize, soybean, sunflower and potatoes. In Denmark and Germany initial results in potatoes and sugar beet show good prospects.

### **Research objectives**

The objective of this project is to investigate the use of ridge till /no till systems as recently developed in the US and Canada, under Dutch conditions. The first step is the collection of all relevant technological data, expert knowledge and required equipment. Stakeholders will be consulted and equipment will be used and tested in cooperation with other projects. In the spring of 2009 a multiple year experiment was designed and initiated in which the efficacy and feasibility of several ridge/ no till systems are investigated in maize.



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The research leading to these results has received funding from the European Union Seventh Framework Programme (FP7/2007-2012) under the survey Programme (FP7/ 2007-2013) under the grant agreement n°265865 -PURE

## **Materials & methods**

## Trial field

The trial was conducted in a field with a history of maize growing (2009 - 2010) and grass (2006 – 2008). The soil is a marine loam (c. 30% clay).

### **Treatments**

Table 1 displays the treatments as carried out in 2011-2013.

Table 1. Treatments compared in 2011-2013.

Trt.	Description tillage system	Weed control
A-W1	Conventional ploughing (. 25 cm depth) & seedbed preparation (rotary harrow)	Chemical
A-W2	Conventional ploughing (. 25 cm depth) & seedbed preparation (rotary harrow)	Mechanical
C-W1	Conservation: deep rigid tine cultivation (25-30 cm depth) & rotary cultivator	Chemical
C-W2	Conservation: deep rigid tine cultivation (25-30 cm depth) & rotary cultivator	Mechanical
D-W1	Ridge tillage; build up ridge after sowing	Chemical
D-W2	Ridge tillage; build up ridge after sowing	Mechanical
E-W1	Direct seeding	Chemical
E-W2	Direct seeding	Mechanical
F-dead	Experimental: strip tillage; grass killed with glyphosate prior to seeding	N.A.
F-alive	2011: Experimental: strip tillage; grass alive; mowed 3 times	N.A.
	2012+2013: Experimental: strip tillage; grass alive; suppression by spray	
	application of rimsulfuron	

### Assessments

Assessments were carried out on 0.75 x 2 m subplots, according to the protocol predefined.

### Statistical analysis

Data were analysed using analysis of variance and Student's t-distribution with the PPAIR procedure in Genstat 16<sup>th</sup> edition. Means in the same column followed by different letters are significantly different.



## **Results & discussion**

### Results 2011

### Weeds

In all reduced tillage treatments high numbers of SOLNI were counted on 22 June (Table 2). Except for the chemical weed control treatment at ridge tillage these treatments showed more SOLNI than the ploughing treatments. At ridge tillage relatively high numbers of STEME were counted, though not at a problematic level. Also numbers of POAAN were quite low. High numbers of SOLNI were not expected but possibly caused by abnormal circumstances: a relatively warm early spring. Even in the strip tillage treatment high numbers of SOLNI were found, virtually in all the cultivated strip. The higher numbers of SOLNI in the glyphosate treated strips is probably a result of the absence of competition with the grass.

Trt.	Tillage	Weed control	Total	SOLNI	STEME	POAAN
A-W1	Ploughing	Chemical	7.6 a	0.7 a	1.3 a .	2.7 a b.
A-W2	Ploughing	Mechanical	2.0 a	0.2 a	0.2 a .	0.7 a
C-W1	Conservation	Chemical	120.0 d	116.7 d	0.7 a .	1.6 a b .
C-W2	Conservation	Mechanical	64.9.bc.	56.2 . b c .	2.7 a b	4.2 . b .
D-W1	Ridge-till	Chemical	46.7 a b	28.2 a b	6.4 . b	10.9 c
D-W2	Ridge-till	Mechanical	110.2 c d	94.4 c d	6.4 . b	2.9 a b.
E-W1	No-till	Chemical	67.3 . b c .	63.8 . b c d	0.4 a .	2.7 a b.
E-W2	No-till	Mechanical	107.6 c d	100.0 c d	0.4 a .	0.4 a
F-dead	Strip tillage	Glyphosate	63.6 . b c .	60.0 . b c .	0.0 a .	1.3 a b .
F-alive	Strip tillage	Mowing	20.0 a b	14.2 a b	0.0 a .	1.1 a b .
LSD (a :	= 0,05)		50.8	54.4	4.8	3.3
F-prob. l	P<0,05		< 0.001	0.001	0.050	< 0.001

Table 2. Weed count on 22 June 2011.

Ridge-till and no-till resulted in a significantly higher percentage of soil covering than the ploughing treatments on 6 October (Table 3), without differences between chemical and mechanical weed control. At conservation tillage mechanical weed control differed from ploughing as well but chemical control did not. The high covering at the experimental strip without chemical control is predominantly the grass that was not killed before seeding the maize, but mowed three times providing competition to the weeds.

Trt.	Tillage	Weed control	Total	Monocotyledons	Dicotyledons
A-W1	Ploughing	Chemical	6.0 a	3.3 a .	2.7 а.
A-W2	Ploughing	Mechanical	4.3 a	1.3 a .	3.0 a .
C-W1	Conservation	Chemical	26.7 a b .	1.0 a .	25.7 a b
C-W2	Conservation	Mechanical	41.7 . b .	1.0 a .	40.7 . b
D-W1	Ridge-till	Chemical	36.7 . b .	1.3 a .	35.3 . b
D-W2	Ridge-till	Mechanical	46.7 . b .	1.3 a .	45.3 . b
E-W1	No-till	Chemical	35.0 . b .	1.0 a .	34.0 . b
E-W2	No-till	Mechanical	35.0 . b .	1.7 a .	33.3 . b
F-dead	Strip tillage	Glyphosate	26.7 a b .	3.0 a .	23.7 a b
F-alive	Strip tillage	Mowing	81.7 c	63.3 . b	18.3 a b
LSD (a	= 0,05)		27.7	3.3	27.5
F-prob.	P<0,05		0.001	< 0.001	0.046

Table 3. Weed covering, 6 October 2011.



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The research leading to these results has received funding from the European Union Seventh Framework Programme (EP7/2007 2012) under the survey Programme (FP7/ 2007-2013) under the grant agreement n°265865 -PURE

### *Crop development*

Placing ploughing with chemical weed control in the position of the standard cropping system, all other systems suffer from plant loss to a certain extent (Table 4). Mechanical weed control is one factor to influence final plant numbers negatively, which is most pronouncedly when comparing weed control methods at ploughing, conservation tillage and ridge-till. Especially building up ridges may cause plant loss if smaller plants are covered with soil, which was the case in the trial. Reason for this was variation in crop emergence caused by dry circumstances.

		P P		,	
Trt.	Tillage	Weed control	16 May	20 May	8 October
A-W1	Ploughing	Chemical	6.5 c	7.2 . b	7.2 e
A-W2	Ploughing	Mechanical	5.6 . b c	7.4 . b	6.8 d e
C-W1	Conservation	Chemical	5.9 c	7.2 . b	6.9 d e
C-W2	Conservation	Mechanical	3.3 a b .	6.2 a b	6.1 c d e
D-W1	Ridge-till	Chemical	4.2 . b c	5.2 a b	5.7.bcd.
D-W2	Ridge-till	Mechanical	1.3 a	3.5 а.	4.8 a b
E-W1	No-till	Chemical	4.6 . b c	6.1 a b	6.0.bcde
E-W2	No-till	Mechanical	4.8 . b c	6.8 . b	6.2 c d e
F-dead	Strip tillage	Glyphosate	4.6 . b c	4.8 a b	5.1 abc
F-alive	Strip tillage	Mowing	4.4 . b c	5.0 a b	3.9 a
LSD (a =	= 0,05)		2.5	2.7	1.3
F-prob. I	P<0,05		0.020	0.105	0.001

**Table 4.** Numbers of maize plants per m row on three dates. 2011.

Despite (significant) variations in plant numbers at harvest (table 4), crop stand was in most cases not significantly influenced (Table 5). Only the lowest plant numbers, at mechanical weed control at ridge-till and at mowing at strip tillage, corresponded with significantly lower crop stand figures.

The level of covering with *Kabatiella zeae* seems inversely proportional to the level of burying crop residues (Table 5). At the ploughing treatments burying is maximal, whereas at no-till minimal, leaving conservation tillage in between. Results show no indication that mechanical weed control - possibly providing entry points for the disease - has enhanced K. zeae.

1 4010	Tuble et erop stalla and fear eo vering with Habalitetta Searc on T beptember, 2011.							
Trt.	Tillage	Weed control	Crop stand	Leaf covering K. zeae				
A-W1	Ploughing	Chemical	8.0 c	12.3 a				
A-W2	Ploughing	Mechanical	7.5 c	9.8 a				
C-W1	Conservation	Chemical	7.8 c	31.7 . b c				
C-W2	Conservation	Mechanical	7.7 c	25.0 a b c				
D-W1	Ridge-till	Chemical	6.8 c	20.0 a b .				
D-W2	Ridge-till	Mechanical	5.2 . b .	20.0 a b .				
E-W1	No-till	Chemical	7.3 c	40.0 c				
E-W2	No-till	Mechanical	7.0 c	41.7 c				
F-dead	Strip tillage	Glyphosate	7.3 c	35.0 . b c				
F-alive	Strip tillage	Mowing	3.7 a	33.3 . b c				
LSD (a	= 0,05)		1.449	18.97				
F-prob.	P<0.05		< 0.001	0.021				

**Table 5.** Crop stand and leaf covering with *Kabatiella zeae* on 1 September, 2011



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The research leading to these results has received funding from the European Union Seventh Framework Programme (FP7/2007-2012) under the event Programme (FP7/ 2007-2013) under the grant agreement n°265865 -PURE

### Yield

Compared with ploughing and chemical weed control all other cultivation systems result in a loss of yield, in fresh weight as well as dry matter and feed value (Table 6). These results may have been the combined result of reduced plant numbers and attack by *K. zeae*. No significant differences were found between chemical and mechanical weed control.

Trt.	Tillage	Weed control	Fresh weight	Dry matter	Feed value
A-W1	Ploughing	Chemical	37162 d	15707 d	14519 d
A-W2	Ploughing	Mechanical	33436 c d	13670 c d	12913 c d
C-W1	Conservation	Chemical	30325 . b c .	12498 . b c .	11104. bс.
C-W2	Conservation	Mechanical	29333 . b c .	11278. bс.	9935 . b
D-W1	Ridge-till	Chemical	28479 bc.	11195 . b с .	10300 . b c .
D-W2	Ridge-till	Mechanical	27248 . b c .	10470 . b	9421 . b
E-W1	No-till	Chemical	26051 . b	10330 . b	9099 b.
E-W2	No-till	Mechanical	27350 . b c .	10114 . b	9006 . b
F-dead	Strip tillage	Glyphosate	27829 bc.	11406 . b c .	10356 . b c .
F-alive	Strip tillage	Mowing	12000 a	5034 a	4650 a
LSD (a =	= 0,05)		6588.8	2767.8	2715.8
F-prob. 1	P<0,05		< 0.001	< 0.001	< 0.001

Table 6. Yield fresh weight, dry matter and yield feed value (VEM), kg/ha, at harvest, 9 October 2011.

### **Results 2012**

Weeds

Except for strip tillage total weed numbers did not differ from the ploughing system at reduced tillage on 26 June (Table 7). Increased weed numbers at strip tillage were virtually completely the result of grass sod residue, both at the chemical and the non-chemical treatment. In contrast, weeds at the remaining systems almost completely consisted of dicotyledons. SOLNI was present in significantly higher numbers at the conservation system (deep tine + superficial rotary cultivator) with herbicide in comparison to the other systems, possibly resulting from sub-lethal doses of herbicide, whereas the mechanical weed control was effective. At the no-till system both weed control strategies were less than optimally effective. At ridge-till with chemical control the highest numbers of CHEAL were counted, significantly differing from the ploughing system and the ridge-till with mechanical control.

Table	7. Wee	ed count on 26	June 20	12.												
Trt.	Tillage	Weed control	То	tal			SO	LN	Ι	CHI	EA	L	Gra	ass*		
A-W1	Ploughing	Chemical	34.0	а			29.3	a		0.9	а		0.2	a		
A-W2	Ploughing	Mechanical	0.4	а			0.4	а		0.0	а		0.0	a		
C-W1	Conservation	Chemical	162.0	а	b		153.3		b	3.6	а	b	0.0	а		
C-W2	Conservation	Mechanical	7.8	а			3.6	а		2.4	а		0.0	а		
D-W1	Ridge-till	Chemical	110.9	а			2.4	а		90.7		b	0.2	а		
D-W2	Ridge-till	Mechanical	4.9	а			0.2	а		1.6	а		0.0	а		
E-W1	No-till	Chemical	64.9	а			50.0	а		0.0	а		0.0	а		
E-W2	No-till	Mechanical	64.9	а			51.1	а		9.1	а	b	0.2	а		
F-dead	Strip tillage	Glyphosate	490.4		b		6.7	а		17.1	а	b	453.3		b	
F-alive	Strip tillage	Rimsulfuron	3394.7			c	22.2	а		15.6	а	b	3333.3			c
LSD (a =	= 0,05)		346.7				92.2			87.3			322.9			
F-prob. I	P<0.05		< 0.001				0.064			0.556			< 0.001			

\* = including sod remains



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Total soil covering by weeds was low after harvest on 24 October (Table 8). Only at the striptill treatments significantly increased soil covering was found, resulting from the grass sod preceding the crop. No differences between chemical and mechanical control occurred. Although not visible from the data escaping weeds may have had a larger soil covering before harvest but were mowed together with the maize.

Trt.	Tillage	Weed control	Total	Monocotyledons <sup>*</sup>	Dicotyledons
A-W1	Ploughing	Chemical	1.3 a	0.3 a	1.0 a .
A-W2	Ploughing	Mechanical	1.3 a	0.0 a	1.3 a .
C-W1	Conservation	Chemical	1.0 a	1.0 a	0.0 a .
C-W2	Conservation	Mechanical	0.3 a	0.3 a	0.0 a .
D-W1	Ridge-till	Chemical	1.0 a	0.7 a	0.3 a .
D-W2	Ridge-till	Mechanical	2.0 a	1.0 a	1.0 a .
E-W1	No-till	Chemical	0.3 a	0.0 a	0.3 a .
E-W2	No-till	Mechanical	2.0 a	1.7 a	0.3 a .
F-dead	Strip tillage	Glyphosate	18.3 . b .	18.3 . b .	0.0 a .
F-alive	Strip tillage	Rimsulfuron	53.3 c	30.0 c	23.3 . b
LSD (a =	= 0,05)		15.1	11.6	6.3
F-prob. H	P<0,05		< 0.001	< 0.001	< 0.001

Table 8. Weed covering, 24 October 2012.

\* = including sod remains

### *Crop development*

Placing ploughing with chemical weed control in the position of the standard cropping system, conservation tillage, ridge-till and no-till did not differ from this reference in final plant numbers (Table 9). The increased plant numbers at the no-till system may be resulting from either a slightly smaller sowing depth, or better soil moisture situation increasing moisture take-up and subsequent germination. The increased plant numbers of strip tillage result from slightly different settings of the machinery used, possibly combined with better germination conditions in the cultivated strip (moisture and/or temperature).

Table 0 Numbers of maize plants par m row on two dates 2012

Table	Table 3. Numbers of maize plants per in row on two dates, 2012.								
Trt.	Tillage	Weed control	25 May	1 June					
A-W1	Ploughing	Chemical	2.8 . b c .	7.1 a.					
A-W2	Ploughing	Mechanical	0.8 a	7.2 a.					
C-W1	Conservation	Chemical	0.5 a	7.4 a.					
C-W2	Conservation	Mechanical	0.3 a	7.2 a.					
D-W1	Ridge-till	Chemical	1.5 ab	7.2 a.					
D-W2	Ridge-till	Mechanical	1.5 ab	6.7 a.					
E-W1	No-till	Chemical	4.8 d	6.4 a.					
E-W2	No-till	Mechanical	3.7 c d	6.7 a.					
F-dead	Strip tillage	Glyphosate	0.6 a	9.3 . b					
F-alive	Strip tillage	Rimsulfuron	0.1 a	8.8 . b					
LSD (a =	= 0,05)		1.6	1.0					
F-prob. I	P<0,05	<	< 0.001	< 0.001					



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### Yield

Compared with ploughing and chemical weed control, conservation tillage and ridge-till resulted in similar yield levels, both gross and regarding yield components (Table 10). No-till resulted in c. 20% (significantly) yield loss expressed as fresh weight, decreasing to c. 15% regarding dry matter and feed value. At both strip tillage systems yield levels were significantly lower than at ploughing, conservation tillage and ridge-till, mutually differing between grass control with glyphosate and mowing. Decreased yield levels at strip tillage are probably due to competition with the grass for water and nutrients. As the glyphosate application was not completely effective (c. 450 grass plants/ $m^2$ , table 7) these dynamics also applied there.

Table 10. Yield fresh weight, yield dry matter and yield feed value (VEM), kg/ha, at harvest, 22 October 2012.

Trt.	Tillage	Weed control	Fresh weight	Dry matter	Feed value
A-W1	Ploughing	Chemical	56207 d	22793 d	22197 d
A-W2	Ploughing	Mechanical	56178 d	22536 c d	22235 d
C-W1	Conservation	Chemical	51052 c d	20920 c d	20338 c d
C-W2	Conservation	Mechanical	53304 c d	21027 c d	20432 c d
D-W1	Ridge-till	Chemical	54756 d	22182 c d	21923 c d
D-W2	Ridge-till	Mechanical	53037 c d	21851 c d	21693 c d
E-W1	No-till	Chemical	45452 . b c .	19448 . b c .	18973 . b c .
E-W2	No-till	Mechanical	46222 . b c .	19555. b с.	19272 . b c d
F-dead	Strip tillage	Glyphosate	39526 . b	16864 . b	16620 . b
F-alive	Strip tillage	Rimsulfuron	27259 a	9987 a	10122 a
LSD (a =	= 0,05)		8219	3164	3105
F-prob. I	P<0,05		< 0.001	< 0.001	< 0.001

### **Results 2013**

In 2013 a mistake was made at sowing, resulting in lower seed rates at the ploughing, conservation and ridge-till systems; the no-till and strip tillage systems were sown with different machines. Instead of the intended seed rate only 56% was sown. When this became clear re-seeding was – all considering – estimated as more influencing on the trial than other options. As the data presented are part of a larger trial it was decided to physically adjust plant numbers at the no-till system, but not at the strip tillage system.

To be able to compare data in this report plant numbers were re-calculated for plots with reduced physical numbers resulting from the sowing mistake.



### Weeds

Total numbers of weeds did not differ from weed control strategies within the ploughing system (Table 11). At the chemical weed control strategy no differences between systems were found in weed presence on 18 July. At conservation agriculture, ridge-till and no-till mechanical weed control showed an increase in total numbers of weeds present, to be attributed to SOLNI and CHEAL.

Table 11. Weed count on 18 July 2013.

Iunic		m on 10 sury 20	15.			
Trt.	Tillage	Weed control	Total	SOLNI	CHEAL	STEME
A-W1	Ploughing	Chemical	0.7 a	0.0 a	0.3 a .	0.0 a .
A-W2	Ploughing	Mechanical	9.7 a b.	5.7 a b .	0.7 a .	2.0 . b
C-W1	Conservation	Chemical	0.7 a	0.0 a	0.0 a .	0.0 a .
C-W2	Conservation	Mechanical	22.7 . b с	14.0 . b c	7.3 a b	1.0 a b
D-W1	Ridge-till	Chemical	0.0 a	0.0 a	0.0 a .	0.0 a .
D-W2	Ridge-till	Mechanical	25.7 . b с	8.0 a b .	17.0 . b	0.3 a .
E-W1	No-till	Chemical	0.0 a	0.0 a	0.0 a .	0.0 a .
E-W2	No-till	Mechanical	34.3 c	25.7 с	7.7 a b	0.0 a .
F-dead	Strip tillage	Glyphosate	0.0 a	0.0 a	0.0 a .	0.0 a .
F-alive	Strip tillage	Rimsulfuron	0.3 a	0.0 a	0.3 a .	0.0 a .
LSD (a =	= 0,05)		18.7	13.7	11.1	1.2
F-prob. I	P<0,05		0.004	0.012	0.062	0.033

Ridge-till and strip tillage resulted in the highest total weed coverings on 23 October, for the former predominantly consisting of dicotyledons whereas at the latter grasses were the major component (Table 12). Except for ridge-till weed covering was higher at mechanical than at chemical weed control.

Table 12	. Weed	covering,	23	October	2013.
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Trt.	Tillage	Weed control	Total	Monocotyledons	Dicotyledons
A-W1	Ploughing	Chemical	2.7 а	0.0 a .	2.7 a b.
A-W2	Ploughing	Mechanical	15.7 a b .	0.0 a .	15.7 a b.
C-W1	Conservation	Chemical	0.7 a	0.0 a .	0.7 a
C-W2	Conservation	Mechanical	4.3 a	0.0 a .	4.3 a b .
D-W1	Ridge-till	Chemical	56.7 c	0.0 a .	56.7 c
D-W2	Ridge-till	Mechanical	28.3 a b c	1.7 a .	26.7 . b .
E-W1	No-till	Chemical	2.0 a	0.0 a .	2.0 a b.
E-W2	No-till	Mechanical	14.3 a b .	0.0 a .	14.3 a b .
F-dead	Strip tillage	Glyphosate	27.3 а b с	20.0 . b	7.3 a b.
F-alive	Strip tillage	Rimsulfuron	38.3 . b c	25.0 . b	13.3 a b.
LSD (a =	= 0,05)		31.7	14.6	25.3
F-prob. I	P<0,05		0.022	0.008	0.006

### Crop development

Mechanical weed control influenced final plant numbers negatively, significantly at ploughing, ridge-till and no-till but not at conservation tillage and strip tillage (Table 13). The more than 30% plant number reduction at mechanical weed control and no-till, as compared with chemical control, may be caused by clods or plant residue damaging maize plants at

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machine passes. The 25% reduction at ridge till is better explained by covering with soil at ridging.

Table	Table 15. Numbers of marze plans per in row on three dates, 2015.						
Trt.	Tillage	Weed control	5 June	19 June	19 October		
A-W1	Ploughing	Chemical	4.7 . b c	6.3 d	6.0 c d		
A-W2	Ploughing	Mechanical	4.5 . b c	5.0 abc.	5.3 a b c d		
C-W1	Conservation	Chemical	4.7 . b c	5.7 . b c d	6.3 c d		
C-W2	Conservation	Mechanical	4.4 a b .	5.7 . b c d	6.0 c d		
D-W1	Ridge-till	Chemical	5.1 . b c	6.2 c d	6.0 c d		
D-W2	Ridge-till	Mechanical	5.0 . b c	4.5 ab	4.2 a		
E-W1	No-till	Chemical	4.8 . b c	6.1 c d	6.5 d		
E-W2	No-till	Mechanical	3.4 a	4.2 a	4.7 a b		
F-dead	Strip tillage	Glyphosate	5.4 c	5.9 c d	5.9 . b c d		
F-alive	Strip tillage	Rimsulfuron	4.5 . b c	6.2 d	5.0 a b c .		
LSD (a =	= 0,05)		1.1	1.2	1.3		
F-prob. I	P<0,05		0.071	0.012	0.023		

Table 13. Numbers of maize plants per m row on three dates, 2013.

### Yield

Strip tillage showed a significant decrease in yield – fresh weight and yield components – in comparison with the ploughing system, even if it was the only system without physically reduced plant numbers (Table 14). At strip tillage grass mowing reduced yield significantly in comparison with the use of glyphosate. At the other systems mechanical weed control compared with chemical control did not reduce fresh weight. At ridge-till however dry matter and feed value yield was lower at mechanical weed control in comparison with herbicide application.

**Table 14.** Yield fresh weight, yield dry matter and yield feed value (VEM), kg/ha, at harvest, 18 October 2013.

Trt.	Tillage	Weed control	Fresh weight	Dry matter	Feed value
A-W1	Ploughing	Chemical	46093 c d	17283 e	17836 e
A-W2	Ploughing	Mechanical	47426 d	17000 d e	17343 e
C-W1	Conservation	Chemical	42407 c d	17074 e	17516 e
C-W2	Conservation	Mechanical	46556 c d	17445 e	16950 c d e
D-W1	Ridge-till	Chemical	38500 . b c .	16523 c d e	17178 d e
D-W2	Ridge-till	Mechanical	33852 . b	12904 . b	12096 a b
E-W1	No-till	Chemical	38630 bc.	15879 c d e	16574 c d e
E-W2	No-till	Mechanical	40259 . b c d	14122 . b c d .	14018 . b c
F-dead	Strip tillage	Glyphosate	33148 . b	13603 . b c	14354 . b c d .
F-alive	Strip tillage	Rimsulfuron	23870 a	9545 a	10111 a
LSD (a =	= 0,05)		8486	2921	2977
F-prob. l	P<0,05		< 0.001	< 0.001	< 0.001



## General results and discussion 2011-2013

### Weeds

Compared to the reference soil cultivation with ploughing the reduced cultivation systems show an increase in weed numbers, though only significantly different at conservation tillage with chemical control and at no-till with mechanical control (Table 15). The latter can hardly be surprising as the mechanical weed control is the only soil cultivation taking place, intrinsically reducing the success chance of the cultivation. At conservation tillage the high SOLNI numbers in 2012 influence the average value greatly. Except for this situation overall weed control was as effective with mechanical means as with chemicals.

Trt.	Tillage	Weed control	Weed count
A-W1	Ploughing	Chemical	14.0 a b
A-W2	Ploughing	Mechanical	4.0 a
C-W1	Conservation	Chemical	94.2 d
C-W2	Conservation	Mechanical	31.8 a b c .
D-W1	Ridge-till	Chemical	52.4 . b c d
D-W2	Ridge-till	Mechanical	46.9 a b c .
E-W1	No-till	Chemical	44.1 a b c .
E-W2	No-till	Mechanical	68.9 c d
F-dead	Strip tillage	Glyphosate	33.6 a b c .
F-alive	Strip tillage	Rimsulfuron in 2012+2013 (2011 grass	27.2 a b c .
		mowed 3 times)	
LSD (a=	= 0,05)		44.4
F-prob. I	P<0,05	0.009	

Table 15. Total number of weeds, average of 2011-2013.

### Crop development

Plant numbers were comparable to ploughing, except for mechanical control in combination with ridge-till (Table 16). This result cannot solely be attributed to the ridging, as with chemical weed control ridging takes place as well. Possibly a combination with damage to plants at other passes explains this result.

Table 16. Num	ber of maize	plants during a	nd after final	emergence,	average of 2	2011-2013
		1 0		0 /	0	

Trt.	Tillage	Weed control	First count	Final count
A-W1	Ploughing	Chemical	4.6 d e	6.9 . b
A-W2	Ploughing	Mechanical	3.6 c	6.5 . b
C-W1	Conservation	Chemical	3.7 c d .	6.8 . b
C-W2	Conservation	Mechanical	2.6 ab	6.3 . b
D-W1	Ridge-till	Chemical	3.6 . b c	6.2 . b
D-W2	Ridge-till	Mechanical	2.6 a	4.9 a .
E-W1	No-till	Chemical	4.7 e	6.2 . b
E-W2	No-till	Mechanical	3.9 c d e	5.9 . b
F-dead	Strip tillage	Glyphosate	3.5 abc	6.7 . b
F-alive	Strip tillage	Rimsulfuron	3.0 a b c	6.6 . b
LSD (a =	= 0,05)		1.0	1.0
F-prob. I	P<0,05		< 0.001	0.012



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### Yield

The reduced tillage systems result in slight (c. 10%) to large (over 50%) reductions in yield (Table17). Reduction differences on average do not differ much regarding either fresh weight or yield components. Only at no-till mechanical weed control reduced yield significantly compared with chemical control, due to the difficult harrowing and hoeing circumstances. Whether the physical yield reductions translated into (direct) economic reductions remains to be worked out.

Table 17. Yield fresh weight, yield dry matter and yield feed value (VEM), kg/ha, at harvest, average of 2011-2013.

Trt.	Tillage	Weed control	Fresh weight	Dry matter	Feed value
A-W1	Ploughing	Chemical	46487 f	18594 f	18184 f
A-W2	Ploughing	Mechanical	45680 f	17735 e f	17497 e f
C-W1	Conservation	Chemical	41261 d e .	16831 d e .	16319 d e .
C-W2	Conservation	Mechanical	43064 e f	16584 c d e .	15772 c d
D-W1	Ridge-till	Chemical	40578 c d e .	16634 c d e .	16467 d e .
D-W2	Ridge-till	Mechanical	38046 c d	15075 . b с	14403 . b c
E-W1	No-till	Chemical	36711 . b c	15219 . b c d	14882 . b c d
E-W2	No-till	Mechanical	37944 c d	14597 . b	14099 . b
F-dead	Strip tillage	Glyphosate	33501 . b	13958 . b	13776 . b
F-alive	Strip tillage	Rimsulfuron	21043 a	8189 a	8294 a
LSD (a =	= 0,05)		4303	1628	1618
F-prob. l	P<0,05		< 0.001	< 0.001	< 0.001



## 4. Task 3.3b: On-farm evaluation of integrated weed management vs. conventional strategies during the first two years of PURE (2011-2012)

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## Introduction

Task 3.3b performs on-farm validation of IPM solutions. This report is aiming at presenting the results obtained during the first two years of the PURE project concerning the on-farm evaluation of the various integrated weed management (IWM) strategies tested against the conventional (CON) approach followed in the different countries under study. More specifically this work aimed at (i) selecting IWM strategies (i.e. using methods and tools already available but not widely implemented) that reduce the reliance and use of herbicides in three important and diverse European grain maize producing regions (southern, central and eastern regions), (ii) testing the efficacy of the selected IWM strategies in on-farm experiments (i.e. real field conditions in commercial or demonstration farms, with natural weed flora) against the conventional approach of each region, and (iii) performing a comparative assessment of their economic sustainability. The content of this report has been used for preparing a manuscript which is submitted to and currently under review by the European Journal of Agronomy.

## **Materials and Methods**

A total of 13 experiments have been carried out to test conventional against IWM methods. In each region, a minimum of two farms were used as the replicates each year (see Fig. 1 for map of the experiments). Two plots (minimum size of 0.5 ha) were created on each farm, where one plot was managed with the CON strategy against weeds (i.e. the one normally implemented in the farm) and the other using different IWM strategies for each region. In order to separate the effects of IWM on the maize grain yields and compare it with the conventional approach, the same crop and pest management was applied to both plots in each farm per region, thus the two plots differed only in the weed management. All on-farm experiments were managed with commercially available or technologically mature equipment suited to field scale applications. Protocol for weed and yield assessments and several



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The research leading to these results has received funding from the European Union Seventh Framework Programme (EP7/ 2007, 2012) under the survey of the su Programme (FP7/ 2007-2013) under the grant agreement n°265865 -PURE

detailed operative procedures were previously agreed and implemented during the experimental season of 2011 and 2012, and can be found in Annex 4 of the previously submitted deliverable (D3.1). Details about the weed management (i.e. herbicide products and doses, mechanical weeding) for CON and IWM strategies for each farm per country are given in Table 1. The 2012 data from the Hungarian experiments were not included in this study due to no machinery being available to apply the band-spraying in the IWM plots (i.e. in 2012 broadcast herbicide spraying was done in both IWM and CON plots with the only difference hoeing in IWM), therefore only the 2011 data are presented.

A cost-benefit analysis of weed management strategies was also performed to identify the economic sustainability. For this, a template was developed and provided to all partners involved in on-farm experiments for data collection of the crop management costs (costs of inputs, e.g. fertilizers and pesticides, and costs of operations, e.g. mechanical weeding, labour costs, fuel) and crop yields. For costs of inputs, the prices that farmers paid were used, whereas the operation costs were based on contract work prices including costs for labour, machinery and fuel, provided by regional contract work companies. Grain maize prices for 2011 and 2012 derived from the Eurostat database. Crop yields, prices and costs of inputs and operations (in  $\in$ ) were used for the calculation of CON and IWM gross margins [gross margin = financial yield (yield × price) – variable costs].



**Fig. 1** Map of experimental locations per country showing the average temperature (°C) and total precipitation (mm) for each growing season (April to October).



			Conventional weed management		Integrated weed management	nt
Farm/ Country	Year	Pre-emergence herbicide	Post-emergence herbicide	Mechanical weeding after post- emergence treatment	Early post-emergence herbicide or other treatment	Mechanical weeding after post- emergence treatment
Herbolzheim (2 farms), DE	2011	NO	topramezone/dicamba (0.04/0.13) + dimethenamid-P (0.58)	NO	topramezone/dicamba (0.04/0.13) + dimethenamid-P (0.58) in band spraying (30 cm) combined with hoeing	Hoeing
	2012	NO	topramezone/dicamba (0.05/0.14) + dimethenamid-P (0.65)	NO	topramezone/dicamba (0.02/0.05); dimethenamid-P (0.22); in band spraying (30 cm) combined with hoeing	Hoeing
Caorle, IT	2011	mesotrione/S-metolachlor/ terbuthylazine (0.15/1.25/0.75)	nicosulfuron (0.04) + dicamba (0.20) + mesotrione (0.06)	Hoeing	scouting & model indicated no application	Hoeing
	2012	same as above	foramsulfuron/isoxadifen-ethyl (0.05/0.05) + dicamba (0.30)	Hoeing	foramsulfuron/isoxadifen-ethyl (0.05/0.05) + dicamba (0.30)	Hoeing
Mogliano, IT	2011	mesotrione/S-metolachlor/ terbuthylazine (0.15/1.25/0.75)	foramsulfuron/isoxadifen-ethyl (0.05/0.05) + dicamba (0.24)	Hoeing	scouting & model indicated no application	Hoeing
	2012	same as above	NO	Hoeing	foramsulfuron/isoxadifen-ethyl (0.05/0.05) + dicamba (0.24)	Hoeing
Ceregnano, IT	2011	mesotrione/S-metolachlor/ terbuthylazine (0.15/1.25/0.75)	NO	Hoeing	scouting & model indicated no application	Hoeing
	2012	same as above	NO	Hoeing	foramsulfuron/isoxadifen-ethyl (0.03/0.03) + dicamba (0.24)	Hoeing
Berra, IT	2011	mesotrione/S-metolachlor/ terbuthylazine (0.15/1.25/0.75)	nicosulfuron (0.04) + dicamba (0.20)	Hoeing	scouting & model indicated no application	Hoeing
	2012	same as above	NO	Hoeing	nicosulfuron (0.04) + dicamba (0.20)	Hoeing
Ravenna, IT	2011	mesotrione/S-metolachlor/ terbuthylazine (0.15/1.25/0.75)	nicosulfuron (0.04) + dicamba (0.20) + mesotrione (0.06)	Hoeing	scouting & model indicated no application	Hoeing
	2012	same as above	NO	Hoeing	scouting & model indicated no application	Hoeing
Debrecen, HU (4 farms)	2011	NO	bentazon/dicamba (0.64/0.18) + nicosulfuron (0.04)	Hoeing	bentazon/dicamba (0.64/0.18) + nicosulfuron (0.04) in band spraying (30 cm)	Hoeing
Jablje, SL	2011	NO	mesotrione (0.02) + prosulfuron (0.02) + S-metolachlor (1.24)	NO	Tine harrowing plus mesotrione (0.06)	NO
	2012	NO	mesotrione/S-metolachlor/terbuthylazine (0.13/1.09/0.66)	NO	Tine harrowing plus mesotrione (0.1) + nicosulfuron (0.02)	NO
Rakican, SL	2011	NO	mesotrione/S-metolachlor/terbuthylazine (0.13/1.09/0.66)	NO	Tine harrowing plus mesotrione (0.1) + nicosulfuron (0.02)	NO
	2012	NO	same as above	NO	same as above	NO

Table 1<sup>a</sup>. Weed management strategies in the different farms of each country under study. <sup>a</sup> Values in brackets indicate the rate of herbicides (kg a.i. ha<sup>-1</sup>)



All statistical analyses were performed with Statistica 10. All data recorded from the weed density assessments were subjected to factorial analysis of variance (ANOVA) considering as main factors the "Strategies" (fixed factor), "Countries", "Stages of weed assessment" and "Years" (random factors), and using farms as replicates within countries. Data from weed biomass and grain yield were analysed without considering the factor "Stages". Effect of subsequent years was analysed with repeated measure ANOVA considering the date (2011 and 2012) as repeated measures (i.e. between years effect) and not as multiple variables as in MANOVA. Means obtained by ANOVA were compared using Fisher's protected LSD test at P=0.05 level of significance. Spearman rank order correlation analysis was performed for yield, weed density and weed dry biomass by pooling data over 3 countries (excluding Hungary) x 2 years, and obtaining six data points for each strategy, and graphical comparison was used to identify general trends.

## **Results and Discussion**

Data collected from the different countries included a very large agro-climatic, weed flora and management variability, with consequent variability in the yield that was normally distributed and with a significant "Country" effect (P<0.01).

The initial weed infestation in the IWM plots, i.e. at the stage before-post as no preemergence herbicides were applied in any country, across the different farms/countries for 2011 and 2012 was variable but generally rather low, ranging from 7 to 157 plants m<sup>-2</sup>, with a low species richness, ranging from 3 to 11 weed species. The most frequent species detected across all experiments/countries was *Chenopodium album*, while *Abutilon theophrasti*, *Amaranthus retroflexus*, *Convolvulus arvensis* and *Echinochloa crus-galli* were commonly observed in Italy, Slovenia and Hungary, and *Chenopodium polyspermum* in Germany, Italy and Slovenia.

Excluding the Italian experiments where pre-emergence herbicides were applied in CON plots (Table 1), statistical analysis showed that in 2011 and 2012 the weed density in the CON and IWM plots at the stage of weed assessment "before-post" in each country was not significantly different. This means that the size and position of plots were properly chosen and that most variability was within (inside) plots, so the effect of the strategies can be highlighted hereafter.

Overall both strategies were effective in significantly reducing weed density (Fig. 2), but CON was significantly more effective than IWM (on average 82% vs. 65% of weed control).



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Effects were stable across countries and years (interactions not significant), despite the greater weed density reduction in 2012 due to the higher initial density. Data analysis of weed dry biomass at the final assessment for Germany, Italy and Slovenia confirms the above main results, with a strong "Country" and "Strategies" effect, and a significant Year x Country interaction. The IWM strategy tested in Germany (i.e. early post-emergence herbicide application combined with hoeing and followed by another hoeing) gave a high weed control similar to the CON only in 2012 (95.4 vs. 97.8% of weed control). In 2011, *C. polyspermum* was not controlled efficiently by the hoeing operations between maize rows in IWM, resulting in high final densities compared to the post-emergence broadcast herbicide application in CON that gave 86% of weed control. Nevertheless, in both years no significant differences were observed in grain yield between IWM and CON strategies in the German trials.



**Fig. 2** Weed density (weeds m<sup>-2</sup>; columns indicate the mean and bars the standard error) at stages of weed assessment "before-post" and "final" as affected by the integrated weed management (IWM) and conventional (CON) strategies tested in the different countries (mean of two growing seasons).

In Italy 2011, the pre-emergence application of herbicides in CON was ineffective because a lack of rain after application failed to activate the herbicides, resulting in similar weed density to that of the "untreated" IWM at the stage "before-post" (6.3 vs. 7.4 plants m<sup>-2</sup>). The scouting and use of the predictive model in IWM indicated no need for post-emergence herbicide application, thus only hoeing was practiced in all five farms, whereas post-emergence herbicide was applied to CON plots in four out of five farms according to the standard practices implemented in these farms (Table 1). This resulted in similar weed control of CON and IWM strategies (final weed density of 9.7 vs. 10.8 plants m<sup>-2</sup>, respectively). In

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2012, the pre-emergence application of herbicides was very effective, with weed control of about 99.9% for CON and a density of 0.5 plants m<sup>-2</sup> compared to IWM that had 44.4 plants m<sup>-2</sup> at the stage "before-post". This high efficacy of pre-emergence herbicide application, when herbicides are properly activated by favourable soil moisture, is typical and explains why farmers in Italy are conventionally adopting this practice. In this case (i.e. 2012), the IWM strategy indicated herbicide application in four out of five farms and resulted in high weed control (92.6%) comparable to that of the CON strategy (98.5%). In both years, there was no significant difference in the grain yields between IWM and CON strategies, but only a "Year" effect because of the extremely dry season that greatly reduced grain yields in 2012, especially in two out of five farms where no irrigation was available.

In Hungary 2011, there was no clear efficacy of the weed control strategies because of low initial and final weed densities observed for CON (10.2 and 11.6 plants m<sup>-2</sup>, respectively) and IWM strategies (23.9 and 19.4 plants m<sup>-2</sup>).

Slovenia had the highest initial weed infestation of all the countries (70 and 161 plants m<sup>-2</sup> in 2011 and 2012, respectively). The tine harrowing combined with reduced herbicide doses provided partial weed control in 2011 compared to the broadcast application of herbicide, whereas the level of control was higher in 2012 when the initial density was higher (56.8% and 83.8% weed control in 2011 and 2012, respectively). Also in Slovenia no significant differences in the grain yields between IWM and CON strategies were observed.

Statistical analysis considering the years 2011 and 2012 as a repeated measure (excluding the Hungarian experiments) showed that the effect of time elapsed on final weed densities was significant (P<0.0001) and also the interaction "Year x Country" (P<0.0001). However, the interaction "Year x Strategy" was not significant. Considering that only data from two years are available, these results suggest that in general the elapsed time for the given strategies does not matter, and in the subsequent years a similar effect can be expected, since both strategies account for the best techniques "here and now".

Correlation analysis of the pooled data showed that final weed density or final weed dry biomass and grain yield are not correlated, nor was an overall strategy effect observed on yield (Fig. 3A and 3B). This is because the natural weed flora in each farm was mixed, with different species of varying competitive ability and time of emergence, so a typical yield lossdensity relationship is unlikely. Furthermore, the final weed densities were low due to the overall good control provided by both strategies, so that other factors were likely of greater importance in reducing yields, e.g. the lack of rainfall/irrigation in the case of Italy and



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Slovenia in 2012. A significant correlation was found only between mean values of weed density and weed dry biomass (n=12, Spearman r=0.61, P<0.05) (Fig. 3C).



**Fig. 3** Correlation analysis of maize grain yields (t ha<sup>-1</sup>) with final weed density (A), weed dry biomass (B) and of weed density and dry biomass (C) as affected by the different strategies per country (excluding Hungary) and years. Empty marker: 2011; Full marker 2012; Circle: conventional; Square: integrated weed management; red: Germany; Black: Italy, Blue: Slovenia.

The IWM compared to the CON strategy (mean of two years) increased costs in Germany  $(+32 \in ha^{-1})$  and Slovenia  $(+6 \in ha^{-1})$ , whereas costs in Italy and Hungary (only 2011 data) were lower (-82 and -24  $\in ha^{-1}$  respectively). The gross margin of IWM was found to be lower than CON in Germany (-105  $\in ha^{-1}$ ) and Italy (-50  $\in ha^{-1}$ ), and higher in Hungary (+61  $\in ha^{-1}$ ; only 2011 data) and Slovenia (+59  $\in ha^{-1}$ ). The differences in total costs and gross margin between the IWM and CON strategies for each year are given in Fig. 4.



**Fig. 4** Difference in total costs (A) and gross margins (B) of integrated weed management (IWM) vs. conventional (CON) strategies tested in the different countries for each growing season (excluding Hungary 2012). Scouting costs apply only in the case of Italy. Where: DE, Germany; IT, Italy; HU, Hungary; SL, Slovenia.



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## **Conclusions and Perspectives**

This study, conducted under real farm conditions, shows that with medium-low weed density (i.e. less than 50 plants  $m^{-2}$ ), IWM strategies tested in the different countries 1) provided sufficient weed control, 2) reduced maize reliance on herbicides, and 3) IWM implementation was economically sustainable as its costs, when averaged over the tested strategies in the different countries, were lower than those of the CON management and no significant reduction in yield was observed. It was highlighted that IWM in Europe, and IPM as a conceptual framework, is not about a unique weed control strategy but is based on general principles that must be adapted to address specific local agro-environmental and social conditions. This should be taken into account by authorities drawing up National Action Plans for implementation of the Directive 2009/128/EC regarding the sustainable use of pesticides. Policy- and decision-makers should provide support by more closely involving the regional advisory services for the general implementation of IPM (e.g. organize open field visits for dissemination of successful IPM strategies, establish farmer training programmes, encourage the formation of farmers groups to exchange experiences of IPM implementation), and enhancing the knowledge on sustainable use of pesticides within the framework of the Directive.

National and regional policies should promote IPM produced maize and the derived end products on the market in order to achieve a positive impact on their price and the willingness of society to pay more, as this could compensate for possible reductions in the farmers' gross margins after implementation of IPM.



## 5. Task 3.3b: On-farm evaluation of innovative integrated pest management strategy during the first two years of PURE (2011-2012)

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## Introduction

Task 3.3b performs on-farm validation of IPM solutions. This report is a summarization of the results obtained during the first two years of the PURE project concerning the on-farm evaluation of the integrated pest management (IPM) strategy tested against the conventional (CON) approach followed in the participating countries. Specifically this work aimed at (i) assessing the efficacy of optimally timed release of *Trichogramma brassicae* to control European corn borer (ECB) in on-farm experiments (i.e. real field conditions in commercial or demonstration farms) against the conventional approach practiced in three important European grain maize producing regions (southern, central and eastern) that are characterised by dissimilar geo-climatic conditions, (ii) performing a comparative assessment of the proposed strategy's economic sustainability, and (iii) providing recommendations to stakeholders involved in European maize production. The content of this report has been used for preparing a manuscript which will be submitted to Pest Management Science in the spring/summer 2014.

## **Materials and Methods**

In 2011 and 2012, fifteen on-farm experiments were conducted to compare the efficacy of IPM and conventional (CON) strategy. Three important European grain maize producing regions (southern, central and eastern regions) were selected for these experiments that represent the range of climatic and edaphic conditions as well as different types of cropping systems in Europe (see Fig. 1 for map of the experiments). In each region, a minimum of two farms were used as replicates each year. Two plots (at least 0.5 ha) were created on each farm, where one plot was managed with the CON strategy (i.e. normal agricultural production for each region) and the other using *Trichogramma*-based IPM strategy against ECB. In order to separate the effects of IPM on the maize grain yields and compare it with the conventional approach, the same crop management was applied to both plots in each farm per region, thus the two plots differed only in the ECB management. All on-farm experiments were managed

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with commercially available or technologically mature equipment suited to field scale applications.



**Fig. 1** Map of experimental locations per country showing the average temperature (°C) and total precipitation (mm) for each growing season (April to October).

ECB biological control strategy involving inundative releases of *T. brassicae* in field corn was performed under normal agricultural practices, i.e. on farms. In order to separate the effects of IPM (biological control with *T. brassicae* ECB egg parasitoids) on the maize grain yields and compare it with the conventional approach, the same crop and weed management was applied to both plots in each farm per region, thus the two plots differed only in the pest management. In Italy, Hungary, Germany various insecticides were used on CON plots. In France and Slovenia no spraying was performed against ECB on CON plots. In all countries no spraying against ECB was performed on IPM plots. For a presentation of different ECB management strategies see Table 1.

In all countries ECB flight was monitored using light traps. Additionally, various ECB-related parameters were observed: total number of plants (final maize stand), plants without ears or cobs, plants with any symptoms of ECB attack (e.g. holes on leaves, stalks or cobs), plants broken above ear and plants broken below ear. Additionally, on ten plants from each subplot, ECB damage and *Fusarium* spp. presence was observed on ears.

Special care was taken to optimize *T. brassicae* release time. The release date was fine-tuned based on the observation of the dynamics of ECB pupation. When the first generation flight was waning, the evaluation of pupation was initiated: ECB-infested plants were cut and ECB larvae and pupae on corn plant leaves and inside corn stalks counted. The release date was



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planned one week after the threshold range of 25-30% of pupation was reached. For exact release dates see Table 1.

Country	Year	Strategy	Pest management <sup>1</sup>	Dates of treatment
Italy (5 farms)	2011	CON	lambda-cyhalothrin, Syngenta; 25 ml ha <sup>-1</sup>	12/7, 16/7, 20/7, 21/7, 22/7,
		IPM	2 x 375,000 <i>T. brassicae</i> ha <sup>-1</sup>	6/7, 20/7
	2012	CON	lambda-cyhalothrin, Syngenta; 25 ml ha <sup>-1</sup>	16/7, 18/7, 19/7, 19/7, 9/8
		IPM	2 x 375,000 <i>T. brassicae</i> ha <sup>-1</sup>	12/7, 25/7
Germany (2 farms)	2011	CON	indoxacarb; 125 g ha <sup>-1</sup>	/
		IPM	2 x 375,000 <i>T. brassicae</i> ha <sup>-1</sup>	15/6, 30/6
	2012	IPM <sup>2</sup>	2 x 375,000 <i>T. brassicae</i> ha <sup>-1</sup>	28/6, 10/7
		IPM	2 x 375,000 <i>T. brassicae</i> ha <sup>-1</sup>	28/6, 10/7
Hungary (4 farms)	2011	CON	lambda-cyhalothrin, Syngenta; 16.5 ml ha-1	14/7
		IPM	2 x 375,000 <i>T. brassicae</i> ha <sup>-1</sup>	15/6, 6/7
	2012	CON	lambda-cyhalothrin, Syngenta; 16.5 ml ha <sup>-1</sup>	9/7
		IPM	2 x 375,000 <i>T. brassicae</i> ha <sup>-1</sup>	2/7,18/7
France (2 farms)	2011	CON	no treatment	no treatment
		IPM	1 x 375,000 <i>T. brassicae</i> ha <sup>-1</sup>	7/7
	2012	CON	no treatment	no treatment
		IPM	1 x 375,000 <i>T. brassicae</i> ha <sup>-1</sup>	17/7
Slovenia (2 farms)	2011	CON	no treatment	no treatment
		IPM	2 x 375,000 <i>T. brassicae</i> ha <sup>-1</sup>	8/7, 20/7
	2012	CON	no treatment	no treatment
		IPM	2 x 375,000 <i>T. brassicae</i> ha <sup>-1</sup>	25/7, 7/8

Table 1. ECB pest management strategies in different countries in the two-year experiment.

<sup>1</sup> Insecticide rate application is expressed as mass active ingredient ha<sup>-1</sup>.

<sup>2</sup> No CON plot was established in Germany in 2012.

*T. brassicae* parasitation was assessed by scouting for ECB egg masses under maize leaves on 100 plants per plot. Each egg mass was categorized as 'fresh' (F), 'parasitized' (P) or 'hatched' (H). Based on these observations, percentage of infestation and parasitation were calculated. Scoutings were carried out twice during the second generation egg laying period: 10 days and 20 days after the first *Trichogramma* release.

Yield was assessed by weighing the entire corn harvest on whole 0.5 ha plots using combine harvester. Grain moisture was determined by standardized methods (ISO 711:1997). Grain yield was expressed in kg ha<sup>-1</sup> of grain with 14 % moisture content.

A cost-benefit analysis of pest management strategies was also performed to identify the economic sustainability. A template was developed and provided to all partners involved in on-farm experiments for data collection of the crop management costs (costs of inputs, e.g. fertilizers and pesticides, and costs of operations, e.g. pesticide spraying, labour costs, fuel) and crop yields. For costs of inputs, the prices that farmers paid were used, whereas the operation costs were based on contract work prices including costs for labour, machinery and fuel, provided by regional contract work companies. For manual work (e.g. application of *Trichogramma*) labour prices were derived from the Farm Accountancy Data Network. Grain maize prices for 2011 and 2012 were derived from the Eurostat database. Crop yields, prices



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and costs of inputs and operations (in  $\in$ ) were used for the calculation of CON and IWM gross margins [gross margin = financial yield (yield × price) – variable costs].

All statistical analyses were performed with Statistica 10 (StatSoft Inc., 2011). All data recorded were subjected to factorial analysis of variance (ANOVA) considering "Strategy", "Country", and "Year" as main factors, and using farms as replicates within countries. When there was lack of replication in time for some countries, their data were analysed without considering the factor "Year". One-way ANOVA was performed only in the case of Germany that had one year data of grain yield. Means obtained by ANOVA were compared using Fisher's protected LSD test at P=0.05. Pearson's product moment correlation coefficient (Pearson's r) was performed to identify the relationship between mycotoxin concentration in grain, ECB damage on ears and *Fusarium* spp. presence on ears by pooling two-year data from four countries where mycotoxin assessments were performed (N=36).

## Results

The ECB pest pressure was evaluated using light traps. The flight dynamics varied strongly between countries. ECB first occurred in France, than Slovenia, Germany, Hungary and last in Italy (Fig. 2).



Fig. 2 Results of ECB monitoring with light traps in 2011 and 2012 in different countries.



The percentage of maize plants damaged by first generation (G1) ECB differed greatly between countries and years (Table 2). The lowest G1 damage was reported from Hungary in 2011 ( $1.0 \pm 0.4$  %, IPM plots) and the greatest in 2012 in France on CON plots ( $16.0 \pm 12.0$  %). The relationship between ECB G1 adults caught in light traps and ECB G1 plant damage was positive ( $r^2 = 0.68$ ). Final maize stand also differed greatly between countries and years (not shown). The minimal number of plants per 20 m before harvest was 48.5 (Italy, 2012, CON plot) and maximum 117.0 (Germany, 2012, CON plot), however no relationship between final maize stand and yield was observed.

**Table 2** Dependence of first generation ECB damage, second generation ECB total egg masses, *Trichogramma brassicae* parasitation of G2 egg masses and corn grain yield, on pest management strategy in different countries in 2011 and 2012. Data presented are means  $\pm$  standard error.

Country	Year	System	ECB G1 attacked	Total ECB G2 egg masses per	ECB G2 parasitism	Average yield (t
			plants (%)	100 plants	rate (%)	ha <sup>-1</sup> )
Italy	2011	CON	14.86 +-3.43	$24.06 \pm 14.53$	-	$12.26\pm0.66$
		IPM	$6.43 \pm 1.49$	$27.48 \pm 9.76$	$82.89 \pm 0.87$	$12.63\pm0.66$
	2012	CON	$10.03 \pm 5.28$	$58.50 \pm 36.77$	-	$7.51 \pm 1.07$
		IPM	$13.30 \pm 5.72$	$53.30 \pm 31.90$	$76.90 \pm 14.20$	$7.39 \pm 1.36$
Slovenia	2011	CON	$3.25 \pm 0.25$	$3.37 \pm 1.75$	-	$10.29\pm0.28$
		IPM	$5.50 \pm 4.50$	$5.00 \pm 3.75$	$80.77 \pm 19.23$	$10.59\pm0.39$
	2012	CON	$5.02 \pm 3.98$	$7.50 \pm 4.38$	_	$7.13\pm0.08$
		IPM	$5.02 \pm 3.48$	$8.44 \pm 5.94$	$0.00 \pm 0.00$	$6.44 \pm 0.43$
France	2011	CON	$4.00 \pm 1.00$	$2.00 \pm 2.00$	_	$13.82\pm0.36$
		IPM	$4.00 \pm 1.00$	$11.25 \pm 1.75$	$62.61 \pm 31.84$	$14.15\pm1.25$
	2012	CON	$16.00 \pm 12.00$	$19.00 \pm 7.00$	_	$11.53 \pm 1.32$
		IPM	$7.00 \pm 7.00$	$12.00 \pm 4.00$	$79.49 \pm 12.82$	$12.48\pm0.92$
Hungary	2011	CON	$1.50\pm0.29$	$1.33 \pm 0.00$	-	$11.30\pm0.06$
		IPM	$1.00 \pm 0.41$	$1.00 \pm 0.33$	nd	$11.55\pm0.18$
	2012	CON	$1.00 \pm 0.42$	$1.33 \pm 0.00$	_	$11.53\pm0.29$
		IPM	$1.25 \pm 0.25$	$0.67 \pm 0.38$	nd	$11.98\pm0.17$
Germany	2011	CON	$1.00 \pm 1.00$	$1.50 \pm 1.50$	-	$14.47\pm0.07$
		IPM	$1.00 \pm 0.00$	$8.50\pm8.50$	$66.67 \pm 0.00$	$14.06\pm0.02$
	2012	IPM	nd	nd	nd	nd
		IPM	nd	$1.25 \pm 0.75$	$50.00\pm0.00$	$12.92\pm0.30$

nd – not determined

Correlation analysis showed a significant relationship of mycotoxin concentration in grain with *Fusarium* spp. presence on ears ( $r^2 = 0.55$ ; *P*<0.001) and ECB damage on ears ( $r^2 = 0.51$ ; *P*<0.001), as well as of *Fusarium* spp. with ECB damage on ears ( $r^2 = 0.54$ ; *P*<0.001) (Figure 3A, 3B, 3C).



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**Fig. 3** Correlation analysis of mycotoxin concentration in grain with *Fusarium* spp. presence on ears (A), ECB damage on ears (B) and of *Fusarium* spp. with ECB damage on ears (C) as affected by the different strategies per country, farms and years. Dots represent the various farms per country and year (see section 2.7. for details). Only farms where mycotoxin concentration was above the EU threshold and where *Fusarium* spp. presence on ears and ECB damage on ears (scale 2) was above 1 % are labelled. Mycotoxins, fumonisin B1 and B2 concentration in grain for France, Italy, Slovenia and DON for Germany 2011; I, Italy; F, France; 1, 2011; 2, 2012; C, conventional; T, *Trichogramma*; dotted lines indicate the confidence intervals at 0.95 probability.

Statistical analysis considering all factors (i.e. without Germany 2011) showed no significant difference in grain yield between strategies but only a "Country", "Year" and their interaction effect since in Italy and Slovenia yields were significantly lower in 2012 due to adverse weather conditions (Figure 4). One-way ANOVA showed significantly higher grain yield in the conventional strategy in Germany 2011 (P<0.05) with 0.4 t ha<sup>-1</sup> of grain more than IPM. When countries were grouped in those using insecticide as their conventional approach and the ones without, CON plots with insecticide yielded 0.05 t ha<sup>-1</sup> more than IPM, whereas CON plots without insecticide yielded 0.30 t ha<sup>-1</sup> less than IPM plots; however no significant difference was obtained in both cases.



**Fig. 4** Grain yield (t ha<sup>-1</sup> adjusted to 14% moisture content; data presented are means  $\pm$  standard error) per year as affected by the integrated pest management (IPM) and conventional (CON) strategies tested in the different countries (only 2011 for Germany). Where: black, conventional 2011; dark blue, conventional 2012; light blue, IPM 2011; white, IPM 2012.



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Compared to the CON strategy the IPM strategy increased costs in all countries, ranging from  $+ 22 \notin ha^{-1}$  in Italy to  $+107 \notin ha^{-1}$  in Slovenia. If only the trials in which a spraying was done in the CON treatment (Italy 2011 and 2012, Germany 2011 and Hungary 2011) were taken into account the total costs increase ranged from  $\notin 10$  to  $\notin 64$  ha^{-1}. The cost increase is mainly due to the costs for the *Trichogramma* product. The gross margin of the IPM strategy was found to be lower than the CON strategy in Germany (-128  $\notin ha^{-1}$ ), Hungary (-5  $\notin ha^{-1}$ ) and Slovenia (-150  $\notin ha^{-1}$ ) and higher in Italy (+6 ha^{-1}) and France (+83 ha^{-1}).

## Discussion

This study conducted in real field conditions, in commercial or demonstration farms in different geographic and climatic regions of Europe demonstrates the diversity of ECB pressure across Europe and in certain extent explains the choice of insecticide applications as the conventional management against this pest in certain countries. Results from ECB adult catches by light traps, maize damage from G1 and the number of G2 ECB egg masses showed clearly that northern Italy suffers the highest ECB pressure among participating countries followed by southern France, Slovenia, southwest Germany and eastern Hungary (Figure 2, Table 2).

Predicting maize plant damage by studying only the abundance of adults is unreliable. The main importance of the first generation is the high reproductive rate of the individuals, because one corn borer female can lay around 300 eggs. The second generation is thus more numerous than the first generation and has as such more direct consequences to maize plant damage. It is only by assessing the number of G2 egg masses the time of *T. brassicae* release could be optimised. Ideally, the timing of egg parasitoid release should anticipate peak ECB oviposition. Asynchrony between parasitoid release and ECB oviposition activity can reduce parasitation efficiency. The method of determining the time of release of *T. brassicae* in this study was satisfactory. Results show that the release dates in the various countries coincided well with the beginning of G1 in Germany and the more damaging G2 for all other countries, since the parasitism rate of *T. brassicae* was above 67% in all countries (without considering Slovenia 2012 and Hungary), reaching even 80% in Italy during the two years of the study (Table 2). The extremely high temperatures in Italy and Slovenia in 2012 principally affected Slovenia (0% parasitism) since no irrigation was done in both farms, and to a lower level also Italy where irrigation was practiced in three out of five farms. Previous studies have

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demonstrated the effect of high temperatures on efficacy of *T. brassicae*. However, it should be taken into account that also the use of insecticides as the conventional pest management, in the respective countries, does not guarantee a 100% ECB control since there is a limited "window" for application. ECB is difficult to control because larvae are only exposed to insecticide sprays from egg hatching until larval tunnelling into maize plants or into the maize ears. Insecticide applications thus have even a smaller time frame for efficient ECB control, than *Trichogramma* spp. release. Systemic insecticides could potentially exhibit a greater efficacy against ECB because of a longer residual time in plant tissue, but unfortunately they could detrimentally affect natural enemies of the pest. In this respect, the use of *Trichogramma* spp. against ECB does not directly damage autochthonous population of generalist predators and other beneficial arthropods, but also alleviates the negative short- and long-term effects of insecticide use on farmers' health and the environment.

Results obtained for ECB damage on final maize stand and ears corroborates that *T. brassicae* release was well timed in the different countries: when IPM strategy was compared to the insecticide treatments in particular, it had the same efficacy against this pest as no significant difference in plant damage was determined between strategies. Additionally, in countries where no insecticide was applied in the conventional management (i.e. France and Slovenia) the average ECB damage on plants and ears was always lower in the IPM plots, however these differences were not significant (Table 2). The same non-significant effect between strategies was observed in grain yields stressing that biological control of ECB could result in similar yields with the conventional management in the respective countries. In fact, when insecticide was sprayed there was overall only a 0.05 t ha<sup>-1</sup> yield increase in CON plots, whereas IPM plots yielded 0.30 t ha<sup>-1</sup> more than CON plots when insecticides were not applied. Several studies worldwide have reported even higher grain yield increases when *Trichogramma* spp. was released against corn borers.

The tested IPM strategy was also effective at preserving mycotoxin levels in grain below the EU threshold for maize destined for human consumption (4000  $\mu$  kg<sup>-1</sup> of grain for fumonisin B1 and B2; 1750  $\mu$  kg<sup>-1</sup> of grain for DON; reference) in the different countries. The only exception was one farm in Italy 2012 where more than 11000  $\mu$ g of fumonisins kg<sup>-1</sup> of grain in both CON and IPM plots were measured. This isolated case was most probably caused by extremely high temperatures and lack of irrigation. Additionally, no significant differences were observed between IPM and the conventional approach on this farm. Investigating the interaction between ECB damage on ears, *Fusarium* spp. presence on ears and mycotoxin concentration, it was identified that all parameters were positively correlated to each other,

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which agrees to previous research demonstrating that ECB can vector fungal infection (i.e. *F. verticillioides*) through kernel wounds and spread the infection during larval movement, thus proportionally influencing the mycotoxin contamination in grain. Therefore, it can be assumed that the parameter ECB damage to ears can be a useful indicator for potential mycotoxin contamination in grain.

The economic impact of the IPM strategy compared to the CON strategy is determined by changes in costs and crop yields. For all trials costs of IPM-strategy were higher than that of the CON strategy (i.e. with and without insecticide), mainly due to the *Trichogramma* product costs.

### **Conclusions and Perspectives**

The 2011-2012 study performed within project PURE, WP3, conducted under real farm conditions in five European countries, shows that biological control using optimally timed mass release of *T. brassicae* egg parasitoids against ECB (i) provided similar efficacy as the insecticide treatments in Italy, Germany and Hungary, and higher (i.e. although not significant) to untreated CON plots in France and Slovenia, (ii) preserved mycotoxin levels below the EU threshold for maize destined for human consumption, (iii) has the potential to reduce the reliance on insecticides in maize production (i.e. where insecticide was applied), (iv) maintained similar or improved grain yields compared to CON, where insecticide was not applied), and (v) its implementation was economically sustainable in France and to a lower extent in Italy, but was not sustainable in other countries.

It is suggested that biological control of ECB using optimally timed mass release of *T*. *brassicae* could be considered agronomically as a sustainable option for IPM programmes in the participating countries. However to achieve this, policy- and decision-makers should provide support by more closely involving the regional advisory services for the successful dissemination and implementation of biological control (e.g. organize open field visits, establish farmer training programmes, encourage the formation of farmer groups to exchange experiences of IPM implementation). Also, a significant improvement of the general adoption of this IPM tool would be via subsidy schemes to motivate farmers for and to compensate them for possible reductions in their gross margins after its implementation. A good example comes from south-western Germany (federal state of Baden Württemberg) where the use of biological control with *T. brassicae* against ECB in maize is currently subsidized with  $60 \in ha^{-1}$ 



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## 6. Task 3.3b: Summary report for on-farm experiments 2013

Compiled by J. Razinger (KIS) based on data kindly provided by: R. Leskovšek (KIS), V.P. Vasileiadis (CNR), M. Giraud (IAS), A. Verschwele (DLO), I.J. Holb (UDCAS), L. Furlan (CNR), M. Sattin (CNR), and G. Urek (KIS)

On-farm experiments were designed to validate integrated weed management (IWM) and integrated pest management (IPM) tools in real field conditions. Within Task 3.3b selected IWM and IPM solutions were tested in Italy, Germany, Slovenia, France and Hungary. In 2013 on-farm trials were conducted in which efficacy of various strategies of IWM and IPM involving Bt spraying (bio-insecticide using Bacillus thurigiensis var. Kurstaki) or Trichogramma release against the European corn borer (ECB) were compared to the conventional (CON) approach. In 2013, the protocols for on-farm trials were modified, therefore different IWM and IPM strategies were validated compared to 2011 and 2012 experiments. In contrast to first two years of IPM on-farm experimentation where Trichogramma was released for biological control of ECB in all countries, in 2013 in Italy, Slovenia and Hungary, spraying of Bt bio-insecticide was selected as a IPM strategy against ECB. France, however, continued with *Trichogramma* evaluation. Germany discontinued its IPM research of ECB control, due to the fact that Trichogramma release is already a standard practice in southern Germany. Specific IWM and IPM protocols are described in section 'Introduction and methodology' for each country. The detailed protocol followed for the assessments and the experimental design can be found in ANNEX.

Encouraging results on weed control by IWM strategy were reported from all participating countries. In Italy, IWM was as efficient as the conventional broadcast herbicide application, but achieved a more than 50% reduction of herbicide use. The obtained grain yield was only 0.3 t/ha lower compared to conventional approach, but this was partially compensated by herbicide savings. In Slovenia efficient weed control was obtained along maize rows using IWM band spraying approach. However, substantial weed infestation was observed at the end of the growing season between the rows. This was probably due to the mechanical treatment being performed too late due to wet conditions. Unfortunately, the effect of IWM on yield is inconclusive in Slovenia, due to severe drought in 2013. Results from Germany show that IWM was able to control the weed infestation within reasonable limits, thus the yield was only slightly and insignificantly lower than in the conventional approach. Purely mechanical weeding, however, did not produce satisfactory results, as the weed density increased after treatment thus significantly decreasing the yield. Here, as in Slovenia, due to adverse weather

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conditions, the mechanical treatment could not be timed and performed optimally. In Hungary the IWM-band application of herbicide plus harrowing seems to be the same efficient as the conventional broadcast application without mechanical weeding, obtaining even higher yields,  $\approx$ 40% reduction of herbicide use for IWM and economic benefits from compensation from herbicide savings. In France, the IWM results are inconclusive regarding their effect on weed density or weed biomass, however in both trials performed in France, IWM increased yield.

Two different IPM approaches were tested against ECB in 2013. In Italy, Slovenia and Hungary *Bacillus thuringhiensis* (Bt) spraying was evaluated as a potential strategy to control ECB. France further evaluated the *Trichogramma* egg parasitoid. Germany discontinued IPM studies of ECB control in maize, since *Trichogramma* use is already a standard method for biological control of ECB in southern Germany.

Italy evaluated the Bt spraying as a single or double application in 2013. Results showed the same efficacy compared to the conventional insecticide since the number of plants above and especially below ear (i.e. causing most of yield loss) did not differ significantly from conventional approach, whereas grain yields were even slightly higher than the conventional approach. Italy additionally evaluated the necessity of using soil insecticides. The results from 2013 indicate that soil insecticide use could be avoided. Results from Slovenia were encouraging. Most of the parameters conveying ECB-related damage were improved by Bt spraying (e.g. parameters plants without cob, plants broken above or below ear, plants with any ECB damage, ECB damage on ears), however this improvement was not statistically significant, most probably due to high data variability. The parameter Fusarium damage on ears, however, was significantly reduced by Bt-spraying. Only parameter percent G1 ECB damaged plants was insignificantly higher on IPM plots. Unfortunately, severe drought resulted in extremely low yields in Slovenia in 2013. In both locations yield on IPM plots was slightly lower than the CON approach. In Hungary, results evaluating the Bt spraying showed significantly lower ECB damage on plants and ears, and higher yields compared to the untreated conventional approach; however costs were higher in this IPM strategy.

France reported that *Trichogramma* release increased yield in both trials performed in 2013. This was most probably by a significant increase of ECB parasitation (54,8 % parasitation increase in Alixan and 54,3 % in Montoison, compared to plots without parasitoid release).



## 7. Task 3.3b: Individual reports for on-farm experiments 2013

## **Results from on-farm experiments in Italy 2013**

Reported by: V.P. Vasileiadis, L. Furlan, M. Sattin (CNR)

### Introduction and methodology

This report is aiming at presenting the first results obtained during 2013 from on-farm experiments (4 trials) conducted in Italy studying the efficacy of integrated weed management (IWM) strategies and of Bt spraying against ECB. In the same study, the effect of soil insecticide against no insecticide treatments was also evaluated to see if soil insecticides could be omitted. Experiments will be repeated in 2014 to achieve replication.

The IPM solutions tested against weeds and ECB were as follows:

- IWM: Pre-emergence herbicide application in band and mechanical weeding with combined rotary tiller with ridging (photo below) or hoeing (less CO<sub>2</sub>) depending on weed density, species and growth stage. Details of the IWM and CON strategies are given in Table 1.
- IPM: Bt spraying when monitoring with light traps and scouting in the field indicates. Details of the IPM and CON strategies are given in Table 2.

TABLE 1.	Convei	ntional weed mana	agement	Integrated weed mar	agement
Farm	Pre- emergence herbicide	Post-emergence herbicide	Mechanical control	Pre-emergence herbicide in band-application (35 cm)	Mechanical control
Diana, IT	LUMAX 4.5 I/ha	Equip 2.5 I/ha+Mondak 1 I/ha	Hoeing	LUMAX 2.1 l/ha	Hoeing
Sasse, IT	LUMAX 4.5 I/ha	NO	Hoeing	LUMAX 2.1 l/ha	Hoeing
Berra, IT	LUMAX 4 I/ha	NO	Hoeing	LUMAX 2 l/ha	Ridging+ rotary tillage
Ravenna, IT	LUMAX 4 I/ha	NO	Rotary tillage	LUMAX 2 l/ha	Ridging+ rotary tillage

Table 1 Details of the IWM and CON strategies in Italy in 2013.



Farm	CONVENTIONAL	IPM
Vallevecchia, IT	KARATE ZEON (lambda-cyhalothrin 10%) 0.2 l/ha	1 & 2 applications of BIOBIT (Bt 6.4%) 1 kg/ha
Diana, IT	KARATE ZEON 0.2 l/ha	1 & 2 applications of BIOBIT 1 kg/ha
Sasse, IT	KARATE ZEON 0.2 l/ha	1 & 2 applications of BIOBIT 1 kg/ha
Berra, IT	CORAGEN (chlorantraniliprole 18.4%) 0.125 l/ha	1 application of BIOBIT 1 kg/ha
Ravenna, IT	CORAGEN 0.125 l/ha	1 application of BIOBIT 1 kg/ha1 application of BIOBIT 1 kg/ha

**Table 2** Details of the IPM and CON strategies in Italy in 2013.

### Results on integrated weed management strategy tested

Overall low and variable weed densities between farms was noticed. Statistical analysis showed significantly lower weed densities along the maize row in the IWM plot compared to the CON plot and significantly higher between rows in the IWM plot compared to the CON, respectively (Figure 1). The same trend was found for weed coverage along and between rows, but with no significant difference between IWM and CON strategies (Figure 2A). Weed biomass was lower along and between maize rows in the IWM strategy compared to CON (although not significant, respectively) which indicates that the higher weed density and coverage between maize rows in IWM was consisted of later emerging weeds that suffered competition by the already established maize crop (Figure 2B). Statistical analysis showed no significant difference in grain yield between strategies (Figure 3), although a slightly lower yield was obtained in the IWM strategy (-0.3 t/ha).



**Fig. 1** Overall weed density along and between maize rows for conventional and IWM strategies (mean of all farms).





**Fig. 2** Overall weed coverage (A) and weed biomass (B) along and between maize rows for conventional and IWM strategies (mean of all farms).



Fig. 3 Grain yield as affected by conventional and IWM strategies (mean of all farms).

### **Results on IPM strategies tested against ECB**

One-way ANOVA showed no significant difference between strategies tested in terms of plants without cobs, broken plants below and above ear and plants with any ECB damage. Only in the case of plants with any ECB damage the Bt double application was almost significant compared to the other treatments (p < 0.07), however this assessment covers any type of damage on maize plants and most probable ECB damage from 1<sup>st</sup> generation caused this result (Figure 4A). Statistical analysis showed no significant difference in grain yield between strategies, whereas a slightly higher yield was achieved under the Bt single and Bt double application compared to the conventional insecticide (Figure 4B). ECB damage on ears was not significantly between strategies with overall low % of damage, whereas

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*Fusarium* spp. damage on ears was significantly higher for the Bt double application (visual scale 2 to 3; 1 to 3%) compared to Bt single and conventional insecticide (visual scale 1 to 2; 0-1%), but also in this case the % damage is low (Figure 5).



**Fig. 4** ECB damage on final maize stand (A) and grain yield (B) as affected by the different ECB management in Italy in 2013 (Conventional insecticide, mean of 5 farms; Bt double application, mean of 3 farms; Bt single application, mean of 5 farms).





### Results on soil insecticide applications against soil insects

Strips without soil insecticide had significantly higher maize stand but also higher plants with wireworm damage than where soil insecticide was applied (Force 7.5 kg/ha; tefluthrin 0.5%), although the latter difference was only of 3 damaged plants compared to the soil insecticide



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(Figure 6A). However, no significant difference was observed when treated and not treated with soil insecticide, showing that soil insecticide treatments could be avoided (Figure 6B).



**Fig. 6** Maize stand and plants damaged by wireworms (A) and grain yield (B) when with and without soil insecticide in 2013 (mean of 3 farms).

### **Conclusions: Italy**

- IWM-Band application seems to be as efficient as the conventional broadcast application. More than 50% reduction of herbicide use for was achieved in IWM. Overall, there was only 0.3 t/ha grain yield reduction compared to CON, which was partially compensated from herbicide savings.
- Results in 2013 evaluating the Bt spraying as single or double application showed the same efficacy compared to the conventional insecticide since the broken plants above and especially below ear (i.e. causing most of yield loss) were not significantly different, whereas grain yields were even slightly higher than the conventional approach.
- Results from 2013 evaluating the use of soil insecticide against soil insects indicate that soil insecticide use could be avoided.


# **Results from on-farm experiments in Germany 2013**

Reported by: Verschwele Arnd (JKI)

# Introduction and methodology

In Germany 2 on-farm trials were conducted in the same area as in 2011 and 2012. The trial design has been changed in contrast to 2011 and 2012: Since the use of *Trichogramma* is a standard method for biological control of ECB for many years, it has been decided to do no further studies in terms of ECB and other pests in maize.

We tested three different weed control treatments:

- a) Conventional (broadcast spraying of herbicide mixture post-emergence, one application)
- b) Integrated: Band spraying combined with hoeing
- c) Mechanical: weed control by repeated harrowing and hoeing

# Results on integrated weed management strategy tested

Due to unfavourable weather conditions the control of mechanical weed control was poor at both sites. Periods with long and heavy rain fall inhibits the correct timing of mechanical treatments. Thus weed control was inefficient in 2013 (Table 3). As expected the high density and biomass of residual weeds reduced the maize yield (Table 4).

site	treatment	before treatment	after treatment
10	conventional	53	1
10	integrated	99	34
10	mechanical	56	61
40	conventional	105	37
40	integrated	103	52
40	mechanical	30	44

Table 3 Weed density before and after treatment at site 10 and site 40, Herbolzheim, 2013.

 Table 4 Maize yield and final weed biomass at site 10 and site 40, Herbolzheim, 2013.

site	treatment	yield (t/ha)	weeds (g/m <sup>2</sup> )
10	conventional	11.24	0.17
10	integrated	11.05	11.74
10	mechanical	7.42	373.83
40	conventional	7.15	0.26
40	integrated	6.84	74.21
40	mechanical	5.20	267.85



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However, the newly tested harrowing equipment developed by the company Treffler (Figure 7) shows high efficacy as long as soil and weather conditions are fine. Additionally the ridging which has been done in combination with the last hoeing was also able to control small weeds by burying them. However, due to late emergence especially *Chenopodium album* was the most serious weed species.

The trials will be repeated in 2014 in the same way and on the same farm.



Fig. 7 Newly developed and test harrow , Treffler Maschinenbau, Germany.

# **Conclusions: Germany**

- The use of *Trichogramma* is a standard method for biological control of ECB for many years in southern Germany. Therefore it was decided to do no further studies in terms of ECB and other pests in maize.
- The conventional approach for controlling weeds was more effective at controlling weed density than integrated approach. Mechanical weeding did not produce satisfactory results, as the weed density increased after treatment.
- Conventional weed management resulted in highest grain yield. Integrated approach produced slightly smaller yields. The weed control by mechanical treatments was inefficient in 2013. Due to high density and biomass of residual weeds the maize yield was severely reduced.



# **Results from on-farm experiments in Slovenia 2013**

Reported by: Robert Leskovšek, Jaka Razinger and Gregor Urek (KIS)

# Introduction and methodology

Two on-farm trials were conducted in Slovenia 2013, where efficacy of strategies of integrated weed management (IWM) and integrated pest management (IPM) involving Bt spraying against the European corn borer (ECB) were compared to the conventional (CON) approach. In 2013, the protocol for on-farm trials was modified, therefore different IPM strategies were validated compared to 2011 and 2012. In contrast to first two years of on-farm experimentation where *Trichogramma* was released for biological control of ECB, for 2013 spraying of Bt bio-insecticide was selected as a IPM strategy against ECB.

The following IPM solutions for weed and pest control were tested and compared with the conventional strategy:

- IWM strategy was band spraying followed by hoeing. Conventional wed management consisted of early post-emergence broadcast herbicide application and no application of insecticides. A description of IWM and CON solutions in given in Table 5.
- IPM strategy tested was Bt spraying based on light trap monitoring and field scouting of ECB. The methodological details are given below.

	Conventional weed management	Integrated weed management			
	Early post broadcast application	Early post band (30	Mechanical		
Location		cm) application	control		
SLO1	Lumax + Kelvin (3.5 L/ha + 0.3L/ha)	1.4 L/ha	Hoeing		
SLO2	Lumax + Kelvin (3.5 L/ha + 0.3L/ha)	1.4 L/ha	Hoeing		

**Table 5** A description of IWM and CON solutions in Slovenia in 2013.

# IPM methodological details tested in Slovenia in 2013:

Two sprayings using *Bacillus thuringiensis* subsp. kurstaki strain ABTS 351 from Valent BioSciences were performed on 29.7.2013 and 5.8.2013. The spraying protocol was as follows:

- working pressure: 3 bar
- spraying speed: 3 km/h
- water application: 600 l/ha
- dose: 1 kg/ha in 600 L of water



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It must be stressed that the Mechanization department of KIS developed especially for PURE a 17-row sprayer (Figure 8).



Fig. 8 A sprayer developed by KIS especially for PURE Task 3.3.b.

Due to the lack of suitable high clearance machinery in Slovenia, such spraying resulted in a 23% loss of grain.

# Results of field trials of integrated weed management (IWM) solutions

In 2013, high initial weed infestation was observed on both SLO 1 and SLO 2 locations. Early post broadcast herbicide application provided excellent weed control on CON plots on both SLO 1 and SLO 2 locations; very low weed densities were observed after the treatment. On IPM plots, weed control was sufficient along the row (band herbicide application) however, very poor weed control was observed after mechanical treatment (hoeing) on IPM plots between the rows on both locations (Figure 9, Figure 10).



Fig. 9 Weed density and weed control efficacy on location SLO 1.



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Fig. 10 Weed density and weed control efficacy on location SLO 2.

Insufficient weed control resulted in high final weed densities on IPM plots between the rows on both locations.

A similar trend was found for weed coverage and final weed dry matter. Weed coverage and final weed dry matter were low on CON plots, however they were higher between the rows on IPM plots (data not shown).

Severe drought resulted in extremely low yields in 2013. On SLO 1 location, maize yield on IPM plot was higher compared to CON plot, however lower on the SLO 2 location (Figure 11).



Fig. 11 Maize yields in 2013 in Slovenia.

# Results of field trials of integrated pest management (IPM) solutions

The final maize stand was approximately 100 plants per 20 m. Approximately 70-75% plants suffered ECB damage. No statistical differences were observed between IPM and CON (Figure 12-left). High data variation was observed in the following parameters: Total egg masses, Plants without cob, Broken plants above and below ear, and Plants with any ECB



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damage. We observed numerical damage reduction on IPM plots, but no stat. differences were calculated between IPM and CON plots (Figure 12-right).



**Fig. 12** Left – Final maize stand and percentage of plants suffering 1<sup>st</sup> generation ECB attack. Right – various ECB-related parameters observed prior to harvest.

The only parameter where a significant reduction of indirect ECB damage was observed was parameter *Fusarium* presence on ears (Figure 13).



Fig. 13 ECB damage on ears and *Fusarium* presence on ears dependent on the strategy of ECB control.

#### **Conclusions: Slovenia**

• Efficient weed control (low final weed coverage and weed dry matter on CON plots and along the rows on the IPM plots) was obtained using IWM approach. Substantial weed infestation at the end of the growing season between the rows on IPM plots was observed (mechanical treatment was performed too late due to wet conditions).



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- The IPM tool resulted in a numerical increase of some ECB related parameters (e.g. % ECB damaged plants) and a numerical decrease of others (e.g. Plants without cob, Plants with any ECB damage), however only parameter *Fusarium* damage on ears was significantly reduced by Bt-spraying.
- Severe drought resulted in extremely low yields in 2013. On SLO 1 location, maize yield on IWM plot was higher compared to CON plot, however lower on the SLO 2 location. In both locations yield on IPM plots was lower than the CON approach.



# **Results from on-farm experiments in France 2013**

Reported by: Marion Giraud (Invivo Agrosolutions - IAS)

# Introduction and methodology

Trials were made in the Drôme area (in the surrounding of Valence in the Rhône Valley, France) on two locations (Montoison, plot size 2.5 ha, variety A46 pioneer, seeding on 18/04/13 and Alixan, plot size 4 ha, variety MAS 52C, seeding on 23/04/2013). The IWM protocol that was adopted in France in 2013 is given in Table 6.

	-	
	CONTROL	IPM WEED
	(Plot A)	(Plot B)
MONTOISON	04/2013 : Spraying	26/05/2013 : hoeing
ALIXAN	24/04/2013: broadcast herbicide	30/05/2013 : hoeing
	application	10/06/2013 : herbicide band spraying + hoeing

**Table 6** IWM protocol followed in France in 2013.

Regarding the IPM protocol, the following tasks were performed:

- ECB flight was monitored using light traps.
- Adult click beetles were monitored Montoison.
- Trichogramma parasitism rate was evaluated.
- ECB pressure was evaluated.
- At harvest damages on cob, grain yield and grain mycotoxin concentrations were determined.

# Results of field trials of integrated weed management (IWM) solutions

In Montoison the weed coverage was higher on IWM plots throughout the season. The opposite was true for Alixan. Weed density was higher in IWM plots in Montoison in June evaluation but lower at harvest. Weed biomass was lower on IWM plots in Alixan, both along and between rows. In Montoison, it was much higher along the rows on IWM plots. The IWM results are given in Table 7 and graphically represented in Figure 14.



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			25/04/13		12/06/13		04/09/13		
			Weed	Weed	Weed	Weed	Weed	Weed	Weed
			coverage	density	coverage	density	coverage	density	biomass
MONTOISON	CONV	Along	0.0	0.0	4.6	2.1	17.3	3.3	2.5
		Between	0.0	0.0	0.7	2.0	21.3	5.3	2.3
	IPM WEED	Along	0.0	0.0	19.7	3.4	50.0	4.9	46.7
		Between	0.0	0.0	1.7	3.0	55.3	1.9	0.6
ALIXAN	CONV	Along	0.0	0.0	4.2	14.5	44.7	8.0	6.0
		Between	0.0	0.0	6.3	18.1	71.0	10.5	14.8
	IPM WEED	Along	0.0	0.0	4.5	15.4	31.7	6.2	2.3
		Between	0.0	0.0	0.7	1.7	61.0	12.3	4.2

#### **Table 7** IWM results in France in 2013.



Fig. 14 IWM results in France in 2013.

# Results of field trials of integrated pest management (IPM) solutions

Second generation ECB pressure evaluated indirectly by counting egg masses was lower in Montoison. *Trichogramma* release increased parasitation rate by 57.8 % Alixan and by 54.3 % in Montoison. *Trichogramma* release date and parasitism rate as well as evaluation of second generation ECB pressure is shown in Figure 15.





Fig. 15 Trichogramma release and parasitism rate and evaluation of second generation ECB pressure.

Due to high data variability, no significant effects were observed at harvest regarding ECB damages and *Fusarium* presence on cobs (Figure 16).





Harvest of each plot could not have been done with a combine. Our plots were not designed to be harvested with a commercial combine at the settlement of the trials. However, we estimated difference between grain yield between the plots A. B and C by sampling 100 cobs on each. Grains were separated from cobs and weighted. The yield on the entire field was calculated from this data. In both trials, IWM and IPM increased yield (Table 8).

Note: Mycotoxin analyses are implemented by INVIVO LABS, 56 ST NOLFF. Analyses are still running at the time of preparation of this report.



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Table 8 Grain yields obtained in France with conventional (CONV). IWM and IPM approach in 2013.

\*Given by farmer.

#### **Conclusions: France**

- IWM results are inconclusive regarding their effect on weed density or weed biomass. •
- Trichogramma release increased parasitation rate by 54.8 % Alixan and by 54.3 % in • Montoison.
- In both trials. IWM and IPM increased yield. •



# **Results from on-farm experiments in Hungary 2013**

Reported by: Imre Holb (UDCAS)

# Introduction and methodology

This report is aiming at presenting the first results obtained during 2013 from on-farm experiments (4 trials) conducted in Hungary studying the efficacy of integrated weed management (IWM) strategies and of Bt spraying against ECB. Experiments will be repeated in 2014 to achieve replication. On-farm experiments were done in four locations (Debrecen1-4) on 5000 m<sup>2</sup> plots per system replicate. Assessments were done in each location on maize density, weather conditions, weed density, weed species, ECB assessments, diseases (*Fusarium*) and yield according to the common protocol agreed. The following IPM solutions for weed and pest control were tested and compared with the conventional strategy in all farms:

- IWM strategy tested was post-emergence herbicide in band spraying (Actual use per hectare due to reduced area treated: Principal plus + Successor T; 300 g/ha + 0.6 l/ha) followed by hoeing against the conventional strategy that consisted only of post-emergence broadcast herbicide application (Principal plus + Successor T; 440 g/ha + 1 l/ha).
- IPM strategy tested was Bt spraying (DiPel ES (3.2 % *Bacillus thuringiensis* (var: kurstaki) at 2 l/ha) based on light trap monitoring and field scouting of ECB against no insecticide spraying as the conventional approach.

# Results of field trials of integrated weed management (IWM) solutions

Statistical analysis showed no significant difference between conventional and IWM strategies for weed density and coverage (Fig. 17), and final dry weed biomass (Fig. 18) when data from all farms were pooled together. Although not significant weed density was slightly lower in CON compared to IWM, whereas the opposite occurred for weed coverage and final dry weed biomass.





**Fig. 17** Overall weed density (A) and weed coverage (B) for conventional and IWM strategies (mean of all farms).



Fig. 18 Overall weed biomass for conventional and IWM strategies (mean of all farms).

#### Results of field trials of integrated pest management (IPM) solutions

One-way ANOVA showed no significant difference between strategies tested in terms of plants without cobs and broken plants below ear. Broken plants above ear were 0% for both strategies. In the case of plants with any ECB damage and ECB damage on ears the Bt spraying resulted in significantly lower damage compared to the untreated conventional approach (P<0.01 and P<0.05 respectively); however, it should be mentioned that % damage in both cases was rather low showing a low pest pressure. *Fusarium* spp. damage on ears was significantly not different between strategies (Fig 19).





**Fig. 19** ECB damage on final maize stand (A) and maize ears (B) as affected by the Bt spraying in comparison to the untreated conventional approach (mean of all farms).

### Yield comparison of IWM and IPM strategies against the conventional

Statistical analysis showed significant lower grain yield in CON strategy compared to the IWM and IPM strategy (P<0.01; Fig. 20).



Fig. 20 Grain yield as affected by conventional. IWM and IPM strategies (mean of all farms).

#### **Conclusions: Hungary**

IWM-Band application seems to be the same efficient as the conventional broadcast application, obtaining even higher yields in 2013, a  $\approx$ 40% reduction of herbicide use for IWM and economic benefits from compensation from herbicide savings.

Results in 2013 evaluating the Bt spraying showed significantly lower ECB damage on plants and ears, and higher yields compared to the untreated conventional approach; however costs are higher in this IPM strategy.



# 8. Task 3.4: Ex-post assessment of on-farm experiments 2013

Wim van Dijk (DLO)

# Introduction

For the implementation of IPM strategies in practice a correct evaluation of the tested strategies in experiments is important. In Pure an ex post and an ex ante assessment is done. The main objective of the ex post assessment is a comparative evaluation of the agronomical, environmental and economic sustainability of IPM solutions and tools identified and tested in the on station and on farm experiments.

The ex post assessment consists of a cost-benefit analysis and estimation of the environmental risks of the use of pesticides. The latter is done with the model SYNOPS. The current report is restricted to the cost-benefit analysis. The calculations with SYNOPS are yet to be done and will be reported in Deliverable 3.3

In this chapter the results of the cost benefit analysis of the IPM tools tested on the on farm experiments in 2013 will be discussed. The results of the cost benefit analysis of the on farm experiments of 2011 and 2012 are summarized in chapters 5 and 6 where an evaluation is given of the first two years on farm testing of IPM tools (2011-2012).

The ex post assessment of the tools tested in the on the station experiments cannot be done yet as crop rotation is part of the tested tools and the results of a complete rotation will only be available in 2014.

# Methodology

# **Experimental** sites

In 2013, 15 on farm experiments were conducted (Germany: 2, Italy: 5, France: 2, Slovenia: 2, Hungary: 4).

# Compared strategies

In the experiments the current conventional strategy (CON) is compared with an advanced strategy for weed control (IPM WEED) and an advanced strategy for ECB control (IPM ECB). Both IPM strategies were tested separately (ECB control in IPM WEED was according



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conventional practice and weed control in IPM ECB according to conventional practice, respectively).

In all experiments the tested IPM WEED strategy consisted of a band spraying of herbicides combined with one or two hoeing operations. In Germany an additional strategy was tested in which the weed control was done completely mechanical by a combination of harrowing and hoeing treatments. In all experiments the conventional weed control consisted of a broadcast spraying of herbicides.

In the IPM ECB strategy the ECB control was done biologically either by a spraying with *Bacillus thuringiensis* (Italy, Slovenia and Hungary) or by release of Trichogramma (France). In Italy in the CON treatment a pesticide spraying was done, in Slovenia, France and Hungary no treatment was done. In Germany no IPM ECB tool was tested as ECB control with Trichogramma is already part of the conventional strategy. Instead, an additional IPM WEED strategy was tested (see above).

More detailed information with regard to the experiments is given in chapter 6.

# Cost benefit analysis

The basis of the cost benefit analysis is the gross margin (= financial yield (yield  $\times$  price) – variable costs). Total variable costs include the costs of inputs (e.g. seeds, fertilizers, pesticides, biological agents) and costs of operations (e.g. mechanical weed control, labour, fuel).

The cost benefit analysis is based on the registration of the inputs, operations and yields of the experiments. For this purpose, a template was developed and provided to all partners involved in the on farm experiments for data collection of the crop management. For costs of inputs, the prices that farmers paid were used, whereas the operation costs were based on contract work prices including costs for labour, machinery and fuel, provided by regional contract work companies. Average grain maize prices for 2013 were derived from the Eurostat database.

# **Results and discussion**

In this section the results of the cost benefit calculations for both the WEED and ECB control are discussed. In the presentation we restrict to the <u>difference</u> in costs and gross margin between the IPM and CON strategy. Furthermore, the results of the separate experiments within the different countries are averaged.



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# Weed control

# Costs

Averaged over all countries the IPM strategy based on a combination of band spraying and hoeing resulted in a small decrease of variable costs (- $\epsilon$ 5/ha) ranging from - $\epsilon$ 50/ha in Italy to + $\epsilon$ 15/ha in Germany (Figure 1, left). The increased costs for operations (band instead of broadcast spraying, hoeing) were not always compensated by reduced costs of herbicides (Figure 1, right). In Italy total costs decrease was relatively high (- $\epsilon$ 50/ha). This is due to the fact that hoeing was also done in the CON strategy in order to incorporate the urea fertiliser. Additionally, in some Italian experiments, in the CON strategy the broadcast spraying was done separately from the drilling while the band spraying was done in combination with the drilling reducing the application costs.

In Germany an additional strategy based on a completely mechanical weed (GE2) control was tested. Total variable cost increased with  $\notin$ 45/ha. Compared with the GE1 strategy (band spraying + hoeing) variable costs were about  $\notin$ 20/ha higher due to costs for extra harrow operations.



**Fig. 1** <u>Difference</u> in total variable costs (left) and costs for inputs and operations (right) between the IPM WEED strategy and the CON strategy of the on farm experiments 2013 (a positive value means an increase in costs of the IPM strategy compared to the CON strategy). Figures refer to averaged values of all experiments within countries (GE1, IT, FR, SLO, HU: IPM = band spraying + hoeing, GE2: IPM = harrowing + hoeing).



Yields

In Figure 2 the grain yields of both the CON and the IPM WEED strategy are plotted for all separate experiments. For the IPM strategy band spraying + hoeing no significant effects on grain yield were observed (all points are relatively close to the y=x line). However, the completely mechanical weed control strategy reduced yields significantly (-25% to -35%).



**Fig. 2** Grain yield (ton/ha) of the CON and IPM WEED strategy of the separate on farm experiments in 2013.

# Gross Margin

The difference in gross margin (financial yield minus total variable costs) between the CON and IPM WEED strategy is given in Figure 3 (right). For comparison also total variable costs are given (Figure 3, left).

Averaged over all countries the effect of the IPM WEED strategy band spraying + hoeing on the gross margin was small (- $\epsilon$ 5/ha), ranging from - $\epsilon$ 200/ha in Slovenia to + $\epsilon$ 130/ha in Hungary. Due to the large negative yield effect (see 3.1.2) the gross margin of the completely mechanical strategy in Germany (GE2) was significantly lower compared with the CON strategy (- $\epsilon$ 600/ha).

Figure 3 makes clear that for the IPM strategy band spraying + hoeing, the variation in gross margin is larger than the variation in variable cost. Relatively small yield effects, although not significant, can have a relatively strong effect on the gross margin.



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**Fig. 3** <u>Difference</u> in variable costs (left) and gross margin (right) between the IPM WEED strategy and the CON strategy of the on farm experiments 2013 (a positive value means an increase in costs or gross margin of the IPM strategy compared to the CON strategy). Figures refer to averaged values of all experiments within countries (GE1, IT, FR, SLO, HU: IPM = band spraying + hoeing, GE2: IPM = harrowing + hoeing).

### Conclusion

Averaged over all countries the costs of the IPM strategy based on an band spraying combined with hoeing were comparable with the CON strategy. Between countries the difference in costs between the IPM and CON strategy ranged from - $\varepsilon$ 50/ha to + $\varepsilon$ 15/ha. No significant yield effect of the IPM strategy was observed.

# ECB control

As mentioned before except for France the IPM ECB control was done with a spraying with *Bacillus thuringiensis* (BT). In Italy there were plots with one and two sprayings. In Slovenia always two sprayings were done, in Hungary always one spraying.

#### Costs

Only in Italy a pesticide spraying was done for ECB control in the CON strategy. For the IPM ECB strategy with one BT-spraying, costs were comparable with the CON strategy. With two BT-sprayings costs increased with  $\notin$ 80/ha due to extra costs for the BT-product and the spraying operation. In Slovenia and Hungary where no treatment was done in the CON strategy, as expected the costs increased (+ $\notin$ 140/ha and + $\notin$ 85/ha respectively). The stronger cost increase in Slovenia compared to Hungary was due to the extra BT-spraying (2 sprayings in Slovenia and 1 spraying in Hungary).



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The release of Trichogramma in the French experiments increased the costs with €45/ha. This is mainly due to the costs of the Trichogramma product. Like Slovenia and Hungary, no treatment was done in the CON strategy.



Fig. 4 <u>Difference</u> in total variable costs (left) and costs for inputs and operations (right) between the IPM ECB strategy and the CON strategy of the on farm experiments 2013 (a positive value means an increase in costs of the IPM strategy compared to the CON strategy). Figures refer to averaged values of all experiments within countries

# Yields

In Figure 5 the grain yields of both the CON and the IPM ECB strategy are plotted for all separate experiments. Overall no significant effects were observed between the CON and IPM ECB strategy. This also applies to the Italian plots were one and two BT-sprayings were compared. The grain yields of the IPM plots of the experiments in Hungary and that of one experiment in France seemed to be somewhat higher than the yield in the corresponding CON plots.



**Fig. 5** Grain yield (ton/ha) of the CON and IPM ECB strategy of the separate on farm experiments in 2013 (for Italy yields of the plots with one and two sprayings are given, IT-1spr and IT-2spr respectively).



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# Gross margin

Averaged over all countries the gross margin of the IPM ECB strategy was  $\notin 25$ /ha lower than that from the CON strategy. Differences between the two strategies ranged from - $\notin 200$ /ha (Slovenia) to + $\notin 115$ /ha (France) (Figure 6). In the French and Hungarian experiments the higher costs of the IPM ECB strategy are compensated by a higher financial yield.



**Fig. 6** <u>Difference</u> in variable costs (left) and gross margin (right) between the IPM ECB strategy and the CON strategy of the on farm experiments 2013 (a positive value means an increase in costs or gross margin of the IPM strategy compared to the CON strategy). Figures refer to averaged values of all experiments within countries.

# Conclusion

In situations where in the CON strategy a pesticide spraying is done, the costs are comparable with the IPM strategy when based on one BT-spraying. If two BT sprayings are done, the costs are  $\in$ 80/ha higher. In situations where no ECB treatment is done in the CON strategy, the higher costs of the IPM strategy ( $\notin$ 45-140/ha) are not always compensated by higher yields.



# 9. Conclusions

#### IPM strategies tested on-station in France and The Netherlands

An overall evaluation of the crop protection strategies tested in maize 2012-2013 on-station in France indicate that the yields of conventional, IPM1 (advanced) and IPM2 (innovative) systems are not significantly different with a good control against weeds for IPM1 that had pre-emergence herbicide application and post-emergence in band application followed by 2 hoeing, and slightly higher densities for IPM2 where only pre-emergence herbicide in band application and three hoeing treatments were used. Both IPM-based strategies tested had a lower pesticide dependence with 30% reduction of herbicides applied/ha (band application) in IPM1, whereas IPM2 was the strategy with the less pesticide input (only pre-emergence in band application, 1/3 of area treated) and therefore with the minimum environmental impact. Damage and pressure from all other pests was insignificant in these 2 years even though the field was historically concerned by high level of wireworms.

In the on-station experiment in the Netherlands, results from the different reduced tillage systems tested against ploughing with chemical or mechanical weed control during 2011-2013 showed an increase in weed numbers, though only significantly different at conservation tillage with chemical control and at no-till with mechanical control. Final maize stand was similar for all treatments except for ridge-tillage with mechanical weed control that had significantly lower stand possibly due to the mechanical control as the ridge-till with chemical control had a good maize stand. The conventional tillage with chemical control resulted in significantly higher yield compared to all other tillage systems with chemical or mechanical control. These yield reductions in the reduced tillage systems ranged from c. 10% to over 50%. In this long-term experiment the sustainability of the strategies tested will be better determined at the end of the project after 4 years of replication.

#### IPM tools tested on-farm 2011-2012

The following IPM solutions against weeds and ECB were tested in 2011-2012 in the different countries and evaluated for their sustainability as listed below:

1. In Italy (five trials), where pre-emergence herbicide was not applied in IPM-weed plots, in 2011 in four out of the five trials scouting and the use of predictive models indicated no early post-emergence herbicide application, thus only hoeing was practiced. In 2012, the



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contrary happened and four out of five trials were treated. Compared to the conventional strategy, this IPM tool resulted in:

- overall good weed control,
- lower environmental impact (i.e. no residual pre-emergence herbicide and overall reduction of herbicide treatments; only five out of ten farms in total were treated during the two years),
- lower total costs (-82 €/ha),
- lower gross margin (-50 €/ha) due to slightly lower yields than the CON (non-significant difference).
- 2. In Slovenia (two trials). the false seedbed plus harrowing at 2-3 maize leaves and low dose of post-emergence herbicide applied in IPM-weed plots resulted in:
  - Overall good weed control as in the conventional strategy,
  - lower environmental impact (50% reduction of herbicides applied),
  - increased total costs (+6 €/ha),
  - increased gross margin (+59 €/ha).
- 3. In Germany (two trials). the hoeing combined with band-spraying of post-emergence herbicide in IPM plots resulted in:
  - partial control (good weed control in 2012 but high final densities in 2011 due not good control of *Chenopodium polyspermum*).
  - lower environmental impact (60% reduction of area sprayed with herbicides and consequently lower herbicide usage per hectare),
  - increased costs (+32 €/ha),
  - decreased the gross margin (-105 €/ha).
- 4. In Hungary (four trials), early post-emergence herbicide in band application plus hoeing (when urea is applied) in IPM-weed plots (only 2011 data since no machinery was available to apply the band-spraying in the IWM plots in 2012 and broadcast spraying was applied in both CON and IWM plots) resulted in:
  - no clear efficacy of the weed control strategies because of low initial and final weed densities observed for CON (10.2 and 11.6 plants m<sup>-2</sup>, respectively) and IWM strategies (23.9 and 19.4 plants m<sup>-2</sup>),



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- lower environmental impact (60% reduction of area sprayed with herbicides and consequently lower herbicide usage per hectare),
- lower total costs (-24 €/ha),
- increased gross margin (+61 €/ha).
- 5. Biological control with Trichogramma brassicae releases against ECB provided:
  - similar efficacy as the insecticide treatments in Italy, Germany and Hungary, and higher (i.e. although not significant) to conventional plots without insecticide in France and Slovenia,
  - preserved mycotoxin levels below the EU threshold for maize destined for human consumption,
  - maintained yield levels in similar or even better levels where insecticide was not applied,
  - its implementation was economically sustainable in France and in Italy (gross margin of +83 €/ha and +6 €/ha, respectively) but not sustainable in all other countries mainly due to high costs of the Trichogramma product.

# IPM tools tested on-farm 2013

Results coming from the new set of IPM tools (new IWM tools, Bt spraying against ECB and Trichogramma in France) tested in 2013 that will be repeated in 2014 are promising in terms of agronomic and economic relevance.

- 6. In Italy, where pre-emergence herbicide was applied as band-spraying and followed by hoeing in IPM-weed plots compared to broadcast herbicide application, resulted in:
  - overall good weed control,
  - lower environmental impact (>50% reduction of area sprayed with herbicides and consequently lower herbicide usage per hectare),
  - lower total costs (≈-50 €/ha),
  - lower gross margin (≈-30 €/ha) due to slightly lower yields than the CON (non-significant difference).
- 7. In Slovenia, where early post-emergence herbicide in band application followed by hoeing was tested against the broadcast early-post herbicide application, resulted in:

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- overall good weed control along maize lines but not between rows due to mechanical treatment being performed too late due to wet conditions,
- lower environmental impact (>50% reduction of area sprayed with herbicides and consequently lower herbicide usage per hectare),
- lower total costs (≈-18 €/ha),
- lower gross margin (≈-200 €/ha) due to lower yields than the CON; however the effect of IWM on yield is inconclusive in Slovenia, due to severe drought in 2013
- 8. In Germany, where band spraying combined with hoeing and only mechanical weeding (i.e. weed control by repeated harrowing and hoeing) was tested against the conventional (broadcast spraying of herbicide mixture post-emergence, one application) showed that:
  - IWM was able to control the weed infestation within reasonable limits, thus the yield was slightly and insignificantly lower than in the conventional approach. Costs increased (+20 €/ha) and gross margin was lower (-45 €/ha). Lower environmental impact (60% reduction of area sprayed with herbicides and consequently lower herbicide usage per hectare).
  - Mechanical weed control, did not produce satisfactory results, as the weed density increased after treatment thus significantly decreasing the yield (i.e. due to adverse weather conditions, the mechanical treatment could not be timed and performed optimally). Costs increased (+45 €/ha) and gross margin was lower (-600 €/ha).
- 9. In Hungary, the IWM-band application of herbicide plus harrowing against the conventional broadcast spraying provided:
  - good weed control,
  - obtained higher yields,
  - ≈40% reduction of herbicide use for IWM and economic benefits from compensation from herbicide savings,
  - higher costs due to the mechanical weeding that is not practiced in the conventional; however partly compensated by the herbicide savings, (+8 €/ha),
  - higher gross margin (+130 €/ha).



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- 10. In France, where only mechanical weeding was tested in one farm and hoeing followed by band application of post-emergence combined with hoeing (IWM) in the other farm resulted in:
  - both strategies had good control
  - higher yields were obtained in both cases compared to the conventional strategy with the IWM yielding 1 t/ha more and the mechanical weeding 0.1 t/ha.
  - averaged costs of these 2 strategies were slightly higher
  - averaged gross margin was higher ( $\approx +110 \notin$ /ha).
- 11. *Bacillus thuringhiensis* (Bt) spraying evaluated in Italy (1 and 2 sprayings), Slovenia (2 sprayings) and Hungary (1 spraying), and Trichogramma releases in France as a potential strategy to control ECB resulted in:
  - the same efficacy compared to the conventional insecticide in Italy and control of ECB in Slovenia and Hungary, where no insecticide was applied in the conventional approach. Good ECB control by Trichogramma was also reported in France compared to the untreated conventional plot.
  - higher yields were obtained in Italy (non-significant) and Hungary (significant), whereas in Slovenia severe drought resulted in extremely low yields so the effect is not so clear. Higher yields were reported with Trichogramma releases in France.
  - higher costs were obtained in all countries due to the products and operations.
  - higher gross margins were obtained only in Hungary and France.



# **10. ANNEXES**

# Task 3.3b: On-farm experimentation 2013-2014

## **Experimental protocol**

Partners involved: KIS, CNR, JKI, UDCAS and Biotop

# **Objectives and perspectives:**

On-farm experiments will test/validate single IPM tools, or one-year solutions to specific problems or "small IPM packages" (e.g. specific weed problems, specific control tools or small IPM packages to manage ECB and/or soil insects) in real field conditions (ideally plots should be at least 5.000  $m^2$ ).

Tools and/or solutions to be tested will be chosen after discussion with the stakeholders groups (the first year will be a bit difficult) or they may arise from on-station experiments.

On-farm experiments will have to be managed with commercially available or technologically mature equipment which is suited for field scale applications.

**Experimental area** (the minimum no. of farms (i.e. replicates) where an IPM tool is tested in each region is 2):

On-farm experiments will be carried out in southern conditions by CNR (5 trials per year) and Invivo (2 trials per year), in central conditions by JKI (2 trials per year - with input of expertise from DLO) and in eastern conditions by KIS (2 trials per year) and UDCAS (4 trials per year), for a total of 14 experiment per year. Replications will be achieved by involving several farms rather than replicating within farms. IPM solutions designed and tested on-station may have to be adapted to the specific farming conditions and constraints.

**Experimental facilities and equipment** - within a common experimental approach, all materials and methods will be locally chosen according to their relevance for IPM implementation (if applicable as many as possible (at least two) different locations with similar experimental design). The trials will run for two years: start 2013 – finish 2014.

Tools that will be assessed (at least 2 IPM tools (or 3 if possible) will be assessed):

- tools to control weeds (See Annex 6 for more details);
- tool to control ECB: Bt spraying against ECB in Italy, Hungary and Slovenia; *Trichogramma* against ECB in France
- tools to predict/control soil insects (see next page).

**Experimental design** – to assess the effectiveness of the proposed IPM solution three plots: A, B and C (each plot will be at least **5.000 m<sup>2</sup>**) will be established on each location (2 IPM tools + conventional should be tested in the same farm) – the plots will be arranged on the same field (the same maize cultivar and the same technology carried out in previous years). The chosen field will be divided on the 3 plots: A, B and C.

**On plot A** (control plot) conventional technology based on existing knowledge and tools (technology conducted according to local practices for maize production – technology that is actually used by farmers).



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On plot B IPM tools for weeds control will be validated (Annex 4: weed protocol):

- SL: Pre-emergence or early post emergence herbicide application in band + hoeing.
- HU: early-post emergence herbicide application in band + hoeing \_
- IT: Pre-emergence herbicide application in band + post-emergence combined rotary tiller with ridging (photo below) or hoeing (less CO2) depending on weed density, species and growth stage.
- DE: 1) early-post emergence herbicide application in band + hoeing and 2) mechanical control
- FR: early-post emergence herbicide application in band + hoeing at ALIXAN & 2-3 mechanical control at MONTOISON (no post-emergence herbicides)

# **On plot C**:

1) Trichogramma release against ECB will be tested in France (Annex 2: Trichogramma **protocol**) which should be situated at least 100 m from the plot A (control plot)

2) Bt spraying (Biobit 1 kg/ha) against ECB in Italy, Hungary and Slovenia (Annex 3: Bt protocol) when the right timing will be assessed based on light traps captures and plant assessement (specific details later). Plots (A+C) in this case will be close to reduce soil variability.

**On plot C** (and A, B where possible) – maize will be sown on alternate strips (with 4-6 rows) treated with soil insecticide (or seed dressing) and 4-6 rows untreated in order to assess the reliability of soil insect alert programme and the actual effect of soil insecticides (Fig. 1). Soil insects will be followed as proposed in **Annex 1**. In regions where soil insecticide application is not permitted or usually not done by farmers all plots will be kept untreated and stand and soil insect damage will be assessed according to Annex 1.

Figure 1: soil insecticide evaluation layout.





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**Experimental conditions** (weather, temperature, precipitations) as well as all other critical parameters (crop variety or hybrid; soil management: water, fertility; planting date; seeding rate/plant population; row spacing; chemical use) will be followed on each location.

**Methodology to measure the effectiveness** of the chosen IPM solution is based on the detection/occurence of the target pests as well as on visual inspection of the damage that is caused by the target pest. Yield of maize will also be measured (**Annex 5**).

Therefore the weed species composition will be determined as proposed in Annex 4.

Additionally the targeted pest species (ECB) will be monitored on each location/plot using light traps (as agreed). Visual inspection to investigate the damage caused by targeted pest will be also conducted (i.e. number of maize stalks infested by ECB, ECB damage to crop ears, yield of maize will also be observed). Materials for monitoring have to be standardised and used by all. If feasible the same materials will be used as in the on-station experiments.

<u>Evaluation of ECB at harvest:</u> total plants; plants without ears; % of attacked ears with a notation of damage by a class system (1-7 levels), and in the same time a notation of the *Fusarium* development also with a class system; number and the position of ECB larvae (in stalks, peduncles or ears); plants broken above ear; plants broken below ear (Annex 2).

### **Demonstation activities**

Field days and demonstration activities will be organized in each participant country (IT, GER, SLO, HUN, NL) on at least one location.



# **ANNEX 1:** Soil insect pressure - Wireworm monitoring

# LARVAE

This will be done in September - October and/or March - April before the swarming period, when soil temperatures above 10°C.

- Bait traps: 6 to 12 bait traps will be placed in each plot according to plot size, provided the soil is bare (traps will only work properly if there is no/low presence of CO<sub>2</sub>-producing roots). Each trap will be made and used according to the description given by Chabert and Blot (1992) a modified version of traps described by Kirfman *et al.* (1986). These comprise a plastic pot 10 cm in diameter provided with holes in the bottom; the pots are filled with vermiculite, 30 ml of wheat seeds and 30 ml of corn (maize) seeds. The pots will be wetted before being placed into the soil just below the surface and covered with an 18 cm diameter plastic lid placed a few cm above the rim of the pot.
- 2. Traps will be checked by hand-sorting the contents after 10 15 day. Count and record the number of larvae found. The manually observed material will be put on Tullgren funnels and processed as described for soil cores. Place all larvae in airtight vials with a little of humid soil, and send to: Dr Lorenzo Furlan, via Q. Sella 12, 30027, San Donà di Piave VE, ITALY, for identification.



• = trap position in the plot (20-40m apart and 10m between)

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Kirfman, G.W., Keaster, A.J. & Story, R.N. 1986. An improved wireworm (Coleoptera: Elateridae) sampling technique for midwest cornfields. *Journal of the Kansas Entomological Society*, **59**, 37-41.

# **MONITORING OF ADULTS**

Use the YATLORf traps with deep bottom if it is going to be used also for the monitoring of Diabrotica adults; baited with the sex pheromones of the various species, products can be supplied by the Plant Protection Institute of Budapest, and place inside a dispenser Kartel 730. The YF trap has to place just above the ground, with the lower rim of the brown trap body, 2-3 cm below the soil level (the deep bottom completely inside the soil).



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# The timing for management of the traps is as follows:

- 1 On **20th March** the trap will be placed, for convenience use an indicator for the place where the trap is, in the centre of the monitoring area with the sex pheromone bait for *A. brevis* in a **low** position with **the top facing below**; (or *A. sputator* in other region, see table 1)
- 2 On **10th April** the captured insects will be taken off<sup>b</sup> and the dispenser with the pheromone for *A. sordidus/rufipalpis (Hungary)* will be added in a **medium** position and with **the top facing below**.
- 3 On **10th May** ca. the captured insects will be at the edge of a field <sup>b</sup> and the pheromone bait<sup>a</sup> for *A. sordidus* (at ca. 30 days) will be substituted with a new one in a **medium** position and with **the top facing below**, but also the bait for *A. litigiosus* will be added in a high position only in Italy.
- 4 On **10th June** ca. the captured insects will be taken off<sup>b</sup> and the bait<sup>a</sup> for *A. brevis* will be substituted with the one for *A. litigiosus* (only Italy) in a **low** position and with **the top facing below;** substitute the bait for *A. litigiosus* in a high position with the bait for *A. ustulatus*; <u>in a high position the pheromone for Diabrotica can also be added; in this case add an insecticide strip at the bottom of the trap</u>.
- 5 On **10th July** ca. the captured insects will be taken off<sup>b</sup> and the bait<sup>a</sup> for *A. ustulatus* will be substituted and placed at the same position. Substitute also the pheromone for Diabrotica.
- 6 On **10th August** the captured insects will be taken off<sup>b</sup> and the trap will be substituted for following year.

# Example procedure; see table 1 for lure combination in each site.

In France, Germany, Slovenia, Hungary and The Netherlands a trap B baited with *A. obscurus* (in low position) and *A. lineatus* (in **medium** position) will be added in early April; the traps will be inspected with lure substitution every month until July (see Table 1).

<sup>a</sup> = capsule Kartel 730 for A. brevis, A. sordidus, A. litigiosus, A. ustulatus; A. lineatus, A. obscurus

- $^{b}$  = insect collection from traps and counting
- 1- the trap is removed from the soil
- 2- Before opening, the trap is placed in a large trasparent bag, then the trap is opened and the insects fall inside the bag.
- 3- the bag should be closed immediately.
- 4- the trap is placed back into the soil.

# Warning: never open lure cap.



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LURE COMBINATIONS	REGION
A. brevis, A. sordidus, (A. litigiosus), A. ustulatus	Italy (North eastern)
A. brevis, A. sordidus, , A. litigiosus	Italy (other regions)
A. brevis, A. sordidus, trap A	France
A .lineatus, A. obscurus trap B	
A. sputator, A.rufipalpis (same lure of sordidus), A. ustulatus - trap A A. lineatus, A. obscurus trap B	Hungary
A. sputator, A. ustulatus - trap A	Slovenia and The
A. lineatus, A. obscurus t <mark>rap B</mark>	Netherlands
A. sputator, A. sordidus, A. ustulatus - trap A A. lineatus, A. obscurus Trap B	Germany

# Assessment of damages by soil insects

Early season (check for soil insects, baklckcutworm, other minor pests) in all the plots (conventional and IPM):

each plot should be scouted by choosing at random 2 areas of 20 m X 6 maize rows per field (20 x 2 central rows per strip in case of alternate treated and untreated strips) and observing all the plants. Plants with typical wireworm or black cutworm damage will be individuated and all the larvae found near the collar will be collected and identified. Please indicate sampling areas used from the beginning till the end of the trial.

The following observations will be done at emergence and 5-7 leaves :

- crop stand (number of normal plants/20 m);
- number of seeds damaged;
- number of emerged plants damaged by wireworms, cutworm or other soil pests per 20 m.



# **ANNEX 2: ECB** assessment

# Biological control against ECB using Trichogramma brassicae

## **Experimental design**

In the same grain maize field, 3 plots A, B,  $C \ge 5000 \text{ m}^2$ 



> 100m

Trichogramma treatment will be done against the second generation of ECB (G2) in France as it is the most damageable for the crop

Sub-task 1: ECB monitoring (see annex 4 of TASK3.3a protocol).

Adults' flight(s) – for G1+G2 - will be followed using a light trap. The trap has to be installed before the beginning of the ECB flight.

# Sub-task 2: Trichogramma release assessment (see ECB Doc 1).

the release date is based on the observation of the **ECB pupation**, which have to be done after the first flight is finished. The release date is planed one week after the threshold of 25-30% of pupation is reached. This information has to be sent to Biotop in order to reactivate *Trichogramma* and to prepare the conditioning. The product will be delivered to each participant by express carriage, in insulated boxes keeping the product in good conditions of temperatures.

# Sub-task 3: Trichogramma treatment (see ECB Doc2)

One release in France where the forecasting system is effective for long time.

<u>Release 1</u>: one week after 25-30 % of pupation. Dose: 375000 T/Ha conditioned in 50 dispensers.

Releases will be done according to the product specifications.

# **Sub-task 4: Pest pressure evaluation and parasitisation assessment** (see ECB Doc 3) In plots A and C:

- In southern countries only: % of attacked plant by the ECB G1 (visual damages). To be done just before the 2<sup>nd</sup> flight beginning, on 100 plants per plot.
- scouting of egg masses under the maize leaves on 100 plants per plot. Each egg mass will be noted as: Fresh (F), Parasitized (P) or Hatched (H). Then percentages of



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infestation and of parasitisation are calculated. Scoutings are carried out at least 2 times during the egg laying period: 10 days and 20 days after the 1<sup>st</sup> release.

#### Sub-task 5: ECB damages assessment at harvest (see ECB Doc 3)

In plots A and C:

At the same sampling areas as indicated in Annex 1, (2 areas of 20 m X 6 maize rows per plot (20m x 2 central rows per strip in case of alternate treated and untreated strips) measure:

- a) Total number of plants (final stand)
- b) Plants without ears/cobs;
- c) Plants with symptoms of ECB attack (e.g. holes on leaves, on cobs);
- d) Plants broken above ear;
- e) Plants broken below ear;

#### On 10 plants from each subplot measure :

f) <u>plants with ECB damage on the cob</u>: each cob of the 10 plants will be classified according to the percentage of surface damaged by ECB using a scale from 1 to 7, which corresponds to: 1 = non attacked, 2 = < 4%; 3 = 5-10%, 4 = 11-25%, 5 = 25-50%, 6 = 50-75%, 7 > 75%.

g) <u>plants with *Fusarium* presence</u> each cob of the 10 plants will be classified according to the percentage of surface covered by *Fusarium* using a scale from 1 to 7, which corresponds to: 1 = non covered; 2 = 1-3 %, 3 = 4-10%; 4 = 11-25 %, 5 = 25-50%, 6 = 50-75%, 7 > 75%.

### **Doc 1:** *Trichogramma* release assessment in France

Principle: the date of release is based on the % of ECB G1 pupation which allows forecasting the beginning of the  $2^{nd}$  flight.





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The evaluation of % of pupation starts when the first flight is finished, by cutting attacked plants and counting larvae and pupae of ECB inside the stalks. For each counting, the total number of larvae and pupae should be at least **30 alive individuals** to be accurate enough, the number of plants to be cut is depending of the infestation (usually < 30 infested plants).

Calculation of the % of pupation: L = number of alive larvae P = number of alive pupae % of pupation = P / (L+P) x 100 with L+P  $\ge$  30 pupae in a maize stalk



A

This work is done in the experimental field, and if possible, in at least 2 others fields in the same area and with the same sowing date (3 fields in total to have an over view of the local situation). If possible, the best is choosing high infested fields to save counting time. Usually several countings are needed, the frequency is once or twice a week depending of the temperatures (total counting 2 to 4), to reach the right percentage (about 25-30% of pupation) to take the decision of the first Trichogramma release (to be placed one week after). Time evaluation: 1 counting for 30 individuals = 1.5 hour

# Doc 2: Trichogramma treatment

Against the 2<sup>nd</sup> generation of ECB only in France 1 release with 375000 T/Ha in 50 dispensers/ha (the product will be ready to use). **Scheme of release:** 



<u>Time evaluation: 1 ha treated = 20 minutes</u>

# Warning!

The product is delivered by express carriage, and has to be used immediately or within 24 hours after delivery (storage between 15 and 20°C, in a ventilated room far away from pesticides or cigarette's smoke).

Do not expose the product to the sunlight and do not leave it in a closed car staying in the sun (very high temperature inside = high mortality of beneficials).

Use the product early in the morning or late in the evening to avoid high temperatures. No risk with rain falls or irrigation.



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# **Doc 3:** pressure and damages assessment of ECB; Trichogramma parasitism evaluation

Only in plots A and C



# a) Pest pressure:

>  $1^{st}$  generation (G1) in southern countries only: before the beginning of  $2^{nd}$  flight: % of damaged plants on 100 plants (all visuals damages on leaves and on stalks).

# Time evaluation: 1 hour/plot

In all countries: 10 days and 20 days (2 times at least) after the 1<sup>st</sup> Trichogramma release, counting of all egg masses found on 100 plants, and notation as Fresh/white (F), Parasitized/black (P) or Hatched (H) egg masses (see pictures below).

# Results: total egg masses/100 plants = (F+P+H)

# Time evaluation: 2 hours/control/plot



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Fresh egg mass under a leave = F



Black head stage = no parasitized = H (Head of larvae visible)



Left: Parasitized (Totally black) = P Right: Fresh



Egg mass just hatching (on left) = H (With young larvae)

**Warning!** Do not mistake parasitized egg mass (totally black eggs) and black head stage that means no parasitized egg mass (small black points are easily visible through the chorion).



## b) Trichogramma parasitism:

## Results: % of apparent parasitism = P/(F+P+H) x 100

NB: this calculation <u>under-evaluate</u> the actual % of parasitism as we do not know if fresh egg masses are parasitized or not (after being parasitized eggs need 4-5 days to turn black).

#### c/ ECB damages assessment at harvest

In plots A and C:

At the same sampling areas as indicated in Annex 1, (2 areas of 20 m X 6 maize rows per plot (20m x 2 central rows per strip in case of alternate treated and untreated strips) measure:

- a) Total number of plants (final stand)
- b) Plants without ears/cobs;
- c) Plants with symptoms of ECB attack (e.g. holes on leaves, on cobs);
- d) Plants broken above ear;
- e) Plants broken below ear;

#### On 10 random plants from each subplot measure :

f) <u>plants with ECB damage on the cob</u>: each cob of the 10 plants will be classified according to the percentage of surface damaged by ECB using a scale from 1 to 7, which corresponds to: 1 = non attacked, 2 = < 4%; 3 = 5-10%, 4 = 11-25%, 5 = 25-50%, 6 = 50-75%, 7 > 75%.

g) <u>plants with *Fusarium* presence</u> each cob of the 10 plants will be classified according to the percentage of surface covered by *Fusarium* using a scale from 1 to 7, which corresponds to: 1 = non covered; 2 = 1-3 %, 3 = 4-10%; 4 = 11-25 %, 5 = 25-50%, 6 = 50-75%, 7 > 75%.





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## ANNEX 3: Bt protocol and ECB assessment

## Biological control against ECB using Bacillus thurigiensis var. kurstaki

## **Experimental design**

In the same grain maize field, 3 plots A, B,  $C \ge 5000 \text{ m}^2$ 



*Bacillus thurigiensis var. kurstaki* treatment will be done against the second generation of ECB (G2) in Italy, Hungary and Slovenia as it is the most damageable for the crop. The treatment date will be based on the weakly observation of at least 200 plants (when to start the assessments will be based on general weather conditions and suggested by Lorenzo Furlan) according the inspection layout described in sub-task 2 (2 subplots of 20 m X 6 maize rows per plot). The treatment date is planned one week after the finding of the first egg mass or just after the finding of first larvae (usually inside the silks). Timing forecast will be also supplied by using a new forecasting ECB model that will run using climatic stations located in Italy; if weather data will be supplied from other sites the model will be run for other locations as well. PLOT C will be treated with Bacillus thuringiensis varietà kurstaki 6,4%, 1 kg/ha using farm equipment, a pressure of 3.5 bars and spray volume of 500-600 l/ha.

Sub-task 1: ECB monitoring (see annex 4 of TASK3.3a protocol).

Adults' flight(s) - for G1+G2 - will be followed using a light trap. The trap has to be installed before the beginning of the ECB flight.

# Sub-task 2: Pest pressure and ECB damage at harvest

In plots A and C:

- Assess the % of attacked plant by the ECB G1 (all visuals damages on leaves and on stalks). To be done just before the 2<sup>nd</sup> flight beginning, on 100 plants per plot.
- Before harvesting, at the same sampling areas as indicated in Annex 1, (2 subplots of 20 m X 6 maize rows per plot (20m x 2 central rows per strip in case of alternate treated and untreated strips) measure:
  - Total number of plants (final stand) per row;
  - Plants without ears/cobs per row;
  - Plants with symptoms of ECB attack (e.g. holes on leaves, on cobs) per row;
  - Plants broken above ear per row;
  - Plants broken below ear per row;



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- <u>plants with ECB damage on the cob</u>: each cob of the 10 plants will be classified according to the percentage of surface damaged by ECB using a scale from 1 to 7, which corresponds to: 1 = non attacked, 2 = < 4%; 3 = 5-10 %, 4 = 11-25 %, 5 = 25-50%, 6 = 50-75%, 7 > 75%.
- <u>plants with *Fusarium* presence</u> each cob of the 10 plants will be classified according to the percentage of surface covered by *Fusarium* using a scale from 1 to 7, which corresponds to: 1

 $\begin{array}{l} \text{= non covered; } 2 = 1 - 3 \%, \\ 3 = 4 - 10\%; \ 4 = 11 - 25 \%, \\ 5 = 25 - 50\%, \ 6 = 50 - 75\%, \\ 7 > 75\%. \end{array}$ 





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### ANNEX 4: Weed assessment

#### **Creating subplots:**

Shortly after sowing of maize we select and mark randomly 15 subplots (sampling areas) per plot A and per plot B (A = control/conventional plot; B = IPM tools for weeds control), of 0,75 m<sup>2</sup>: width 0,75 m (rows distance from plant to plant) x length 1 m. In each of these subplots, weed assessments will be done in 2 quadrates (size of 0.33 x 0.33cm each), placed 1 along and 1 between rows so the effect of band-application can be determined (see figure below on positioning).

These subplots should be selected according the weed distribution/condition in the field (a short scouting across the plot will give you the idea of distribution) in order to get the best estimation of the weed density in each plot.

#### Example of quadrats positioning inside a subplot (1m x 0.75cm)



#### The following parameters should be reported:

- a) weed species (according to the EPPO-Code, see <a href="http://cipm.ncsu.edu/names/index.cfm">http://cipm.ncsu.edu/names/index.cfm</a>)
- b) weed density/number per species
- c) total weed coverage (%)
- d) total weed biomass (dry matter), only estimated at the last evaluation

### Weeds should be estimated at 3 times:

a) just before the post-emergence treatment after maize emergence (herbicide or other method)

b) 3 weeks after the last treatment (herbicide or other method)

c) cca. 3 months after the (last) treatment, when weed biomass is at maximum, e.g. more than 50 % of the weeds are flowering (BBCH 61-65) - before harvesting

**Weed density/number assessment**: Weed seedlings/species should be counted from all **15** subplots/quadrates/plot. **Quadrates sampled within each subplot should be coded "Along" and "Between" so we distinguish the effect of band application.** 

Weed biomass assessment: For each plot (A + B), total weeds will be cut at the soil surface from the quadrates sampled within each subplot and placed in 2 bags coded "Along" and "Between" so we distinguish the effect of band application. Total weed biomass/plot will be dried in an oven and weighed (kg/ha).



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## ANNEX 5: Yield estimation & mycotoxins

Yield shall be assessed at all experimental field/plots (control, weed and ECB) separately.

The yield shall be estimated using combine harvester. The weight of harvested grains shall be separately assessed for each experimental field/plot, whereas a random grain sample of 500 g for the moisture content determination and one of 2-3 kg for the mycotoxin analysis will be collected as follows:

- With a specific container you take in successive moments small grain samples that come from the cochlea of the harvester (at least 10 samples of 200 gr or better 20 samples of 100 gr and put them together in a plastic bag;
- Close the bag in an air tight way so you avoid air as much as possible inside the bag;
- place a tag inside and one outside the bag;
- in maximum 6 hours place the samples in a freezer (-18°C).

Calculation of grain yield shall be expressed in tonnes per hectare grain with 14 % moisture content.

#### \* Alternatively (not obligatory – see below):

On each field/plot 4 subplots (randomly chosen) shall be created.

<u>Harvest</u>

Subplots shall be randomly determinate in all experimental fields/plots

Number of subplots in each field/plot: 4

Dimension of subplots: cca. 10 m<sup>2</sup>, (2 rows x 7 m length)

Type of harvest: manually - all the cobs from each subplot shall be detached from the plants and immediately taken from the field for further evaluation

# **Yield estimation**

Separation of grain from all cobs harvested from each subplot with parcel grain harvester (if applicable; if not grain shall be separated by hand from the sample of 10 cobs. From the ratio of grain and ears the grain yield is assessed – weight all cobs from separated subplot and using the ratio of grain and ears calculate the grain yield  $\rightarrow$  *Grain yield* = *weight of cobs x ratio of grain and ears of the sample of 10 cobs*)

Grain moisture determination (ISO 711:1997)

Calculation of grain yield which shall be expressed in kg per hectare grain with 14 % moisture content.



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## ANNEX 6: Tools to be tested in 2013-2014

### IPM tools to be tested in Italy against the conventional approach 2013-14:

1) BT spraying when monitoring indicates

2) Pre-emergence herbicide application in band + post-emergence combined rotary tiller with ridging (photo below) or hoeing (less  $CO_2$ ) depending on weed density, species and growth stage.



## IPM tools to be tested in Slovenia against the conventional approach 2013-14:

- 1) BT spraying when monitoring indicates
- 2) Pre-emergence or early post emergence herbicide application in band + hoeing

### IPM tools to be tested in Hungary against the conventional approach 2013-14:

- 1) BT spraying when monitoring indicates
- 2) early-post emergence herbicide application in band + hoeing

### IPM tools to be tested in France against the conventional approach 2013-14:

1) Trichogramma when monitoring indicates

2) early-post emergence herbicide application in band + hoeing at ALIXAN & 2-3 mechanical control at MONTOISON (no post-emergence herbicides)

### IPM tools to be tested in Germany against the conventional approach 2013-14:

early-post emergence herbicide application in band (early post) + hoeing
mechanical weed control



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