

Identification of critical points from validated results: Effect of structural variables on GM admixture in non GM harvest based on simulation results - Alsace, Switzerland case studies - Maize

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D7.2 Identification of critical points from validated results

- Effect of the structural variables on GM admixture in non GM harvest based on simulation results -

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PU	Public				
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EXECUTIVE SUMMARY - SIGMEA Deliverable 7.2

This deliverable aims at identifying the effects of the structural variables on gene flow in various regional case studies, using gene-flow model (GENESYS for OSR and MAPOD for maize).

The simulations took the characteristics of landscapes and cropping systems described in the D7.1 as inputs. From this starting point, different factors affecting gene flow were assessed. The following table synthesizes the studied factors for each case study. The analyses allowed comparison between case studies and also within each study. GM varieties were considered isogenic to non GM. Several random allocations of GM varieties to the fields were used, without incorporating any additional measures to decrease cross-pollination. Maize cases studies focussed on spatial GM dissemination schemes, whereas OSR case studies dealt also with return to non GM after GM OSR in the rotation.

		Maize			OSR	
Factors	Alsace	Aragon	Swiss	Beauce	Fife	Germany
field pattern (number of spots)	2	2	1	1	1	1
% crop / AUA	25 to 100%	11 to 100%		21 %	11 %	17%
ratio GM / non GM crops	10%, 30%, 50%, 70%	10%, 30%, 50%, 70%		10%, 50%, 75%	10%, 50%	10%, 50%, 75%
crop distribution	random selection and historical references	random selection and historical references		fixed (historical references)	fixed (historical references)	fixed (historical references)
GM field random allocation of GM distribution maize to the fields		ocation of GM the fields	Clustering of GM and non-GM maize	random allocat	ion of GM OSR	to the fields
Distribution of wind speed and direction during flowering period	Average distribution from nearby station	Average and yearly distributions from nearby station	Average distribution from nearby station	not taken ir	nto account by th	ne model
Crop management	/	flowering time lag or synchronicity	/	several practices affecting gene flow by pollen	/	/
Crop sequence	No volunteers	Various hypothesis on gene flow due to volunteers	/	several practices affecting gene flow by volunteers	several practices affecting gene flow by volunteers	

Table 1: list of studied factors taken into account by geneflow models, affecting gene flow for each case study

Two indicators were used to assess the effects of regional crop coexistence on grain quality. At the field level we considered the **percentage of non GM crop area above the threshold**. At the landscape level we considered the **average content of GM material in the bulked**

non GM harvest (for example the grain quality at the silo level). The GM content was expressed as a percentage of grains. The main threshold used for the assessment was the legal one defined by the EU directive (0,9%). Other thresholds were used in maize case studies corresponding to real cases seen in starch or semolina industry (0,1%) and 0,01%).

These studies showed how gene flow varies according to the structural variables describing landscapes and cropping systems. Variations were identified between case studies, and also within each of them, according to the conclusions of D7.1. Table 2 gives an overview of sensitivity to gene flow, taking into account current cropping systems and several allocations of GM and non-GM crops to the fields. The results depended on the scale used to assess coexistence. Generally, complying with coexistence would be much easier at the silo level than at the field level, due to a dilution effect. This effect is more pronounced with the size of the silo

Table 2 : Risk of exceeding GM thresholds (percentage of GM grains) at the field and landscape level with a 50% GM introduction rate in current cropping systems without coexistence measures (min, median, max)

Region	Spot	Proportion of non-GM area not complying with the 0.9% threshold (%)			GM advent GM harves (%) under threshold	itious presence at at the lane (1) or above (e in the non- lscape level <mark>2</mark>) the 0.9%
		min ¹	median ¹	max ¹	min ¹	median ¹	max ¹
			Ma	ize			
Alsace	Ensisheim 2005 ² (70% of maize)	19.8	28.9	37.5	1	1	1
	Heiwiller 2005 ² (85% of maize)	39.4	44.7	56.7	2	2	2
Aragon	Sariñena 2005 (42%) of maize) ³	0.8	3.6	18.2	1	1	1
	Gurrea de Gallego 2003 (34% of maize) ⁴	7.5	14.5	22	1	1	1
			OS	R			
Beauce	2004 (21% OSR) ⁵	0	8	52	1	1	2
Fife	2005 (11% OSR) ⁵	0	0	100*	1	1	2

* distribution with only one non GM field

¹ among many repetitions (allocation of GM crops to fields), other things being equals.

² assuming that GM and non-GM maize flowered simultaneously and there is no maize volunteers

⁵ co-existence during 20 years between cropping systems supporting GM or conventional non-GM OSR, focusing mainly on spatial aspects where a conventional OSR cannot be grown after a GM OSR and vice versa

³ assuming that GM and non-GM maize flowered simultaneously and there are maize volunteers (responsible for an extra-GM adventitious presence in the non-GM harvest).

⁴ assuming flowering time-lag and that there are maize volunteers (responsible for an extra-GM adventitious presence in the non-GM harvest).

Table 2 shows the diversity of risks of exceeding GM thresholds between regions but also inside regions. This diversity is explained by structural variables: field pattern, cropping system and field distribution. All factors being equivalent, the variability of risk is linked to GM and non GM spatial distribution and also flowering dates.

Additional results from simulations not shown in the table 2 are described in the full report

- The annual variability of structural variables like wind distribution and crop proportion results in subsequent variability of crops being above the threshold.
- The Swiss case study shows that a cropping block strategy decreases the average admixture at harvest, but additional measures may be required to manage transboundary problems.
- The maize case studies show that lower thresholds greatly increase the probability for a field to be above the threshold. It will thus be difficult to achieve with lower thresholds (0,1 and 0,01%) without isolation measures between crops.
- Volunteers are the major issue in cases of returning to cultivation of non GM OSR following GM cultivation on the same field.
- Few cumulative effects arose under current cropping systems in the OSR case studies, due the good volunteers' management in set aside and field margins.

D 7.2 Identification of critical points from validated results

- Effect of the structural variables on GM admixture in non GM harvest based on simulation results -

Alsace case study - maize

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SUMMARY

Introdu	iction3
I. [.]	The Mapod Model
а-	MAPOD model structure
b -	General parametric choices5
b.1	1. Two different measures of GM adventitious presence rate in non GM crop5
b.2	2. Gene characteristics of GM maize varieties
C -	Limitations of current MAPOD model7
II. ·	The simulations in Alsace9
а-	Material9
a.1	1. Choice of simulation areas9
a.2	2. Meteorological data
a.3	B. Distribution of conventional and transgenic maize in the studied islands
a.4	4. Crop practices and varietal characteristics
b -	Methodology for the treatment and presentations of model outputs
b.1	1. The output indicator, the thresholds
b.2	2. The spatial units considered for analysis15
III.	Results17
a -	Impact of introducing GM Maize in the current crop rotations on the non GM area to
be do	owngraded17
b -	Effect of the maximum threshold for cross-pollination with GM maize and of the
spati	al distribution of GM maize on the non-GM area to be downgraded
с -	Effect of the spatial unit for analysis on the non GM area to be downgraded21
d -	Effect of the study area delimitation and of the way to allocate GM maize (by farms
or by	r fields) on the "non-GM" area to be downgraded23
d.1	1. Effect of the delimitation of study area (with or without edge fields) on the "non-
GN	A" area to be downgraded23
d.2	2. Effect of the way to allocate GM maize (by farms or by fields) on the "non-GM"
are	ea to be downgraded23
е-	Impact of non GM and GM maize levels in the landscape on the non GM area to be
dowr	ngraded and on the average GM adventitious presence in the total "non-GM" harvest
	25

 Appendixes
 30

 Appendix 1: Variation of the part of total non-GM area downgraded according to the delimitation of the area used for treatment (with or without edge fields) and the maximum threshold for GM adventitious presence
 30

 Appendix 2: Impact of the spatial unit for analysis on the non GM area to be downgraded
 32

 Appendix 3: Part of downgraded "non-GM" area considering two ways for the introduction of GM maize in the landscape: by fields or by farms
 34

 Appendix 4: Impact of non GM and GM maize levels in the landscape on the non GM area to be downgraded
 36

Introduction

In that task the general purpose is to identify the critical points for GM dissemination within different landscapes. For that, two steps have been managed for each couple of region-model – Alsace-MAPOD (maize) and Beauce-GENESYS (rapeseed):

In a first step, WP4 models are used to assess, for each regional case study and with no change into agricultural practices of the farmers, the impact of introducing GM varieties in the landscape on GM adventitious presence in non GM harvest.

The second step consists in organizing, in each region-crop case, stakeholder working groups. Those groups aim at discussing the results of simulations and also defining and classifying the difficulties to ensure coexistence between GM and non GM crops in study areas

In this chapter we give in a first part (I1), a small description of Mapod model (Structure and limitations) and the main characteristics of parametric choices. In a second part (I2) we present the Alsatian databases and methodologies: (a) the Alsatian databases allowing to describe spatial distribution of fields, mean crop practices and genotypic characteristics used as inputs in the model; (b) the output indicators and statistical methodologies used to calculate them. The results are presented in a third part.

I. The Mapod Model

a - MAPOD model structure

MAPOD maize (Klein *et al*, 2003; Angevin *et al.*, 2001) is a gene flow model based on two main modules (Figure 1): a flowering dynamics module and pollen dispersal module. The input data required for the functioning of each module have been reduced to a minimum so that the model is easy to use in a large number of situations. Figure 9.1 presents a simplified flow-chart of the model. Input data can be classified as follows, according to the information provided:

- Field plan (in pink): Form and size of the fields Location of GM plants
 - Climate (daily data) (in blue): Temperature; rainfall; wind: speed and direction
- Cropping systems (in red): Sowing dates and densities

Drought stress before flowering

Drought stress during flowering

• Variety (in green): Quantity of pollen per plant

Pollen sensitivity to high temperature Temperature requirements between sowing and female flowering Genotype of the GMO: homozygous or heterozygous Tassel height Cob height of the non-GM variety



* Ratio of pollen emitted by non-GM plants to pollen emitted by GM plants

** Height difference between tassel of each variety and cob of non-GM variety.

*** Equation used to simulate pollen dispersal for one maize plant (Klein, 2000).

**** Temperature needs between sowing and female flowering.

The first module uses information about climate, cropping systems and varieties to determine, on a day-to-day basis, the flowering dynamics of GM and non GM panicles and Non-GM silks. Flowering date for female flowers is expressed in degree-days, as a function of climate and sowing date.

Most of the varieties currently used display protandry, which means that male flowering begins several days before female flowering. The duration (in days) of this time lag can be used to calculate flowering time for the male flowers. Drought stress and sowing density affect protandry.

Once flowering date is known, the dynamics of male and female flowering can be modelled to estimate the amounts of pollen produced by GM and non-GM varieties, respectively, and the number of recipient silks for non-GM varieties. Factors affecting the viability of pollen and the receptivity of silks are taken into account. The composition of the pollen cloud in the air around the plants is therefore known on a day-to-day basis for the entire flowering period.

The second module uses flowering dynamics and other information concerning field plan, climate and varieties to predict spatially defined GM adventitious presence in non-GM maize every day and at harvest.

Pollen dispersal is simulated by Klein's equation (2000 & 2003). It is a statistical function of distance from the emitter. The parameters used for its calculation are the direction and mean speed of the wind during flowering and the difference in height between the panicle from which the pollen is emitted and the recipient silks. The composition of the pollen cloud at a given site in a non-GMO field is determined by the pollen dispersal curves for all plants in the neighbourhood, whether close or further away.

The frequency of GM seeds is calculated daily as a ratio of the number of non-GM ovules fertilised by GM pollen to the total number of ovules fertilised. These daily results are pooled to provide a total frequency of GM seeds in the harvest.

b - General parametric choices

b.1. Two different measures of GM adventitious presence rate in non GM crop

Throughout this study, GM adventitious presence rates have been evaluated as percentage of harvested grains carrying the transgene. This way of quantification cannot be directly related to the one obtained from a genetic quantification of the GM adventitious presence.

In GMO quantification using PCR-based methods, GM proportion in a given substrate is estimated by calculating the transgenic genome copy number in the total maize genome copy number. Thus, two PCR reactions are carried out, one amplifying the transgene to determine its number of copies, and another one amplifying an endogenous gene in maize to determine the total number of genome copies of maize in the sample.

Maize kernels used for PCR analysis, are made up, mainly, of a tegument, an embryo and an endosperm. Trifa and Zhang showed in 2004 that the proportion of these elements depends on the variety. The DNA origins of those tissues are presented in the table 1. Generally, tegument DNA can be neglected (maximum 3.5% of total DNA). Endosperm and embryo DNA ratio were globally similar since, in this study, these ratios range from 36.27% to 59.41% for the endosperm and from 38.56% to 59.55% for the embryo.

In addition to the genetic structure of the GM material (homozygous, heterozygous, stacked), it is thus important to know the relative DNA content ratio of the different tissues to total DNA content in order to be able to express the relation between the results given by the model (% of seeds) and the one given by PCR methodology.

Table 1: DNA origins of grain tissues

Tissue	Number of genome copies	Origin of the copies
	2 (diploid)	1/2 Maternal + 1/2 Paternal (fusion of one haploid
Embryo cens	2 (dipiola)	maternal nucleus and one haploid paternal nucleus)
Endocrorm	2 (triploid)	2/3 Maternal + 1/3 Paternal (fusion of two maternal
Endosperm	3 (1101010)	polar nuclei with one sperm nucleus)
Teguments	2 (diploid)	Maternal

Table 2: Consequences of GM impurities in seed batches

GM impurities in seed: 0.3 %	Rate of GM adventitious presence in the harvested grain estimated by MAPOD
Homozygous	~ 0.6 %
Heterozygous	~ 0.3 %

In these simulations, the mean wind speed is 4 m.s⁻¹ and the wind blows throughout the flowering period. The contaminating GM seeds are distributed randomly over the plot, as would occur during sowing in the real situation.

As an example, a situation where a maize silk from a non GM female plant is pollinated by a pollen grain carrying one copy of a transgene can be considered. In these conditions, half of embryo's DNA and one third of endosperm's DNA would be GM. With the hypothesis that the harvested grain presents the following relative DNA content ratio of tissues: Embryo = 48% of DNA; Endosperm = 49% of DNA; Tegument = 3% of DNA.

The percentage of DNA bearing the transgene in the grain is:

 $48\%^{*}(1/2)+49\%^{*}(1/3)+3\%^{*}0 = 40.3\%$.

Thus, in the case of heterozygous GM maize, the GM adventitious presence rates expressed as percentage of seeds as evaluated by the model should be multiplied by 0.405 to obtain the genetic quantification that would obtain by PCR methods.

b.2. Gene characteristics of GM maize varieties

In this study, we have considered an heterozygous GM maize with a very simple molecular characterization: one transgene on one chromosome. This hypothesis is consistent if, during the multiplication process of the variety, the modified gene is carried by only one of the two parents. In fact, it is the case for most of Bt maize varieties. According to this hypothesis, only one pollen grain over two carries the transgene.

However, some GM maize varieties can carry many transgenes coding for one or several actions (insect resistance, tolerance to environmental condition, tolerance to an herbicide, etc.). Those genes may be carried by one or some chromosomes and the industry producing the GM variety can decide to multiply it by crossing two parents carrying both transgenes or by crossing a GM parent carrying all the transgenes with a non GM parent. Then, the proportion of GM pollen emitted by the GM variety will vary from 50% to 100% according to the number of transgenes and their distribution on the chromosomes.

This proportion of GM pollen is of the utmost importance since it is directly linked to the rate of GM adventitious presence in non GM crops. For instance, if 100% of the pollen is genetically modified, the rates of GM adventitious presence in non GM maize crops are twice as big as the one obtained with only 50% of the pollen carrying the transgene.

c - Limitations of current MAPOD model

Some limits of the model, in its current state of development, have to be kept in mind.

The discontinuities that may exist in an agricultural landscape (bare soil, other crops, roads and paths) are represented as totally sterile maize plants and the model doesn't take into account the pollen carried away on long distance in the atmosphere.

Two sources of genes can be considered as negligible in European situations: the presence of "wild maize" and maize volunteers. A third one has to be more discussed: varietal impurities in seed lots are not directly taken into account by MAPOD. Simulations were carried out to determine the effects of seed impurities, taking into account a field planted with a non-GM variety in an agricultural area without GMOs. Two cases were considered, according to the genetic make-up of the GMO (homozygous or heterozygous).

We can assume that the impurity rate of the seed is due to cross-pollination of the female plants in a seed production field by pollen from GM fields. In this case, GM seeds are heterozygous and, according to the results shown in table 2, the effect of varietal impurities on seed lots is taken into account in an additive manner (1:1 ratio) in the estimate of the total frequency of GMOs in the harvest as well as the GM adventitious presence due to machinery.



Figure 2: Study landscape (communes¹)

Figure 3: Wind distributions

Ensisheim study area



Heiwiller study area



¹ French administrative district

II. <u>The simulations in Alsace</u>

Alsace presents high level of maize in the crop rotations. This maize is collected by a few elevator firms. The main outlets are animal feed and mostly human feed (starch and semolina industries). In order to be the most representative of Alsatian field pattern characteristics, two contrasted areas (several hundreds of hectare each) (a) have been used to run three batches of simulations (b).

a - Material

a.1. Choice of simulation areas

The landscape used consists in 108 communes, which represent the collection areas of 3 storage silos of the CAC^2 (silos of Ensisheim, Huningue and Heiwiller, see Figure 2).

Those data supplied by ONIC³ have been extracted from database of CAP⁴ declarations of crops grown by farmers in 2005. They consist in two types of information:

- GIS information: Spatial unit in France for those declarations is not the field but the "production island". A *production island* is defined as a group of contiguous fields owned by the same farmer and limited by permanent physical limits or other farms. The delimitations of *production island*s of the 108 communes have been supplied. That represents about 24,000 islands and 2,012 farmers.
- Production information: ONIC has also given information about the surfaces of the crops grown on those islands.

A small study on island areas by commune allowed us to differentiate three types of island pattern (Figure 4). The first one around Ensisheim ("Plaine du Rhin" and "Hardt" regions) consists in large production islands (mean island area over 4 ha). The second one around Heiwiller ("Sundgau" region) is made up of smaller islands (mean island area between 1 and 3 ha). The last type of island pattern around Huningue ("Sundgau" region) is made up of very small islands (mean area below 2ha). For practical reasons, we only considered the two first types of patterns since we have not been able to meet farmers from the third area. In fact, Root Worm has been detected near Huningue in 2003, and thus, maize has been forbidden the last two years in this region.

In accordance with the capacities of the model in terms of simulation time, we have extracted from the whole island pattern two areas of simulation (about 6 to 9 km²) around Ensisheim and Huningue. At this level, the production islands have been reshaped (moved, cut and sometimes deleted) according to Aerial photographs (orthophotos) of Haut-Rhin agricultural area so that they overlap the best with the cultivated fields of the area. Thus, after this work of updating, the spatial unit is no longer the "production island" but the field. Those simulation areas contrasted in terms of field characteristics (shape, surface...) are presented in figure 5.

a.2. Meteorological data

Wind distributions used for the simulation (Fig 3) have been established with meteorological data recorded in the months of July and August on: (1) The Meteo-France's station of Meyenheim between 1981 and 1999 for the Ensisheim study area; (2)The meteorological station on the airport of Basel-Muhlouse between 1996 and 2005 for the Heiwiller study area.

² Coopérative agricole de céréales: storage organism in Haut-Rhin department

³ Office National Interprofessionnel des Céréales

⁴ Common Agricultural Policy



Figure 4: Mean area of production islands by commune

Figure 5: Island patterns considered for simulations





In both cases, for each direction the same average wind velocity of 3 m/s has been considered.

a.3. Distribution of conventional and transgenic maize in the studied islands

It is specified in the task 2 of the workpackage 7 that the level of maize in the AUA used for simulations must be representative of the current state of maize production in study areas. Thus, we should consider the maize levels evaluated during the French agricultural census (2000), which means 60% of the AUA sown with maize in "Rhin Plain" and only 50% in "Sundgau". However, those levels of maize, which have been evaluated for one year at a large scale (numerous communes), hide great variations in space and time. For instance, in the areas of simulation (600 to 900 ha), according to 2005 CAP statements of farmers, 70% of AUA have been sown with maize near Heiwiller and 85% near Ensisheim (40% larger than the areas of the agricultural census of 2000). As previous studies on coexistence between GM and non GM maize varieties have shown that cross-pollination is a very local phenomenon (few hundreds of meters), those spatial variations have to be taken into account. To do it we ran a first batch of simulations where maize area moved from 25 to 100% of the AUA. However, two other batches were implemented where maize was allocated according to, on the one hand, the levels of maize evaluated during the 2000 French agricultural census, and, on the other hand, the PAC statements of farmers in 2005 (batch 3)

Concerning the **rate of transgenic varieties**, the levels of 10%, 30%, 50% and 70% have been implemented.

For each combination of GM/non GM maize levels, many repetitions were performed in order to have an idea of the variability due to GM and non-GM maize distribution.

As specified in the project work plan, GM maize has been randomly introduced in the fields (50% of the repetitions) or in the farms (50% of the repetitions). This last scenario aims at simulating an introduction of GM maize by a limited number of farmers.

The simulated combinations of GM/non GM maize levels in the AUA as well as the ways to allocate each type of maize and the number of repetitions are precisely described in table 3.

a.4. Crop practices and varietal characteristics

In this task 7.2, cultural practices are the useful ones for the region, described in the task 7.1, except for the part of GM varieties supposed to be sown. The following assumptions have been made for those first simulations:

- GM and non-GM varieties flower at the same time;
- GM and non-GM varieties produce similar amounts of pollen;
- GM and non-GM varieties are sown at similar densities.

It will be possible to simulate the effect of each of these factors on cross-pollination rates in a second step of the study.

Batch of simulation	levels of maize (and number of repetition) on Study area of		Way to allocate (and number of		Way to allocate GM maize	Each simulation
number	Ensisheim	Heiwiller	maize in fields repetition)		in fields sown with maize	=
	25% (40)	25% (40)		10% (10)		1 random allocation of
	50% (40)	50% (40)	Random	30% (10)	Random selection of	maize in fields
1	75% (40)	75% (40)	selection of	50% (10)	maize fields	×
	100% (40)	100% (40)	fields sown with	70% (10)	(50% of repetitions)	1 random allocation of
2	FAC° level = 60%	FAC level = 50%	maize	10% (80)		GM maize in maize
2	(160)	(160)		50% (80)	• Random selection of	fields
			maize allocated		farmers growing maize	The allocation of maize
			according to	400/ (40)	and attribution of	in 2005
2	CAP Ievel = 85%	CAP level = 70%	CAP statements	10% (40)	the fields where those	x 1 random allocation of
3	(80)	(80)	of farmers in	50 % (40)	farmers grow maize	GM maize in maize
			2005		(50% of repetitions)	fields
	Field patte	rn	Sna	tial maize distribution	Spatia	distribution of GM maize
Example of non GM and GM maize allocation on study area near Ensisheim	100 0 200	W E S Mi distri in finto	aize bution	aize her crops	Non GM maize distribution in maize fields	maize bBM maize en crops
	700 0 700	1400 Metres	700	0 700 H400 Metre	s 700	0 700 H00 Metres

Table 3: Levels and distribution of non GM and GM maize in the landscape

⁵ French Agricultural Census (2000)

b - Methodology for the treatment and presentations of model outputs

b.1. The output indicator, the thresholds

In order to assess the impact of the sowing transgenic varieties in study areas, we have evaluated, for each simulation, the part of total area sown with "non-GM maize varieties" where GM adventitious presence was over a specific threshold so that the harvested grain could be rejected or downgraded.

The previous threshold can be defined by law or a contract between stakeholders. In fact, the legal maximum threshold for GM adventitious presence in non GM harvest to avoid the labelling "contain GMO", is 0.9%. However, the farmer can be asked for lower GM presence levels. For instance, no trace of GM maize must be detected in an organic harvest and the threshold for detection is 0.01%. Conventional producer may also be subjected to stringent purity specifications. In Alsace, starch industries ask for GM adventitious presence below 0.1%.

Furthermore, MAPOD outputs cannot be used directly. In fact, the model evaluates GM adventitious presence due to cross-pollination and other sources of admixture do exist in the production chain and must be added to cross-pollination rates between GM and non GM fields in order to obtain the total GM adventitious presence:

- GM presence in seed may be important (thresholds of 0.3% and 0.5% are under discussion) (see I1c).
- Machinery is a non negligible source of admixture. The pieces of equipment presenting a risk in terms of the introduction of GM grains in non-GM harvests are seed drills, combine harvesters and transport machinery (grain trailers and trucks). However the main risk consists in harvesting stage. Not cleaning the harvester between one GM and one non GM field may cause GM presence in the first trailer of about 0.4%. As the complete removal of all grain is extremely difficult and time-consuming, an alternative strategy based on flushing with non-GM grain may be of value. Various limited studies have indicated that flushing with a partial tank load of grain can reduce GM adventitious presence levels to below 0.1%, even without thorough machine cleaning⁶.
- Some farmers may dry and store their grain themselves. At this stage admixture with GM grain may occur due to handling mistakes or poor cleaning of drying and storage elements.

For those reasons, a large range of maximum GM adventitious presence due to crosspollination has been considered: 0.01%, 0.1%, 0.3%, 0.4%, 0.5%, 0.6%, 0.7% and 0.9%. The paragraph I1b showed that, under particular assumptions of DNA content in embryo, tegument and endosperm, there is a ratio of 0.405 between the percentage of GM grains in the harvest (model output) and the results given by a PCR analysis. For instance, when the MAPOD model evaluates a threshold of 2.25% it corresponds to 0.9% by PCR quantification. Thus, we also consider the following thresholds in the simulations previously presented: 0.025%, 0.25%, 0.75%, 1%, 1.25%, 1.5%, 1.75%, and 2.25%.

⁶ www.machinerylink.com/resources/ipg/article/harvester_clean_out.asp

Field pattern	Definition	GM adventitious presence due to cross- pollination evaluated for the whole non-GM maize area
2 *	Controls (number of control points)	On the whole grain of a storage silo (Few)
	Consequences (Economic risk in case of rejection)	If GM adventitious presence is over defined thresholds, rejection of the grain harvested on all the fields (High)
Field	Definition	GM adventitious presence due to cross- pollination evaluated for each field
*	Controls (number of control points)	Controls are made on the whole field harvest (in the trailers for instance) (Average)
Consequences (Economic risk in case of rejection		Rejection of all the fields where GM adventitious presence is over defined thresholds (medium)
Intra-field	Definition	GM adventitious presence due to cross pollination evaluated on each point of the fields
*	Controls (number of control points)	Controls are made locally in the field (Numerous)
	Consequences (Economic risk in case of rejection)	Rejection of the parts of fields where GM adventitious presence is over defined thresholds (low)

Table 4: Description of the three spatial unit for analysis we considered

Figure 5: Location of edge fields





b.2. The spatial units considered for analysis

A spatial unit for analysis is the smallest spatial element on which we have information about GM adventitious presence in non GM grain. This information requires controls and will allow some stakeholders (farmer, coops...) to make a decision on the future of grain batch (accepted, downgraded or even rejected). Three different units for analysis have been used to calculate the output indicator in this study: The whole study area, the field and the intra-field unit (the smaller unit for calculation is 100 m²). The table 4 gives a description of those units and provides peaces of information about the controls required and the economic risk run in case of one grain batch rejection.

b3- Statistics and presentation of the results

In the next results we adopted three principles.

Firstly, for each simulation we evaluated the part of total non GM maize area over a given threshold. For each situation, results are represented by a box plot tool that must be read as presented in figure 6. It gives not only the median value but also a measure of the dispersion of the different repetitions around this median for each situation.



Figure 6: How to read a box plot?

Secondly, the parallel presentation of results in both zone (Ensisheim and Heiwiller) give the opportunity to comment the regional effect on downgraded risks. This regional effect is made of intrinsic characteristics of fields, spatial arrangement of fields and climate.

Lastly, the field patterns have been considered from two points of view: the total area or the central fields only. In fact, the cross-pollination evaluated on the fields at the edge may be underestimated, as we have no information on the surrounding area. That's why some of the results presented in the following report are given taking into account or not those edge fields presented in the figure 5. The results with edge fields may be underestimated and the one without edge fields are certainly a bit overestimated.

Figure 7: Part of total non-GM area where cross-pollination with GM varieties exceeds 0.9%

Assumptions:

• Maize distributed according to:

2005 CAP declarations: 85% (Ensisheim) and 70% (Heiwiller) of maize in the AUA 2000 French Agricultural Census: 60% (Ensisheim) and 50% (Heiwiller) of maize in the AUA

- Part of total maize area sown with GM varieties: 10% and 50%
- Spatial unit for analysis: Field



III. <u>Results</u>

In a first step, we assessed, for the current regional maize levels (batch 2 and 3), the effect of introducing various levels of GM maize (with different repartition in the landscape) on the "non GM" areas downgraded if implementing a 0.9% maximum threshold for GM presence in non GM harvests (legal threshold) (§a).

In a second step, we assessed, still considering the current regional maize levels (batches 2 and 3) the impact on the non GM area downgraded of:

- The maximum threshold implemented for GM presence in non GM harvest (§b) (Do I decrease substantially non GM downgraded areas when lightening the constraints on the threshold? What happens if more stringent thresholds are specified in contracts?);
- The spatial unit used for decision making (§c);
- The way to calculate the non GM downgraded area (considering or not the edge fields) and the way to allocate GM maize (by field or by farm) (§d).

In a third step (§e), we used the batch of simulation 1 to test the effect of GM and non-GM maize presence in the AUA (variation of both %maize and %GM maize and their repartition in the landscape) on two indicators: the part of non GM area downgraded and the average adventitious presence in the whole non GM area.

a - Impact of introducing GM Maize in the current crop rotations on the non GM area to be downgraded

Based on the results of the batches 2 and 3, the figure 7 considering a maximum threshold for cross-pollination with GM varieties of 0.9% shows that:

- More "non-GM area" is downgraded if maize is distributed according to 2005 CAP declarations. It is due to a higher density of maize in AUA. As the batch of simulation 3 presents the highest GM adventitious presences, it has been used to illustrate the following results.
- A very small part of the "non-GM area" has to be downgraded (less than 8%) if GM maize represents 10% of total maize area,
- If we consider the risky situation of the year 2005 with equivalent areas in GM and non-GM varieties, about 45% of "non-GM area" has to be downgraded in Heiwiller (75% of repetitions are below 50%). This means that, in that study area, if half of the maize area is declared transgenic and if a percentage of GM grains is measured in the non-GM harvests, only a quarter of the Maize area could be considered as non GM according to a maximum threshold for GM presence of 0.9% (legal threshold).

Those results are less drastic for Ensisheim study area since only 26% (75% of the repetitions below 32%) of non GM area present GM adventitious presence over 0.9%. It is due to the field characteristics which lead to lower cross-pollination rates between GM and non GM varieties. Actually, as shown on the Figure 4, the mean area of the fields is smaller in Heiwiller study area (1.85 ha) than in Ensisheim study area (9.51 ha). Thus, pollen protection of non-GM fields and average distance between GM and non GM maize are bigger near Heiwiller, which explains the lower rates of cross-pollination.

<u>Figure 8</u>: Part of downgraded "non-GM" area according to maximum threshold for cross-pollination, considering the 2005 maize allocation in AUA (CAP databases, 2005) and two rates of GM maize (A-10% and B- 50%)



Table 5: probabilities that the average GM presence in total non GM area is over specific thresholds for two rates of GM maize (A-10%, B-50%) A

Г	Study area	Part of total num	ber of simulations wh	ere GM presence in	non GM grain harve	sted on the total non	GM area is over:
	of	0.01%	0.10%	0.40%	0.60%	0.90%	2.25%
	Ensisheim	1	0.825	0	0	0	0
	Heiwiller	1	1	0	0	0	0

В

Study area	Part of total num	ber of simulations wh	nere GM presence in	non GM grain harve	sted on the total non	GM area is over:
of	0.01%	0.10%	0.40%	0.60%	0.90%	2.25%
Ensisheim	1	1	1	0.8	0	0
Heiwiller	1	1	1	1	0.975	0

b - Effect of the maximum threshold for cross-pollination with GM maize and of the spatial distribution of GM maize on the non-GM area to be downgraded

The previous results take into account a maximum threshold of GM adventitious presence in non GM harvest of 0.9%. However, this GM adventitious presence is expressed as percentage of seeds and a genetic quantification may lead to different conclusions. Indeed, according to hypothesis made in paragraph 11b, a genetic adventitious presence of 0.9% corresponds more or less to 2.25% of grains bearing the transgene. In that case, less than 15% of the non GM area has to be downgraded in the worst situation (Heiwiller – 50% of GM maize – 2005 maize distribution) (Figure 8). This result shows the importance of the way to quantify GM presence in non-GM harvests.

Nevertheless, many contracts are much more stringent than the EEC regulations concerning maximum GM presence in non GM products (see § I2b). Analysing the figure 8 for lower thresholds than 0.9% we notice that:

- If 10% of maize area is transgenic, more than 70% of the "non-GM maize area" could be downgraded if implementing a 0.01% threshold (limit for detection). On the contrary for threshold over 0.4%, downgraded "non GM area" remains feeble (below 15%).
- If 50% of maize is transgenic, at least half of "non GM area" is downgraded if the threshold is below 0.9%. If implementing a 0.1% threshold, which is currently asked for by starch industry, more than 90% of the non GM area could be downgraded.

By analysing the median values, the benefit of implementing a higher threshold appears to be heavily dependant on the percentage of GM maize in maize area. In fact, for situations with 50% of GM maize, increasing the threshold, step by step, from 0.01% to 0.9%, is always valuable since the decrease of rejected area is always sensible. However, the rejected area reminds important even for thresholds of 0.9% (over 20% of non GM maize area). On the contrary, for situations with 10% of GM maize, increasing the threshold over 0.4% is not always of value since the decrease of downgraded area remains feeble.

More generally, the figure 9, established according to the graphs presented in appendix 1, shows a theoretical evolution of the part of non-GM maize area downgraded according to the maximum threshold for cross-pollination. The sigmoid form show two inflexions points x1 and x2 which divide the curve into three intervals according to the slope. In the A interval below x1 or in the C interval beyond x2, increasing the maximum threshold hardly reduces the rejected non-GM area but implicates proportional additional effort to ensure segregation of GM and non-GM maize at the other stages of the production chain (seed production, post harvest process...). In fact, the room for manoeuvre at those stages will be smaller. The threshold values x1 and x2 are of great interest and their values depend on the GM/non GM proportion we consider.

The variability of the results for each maximum cross-pollination threshold can be important. For instance, the maximum part of downgraded non GM area can be more than twice as big or twice as small as the median value. This variability is due to the strong impact of GM and non GM maize distribution in field pattern. Thus, the allocation choice of GM and non GM maize seems to be an effective way to limit cross-pollination. This variability is more important in Ensisheim due to the greatest variability of field area in this study case.



<u>Figure 9</u>: Theoretical evolution of downgraded area according to the maximum threshold we consider for cross-pollination

Figure 10: Impact of spatial unit for analysis on downgraded area



	X1	X2
Eins	sisheim study a	area
10%	0.01%	0.6%
50%	0.01%	2.25%
He	iwiller study ar	ea
10%	0.01%	0.6%
50%	0.1%	2.25%

From annendix 2

c - Effect of the spatial unit for analysis on the non GM area to be downgraded

The previous results have been obtained by using a field scale for analysis (Yellow boxes in figure 8). Using a smaller unit (100m²) to calculate the downgraded areas (white boxes plots in figure 8) may leads to a 20 to 35% decrease of rejected area. However, this decrease of rejected non GM area is sharply reduced:

- When a large part of non GM maize is downgraded (situations with 50% of GM maize and a maximum threshold of 0.01% for instance)
- When very few non GM maize is downgraded (situations with 25% of GM maize and a maximum threshold over 0.5% for instance). In this situation, implementing an intra-field unit for analysis may lead to a limited increase of rejected area (situations with a maximum threshold of 2.25% for instance).

The figure 10, established according to the graphs presented in appendix 2, shows a theoretical evolution of the part of downgraded non-GM maize area according to the maximum threshold for cross-pollination when considering field and intra-field units for analysis. Once again, two strategic values (x1 and x2) can be defined. Those values, which strongly depend on the characteristics of GM and non-GM area, delimit the threshold interval where implementing a coexistence management plan based on an intra-field unit for analysis may be of interest.

The graphs in appendix 2 show that the variability of non GM area where cross-pollination exceeds defined thresholds depends on the spatial unit we consider. In fact, this variability is clearly more important for a field spatial unit. Therefore, the allocation of GM and non GM varieties in the fields has a smaller impact on rejected non-GM area when considering an intra-field unit for analysis.

This difference of variability when considering one or another unit for analysis and the crossing of the curves in figure 10 are mainly due to the fact that, at an intra-field scale of analysis, we don't take into account the effect of GM grain dilution in the whole non GM field harvest which may be important at a field scale. On the one hand, implementing a management plan for coexistence based on an intra-field unit for analysis allows, under certain conditions, to decrease sensibly the downgraded non GM area, by rejecting only the areas where cross-pollination exceeds defined thresholds. On the other hand, for higher thresholds (0.9%), this smaller spatial unit for analysis also makes it more difficult to achieve the objective of 0% of non GM maize downgraded. Actually, very local high levels of GM presence due to cross-pollination (in the edge rows of non GM maize field for instance) are not diluted in the whole field harvest.

This phenomenon of GM grain dilution into non GM harvest is even more important in the case of a landscape scale for analysis. The table 5 shows that, by mixing the grains from all the non GM fields, thresholds of 0.4% are possible to implement if 10% of maize area is transgenic and even for high levels of maize in the AUA (85% near Ensisheim and 70% near Huningue). Furthermore, near Ensisheim, a threshold of 0.9% is also possible to implement for 85% of maize in the AUA and 50% of GM maize. It is not the case near Heiwiller. Even if maize presence is lower in this study area, the field characteristics lead to higher cross-pollination rates between GM and non GM varieties.

Nevertheless, whatever the study area, the field characteristics and the presence of GM maize, the landscape unit is not adapted to implement very stringent maximum thresholds for GM adventitious presence in non GM grain (below 0.1%).

<u>Figure 11</u>: Variation of the part of total non-GM area downgraded according to the delimitation of the area used for treatment (with or without edge fields)

The edge fields are defined in figure 5 Assumptions:

- Maize distributed according to: 2005 CAP declarations;
- Part of total maize area sown with GM varieties: A-10% and B-50%;
- Spatial unit for analysis: Field

A Ensisheim study area 2005 maize distribution 10% of GM maize 1.0 0.1 with edge fields
 without edge fields 0.0 80 part of downgraded non GM area part of downgraded non GM area 0.0 0.6 0.4 0.4 0.2 0.2 0.0 0.0 0.01% 0.1% 0.3% 0.4% 0.5% 0.6% 0.7% 0.9% thresholds



Heiwiller study area



22

These results insist on the strategic question of the scale adopted to appreciate the effect of sowing GM maize on non GM maize areas. However, the relevance of using a small spatial unit for analysis (intra-field or field) instead of a larger one (field or landscape) will be evaluated by farmers or storage organisms according to the savings of non GM grain forecasted but also according to the price of controls and the heaviness of sampling processes. Those points must be included into a coexistence management plan.

d - Effect of the study area delimitation and of the way to allocate GM maize (by farms or by fields) on the "non-GM" area to be downgraded

d.1. Effect of the delimitation of study area (with or without edge fields) on the "non-GM" area to be downgraded

The previous results have been calculated on the whole fields of each study area. However, the impact of the edge fields on downgraded non-GM maize area is sensible in both study case (figure 11). In fact, cropping area is less important around edge fields so that level cross-pollination with surrounding GM maize is generally lower. However, the impact of those edge fields is more important in Ensisheim case. In fact, as we evaluate the part of non GM area where GM presence is over defined threshold (and not a part of total number of non GM field), larger fields have a greater importance in this calculation than smaller ones. Furthermore, the larger the field is, the smaller the cross-pollination rate (dilution effect). Thus the fact that more than one field on two and most of the bigger fields (over 25 ha) is situated at the edge of Ensisheim study area (although, in Heiwiller study area, only one field on three is edge situated and bigger fields are situated in the centre area) may explain the bigger impact of edge fields in this case.

d.2. Effect of the way to allocate GM maize (by farms or by fields) on the "non-GM" area to be downgraded

The project work plan specifies that two scenarios must be implemented for the introduction of GM varieties in field patterns. In the first scenario, the maize is randomly introduced in the fields and, in the second one, it is randomly introduced in the farms. This last scenario aims at simulating an introduction of GM maize by a limited number of farmers and evaluating the consequences of this type of introduction on GM-non GM cross-pollination rates and on the extent of downgraded non-GM area.

Random introductions of GM maize in the farms lead most of time to a decrease of downgraded non GM area in comparison with a random introduction in fields (Fig 13 & appendix 3). In fact, this phenomenon can be observed for all the threshold, GM and non-GM levels in the study area of Heiwiller and in most of cases in the study area of Ensisheim.

However, this reduction of downgraded non-GM area is relatively small since it seldom exceeds 15% and remains, most of time below 10%. This small impact of the way to introduce GM maize in the landscape is very typical of the Alsatian context. As clearly shown on the figure 11, farm field patterns are scattered in both study areas. Thus, because of this field pattern characteristic, the spatial gathering of GM maize in case of an introduction by farm is limited and the decrease of cross-pollination between GM and non GM field also.

Furthermore, the fields of each farm are dispersed at a very large scale so that our study areas don't ever bear all the fields of one farm. This scattering of farms makes a common management between farmers necessary and particularly difficult in case of GM introduction in the landscape. We will have to keep that information in mind when building scenarios for coexistence.



Figure 12: Owner Identification of each field

Figure 13: Part of downgraded "non-GM" area considering two ways for the introduction of GM maize in the landscape: by fields or by farms

Assumptions:

- Maize distributed according to: 2005 CAP declarations;
- Part of total maize area sown with GM varieties: 10%
- Spatial unit for analysis: Field
- Total area used for treatment (with edge fields)



e - Impact of non GM and GM maize levels in the landscape on the non GM area to be downgraded and on the average GM adventitious presence in the total "non-GM" harvest

The figures 14 and 15 show the effect of different levels of GM and non GM maize in the landscape respectively on the average adventitious presence in the non-GM grain harvested in the whole landscape and on the non-GM downgraded area if a 0.9% maximum threshold is implemented (results for other thresholds are given in the annex 4).

It appears clearly on the figure 14 and 15 that the level of downgraded non GM area as well as the average GM adventitious presence in total non GM harvest rises sensibly and almost linearly when increasing the area in maize sown with GM varieties. This first result is clearly obvious but the graphs show also that those indicators (downgraded non GM area and average GM adventitious presence) rise when the non GM area increases for a constant percentage of maize sown with GM varieties. This rise is due to a higher density of maize fields and thus a global reduction of isolation distances between non GM and GM fields when increasing percentage of maize in AUA.

Once again a clear difference appears between Ensisheim and Heiwiller study cases. Actually, due to smaller fields near Heiwiller the median downgraded areas and the average GM adventitious presences are more important in this study area. Furthermore, the figures 14 and 15 show that for a constant level of GM maize in total maize area, the average GM adventitious presence in the silo and the downgraded "non-GM" area are bigger near Heiwiller for 50% and 75% of maize in the AUA than near Ensisheim for respectively 75% and 100% of maize in the AUA.

A general analysis of the graphs in annex 4 shows that, at a field scale:

- Maximum thresholds for cross-pollination of 0.01 and 0.1 % area not possible to implement without any changes in actual practices in both study areas. In fact, when implementing thresholds of 0.1% and 0.01%, median rejected area is over respectively 10% and 40% whatever the GM and non GM maize levels in the landscape, and over 50% and 90% if maize represents at least 50% of the AUA and transgenic maize at least 30% of total maize area.
- On the contrary, the non GM area downgraded in case of implementing a threshold of 2.25% for cross-pollination, remains low (<10%) even for high levels of non GM and GM maize in the field patterns (100% of maize and 70% of GM maize in Ensisheim study case and 75% of maize and 50% of GM maize in Heiwiller study area).
- For the other thresholds, they are most of time possible to implement with current practices, if only 25% of the AUA is sown with maize (GM presence of 70% for Ensisheim case and 30 to 50% in Heiwiller case), or if only 10% of maize area is transgenic (from 50 to 100% of maize in the AUA according to the threshold considered).

Figure 14: Average GM adventitious presence in harvested "non-GM" grains evaluated at a landscape scale according to the percentage of maize in the AUA and the percentage of GM maize in the total maize area



Part of AUA sown with maize

Figure 15: Part of "non-GM" area where GM adventitious presence evaluated at a field scale exceeds 0.9% according to the percentage of maize in the AUA and the percentage of GM maize in the total maize area See also appendix 4



Part of AUA sown with maize

Comparing the figure 14 with the graphs in annex 4, we notice that the average adventitious presence in the non-GM grain harvested in the whole landscape is well correlated with the "non-GM" downgraded area whatever the threshold we consider. Additional studies have shown that this correlation is observed whatever the spatial unit we used to assess the downgraded "non-GM" area (Figure 16).

Thus, for each maximum threshold for GM adventitious presence, we calculated the maximum part of non-GM area downgraded which doesn't lead to a rejection of the whole non-GM grain harvested in the landscape if it is mixed in the same silo. The results are presented in table 6.

This table shows that mixing all the non-GM harvest in a same silo is an interesting strategy only if the part of total non-GM maize area potentially rejected is limited. For a threshold of 0.9%, this potentially rejected area must represent less than 40% of non-GM maize area if this rejected area is calculated according to a field spatial unit and less than 30% if it is calculated according to an intra-field unit.

The maximum potentially rejected non-GM area that can remains non-GM if diluted in the non-GM grain harvested on the whole landscape depends not only on the spatial unit for analysis but also on the threshold we consider. Actually, this area decreases with the threshold. That's why a landscape scale strategy mixing all the non-GM harvests in the same silo is not valuable if maximum threshold for GM adventitious presence in the non GM harvests is too low (0.1% and 0.01%).

Figure 16: Graphic representation of the correlation between GM adventitious presence in the total non-GM harvest and the part of downgraded non GM area if implementing a 0.9% maximum threshold for GM adventitious presence

Assumptions:

- Study area: Heiwiller
- Simulations used: batch 1



GM adventitious presence in the total non GM harvest

Table 6: Maximum part of total maize area over defined thresholds that doesn't lead to
a rejection of the whole non-GM grain harvested in the landscape if it is mixed in the
same silo

Spatial unit for analysis	Maximum threshold for GM adventitious presence			
	0.90%	0.60%	0.40%	0.10%
	Ensisheim study area			
Field	0.38	0.34	0.33	0.21
Intra-field	0.28	0.24	0.22	0.15
	Heiwiller study area			
Field	0.32	0.32	0.27	0.23
Intra-field	0.24	0.22	0.19	0.16

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Appendixes

Appendix 1: Variation of the part of total non-GM area downgraded according to the delimitation of the area used for treatment (with or without edge fields) and the maximum threshold for GM adventitious presence

Assumptions:

- Maize distributed according to: 2005 CAP declarations and 2000 agricultural census;
- Part of total maize area sown with GM varieties: 10% and 50%;
- Spatial unit for analysis: Field.




Appendix 2: Impact of the spatial unit for analysis on the non GM area to be downgraded

Three spatial units:



The field and the intra-field unit (see graphs) The landscape unit (whole non-GM maize area) (see tables)







Appendix 3: Part of downgraded "non-GM" area considering two ways for the introduction of GM maize in the landscape: by fields or by farms

Assumptions:

- Maize distributed according to: 2005 CAP declarations and 2000 agricultural census;
- Part of total maize area sown with GM varieties: 10% and 50%;
- Spatial unit for analysis: Field
- Total area used for treatment (with edge fields)





Appendix 4: Impact of non GM and GM maize levels in the landscape on the non GM area to be downgraded

Assumptions:

- Percentage of maize in AUA: 25%, 50%, 75% and 100%;
- Part of total maize area sown with GM varieties: 10%, 30%, 50% and 70%;
- Spatial unit for analysis: Field
- Total area used for treatment (with edge fields)













D 7.2 Identification of critical points from validated results - Effect of the structural variables on GM admixture in non GM harvest based on simulation results -

Switzerland case study - maize

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Figure 1: Geographic situation of Swiss study area

Figure 2: Main characteristics of the study field pattern



I. Main objectives of the study

This study aims at analysing the trans-boundary coexistence issues. Actually, neighbouring countries may manage coexistence between GM and non GM maize differently according, for instance, to the type of maize produced (organic maize, maize for starch, semolina or animal feeding...) and the purity required for the harvest (0.01%, 0.1%, 0.5%, 0.9% ...). However, even if a regulation for coexistence does exist in each country, common rules may not have been defined in the frontier areas. Thus, it is of utmost importance to assess the risks of cross-pollination between countries in those frontier areas.

Moreover, this study gives also information on the pertinence to organize GM and non-GM production into clusters. In fact, the coexistence issues at the limits of each cluster will be very similar to the ones at the frontier between countries.

It is admitted that regrouping non GM field in space allows to achieve higher purity rates. This strategy is commonly implemented in seed production for instance. However, it is also assumed that those high purity rates are mainly due to pollen protection from neighbouring fields. Thus, in a first step, we will verify this assumption by assessing this pollen protection ensured when grouping non-GM fields (results presented in paragraph III.1).

In a second step, we will try to identify the non-GM fields at risk in terms of cross-pollination with GM maize if a GM cluster is grown in non-GM area and if a non-GM cluster is grown in a GM area (results presented in paragraph III.2).

In a third step, we will assess the GM presence in non-GM maize in Switzerland if GM maize is grown in France with no trans-boundary rules for coexistence (results presented in paragraph III.3).

II. Description of the simulations implemented

II.1. Material

II.1.a. Description of simulation area

This study is based on simulations run on a frontier area between Alsace French region and Switzerland. More precisely, we focused on the Rodersdorf territory situated in Basel-Town Swiss region. We have chosen this area because of its geographic situation. Actually, Rodersdorf territory is very enclosed into Alsatian region (see figure 1), which may lead to intense gene flow between countries and to great trans-boundary coexistence issues.

FiBL¹ has provided Swiss field pattern as well as aerial photographs which were used to draw French field patterns 1 km around Rodersdorf territory. The field pattern used for simulations is presented in figure 2. This figure shows only the fields of AUA where maize can be grown. The Fields are a bit larger in Switzerland (average area: 2.2 ha) than in France (average area: 1.6 ha). However, the variability of field area is a more important in France (standard deviation: 1.8 ha) than in Switzerland (standard deviation: 1.5 ha). Furthermore, the whole field pattern situated in the south of Sundgau region in Alsace is quite similar to the one met in Heiwiller study area (North of Sundgau). In fact, both field patterns present an average field area of about 1.8 ha and a standard deviation moving between 1.8 and 1.9 ha. A comparison of the results obtained in each region will be of interest in order to assess the pertinence of a cluster strategy to isolate GM and non-GM maize.

¹ Forschungsinstitut für Biologischen Landbau



Figure 3: Wind distribution (Source: MeteoSchweiz)

The presence of maize in AUA is higher in the south of Sundgau (50% of AUA is grown with maize, 35 to 40% for grain production and 10 to 20% for animal feeding) than in the Rodersdorf territory (maize represent less than 20% of the AUA).

II.1.b. Meteorological data

Wind distribution used for simulation has been established with meteorological data recorded near Basel Town by MeteoSchweiz during the months of July and August between 1986 and 2005. The figure 3 shows that westerly wind is the most frequent. Thus, this real wind distribution does not give maximum cross-pollination rates between the two neighbouring territories. In fact, in the west part of study area Swiss fields are isolated from French fields by a distance of at least 300 to 800 meters (see Figure 2).

The spatial distribution of the fields presented on figure 2 shows that the winds having the highest potential for trans-boundary cross-pollination are North-East and South-West ones. That's why, in order to identify the fields at risks in critical situations, we will also simulate trans-boundary cross-pollination considering only South-West or North-East winds. For each direction the same average wind velocity of 3 m/s has been considered.

II.1.c. Distribution of conventional and transgenic maize in the fields

As this study only considers trans-boundary coexistence issues, we adopted a scenario where each country produce only one type of maize (GM or conventional). Thus, GM and non GM maize will never coexist in a same country and for each simulation we will define a "GM country" producing GM maize and a "non-GM country" producing conventional maize.

We ran in this study two batches of simulations differing by GM and non GM levels in AUA:

<u>Batch 1</u>: This batch aims at assessing pollen protection on each non GM field, at identifying fields where cross-pollination may exceed various defined levels and at assessing the GM presence in Swiss non-GM harvest when GM maize is grown in France. To do it we considered risky situations where the "GM country" (successively France and Switzerland) produces GM maize on 100% of the AUA.

Concerning maize distribution in "non-GM country", we ran:

- One simulation with 100% of AUA sown with conventional maize. That simulation assesses the cross-pollination for each non-GM field if pollen protection is minimal.
- n simulations (n = number of fields in "non-GM country": 92 for Switzerland and 261 for France) with one single field sown with maize. Those simulations assess the crosspollination for each non-GM field if pollen protection is minimal.

<u>Batch 2</u>: This batch aims at assessing cross-pollination levels on Swiss non-GM fields if various levels of GM maize are cultivated in France: 25% (40 repetitions), 50% (40 repetitions), 75% (40 repetitions) and 100% (1 possibility) of the AUA. Cross-pollination rates have been calculated for each non-GM field in Switzerland considering a minimum and a maximum pollen-protection.

II.1.d. Crop practices and varietal characteristics

As for Alsatian study cases, the following assumptions have been made for those first simulations:

- GM and non-GM varieties flower at the same time;
- GM and non-GM varieties produce similar amounts of pollen;
- GM and non-GM varieties are sown at similar densities.

<u>Box 1</u> : Descri	ption of the indicator used to assess pollen protection on each non-GM field <pre>PPmax[i] = (CPmax[i] - CPmin[i]) / CPmax[i]</pre>
PPmax[i]:	Maximum pollen protection on field i
CPmax[i]:	Maximum Cross-Pollination with GM maize in non-GM field i (non-GM maize is grown in the field i only: no pollen protection)
CPmin[i]:	Minimum Cross-Pollination with GM maize in non-GM field i (non-GM maize is grown in all the fields of the non-GM country: maximum pollen protection)

II.2. Methodology for the treatment of model outputs

II.2.a. The output indicator

By using the simulations from batch 1 where non-GM maize is cultivated in Switzerland only, we have assessed for each Swiss non-GM field an indicator we called "maximum pollen protection". This indicator can be defined as the maximum reduction of cross-pollination with GM maize due to the emission of protective pollen by neighbouring non-GM fields. It has been calculated as described in box 1.

Afterwards, those maximum cross-pollination rates (batch 1) have been used to identify the fields where cross-pollination may exceed various defined levels and to calculate the maximum percentage of AUA where growing a non-GM maize is at risk in terms of coexistence.

Finally, GM-presence in Swiss non-GM maize has been assessed by the two indicators already used in Alsatian study case: the non-GM area where cross-pollination exceeds defined thresholds and the GM presence in total non-GM harvest.

- In a first step, those two indicators have been assessed in a risky situation (Batch 1: GM maize grown on 100% of French AUA) for various levels of non-GM maize in Swiss AUA (15%, 25%, 50%, 75% and 100%).
 The median value of each indicator and its variability have been assessed by choosing at random the fields sown with non-GM maize (100 repetitions have been done for each non-GM maize level in AUA).
 Then, non-GM maize fields have been chosen specifically in order to assess (for each non-GM maize level) the minimum and maximum values of each indicator.
- In a second step, the indicators have been assessed for various levels of GM maize in French AUA (25%, 50%, 75% and 100%) by using the results of simulations from batch 2. Those results have been used to assess the minimum and maximum values of crosspollination risk for each non-GM maize level previously defined.

II.2.b. The thresholds

For the same reasons than those described in Alsatian study case, we considered various maximum thresholds for cross-pollinations: 0.01% (asked by organic farmers and semolina industries), 0.1% (asked by starch industries), 0.4% (if 0.5% of impurities in the seeds), 0.6% (if 0.3% of impurities in the seeds), 0.9% (legal threshold) and 2.25% (corresponds to a 0.9% presence in case of a PCR analysis according to the assumptions described in Alsatian report).

II.2.c. The spatial unit for analysis

Two different units for analysis have been used to calculate the output indicator in this study: The whole study area and the field unit (those units for analysis are described in the table 4 of the Alsatian report). Table 1: Classification of fields according to the maximum pollen protection they can benefit

Assumptions:

- Non-GM country: Switzerland
- 3 Wind distributions: Real, South-West wind only, North-East wind only

		Max	imum Poller	n protection	level
_		<5%	<10%	<15%	<20%
ion	Real	39%	83%	99%	100%
Wind tribut	South-West	35%	79%	97%	100%
dis	North-East	38%	85%	98%	100%

Figure 4: Description of the fields according to the maximum pollen protection they can benefit

Assumptions:

- Non-GM country: Switzerland
- Real wind distributions



III. Results

III.1. Assessment of pollen protection ensured by clustering non-GM fields

The table 1 shows that the pollen protection hardly depends on the wind distribution and is quite feeble. In fact, for the major part of the fields (about 80%) the maximal reduction of cross-pollination due to pollen protection remains below 10% and almost never exceeds 15%.

Furthermore, we observed that generally, the biggest maximal pollen protections are observed on fields presenting low levels of cross-pollination with GM maize. Thus, this maximal pollen protection is most of time below 9% and 2% when cross-pollination rates are respectively over 0.5% and 1%. Those values of 2% 9%, 10% and even 15% are still reasonable and in accordance to the precision of MAPOD model.

The potential pollen protection for each field depends on:

- The characteristics of the field. The factor having the main impact on potential pollen protection is the area of the receptive field. In fact, the proportion of foreign pollen in the pollen cloud is smaller over large recipient fields than over small ones. In the Swiss study case, a graphic analysis showed that the maximum reduction of cross-pollination due to pollen protection remains below 11%, 9% and 7% in fields which areas are respectively over 2, 3 and 4 ha. However, other field characteristics not taken into account in this study like their shape may have sensible impact on pollen protection level.
- The characteristics of the environment close to the field. The figure 4 shows clearly that the fields presenting the lowest pollen protection (<5%) are generally at the edge of the non-GM production area. In fact, the density of non-GM maize around those fields is smaller as well as the amount of protective pollen. Furthermore the non-GM fields at risk contiguous to GM ones are very little protected by pollen emitted by non-GM neighbouring fields (pollen protection below 2%), particularly if the wind blows from the GM field to the non-GM one.

As a conclusion, the protection ensured by pollen emitted by non-GM neighbouring fields remains quite limited. Thus, the reduction of cross-pollination observed when grouping non-GM fields is probably more the results of a general increase in isolation distance than of this pollen protection.

In the following study, we will use for data processing the minimum cross-pollination rates (maximum pollen protection) if presence of non-GM maize in non-GM country is over 75% and the maximum cross-pollination rates (no pollen protection) in the other cases.

III.2. Identification of the fields at risk in terms of cross-pollination

In this part, we have used simulations from batch 1 to identify in Switzerland (GM maize cultivated in France) and in France (GM maize grown in Switzerland) all the non-GM fields where the cross-pollination rate may exceed various thresholds. Growing non-GM maize in those fields would require information on crop cultivated in neighbouring area and probably the implementation of adapted coexistence strategies. As the part of maize in the AUA remains below 75% in Alsace (50% of the AUA) and near Rodersdorf (18% of AUA), we considered for each field maximum cross-pollination rates (risky situation where pollen protection is minimal).

Figure 5: Maximum cross-pollination assessed for a real wind distribution

Assumption: GM maize cultivated in all the fields of the GM country



Figure 6: Part of Swiss AUA where cross-pollination rates with GM fields may exceed various thresholds

Assumption:

GM maize cultivated in all the French fields



III.2.a. If GM maize is grown in a non GM area

It appears on the figure 5 that the cross-pollination risk depends on the distance between GM and non-GM fields, on the area and shape of non-GM field but most of all on the spatial position of non GM field according to the main wind directions. In fact, the figure 5.A shows that the fields situated in the North-East of GM area are clearly more cross-pollinated than those situated in the South-West. It is due to the fact that South-West winds are seven times as frequent as North-East winds (see figure 3). Thus, the identification of the fields at risk in terms of cross-pollination with GM maize will clearly depends on the wind distribution. The figure A in appendix 1 shows that a wind blowing from South-West during all the flowering period will clearly disadvantage fields on the North-East of GM area. On the contrary, 0.01% threshold will be achievable in most of South-West non-GM fields without implementing any strategy for coexistence.

The first table in Appendix 2 gives a brief description of the French fields where crosspollination may exceed various thresholds. For a real wind distribution, cross-pollination rates over 0.9% can be observed in a limited number of fields all contiguous to GM maize. On the contrary, the cross-pollination levels remains sensible some hundreds of meters away from GM area. Thus, implementing thresholds of 0.1 or 0.01% will be possible without any strategies for coexistence only in the fields separated to GM area by minimal distances moving respectively between 150m and 300m and between 350m and 700m according to the position of the recipient non-GM field and the main wind directions. Furthermore, in a risky situation (fields situated on the North-East of GM area when a South-West wind blows during all the flowering period), all the fields situated at less than 100m away from GM area may require coexistence strategies to achieve cross-pollination rates below 0.9%.

III.2.b. If non GM maize is grown in a GM area

The figure 5.B and the figure B in appendix 1 show once again the impact of wind distribution on spatial layout of the fields at risk in terms of cross-pollination. In fact, the South-West wind gives more contrasted results than a real wind distribution. Indeed, the maximum cross-pollinations calculated at the scale of the field move between 0.001 and 4.59% for a South-West wind and only between 0.017% and 2.75% for a real wind distribution.

However, the figure 6 shows that for most of thresholds, a South-West wind increase the risk of cross-pollination since the part of AUA where cross-pollination may exceed various thresholds is generally bigger in this case than for a real wind distribution. This figure also shows that grouping all the non-GM maize into a 200 ha area is generally not sufficient to implement maximum thresholds for cross-pollination below 0.1%. On the contrary, this strategy appears well adapted to implement higher thresholds (over 0.4%) since the maximum part of AUA where a coexistence strategy may be necessary remains feeble (below 20%).

The characteristics of the Swiss fields at risk in terms of cross-pollination are given in the second table of the appendix 2. Comparing this table with the first table (appendix 2), we can notice that the characteristics of the fields at risk are quite similar in Switzerland and in France (see paragraph III.2.a). It is certainly due to the fact that wind distribution is the same and field characteristics are quite similar.

Those first results show that the strategy of grouping the GM and non-GM crops into clusters may be of value if the size of the cluster is adapted to the threshold to be implemented. For instance, in our case, a non-GM cluster of 200 ha does not allow to achieve purity rates over 99.99%. However, whatever the threshold considered some fields will be at risk in terms of cross-pollination and rules for coexistence are necessary on those fields. The characteristics of the fields at risk are similar in both countries and their identification clearly depends on the environment (distance to the GM area and wind distribution).

Figure 7: Part of non-GM Swiss area where GM presence due to cross-pollination exceeds...

A: ... a threshold of 0.1%

B: ... a threshold of 0.9%



Assumptions:

- GM maize is grown on 100% of the French AUA
- Real wind distribution

Figure 8: Mean cross-pollination in Swiss non-GM maize area

Assumptions:

- GM maize is grown on 100% of the French AUA
- Real wind distribution



<u>Table 2</u>: Part of total number of simulations where cross-pollination rate calculated at a landscape scale exceeds various thresholds

Assumptions:

- GM maize is grown on 100% of the French AUA
- Real wind distribution

		Part of	swiss A maize	\UA sov (in %)	wn with
		15	25	50	75
	0.01	100%	100%	100%	100%
(% נ	0.1	80%	94%	98%	99%
ds (ir	0.4	10%	2%	0%	0%
lohse	0.6	1%	1%	0%	0%
Thr€	0.9	1%	0%	0%	0%
	2.25	0%	0%	0%	0%

III.3. Assessment of GM presence in Swiss maize if no rules are implemented to reduce trans-boundary gene flow

In the part III.2, we worked on extremely pessimistic situations in order to give an exhaustive identification of the fields at risk in terms of coexistence. Thus, the results presented in figure 6 calculated on a particularly risky situation (100% of French AUA grown with GM maize and a minimum pollen protection on each non-GM field) are not representative of French and Swiss crop allocation.

In this part, we have tried to give a more realistic assessment of the GM presence in Swiss non-GM maize if no trans-boundary rules for coexistence are implemented. To do it, we moved the level of non-GM maize in Swiss AUA between 15% and 100% (Paragraph III.3.a) and the level of GM maize in French AUA between 25% and 100% (Paragraph III.3.b).

III.3.a. Impact of non-GM maize level in Swiss AUA on GM presence in Swiss maize

The Figure 7 shows on two maximum thresholds for cross-pollination (0.1% and 0.9%) that the average part of non-GM area downgraded does not depend on the percentage of maize in Swiss AUA. In fact, statistical tests showed that the means obtained with 15%, 25%, 50% and 75% of maize in Swiss AUA are not different from the one obtained if maize is cultivated on the whole Swiss area. Thus, to determine the average parts of non GM area where cross-pollination exceeds various thresholds, we can refer to the figure 6.

However, the variability of the simulation outputs (due to maize allocation) clearly increases when the maize area in Switzerland decreases. Indeed, if we consider a 0.1% maximum threshold, the downgraded non-GM area in Switzerland moves from 0% to 100% if maize is cultivated on less than 25% of the AUA and only from 15% to 50% if maize is cultivated on 75% of the AUA. Furthermore, the probability to downgrade more than 10% of non-GM maize when implementing a 0.9% threshold is null if at least 50% of AUA is cultivated with maize and exceeds 15% if maize is grown on only 15% of the Swiss AUA.

Those results are confirmed if we consider the GM presence in total non-GM harvest (see figure 8). The mean is quite stable and the variance increases when non-GM area decreases. The table 2 shows that even in a risky situation (GM maize cultivated in all the French AUA), grouping the non-GM fields and harvesting the whole non-GM grain without any segregation is an efficient strategy for thresholds over 0.4%, but not for thresholds below 0.1%. In this last case, the probability to reject the grain harvested in the whole Swiss area is over 80%.

Comparing for each simulation the part of downgraded non-GM area and the average GM presence in the grain collected in the whole Swiss field pattern, it appeared that mixing the whole grain in the same silo is of interest only if the part of downgraded non-GM area is limited (below 15% and 25% of the total Swiss non-GM maize area for maximum thresholds of respectively 0.1% and 0.4%).

III.3.b. Impact of GM maize level in French AUA on GM presence in Swiss maize

The tables 3 are based on simulations where non-GM maize is grown on all the fields in Switzerland. Nevertheless, according to the previous paragraph (III.3.a), those results should be the same whatever the presence of maize in Swiss AUA if we considered an average downgraded non-GM area.

Tables 3: Part of total number of	simulations where the Swiss	downgraded non GM-area remains:
		5

	A: Nu	II					В	: Belo	w 5% (of total	non-G	SM are	a
Part of French	Max	imum th	reshold	for cros	s-pollin	ation	Part of French	Max	imum th	reshold	for cros	s-pollin	ation
GM maize	0.01%	0.1%	0.4%	0.6%	0.9%	2.25%	GM maize	0.01%	0.1%	0.4%	0.6%	0.9%	2.25%
25%	NP	NP	20%	23%	33%	73%	25%	NP	23%	ET	ET	ET	ET
50%	NP	NP	NP	3%	8%	25%	50%	NP	NP	48%	93%	ET	ET
75%	NP	NP	NP	NP	NP	3%	75%	NP	NP	NP	53%	ET	ET
100%	NP	NP	NP	NP	NP	NP	100%	NP	NP	NP	NP	ET	ET
	C: Be	low 10)% of to	otal no	n-GM	area): Belo	w 20%	of tota	al non-	GM ar	rea

Part of French	Max	imum th	reshold	for cros	s-pollina	ation		
GM maize	0.01%	0.1%	0.4%	0.6%	0.9%	2.25%		
25%	NP	73%	ET	ET	ET	ET		
50%	NP	3%	ET	ET	ET	ET		
75%	NP	NP NP 88% ET ET ET						
100%	NP	NP	NP	ET	ET	ET		

Part of French	Maximum threshold for cross-pollination							
GM maize	0.01%	0.01% 0.1% 0.4% 0.6% 0.9% 2.25%						
25%	NP	ET	ET	ET	ET	ET		
50%	NP	25%	ET	ET	ET	ET		
75%	NP	NP NP ET ET ET ET						
100%	NP	NP	ET	ET	ET	ET		

Legend: NP: Not Possible ET: Every Time

Assumptions:

- Non GM maize is grown on 100% of Swiss • AUA
- Real Wind distribution

Tables 4: Part of total number of simulations where GM adventitious presence in total Swiss non-GM harvest exceeds various thresholds

A: Mean GM adventitious presence

Part of French	Max	imum th	reshold	for cros	s-pollina	ation
GM maize	0.01%	0.1%	0.4%	0.6%	0.9%	2.25%
25%	NP	ET	ET	ET	ET	ET
50%	NP	38%	ET	ET	ET	ET
75%	NP	NP	ET	ET	ET	ET
100%	NP	NP	ET	ET	ET	ET

B: Minimal GM adventitious presence

Part of French	Max	imum th	reshold	for cros	s-pollina	ation
GM maize	0.01%	0.1%	0.4%	0.6%	0.9%	2.25%
25%	ET	ET	ET	ET	ET	ET
50%	8%	ET	ET	ET	ET	ET
75%	NP	ET	ET	ET	ET	ET
100%	NP	ET	ET	ET	ET	ET

Legend: NP: Not Possible ET: Every Time

C: Maximum GM adventitious presence

Part of French	Max	imum th	reshold	for cros	s-pollina	ation		
GM maize	0.01%	0.1%	0.4%	0.6%	0.9%	2.25%		
25%	NP	8%	90%	ET	ET	ET		
50%	NP	NP	25%	68%	ET	ET		
75%	NP	NP NP NP 13% ET ET						
100%	NP	NP	NP	NP	NP	FT		

Assumptions:

- Non GM maize is grown on 15% of Swiss AUA
- Real Wind distribution

Tables 3 show that the possibility to make coexist GM and non-GM maize clearly depends on the part of non-GM maize that is accepted to be downgraded by the farmers. If coexistence must be ensured on each field (table 3.A), even a 2.25% maximum threshold is hard to implement with current level of maize in French AUA (50%). On the contrary, in the same conditions, if 10% or 20% of downgraded non-GM area is tolerated, threshold of 0.4% is always feasible and threshold of 0.1% may be achieved under certain conditions of GM maize distribution (tables 3.C & D). However, threshold of 0.01% is not feasible without implementing any strategy for coexistence whatever the risk accepted by farmers.

We saw in the previous paragraph that a bit more than 20% of non-GM grain downgraded can be mixed to the whole non-GM harvest without leading to the rejection of this harvest. That is the reason why the mean GM adventitious presence due to cross-pollination and calculated at a landscape scale never exceeds 0.4% (table 4.A). Furthermore 0.1% threshold is always possible to implement if GM maize is grown on less than 25% of French AUA.

However, if we consider current levels of maize in Swiss AUA (15%), GM presence in non-GM grain assessed at a landscape scale may vary strongly according to the distribution of maize in Swiss field pattern.

In fact, if maize fields in Switzerland are well protected from French GM maize, this GM presence may remain below 0.1% whatever the level of GM maize in French AUA and below 0.01% if GM maize is grown on less than 25% of French AUA (Table 4.B).

On the contrary, if non-GM maize fields are situated at the edge of the Swiss territory and if maize is grown on 50% of French AUA, maximum thresholds of 0.6% and 0.4% are not possible to implement in respectively 75% and 30% of the cases.

III.3.c. Impact of grouping non-GM field on GM presence in non-GM harvest

In order to assess the pertinence to organize non-GM production into clusters, we have compared some of the results obtained in this study with some obtained in the Sundgau region of the Alsatian case. Those two study areas can be used for comparison since their characteristics are very similar (see paragraph I.1.a).

To perform this comparison, the presence of maize in total AUA and the percentage of GM maize in total maize area must be equal in both study cases. That is why we focused on two situations:

- Situation 1: non-GM maize is grown on 100% of Swiss AUA and GM maize is grown on 25% of French AUA. Under those conditions, maize area represents 50% of total AUA and GM maize is about 30% of total maize area.
- Situation 2: non-GM maize is grown on 100% of Swiss AUA and GM maize is grown on 100% of French AUA. Under those conditions, maize area represents 100% of total AUA and GM maize is about 70% of total maize area.

The table 5 shows that organizing non-GM production into clusters can lead to a 90% reduction of the mean cross-pollination rate evaluated at a landscape scale. Furthermore, it appears that the part of non-GM area downgraded is sharply reduced when grouping non-GM fields if the maximum threshold for cross-pollination is over 0.1%. However, purity rates over 99.99% remain impossible to achieve with or without any spatial organization of GM and non-GM fields.

Table 5: Impact of grouping non-GM field on GM presence in non-GM production

Situation Study case		Mean cross- pollination rate	. Part of non-GM area where cross-pollination te exceeds:						
Situation	Study case	in the whole non- GM grain	0.01%	0.1%	0.4%	0.6%	0.9%	2.25%	
1	Alsace	0.475%	99%	69%	32%	23%	14%	3%	
1	Switzerland	0.053%	62%	7%	2%	1%	1%	0%	
2	Alsace	2.098%	100%	100%	97%	94%	84%	35%	
2 Switzerland		0.208%	100%	34%	11%	7%	5%	1%	

Situation 1: Maize grown on 50% of total AUA

GM maize grown on 30% of total maize area

Situation 2: Maize grown on 100% of total AUA GM maize grown on 70% of total maize area

Alsatian study case: No cluster Organization of non-GM maize production Swiss study case: Non-GM maize grown in a cluster Appendices

Appendix 1: Maximum cross-pollination assessed for a South-West wind

Assumptions: GM maize cultivated in all the fields of the GM country



<u>Appendix 2</u>: Rapid description of the characteristics of the fields where crosspollination rates may exceed various thresholds

The results are given for a real wind distribution and for a wind blowing all the time from the South-West to the North-East.

Threshold	Real wind distribution	South-West wind
2.25%	North-East fields contiguous to GM maize & Area < 0.5ha South-West fields contiguous to GM maize & particullarly thin	North-East fields contiguous to GM maize South-West fields contiguous to GM maize & particullarly thin
0.9%	North-East fields contiguous to GM maize South-West fields contiguous to GM maize & Area < 2ha	Fields less than 100 to 150 m North-East of GM maize South-West fields contiguous to GM maize & particullarly thin
0.6%	Fields less than 50 m North-East of GM maize South-West fields contiguous to GM maize & Area < 2ha	Fields less than 250 m North-East of GM maize South-West fields contiguous to GM maize & particullarly thin
0.4%	Fields less than 50 m North-East of GM maize South-West fields contiguous to GM maize & Area < 2ha	Fields less than 350 m North-East of GM maize South-West fields contiguous to GM maize & particullarly thin
0.1%	Fields less than 300 m North-East of GM maize Fields less than 150 m South-West of GM maize	Fields less than 700 m North-East of GM maize South-West fields contiguous to GM maize & Area < 1ha
0.01%	Fields less than 700 m North-East of GM maize Fields less than 350 m South-West of GM maize	All the North-East fields South-West fields contiguous to GM maize

Situation 1: A GM cluster (Switzerland) is grown in non-GM area (France)

Situation 2: A non-GM cluster (Switzerland) is grown in a GM area (France)

Threshold	Real wind distribution	South-West wind
2.25%	South-West fields contiguous to GM maize & Area < 2ha	South-West fields contiguous to GM maize
0.9%	South-West fields contiguous to GM maize & Area < 3ha	Fields less than 100 m South-West of GM maize
0.6%	Fields less than 50 m South-West of GM maize	Fields less than 250 m South-West of GM maize
0.4%	Fields less than 70 m South-West of GM maize North-East fields contiguous to GM maize	Fields less than 350 m South-West of GM maize
0.1%	Fields less than 300 m South-West of GM maize Fields less than 200 m North-East of GM maize	Fields less than 750 m South-West of GM maize North-East fields contiguous to GM maize
0.01%	All the fields	All the fields with a few exceptions

D 7.2 Identification of critical points from validated results - Effect of the structural variables on GM admixture in non GM harvest based on simulation results -

Aragon case study - maize

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Table of contents

I. Main objectives of the study		
II. Description of the simulation implemented	3	
 <i>II.1. Material</i> II.1.a. Description of the simulation area II.1.b. Meteorological data II.1.c. Relative share of maize crops and distribution pattern in th area 5 	3 3 e agricultural used	
II.1.d. Crop practices and varietal characteristicsII.1.e. Maize volunteers	7 7	
II.2. Methodology for the treatment of model outputs	9	
III. Results	11	
III.1. Effect of various rates of transgenic varieties introduction in maize in Aragon and dynamic of flowwering	real allocation of 11	
III.2. Effect of maize volunteers (with flowering periods synchrono	us) 15	
III.3. Effect of the geographical and inter-annual variability of the (no volunteers / synchrony of flowering)	wind characteristics 15	
 III.4. Effect of the landscape (no volunteers / synchrony of flowerin III.4.a. Comparison between Gurrea de Gallego and Sariñena III.4.b. Comparison between Alsace and Aragon 	ng) 19 19 19	
IV. Conclusion on Aragon case	21	
Bibliography	21	
Appendixes	22	

I. Main objectives of the study

The overall objective of SIGMEA is to assess the impacts of introduction of GM maize (Bt maize) in selected SIGMEA regions. Several regional case studies have been chosen for maize (Alsace (France), Basel-Town Swiss region and Aragon (Spain)) in order to cover various situations (landscape fragmentation, trading and farming systems as well as agricultural practices). We assessed the effect of regional structural variables on the GM adventitious presence in non GM harvest:

- <u>Level of maize crop</u>: The level of maize in agricultural used area (AUA) is quite sensitive to the region. It is much higher in Alsace than in Aragon. We assessed the effect of share of maize crops in total agricultural used area.
- <u>Synchrony in flowering time</u>: In the Alsatian case study and for a majority of the simulations implemented in Aragon, we assumed that GM and non GM varieties flower at the same time. The synchronization of pollen dispersal and silking has been demonstrated to be crucial in determining the extent of out-crossing in maize. This assumption leads us to study the worst-case. Nevertheless, the flowering period is quite large in Aragon. We assessed the effect of flowering time lag between GM and non GM maize, without any containment.
- <u>Presence of maize volunteers:</u> Under Alsatian climatic conditions, the presence of maize volunteers is practically non-existent, due to the low resistance of maize to frost. Nevertheless, it cannot be considered as negligible in Aragon. This study aims at studying the effect of various levels of cross-pollination rate due to maize volunteers.
- <u>Distribution of wind direction and speed:</u> Maize pollen is essentially dispersed by wind. Hence MAPOD takes into account the distribution of wind direction and wind speed. We assessed the effect of the variability of the wind characteristics (direction and speed) on the GM adventitious presence in non-GM maize, taking into account various meteorological stations, over different years.

Last, the Aragon case study gave the opportunity of increasing the number of different landscapes on which simulations are implemented. We assessed the effect of the landscape on the GM adventitious presence in non GM harvest, other things being equals. By "landscape", we mean the field pattern (number, area and shape of the fields) and the wind characteristics (distribution of wind direction and wind speed).



	-							
Total area						1081 ha		
Number of fields						106		
Mean area						10.20 ha		
20	Distributio	on of fields	accordi	ng to t	their are	a		
10 - Isoco	0.5.11 [1:2] [2:3] [3:4]	[4:5] [5:6] [6:7]	19:51 19:51	(9;10) 0:151	15/201 10:301	los:os		

Gurrea study area Total area 964 ha Number of fields 489 Mean area 1.97 ha Distribution of fields according to their area Part of total number of fields (%) 8 2 (0.0.5) (0.5) (1.2) (2.3) (3.4) (4.5) (5.6 area (ha)



<u>Figure 1.</u> Field patterns considered for simulations

II. Description of the simulation implemented

II.1. Material

II.1.a. Description of the simulation area

Two small areas, in the municipalities of Gurrea de Gallego and Sariñena, were chosen to implement simulations.

From the database of SIGPAC¹, FEGA² provided an island pattern, with the delimitations of the production islands. A production island is defined as a group of contiguous fields owned by the same farmer and limited by permanent physical limits. The surface of the crops grown between 2003 and 2004 was known for every production island. According to the production data and aerial photographs (orthophotos), the island pattern was reshaped in order to delimitate all the fields. All the fields of the two selected areas are irrigable.

The field patterns used for Aragon simulations are presented in Figure 1. They are quite contrasted in terms of field area. In Gurrea de Gallego, the area is divided into a lot of small fields (in average 2 ha), whereas the fields are bigger and more heterogeneous in terms of area in Sariñena (in average 10 ha). According to the field area distribution, the types of field patterns are quite similar in Aragon and Alsace: we distinguish a first type of large fields (Ensisheim and Sariñena) and a second type made up of smaller fields (Heiwiller and Gurrea de Gallego) (Alsace field patterns in Appendix 1).

II.1.b. Meteorological data

Direction and mean speed of the wind used for the simulations have been established with meteorological data recorded between the first of July and the fifteenth of August (flowering period). Two meteorological stations, with contrasted wind characteristics, have been considered: the stations of Saragosse and Lleida. Saragosse station is around 62km and 41km far from Sariñena and Gurrea de Gallego respectively whereas Lleida station is around 68km and 123km far from Sariñena and Gurrea de Gallego respectively.

In a first step, we considered an average wind: distribution of direction and speed of the wind were calculated over 10 years (1996 - 2005) for the station of Saragosse and over the past 3 years (2004 – 2006) for the station of Lleida. The data used for the simulations are presented in Figure 2 & 3. In Lleida, the wind is blowing with three prevailing directions and a moderate speed. In Saragosse, the north-west wind is by far the most frequent; the wind speed depends on the direction, and ranges from 2 to 8 m/s. That's why, the wind velocity for each direction has been considered. On the contrary, in Alsace, the same average wind speed (3 m/s) was taken into account, according to the meteorological data.

In a second step, in order to assess the effect of the variability of the wind over the years, we simulated the introduction of transgenic varieties, considering an average wind distribution for each separated year, instead of considering a ten (Saragosse) or three (Lleida) years average wind. Only the data obtained from the Saragosse station were taken into account since the number of years of data is not sufficient in Lleida. The distribution of direction and speed of the wind for each year at the Saragosse station are presented in appendix 2. They are quite different between years: there is one clear dominating wind direction all over the years (North West), but its frequency ranges from 37% to 70%.

¹ Sistema de Información Geográfica de la PAC

² Fondo Español de Garantía Ágraria


<u>Figure 2.</u> Distribution of wind direction and wind speed during the flowering period A/ Meteorological station of Lleida (2004 – 2006)

B/ Meteorological station of Saragosse (1996 – 2005) Wind direction Wind speed (m/s)



Table 1.	Levels	of	maize	in	the	AUA	in	the	selected	areas	(Alsace	and	Aragon),
according	g to CAl	P sta	atemen	ts o	f the	farme	rs						

Area	Year					
	2003	2004	2005			
Heiwiller			70%			
Gurrea de Gallego	34%	30%	11%			
Ensisheim			85%			
Sariñena	65%	64%	42%			

In Alsace, the CAP statements of the farmers was provided only for 2005

II.1.c. Relative share of maize crops and distribution pattern in the agricultural used area

The share of maize in Agricultural Used Area (AUA) must be representative of the current state of maize production in the areas. The maize crops distribution patterns in the selected areas according to CAP statements of the farmers from 2003 to 2005 were thus taken into account. The Table 1 presents the level of maize in AUA of the selected area in Aragon and Alsace, according to CAP statements. The level of maize is in average lower in Aragon than in Alsace. But we should notice than density of maize in Alsace may be overestimated, due to attacks from root worm (Diabrotica virgifera virgifera). Since its larvae can only survive in maize roots and there's no real efficient pesticide, the most efficient measure to limit its expansion is to limit the single-maize crop farming. What's more, the two Spanish areas are contrasted in terms of maize level in AUA: in 2003, in Sariñena, maize was sown on 65% of irrigated AUA for only 34% in Gurrea de Gallego. Moreover, the density of maize in AUA varies according to the year and decreases between 2003 and 2005, due to climatic conditions. In 2005, the level of maize is three times as low as in 2003 in Gurrea. Indeed, in 2005, the soil useful water and precipitations fallen before sowing were faint. The amount of water available for irrigation was thus lower, which didn't allow sowing maize on the same area than the previous years.

How did farmers allocate maize in crops rotation? The allocation of maize in fields is presented in appendix (appendix 3). In Sariñena, maize is regularly distributed in the landscape. In Gurrea de Gallego, maize was little drilled in the North-East of the selected area. Moreover, while reducing the density of maize, the farmers seem to sow maize preferentially on fields on which maize was cultivated the previous years. That's particularly true in Sariñena, where percent of total area sown with maize over the 3 years (31%) represents the great majority of the fields sown with maize in 2005 (42% of AUA). It is clear that farmers do not allocate crops and particularly maize at random. But it is not easy to identify their decision rules. And we should handle the data with care, all the more that crop rotation is known only over three years and maize is cultivated in rotation with alfalfa, which is a perennial crop, harvested during the four years after sowing.

In a second step, in order to assess the effect of the relative share of maize crops, other things being equals, we ran simulations where **maize area moved from 25% to 100% of the AUA**. On the contrary to the previous batch, maize was randomly allocated in the fields, with 10 repetitions for each level of maize.

About the **rates of transgenic varieties**, the levels of 10%, 30%, 50% and 70% of GM maize in total maize area were implemented. GM maize was randomly introduced in the fields. We did not introduce GM maize by farmers, as the farmer of the fields was not known.

In order to assess the effect of the distribution of GM and non GM maize in field pattern, **many repetitions** were performed:

- When maize is distributed according to CAP statements of the farmers, 40 random distribution of GM maize in maize field are simulated (only one maize crops distribution corresponding to what the farmers really did);
- When maize is randomly distributed (relative share of 25% to 100%), 10 random distribution of GM maize and non GM maize are simulated (one distribution of GM maize for one distribution of maize in field pattern).



Figure 3. Distribution of flowering date – 2004 (from Messeguer *et al*, 2006)





Number of days after flowering onset

II.1.d. Crop practices and varietal characteristics

Firstly, we assumed that GM and non-GM varieties are sown at similar density and produce similar amount of pollen. In addition, we assumed that **GM and non GM varieties flower at the same time**, in order to consider worse case scenarios. All the pollen and silks are emitted during one single day. With this assumption, we considered the worst case.

In a second step, simulations were run with **asynchronous flowering**. In fact, according to climate and sowing dates, there can be a wide range of flowering dates and previous studies showed that flowering time lag can have a strong impact on cross-pollination between GM and non GM varieties. Two elements must be considered:

- Onset of male and female flowering: In the WP2 of SIGMEA, a trial was conducted on maize pollen dispersal in two areas containing Bt and conventional maize fields in Catalunya (Messeguer *et al*, 2007). We considered the distribution of flowering dates from one area (Termens) (Figure 4). We distributed the onset of male flowering of the GM and conventional maize fields in the same proportion. We assumed that there was one day protandry, so that the female flowering begins one day after the onset of male flowering (between 0 and 5 in Angevin *et al*, *accepted*).
- Male and female flowering dynamics: we took into account the flowering dynamics used in MAPOD (flowering period of 12 days with a peak on the 4th day in Angevin *et al, accepted*).

II.1.e. Maize volunteers

In Alsace, the presence of maize volunteers has been considered as negligible. This assumption is not valuable in Aragon. As MAPOD model does not take directly into account the occurrence of maize volunteers to calculate gene flow, we considered the effect of maize volunteers in an additive manner. The real crop rotation between 2003 and 2005 is considered.

Based on results of current research in Aragon, we considered the effect of maize volunteers:

- Coming from GM maize in the seed bank (maize volunteers coming from conventional maize pollinated by GM pollen are considered negligible).
- Resulting from the seeds remaining in the fields from the harvest of the previous year.

Thus, for the conventional maize fields sown with GM maize the previous year, the GM adventitious presence in non GM harvest is equal to the GM adventitious presence predicted by MAPOD plus the GM adventitious presence due to maize volunteers. We made different hypothesis on the rate of GM adventitious presence due to maize volunteers: 0.0% / 0.2% / 0.5%, according to Spanish study (Peñas *et al*, 2007).

The various simulations implemented are summarized in **table 2**.

<u>Table 2.</u> Overview of the implemented simulations

	Distribution (number of sin	of maize nulations)	Levels of GM maize (number of simulations)	Distribution of wind direction and wind speed	Flowering dynamics
Gurrea de Gallego		2003 = 34% (160) $2004 = 30% (160)$ $2005 = 11% (160)$	10% (40) 30% (40) 50% (40) 70% (40)	Lleida Station 3 years distribution (2004 – 2006)	Synchronicity
	CAP (maize is distributed according to CAP statements of the farmers)	2003 = 34% (160) $2004 = 30% (160)$ $2005 = 11% (160)$	$ \begin{array}{c} 10\% (40) \\ 30\% (40) \\ 50\% (40) \\ 70\% (40) \end{array} $	Saragosse Station 10 years distribution (1996 - 2005)	Synchronicity
		farmers) $2003 = 34\%$ (800)		Saragosse Station Distribution of each year from 1996 to 2005	Synchronicity
		2003 = 34% (40)	50% (40)	Lleida Station 3 years distribution (2004 – 2006)	Flowering time lag
	Random selection of fields sown with maize	25% (40) 50% (40) 75% (40) 100% (40)	10% (10) 30% (10) 50% (10) 70% (10)	Lleida Station 3 years distribution (2004 – 2006)	Synchronicity
	CAP (maize is distributed	2003 = 34% (160) $2004 = 30% (160)$ $2005 = 11% (160)$	10% (40) 30% (40) 50% (40) 70% (40)	Lleida Station 3 years distribution (2004 – 2006)	Synchronicity
Sariñena	statements of the farmers)	2003 = 34% (160) 2004 = 30% (160) 2005 = 11% (160)	10% (40) 30% (40) 50% (40) 70% (40)	Saragosse Station 10 years distribution (1996 - 2005)	Synchronicity
	Random selection of fields sown with maize	25% (40) 50% (40) 75% (40) 100% (40)	10% (10) 30% (10) 50% (10) 70% (10)	Lleida Station 3 years distribution (2004 – 2006)	Synchronicity

II.2. Methodology for the treatment of model outputs

In order to assess the impact of the sowing transgenic varieties in the studied areas, we considered two kinds of indicator:

- The **mean cross pollination rate of the whole area**, expressed in percentage of GM grains in non GM harvest. It corresponds to the mean cross-pollination rate of a silo, assuming that all the non GM harvests of the area are mingled in the same silo.
- The **part of total area** sown with "non GM maize varieties" where GM adventitious presence is over a specific threshold and is thus **potentially** rejected or **downgraded**.

For the same reasons as those described in Alsatian study case, we considered various maximum thresholds for cross-pollinations: 0.01% (asked by organic farmers and semolina industries), 0.1% (asked by starch industries), 0.4% (if 0.5% of impurities in the seeds), 0.6% (if 0.3% of impurities in the seeds), 0.9% (legal threshold) and 2.25% (corresponds to a 0.9% presence in case of a PCR analysis according to the assumptions described in Alsatian report).

Two different units for analysis have been used to calculate the output indicator in this study: the whole study area and the field unit (those units for analysis are described in the table 4 of the Alsatian report – see D7.2 Alsace).

<u>Figure 5</u>. Part of downgraded non GM maize area according to various maximum thresholds for cross-pollination in Gurrea de Gallego



2003 maize crop distribution pattern (34% of maize in AUA); Wind characteristics from Lleida station; Synchrony of flowering period.





50% of GM maize in total maize area; 2003 maize crop distribution pattern (34% of maize in AUA); Wind characteristics from Lleida station; Synchrony of flowering period. Apart the 40 implemented simulations, those maps present two opposite situation: minimum (A) and maximum (B) of the part of downgraded non GM area with the 0.9% threshold.

III. Results

In a first step, we consider the area near Gurrea de Gallego. The wind distribution of speed and direction established in Lleida station were used for simulations.

III.1. Effect of various rates of transgenic varieties introduction in real allocation of maize in Aragon and dynamic of flowwering

Assuming that all the flowering periods are synchronous

Let's consider the **part of downgraded area**, with the 2003 distribution of maize (34% of maize in AUA). With the legal threshold (0.9%), a very small part of non-GM area is to be downgraded if GM maize represents 10% of the total maize area (Figure 5A). With 50% of GM maize (Figure 5B), around 15% of the non-GM area exceeds the 0.9% threshold. The part of downgraded area increases if considering more severe thresholds:

- From 0.4% to 0.9% threshold, the relative increase of the part of downgraded non GM area is quite similar for both level of GM maize in total maize area (the median value increases of 33% and 40% respectively with 10% and 50% of GM maize). Nevertheless, the larger the GM area, the higher the part of downgraded non GM area: in average, with 50% of GM maize, it is five times as high as with 10% of GM maize. We have already seen that the lower thresholds enable to take into account seed impurities (see II.2): assuming a 0.5% GM impurities rate in seed lots, we should consider the result predicted by MAPOD with 0.4% threshold in order to take into account the GM impurities in seeds in an additive manner. As a consequence, when GM impurities in seeds increase from 0% to 0.5%, the part of downgraded non GM area increases more with 50% of GM varieties than with 10% of GM varieties. Thus, the issue of GM impurities in seeds is more complicated with a high level of GM maize. In addition, a low level of GM impurities in seeds is more difficult to achieve with a high level of GM maize (non GM pollen protection is lower).
- The part of downgraded area is **quite high with very low thresholds**. For instance, the 0.01% threshold corresponds to current requirements of semolina industries. A vast majority of the non GM fields exceed the 0.01% threshold, even with 10% of GM maize.

The previous results were obtained with a field spatial unit for analysis. The part of downgraded area may also be computed with a smaller unit (10m * 10m). The results are presented in appendix 4. The part of downgraded area is lower if considering an intra-field spatial unit. It is due to the fact that contamination is not homogeneous in the fields. With a field spatial unit, we consider the mean cross-pollination rate of the whole field, which may hide great variability of the cross-pollination rate within the field. However, as shown in Alsace, the decrease of rejected non GM area is reduced when a large part of non GM maize is downgraded (50% of maize and 0.01% threshold) and when very few non GM maize fields is downgraded (10% of GM maize and 2.25% threshold).

For a given threshold, figure 5 shows **a large variability** of downgraded non GM area. For instance, with 50% of GM varieties and the 0.9% threshold, the median part of downgraded non GM area is equal to 13%, but ranges from 3.8% to 24,8%. This is due to the strong impact of GM and non GM maize distribution in field pattern. Among the 40 implemented simulations, the maximum and minimum of rejected area with 0.9% threshold are represented in figure 7. In the most favourable situation (minimum of rejected area), a great part of GM maize were sown in the south east of the area, and thus the density of GM maize is lower in

<u>Figure 7.</u> Average GM adventitious presence in harvested non-GM grains evaluated at a landscape scale in Gurrea de Gallego from 2003 to 2005



Real maize distribution pattern according to CAP statements of farmers; Wind characteristics from Lleida station; Synchrony of flowering period.

<u>Figure 8.</u> Variation of the GM adventitious presence in harvested non GM grains in Gurrea de Gallego according to the flowering dynamic



A/ Cross pollination rate at landscape scale

B/ Part of downgraded non GM area

50% of GM maize in total maize area; 2003 maize crop distribution pattern (34% of maize in the AUA); Wind characteristics from Lleida station.

the rest of the area. In the most unfavourable situation (maximum of rejected area), non-GM maize are more regularly distributed in the area. Not only does the part of downgraded area depend on the level of GM varieties and the threshold required, but also on the GM and non GM maize distribution in field pattern. The allocation choice of GM and non GM maize could be an effective way to limit cross-pollination. The variability of the results is lower with the intra-field spatial unit for analysis (appendix 4). It is due to the fact that the effect of GM grain dilution is less important at an intra-field scale.

Let's consider now the **mean cross pollination rate at the landscape scale** with the real allocation of maize from 2003 to 2005 (Figure 7).

First of all, with 50% of GM maize, the mean cross pollination rate for the whole area is always below 0.9%, even if the previous results showed that 13% of the non GM area is over this threshold (median value). It is the case when fields are blended in a same silo. Nonetheless, the most severe threshold (0.01%) is never achieved at the landscape scale, even for 10% of transgenic varieties.

For a given level of maize in total maize area and a given level of GM varieties, the variability of the results might be important, as observed for the part of downgraded non GM area. In 2003 and 2004, the 0.9% threshold might be or not be possible to achieve for the whole area, according to the space allocation of GM and non GM maize in fields. In some cases, the variability is so high that box plots corresponding to two contiguous rate of GM varieties overlap. For example, in 2003, for 12.5% of the situations with 70% of GM varieties, the cross-pollination rate is below the maximum cross pollination rate of the whole area with 50% of transgenic varieties. This overlapping rate depends on the density of maize: it is all the larger as the level of maize in total maize area is high.

In addition, a decrease of the cross pollination rate is clearly observed from 2004 to 2005 (on average 32% for the median value of the mean cross-pollination rate). It is due to a decrease of the density of maize in AUA (35%), which is related to drought conditions (see II.1.c). Thus, the maximum density of GM maize for which the mean cross-pollination rate is below 0.9% depends on the year: 50% in 2003 and 2004 and 70% in 2005 in Gurrea de Gallego.

Assuming time lag flowering

In the previous simulations, we assumed that all the maize flowered simultaneously. Thus, the risk of cross-pollination between GM and non GM is high, as all the non GM silks are receptive while all the GM pollen is emitted. In figure 8, we assess the effect of time lag flowering on cross pollination rate. The same GM maize distributions in field pattern are used for simulations. As expected, taking into account time lag flowerings leads to a decrease of GM adventitious presence in non GM harvest. In average, a **30% decrease of the cross-pollination rate** is observed (Figure 8A). The variances of the cross pollination rates are not significantly different. In the same way, the rejected area is lower with time lag flowerings (Figure 8B). This decrease is reduced when a large part of non GM maize is to be rejected (high threshold). With a 0.9% threshold, the median value is 44% lower with time lag flowering compared with situation where flowerings are synchronous. The hierarchy between the various GM and non GM distributions is modified by taking into account time lag flowering. As a conclusion, not only the distance between GM and non GM fields has an effect but also the time lag flowerings between GM and non GM fields.

<u>Figure 9.</u> Variation of the GM adventitious presence in harvested non GM grains in Gurrea de Gallego according to various hypothesis on the effect of GM maize volunteers



Hypothesis on the effect of volunteers: 1: no effect of maize volunteers; 2: for a GM maize – non GM maize sequence, the cross-pollination rate predicted by MAPOD is put up of 0.2%; 3: for a GM maize – non GM maize sequence, the cross-pollination rate predicted by MAPOD is put up of 0.5%

Figure 10. Variation of the GM adventitious presence in harvested non GM grains in Gurrea de Gallego according to the wind characteristics from two different meteorological stations

2004 maize crop distribution pattern (30% of maize in AUA); Synchrony of flowering period



III.2. Effect of maize volunteers (with flowering periods synchronous)

On the contrary to Alsace, maize volunteers can not be considered as negligible in Aragon. Few data are available on the effect of maize volunteers on GM adventitious presence in non GM harvest. In fact, in experimentation, it remains difficult to evaluate the GM adventitious presence in non GM harvest that is only due to maize volunteers. That's why we made different hypothesis based on current research (Peñas *et al*, 2007). We considered the crop sequence from 2003 and 2005 (Figure 9).

We have already seen that the mean cross-pollination rate decreased from 2003 to 2005, due to the decrease of the level of maize in the AUA. Let's compare the mean cross-pollination rate for the same year with the different hypothesis. It doesn't move in 2003 as it is the beginning of the considered crop sequence. As expected, taking into account the presence of maize volunteers leads to an increase of the cross-pollination rate. Considering the median value of cross pollination rate, the increase ranges from 10% to 19% with the second hypothesis (0.2%) and from 24% to 44% with the third hypothesis (0.5%).

If we compare the results between years: he effect of GM maize volunteers is more important in 2005 (orange plots) than in 2004 (yellow plots), whatever the hypothesis. Indeed, maize single crops farming is more common from 2004 to 2005: 73% of the maize drilled with maize in 2005 had been drilled with maize in 2004 (57% between 2003 and 2004). Thus, the probability of having GM maize volunteers in a conventional maize field is higher in 2005 than in 2004. The increase of GM adventitious presence in non GM harvest due to GM maize volunteers is in direct proportion to the total area drilled with non GM maize after GM maize, which depends on: (i) the level of fields cultivated in maize two succeeding years, (ii) the level of GM maize of the previous year, and (iii) the level of GM maize fields of the current year.

III.3. Effect of the geographical and inter-annual variability of the wind characteristics (no volunteers / synchrony of flowering)

Geographical variability of the wind characteristics

Previous results were obtained with wind characteristics from Lleida station. In the figure 10, we assess the geographical variability of the wind characteristics, using wind data from two meteorological stations: Lleida and Saragosse stations. The mean cross pollination rate is higher with the wind characteristics from Saragosse station (around 30% higher for the median value). In fact, in Saragosse, the wind speed is in average twice as high as in Lleida. Pollen dispersal, including GM pollen dispersal, is thus higher. The variances of the mean cross pollination rate are significantly different and higher with distribution of wind speed and direction from Saragosse station. As there is one prevailing direction in Saragosse, the distribution of GM and non GM maize in fields is of utmost importance. This distribution is still but less important with wind characteristics of Lleida, since the prevailing wind directions are more numerous. In addition, the GM and non GM maize risky distributions are not exactly the same with both wind characteristics. We have previously shown that there is a strong impact of GM and non GM maize in fields. The results show that this impact is due on the one hand to the distance between GM and non GM maize and on the other hand to the relative position of GM and non GM maize compared to the wind direction. The part of downgraded non GM area is also greater with wind characteristic from Saragosse station, whatever the threshold (appendix 5).

Figure 11. Variation of the cross-pollination rate in Gurrea de Gallego according to the interannual variation of the wind characteristics



A/10% of GM maize

B/ 50% of GM maize





The wind characteristics (distributions of direction and speed) of each year from 1996 to 2005 are considered for the simulations

Saragosse and Lleida are both near the area of simulation. It is important to take into account local wind characteristics, as distribution of wind direction and speed is a quite local phenomenon and pollen dispersal is sensitive to the wind direction and speed.

Inter annual variability of the wind

In the figure 11, we **assess the effect of inter annual variability of the wind** on the mean cross-pollination rate at the landscape scale. The variability of the mean cross-pollination rate is quite important. We showed previously that there is a great variability of the results due to GM and non GM maize distribution in field pattern (see III.1 and Alsace case study). Here, we shown that the variability of the cross pollination rate due to inter annual variability of wind is in the same order that the variability due GM and non GM space allocation. The mean cross-pollination rate for each distribution of GM and non GM maize in field pattern with the wind characteristics of each year is presented in appendix 6.

Hence, we should compare a frequency analysis with an analysis base on average distribution. For instance, if 50% of the maize area is sown with transgenic varieties (Figure 11B), the

Figure 12. Average GM adventitious presence in harvested non GM grains (median value) in Gurrea de Gallego according to the wind speed of the prevailing direction.



median cross-pollination rate is below 0.9% with the 10 years average distribution of wind speed and direction. According to the climatic frequency analysis, the 0.9% threshold is achievable 6 years out of 10 (median value). The GM presence in non GM grain exceeds 0.9% in more than 75% of the situations with 2002 wind distribution and is always below 0.9% with 2003 wind distribution.

The figure 12 shows that the median value of cross pollination is well correlated to the wind speed in the prevailing direction. The median cross pollination rate

increases with the wind speed in prevailing direction, all the more that the share of GM maize in the total maize area is high.



<u>Figure 13.</u> Variation of the GM adventitious presence in harvested non GM grains in Gurrea de Gallego and Sariñena from 2003 to 2005

Real maize distribution pattern according to CAP statements of farmers; Wind characteristics from Lleida station; Synchrony of flowering period.





GM and non GM maize are allocated at random; wind characteristics from Lleida station; Synchrony of flowering periods

III.4. Effect of the landscape (no volunteers / synchrony of flowering)

III.4.a. Comparison between Gurrea de Gallego and Sariñena

The figure 13 presents the mean cross-pollination rate in Sariñena and Gurrea de Gallego from 2003 to 2005 with various rates of transgenic varieties. For a given year with the same rate of transgenic varieties, the median levels of out-crossing are always higher in Gurrea de Gallego than in Sariñena, even if the density of maize in AUA is lower in Gurrea. In average, the **median cross pollination is 30% higher** in Gurrea de Gallego. The overall levels of cross-fertilization remain always under 0.9% in Sariñena, whatever the rate of transgenic varieties. On the contrary, the overall levels of cross-fertilization exceed 0.9% with 70% of transgenic varieties in 57.5% of the simulations in 2003 and 2004. In fact, the smaller size of the fields in Gurrea de Gallego leads to a decrease of pollen protection of non-GM fields and of average distance between GM and non-GM fields. As a consequence, for a same region (Gurrea de Gallego and Sariñena are not so far), the out-crossing rates may be quite variable, owing to the characteristics of the field patterns.

In the same way, the non compliant area with 0.9% threshold is in average twice as high in Gurrea de Gallego as in Sariñena. The difference remains fainter for the 0.1% threshold: for both areas, the more severe thresholds are difficult to achieve (see appendix 7).

In the figure 14, we compare both areas, other things being equals (including levels of maize in the AUA). As previously, the cross-pollination rate is higher in Gurrea de Gallego, for the same share of GM and non GM maize. Generally speaking, the relative differences between two box plots are quite similar for both areas: these areas stand up the same way to the decrease of relative share of maize in the AUA or to the decrease of relative share of GM maize in total maize area.

III.4.b. Comparison between Alsace and Aragon

In the figure 15, we compare the landscapes which are quite similar in terms of distribution of field area. The mean cross-pollination rate is quite similar between Gurrea de Gallego and Heiwiller on the one hand and between Sariñena and Ensisheim on the other hand, whatever the share of maize in the AUA and the share off GM maize. The same observation goes for the part of downgraded non GM area (see appendix 8). As a conclusion, according to MAPOD model, the cross pollination rate and the part of downgraded area of two landscapes with same distribution of field areas and more or less same wind characteristics are similar, whatever the region. According to MAPOD, the distribution of field area is a great indicator of the risk of GM adventitious presence in non GM harvest of a given landscape.

<u>Figure 15.</u> Comparison of the average GM adventitious presence in harvested "non-GM" grains in Aragon and Alsace according to the percentage of maize in the AUA and the percentage of GM maize in the total maize area



A/ Gurrea de Gallego and Heiwiller



B/ Sariñena and Ensisheim

GM and non *GM* maize are allocated at random; Wind characteristics from Lleida station for Aragon simulations; Synchrony of flowering periods

IV. Conclusion on Aragon case

Main differences between Alsatian and Aragon cases in term of cross-pollination risk with hazard distribution of GM and non GM maize fields are not due to the landscape shape differences. They are mainly due to: (i) the characteristics of local wind with high differences between climatic stations and inter annual variability of wind, (ii) the presence of volunteers which is for the moment poorly informed, (iii) the flowering time lag which open potentially a larger window for management of cross pollination in Aragon than in Alsace.

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Meteorological data from website: <u>http://www.wunderground.com/</u>

Appendixes

Appendix 1. Main characteristics of the Alsatian field patterns.



Ensisheim study area						
Total area 885 ha						
Number of fields	93					
Mean area	9.5 ha					





Heiwiller study area						
Total area 660 ha						
Number of fields	356					
Mean area	1.9 ha					



Distribution of fields according to their area





(a) Wind direction











SE Wind

(b) Wind speed

%





















N

ω

<u>Appendix 3.</u> Crop rotations

A/ Gurrea de Gallego

Rotation	Percent of total area
maize / maize / maize	6%
maize / maize / other crop	10%
maize / other crop / maize	1%
other crop / maize / maize	1%
only one maize	30%
no maize	51%



B/ Sariñena

Rotation	Percent of total area	ROTATION maize / maize / maize maize / other crop maize / other crop / maize determed (maize / maize
maize / maize / maize	31%	only one maize
maize / maize / other crop	21%	no maize
maize / other crop / maize	1%	
other crop / maize / maize	5%	
only one maize	24%	
no maize	18%	
		500 250 0 500 Mètres

Appendix 4. Impact of the spatial unit for analysis on the non GM area to be downgraded

Three spatial units:

- The field and intra-field unit (see graphs)
- The landscape unit (whole non GM maize area) (see tables)

2003 maize crop distribution pattern (34% of maize in AUA); Wind characteristics from Lleida station; Synchrony of flowering period.

A/10% of GM maize



Threshold

100%

Threshold

	Part of total number of simulations where GM presence in non GM grain harvested on the total non GM area is over							
Threshold	0.01%	0.10%	0.40%	0.60%	0.90%	2.25%		
10% of GM maize	100%	100%	0%	0%	0%	0%		
50% of GM maize	100%	100%	100%	92.5%	0%	0%		

<u>Appendix 5.</u> Variation of the part of downgraded non GM area in Gurrea de Gallego according to the wind characteristics from two different meteorological stations and considering various maximum thresholds for GM adventitious presence



A/ 10% of GM maize in total maize area

B/ 50% of GM maize in total maize area



2004 maize crop distribution pattern (30% of maize in AUA); Synchrony of flowering period.

<u>Appendix 6.</u> Impact of interannual variability of wind on the GM adventitious presence in non GM harvest for each GM and non GM maize distribution pattern in Gurrea de Gallego

2003 maize crop distribution pattern (34% of maize in AUA); Synchrony of flowering period. A/ 10% of GM maize



B/ 50% of GM maize



40 GM maize space distributions

40 GM maize space distributions

Legend

- Boxplot: each of the 10 years wind characteristics from 1996 to 2005 is considered
- In red: the 10 years average wind characteristics is considered.

<u>Appendix 7.</u> Variation of the part of downgraded non-GM area in Gurrea de Gallego and Sariñena from 2003 to 2005 for two maximum thresholds for cross-pollination

Real maize distribution pattern according to CAP statements of farmers; Wind characteristics from Lleida station; Synchrony of flowering period.



A/ Maximum threshold for cross-pollination: 0.9%



B/ Maximum threshold for cross-pollination: 0.1%

Appendix 8. Comparison of the part of downgraded non-GM area in Aragon and Alsace according to the percentage of maize in the AUA and the percentage of GM maize in the total maize area for two maximum thresholds for cross-pollination cross-pollination

GM and non GM maize are allocated at random; Wind characteristics from Lleida station for Aragon simulations; Synchrony of flowering periods



A/ Maximum threshold for cross-pollination: 0.9%





B/ Maximum threshold for cross-pollination: 0.1%

D 7.2 Identification of critical points from validated results

- Effect of structural variables on GM admixture in non GM harvest – - Simulation results -

Aquitaine case study - maize

B. Lécroart, M. Le Bail - INRA

Foreword by WP7 coordination: this work was carried out at the very end of the SIGMEA project. Thus, the report focuses on simulation results without extensive analysis.

Report on simulations implemented for the South West of France case study

I. Material and methods

We used MAPOD, a gene flow model for maize (Angevin *et al.*, 2008), to simulate cross-pollination between genetically modified (GM) and non-GM maize in an agricultural landscape in the South west of France.

I.1. Input data

In those simulations, the GM variety is heterozygous for the GM character.

I.1.a. Field pattern

Figure 1. Field area distribution



The field pattern supplied by ARVALIS was used (Appendix 1). Figure 1 shows the resulting distribution of field area.

This type of field pattern is intermediate between the two types identified in Alsace and Aragon. In this case, mediumsized fields are most common, whereas very large fields (like in Ensisheim) or very small fields (like in Heiwiller) are little represented.

I.1.b. Distribution of wind speed and direction

The distributions of wind speed and direction during the flowering period were obtained from local meteorological stations (<u>sources:</u> Météo-France and <u>www.underground.fr</u>). Simulations were run with average wind distributions, from 1985 to 2005 (Figure 2). In average, the wind is mainly blowing from North-West and West with a moderate speed.



Figure 2. Distribution of wind speed and direction during the flowering period

I.1.c. Relative share of maize crop in the agricultural used area

According to the statements of the farmers, which had been surveyed, 67% of the agricultural used area (AUA) was sown with maize in 2004 and 2005. There was a slight decrease in 2006 with 60% of the AUA sown with maize. A very high proportion of seed maize production was recorded. The simulation area is located in the agricultural regions of the Béarn hillsides and Gave du Pau valley: maize was grown on **60%** of the AUA of those two regions in 2004 (CAP data).

For the simulations, we considered only the maize grain production to fulfil SIGMEA project goals. Moreover, in order to be representative, maize was randomly allocated to the fields, considering the average proportion of maize in the AUA of the region. We took into account various frequencies of transgenic varieties (10% and 50% of the total maize area).

I.1.d. Flowering period

Figure 3. Distribution of flowering period used for the simulations



In order to take into account the natural flowering time lag between maize fields. results from experiments carried out in the study zone were used (trials established by Arvalis as part of the CTPS 2002-2005 call for tender). The difference between the earliest and latest flowering fields was 15 days. Thus the onset of female flowering was randomly allocated, according to a normal distribution with a standard deviation of 3 (Figure 3).

On average, the difference between female and male flowering dates was around 1 day. Simulations therefore included one day of protandry. The MAPOD flowering dynamics were then applied (Angevin *et al.*, 2008).

It was assumed that GM and non-GM varieties produced the same quantity of pollen and were sown at the same density.

In the end, **80 simulations** were carried out as part of the South West case study: 40 simulations with 10% GM maize and 40 simulations with 50% GM crops. For each of those simulations, conventional and GM maize were randomly allocated to the fields, and flowering date of each field was also randomly allotted according to a normal distribution. A simulation lasted on average 4 to 5 hours.

I.2. Methodology for the treatment of model outputs

Some of the elements of the methodology are here briefly summed up. They were already detailed in the deliverable 7.2 Alsace as well as in the IPTS report (Messéan et al., 2006).

Two indicators were considered to assess the impacts of the introduction of GM varieties on the non-GM production:

- The mean cross-pollination rate, measured as a proportion of non-GM grains in the non-GM harvest.
- The **proportion of the non-GM maize area not complying** with a given threshold

Several thresholds were defined in order to take into account the actual **stakeholder requirements**. Indeed, in addition to the 0.9% legal threshold, starch and semolina industries are currently asking for higher purity rates, respectively 0.1% and 0.01%. Furthermore, these thresholds allow taking into account **other sources of GM admixture**. MAPOD evaluates the adventitious presence of GM grains due to pollen flow between fields. Actually, other sources of GM adventitious presence had been identified, such as seeds and machinery (drill, combine harvester and transport). In the case of heterozygous varietal impurity, Messéan *et al* (2006) showed that the effect of varietal impurities in batches can be taken into account as an additive effect using a 1:1 ratio when estimating the overall GM proportion in the harvested crop. For example, using the hypothesis that the level of seed impurity is 0.4%, the threshold must be set at 0.5% to ensure the harvested crop complies with the legal 0.9% threshold (0.4% + 0.5% = 0.9%).

Finally, adventitious GM presence rates were assessed as the percentage of grains carrying the transgene. This way of quantification is not directly equivalent to the result obtained from DNA-based quantification of the adventitious GM presence using PCR methodology. Maize kernels used for PCR analysis are made up, mainly, of a tegument, an embryo and an endosperm. The endosperm is triploid whereas the tegument and the embryo are diploid. In 2004, Trifa and Zhang showed that the proportion of these elements depends on the variety. Based on their work, we can use the following hypothesis for the relative DNA proportion between tissues: 48% DNA in the embryo, 49% in the endosperm, and 3% in the tegument. The percentage of DNA carrying the transgene in the grain is then 40.3% (Messéan *et al.*, 2006). In the case of heterozygous maize, the adventitious GM presence rates calculated by the model should be multiplied by 0.403 to obtain the genetic quantification that would be obtained by PCR methods. For example, the 2.25% of GM grain threshold corresponds to a 0.9% threshold when using a PCR quantification method. Nevertheless, Messéan (2006) added that for more complex genetic structures, such as stacked genes, case-by-case studies should be performed to relate the percentage of GM seeds to the DNA quantification by PCR.

We considered several scales for the analysis:

- the whole study zone: all the non-GM crops are blended in a same silo,
- the field,
- the intra-field: 10 m x 10 m square.

The results are illustrated using box plots:



II. Results

In this section, we present the results of simulations, without any analysis.

Example of MAPOD output

Figure 4. Example of MAPOD output



10% of GM maize in the total maize area

GM adventitious presence evaluated for the whole non-GM maize area

Figure 5. GM adventitious presence evaluated for the whole non-GM maize area (60% of maize in the AUA)



GM adventitious presence in the non-GM area evaluated at field scale

A/ 10% of GM maize





Mean cross-pollination rate

B/ 50% of GM maize



Figure 7. Distribution of the non-GM adventitious presence in the non-GM area evaluated at field scale - 50% of GM maize

The rate of cross-pollination varies significantly between the fields. The maximum crosspollination rates observed are 6.3% and 10.3% respectively with 10% and 50% of GM maize in the total maize area. Such high values are found in simulations where the non-GM field is small, close to GM fields, and when GM and non-GM flowering stages occur at the same time. The factors which have an effect on the rate of cross-pollination within an emitting field/receiving field pair are known (Messéan *et al* 2006): distance between the fields, wind direction compared with direction of the field pair, area of the emitting field, area of the receiving field, and difference in timing of flowering stage between emitting and receiving fields. In addition, in our case, the additive effect of several GM fields explains the results that were obtained.
2.25%

0%



> Proportion of the non-GM maize area not complying with various thresholds

Threshold .								
Proportion of total number of simulations where GM presence in the whole non-								
			GM harve	est is over:				
Threshold	0.01%	0.1%	0.4%	0.6%	0.9%	2.25%		
Proportion	100%	67.5%	0%	0%	0%	0%		



100%

B/ 50% of GM maize

Proportion

100%

12.5%

0%

100%

Cross-pollination per field

A/ 10% of GM maize

Three different simulation results are represented: (i) minimum value, (ii) near the mean value, and (iii) maximum value of cross-pollination rate of the whole non-GM area.

1) Minimum (mean cross-pollination rate: 0.07%)



	Propor	tion of the no	n-GM area no	t complying w	ith a given th	reshold
Threshold	0.01%	0.1%	0.4%	0.6%	0.9%	2.25%
Proportion	66%	15%	3%	1%	1%	0%

2) Mean (mean cross-pollination rate: 0.11%)



Proportion of the non-GM area not complying with a given threshold							
Threshold	0.01%	0.1%	0.4%	0.6%	0.9%	2.25%	
Proportion	80%	23%	6%	5%	1%	0%	

3) Maximum (mean cross-pollination rate: 0.15%)



	Propor	tion of the no	n-GM area no	t complying w	ith a given th	reshold
Threshold	0.01%	0.1%	0.4%	0.6%	0.9%	2.25%
Proportion	84%	27%	12%	8%	2%	0%

B/ 50% of GM maize

1) Minimum (mean cross-pollination rate: 0.41%)



Proportion of the non-GM area not complying with a given threshold							
Threshold	0.01%	0.1%	0.4%	0.6%	0.9%	2.25%	
Proportion	100%	81%	33%	20%	11%	1%	

2) Mean (mean cross-pollination rate: 0.54%)



	Propor	tion of the no	n-GM area no	t complying w	ith a given th	reshold
Threshold	0.01%	0.1%	0.4%	0.6%	0.9%	2.25%
Proportion	100%	93%	36%	25%	16%	3%

3) Maximum (mean cross-pollination rate: 0.65%)



	Propor	tion of the no	n-GM area no	t complying w	ith a given th	reshold
Threshold	0.01%	0.1%	0.4%	0.6%	0.9%	2.25%
Proportion	100%	94%	60%	41%	19%	2%

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Appendix 1. Field pattern used for the simulations with MAPOD

D 7.2 Identification of critical points from validated results - Effect of the structural variables on GM admixture in non GM harvest based on simulation results -

Beauce case study - oilseed rape

C. Sausse - CETIOM

TABLE OF CONTENTS

Introduction	3
1 Review of the critical points initially identified	3
1.1 The GENESYS oilseed rape model	3
1.2 Critical points in cropping systems identified by GENESYS	4
1.3 Critical points associated with farm systems and types of harvest collection but not taken into	
account by GENESYS	7
2 Simulation for Beauce	7
2.1 The characteristics of cropping and production systems in Beauce	8
2.2 Choice of situations to simulate	8
2.3 Method for processing and analysing the results of the simulations	11
3 Simulation results	12
3.1 Evaluation of the initial situation, with no change in practices other than the introduction of GM	1
OSR	12
3.4 Contribution of basic practices	19
3.5 The case of organic OSR production	26
4 Identification of critical points: summary	28
References	31
APPENDIX 1 : rotations	32
APPENDIX 2 : agricultural practices	33

Introduction

The aim of this report is to identify landscape and agricultural factors posing problems to the coexistence of genetically modified (GM), conventional and organic oilseed rape (OSR) crops in the small "Beauce" region. A three-step method was used for this analysis: (1) assessment of current knowledge; (2) determination of the status of the region for the identified elements and (3) model-based simulation to quantify gene flow in a realistic landscape and to determine the contribution of certain key practices. Only issues associated with cropping systems and farms were analysed with these simulations.

1 Review of the critical points initially identified

1.1 The GENESYS oilseed rape model

We used the GENESYS model for oilseed rape (Colbach *et al.*, 2001a and b). This model describes the spatial pattern of gene flow, via pollen and seed, and the temporal pattern of gene flow in the form of volunteers (figure 1).



Figure 1: Model of the annual life cycle of oilseed rape in each field (____) and in the border (____) of a group of fields, from flowering to harvest (Adultes = Adults, fleurs = flowers, pollen, production semencière = seed production, graines récoltées = harvested seeds, graines non récoltés = non-

harvested seeds, stock semencier = seed bank, graines de colza semées = oilseed rape seeds sown, plantules = plantlets)

We used the version of the model updated on February 20th 2006. The input variables were:

- Characteristics of oilseed rape varieties: genotypic composition, self-pollination rate, pollen production rate and relative yield
- Daily temperatures
- Latitude
- Field pattern (fields and borders), in the form of polygons
- For each field and each year of simulation:
 - o The crop grown
 - Soil tillage practices (nature and date)
 - o Sowing date and density
 - The proportion of the area harrowed
 - o Weeding practices, expressed as a percentage of volunteers destroyed, and dates
 - Harvesting dates
 - For the oilseed rape crop: possible mixture of varieties, use of farmers' seed, yield losses and possible routing
 - o For fallow and borders: dates of cutting and grazing

1.2 Critical points in cropping systems identified by GENESYS

The aim of this part of the text is to list known critical points, particularly those identified in the COEX1 and COEX2 studies (Bock *et al.*, 2002, Messéan *et al.*, 2006) and in analyses of the sensitivity of the model (Colbach *et al.*, 2005a and b). This information is organised in table 1.

1.2.1 Critical points relating to landscape organisation

Field layout

Scattered fields with small fields are much more sensitive than grouped fields with larger fields to pollen dispersal.

Table 1: List of the critical points identified in the COEX1 study (red: accessible via GENESYS)

	Critical point	Main source of	Possible action	Which farmers ?	spatial	return to
Step	-	contamination			coexisten	conv.
-					ce	
	field pattern	Pollen	isolation distance, discard	all	x	
Site selection			widths			
	field pattern	Pollen	gathering fields	conv	x	
Recention of coords	seed purity	Seed	no farm saved seeds	conv	x	х
Reception of seeds	seed purity	Seed	pure commercial seeds	conv	х	х
Site preparation	soil tillage	Seed bank	plough	conv		х
	seed purity	Seed	clean machinery	conv and GM on the	х	х
Sowing				same farm or shared		
				equipment		
	sowing practices	Volunteers	sowing conv before GM	all	x	
	variety	Volunteers	no CHL conv	conv	х	х
	variety	Volunteers	no 1/2 dwarf conv	conv		х
	sowing practices	Volunteers	increase conv sowing density	conv		х
OSR management						
	seed loss	Seed loss	special cutter bar	GM		х
	seed loss	Seed loss	clean harvest machinery	conv and GM on the	х	х
				same farm or shared		
				equipment		
Field border	border management	Pollen	early cutting	all	х	
management	border management	Pollen	no glyphosate	all	х	
management	border management	Seed bank	late cutting	all	х	
First intercronning	intercropping management	Seed bank	no plough	GM		х
noriod	intercropping management	Seed bank	soil tillage after volunteer	GM		х
period			emergence			
Other crop management	weed control	Volunteers	appropriate herbicide or	all		х
Other crop management			mechanical tillage			
	rotation	Seed bank	add spring crops	conv		х
Rotation management	delay for OSR return	Seed bank	increase rotation length	conv		х
	rotation	Seed bank	add spring sown set aside	conv		х
	transportation	Seed loss	truck covering	GM	х	х
Transportation	transportation	Grain	clean	conv and GM on the	x	х
Transportation				same farm or shared		
				equipment		
	storage	Grain	dedicated storage	conv and GM on the	x	x
				same farm or shared		1
Storage on farm				equipment		
	storage	Grain	clean storage area	conv and GM on the	x	x
1				same farm or shared		
				equipment		

Field border management

Field border management is an important way of reducing unfavourable selection pressure (through the use of glyphosate), and preventing seed production. Late cutting gives the best long-term results, as it prevents seeding.

1.2.2 Critical points relating to the cropping system

Management of sowing

This point was dealt with in the COEX2 study. The use of contaminated seed lots has an additive effect: the impurity rate of the seeds used adds to the final proportion of GM OSR in the non-GM OSR crop. By contrast, the use of farmer's seeds has a cumulative effect over time.

Rotations

Long rotations (seven years) were used in the COEX1 study, greatly reducing the effect of the stock of seeds from the previous OSR crop. Non-sown fallow has a major effect, and the introduction of spring crops may improve harvest quality, by suppressing volunteers.

In principle, shorter rotations (three to four years) are associated with a greater risk of volunteers, and also of pollen dispersal, due to the higher density of OSR in the landscape.

Cropping practices

Cropping practices affect the dynamics of volunteer growth in several ways:

- Control of volunteers in cereal crops by chemical or mechanical weeding;
- Decreasing yield losses by using appropriate material and practices (Sausse et al. 2006);
- Control of volunteers during intercrop periods;
- Management of seed stocks according to the type of tillage: ploughing after the OSR crop results in young seeds being buried (negative effect). Ploughing before the planting of OSR unearths old seeds with a lower germination capacity (positive effect).

1.3 Critical points associated with farm systems and types of harvest collection but not taken into account by GENESYS

GENESYS represents only the flow of genes linked to the management of cropping systems, in the strict sense of the term. Other types of gene flow identified at farm level are not modelled.

Equipment cleaning

Gene flow and mixing may occur if the following items of equipment are not cleaned between uses for different types of production: soil tillage equipment, seed drills, combine harvesters, tip lorries and storage cells. Such mixing may occur if coexistence occurs within a given farm or if material is shared by several farms.

Transport

The projection of seeds during transport may result in the establishment of a population of OSR outside the field.

Storage

Mixing may occur during storage on the farm, if more than one type of OSR is grown on that farm.

2 Simulation for Beauce

Deliverable 7.1 described the small "Beauce" region in terms of the critical points described above. The farms in this region are highly diverse, leading to differences in sensitivity to gene flow (2a). Based on these observations, we chose situations for simulation so as to represent as accurately as possible the current organisation of the landscape and cropping practices (2b) and adopted a simulation approach adapted to issues concerning the spatial dissemination of genes and their evolution over time (2c).

2.1 The characteristics of cropping and production systems in Beauce

Three principal sources of variation, affecting different types of risk associated with the coexistence of GM and non-GM OSR can be identified:

- The shape, size and overlap of fields has a major impact. Casagrande (2005) identified three different types of field patterns. Different types of pattern may occur together on certain farms.

Type A: small fields (< 5 ha) have extensive borders with neighbours (in areas in which fields have not been regrouped) \rightarrow high risk and control rendered difficult by the large number of neighbours;

Type B: large fields (5 to 15 ha) with extensive borders with neighbours \rightarrow moderate risk and control quite difficult;

Type C: large fields (> 10 ha) with small borders with neighbours \rightarrow low risk and autonomous control possible.

- Possibilities for irrigating seed crops determine the complexity of cropping systems: (1) three year rotation of the type OSR/winter cereal/winter cereal; (2) rotation including OSR once every six to eight years, together with spring crops. The longer of these two rotations is associated with a lower risk, all other factors being equal.

- A given co-operative may dominate, but farmers have the possibility of delivering to other operators: (1) on-farm storage with sales to a number of clients or (2) a single client. The second of these two options is less risky, all other things being equal.

Other factors influencing gene flow are more homogeneous, particularly as concerns volunteer control. Rotations only exceptionally include bare fallow. Permanent set aside fields are cut regularly. Intercrop management practices are relatively intensive in terms of the number of passages of heavy equipment, resulting in a reduction of seed stocks. Tillage practices depend on many factors, but tend to be alternated, with direct drilling techniques included in the rotation. The field borders are generally managed with late cutting to prevent seeding, therefore reducing gene flow. Glyphosate may be used. The use of this herbicide as the sole means of weed management may be potentially dangerous, but this practice is of concern only to the *Direction Départementale de l'Equipement* (the planning department). The combination of these factors tends to encourage the effective control of volunteers.

Only one of the 20 farmers interviewed in 2005 stated that they used farm-saved seed. The true extent to which farm-saved seed is used is difficult to estimate (30% nationally according to the AMSOL).

The machinery used was generally clean, and this was particularly true of harvesting equipment. However, the issue of cleaning remains important if coexistence occurs on an individual farm.

The harvest is generally delivered directly to the co-operative or grain merchant. This is a critical point in that farmer is responsible for transporting his crop to the country elevator, with the possibility of losses depending on the equipment used (use of an awning) and the duration of transport.

There is no direct link between the diversity of farm systems¹ and the risk of gene flow or mixing on the farm. Within a given type of farm, the risks may vary, as farm typing does not usually take into account discriminant factors, such as the type of field pattern (Casagrande 2005). These risks may vary on a single farm, particularly in cases in which the farm has been extended by buying field blocks located at some distance from the historical heart of the farm.

2.2 Choice of situations to simulate

The scale at which GENESYS operates is not compatible with analyses taking into account all the diverse situations encountered in Beauce. The unrepresentative nature of the scale used for simulation makes it necessary to consider highly contrasting situations in terms of the critical points identified above. This strategy aims to maximise the diversity of the landscapes studied. In the Beauce case study, we decided to work on one landscape potentially at risk (Marolles) and another with a lower level of risk (Selommes). Due to a lack of data, priority was given to Marolles, and the results presented in this report concern this zone only.

¹ Considered as coherent units in terms of production and production factors (labour, area, capital, mechanisation etc.)

2.2.1 Input data

The "landscapes" were translated into input files for GENESYS containing the following information:

- Field map: fields cultivated and borders
- For each field and border and for each year: crop grown and cropping schedule.

The initial field map was extracted from a collection of CAP islets supplied by the ONIC². This dataset was rendered compatible with GENESYS as follows: islets, containing several crops, were spilt into fields, fields were split into four-sided polygons and overlaps were eliminated. Data concerning rotations and cropping practices were obtained through surveys carried out in 2005. GENESYS uses meteorological data to determine flowering time. We chose to work with a mean year calculated from data collected at the Blois station over a ten-year period.

The landscape studied had the following characteristics:

- Area of 243 ha

- Small, highly dispersed fields.

- Cropping systems without seed production, of the type, head-of-rotation crop/wheat/wheat/(wheat or barley), with OSR the principal head-of-rotation crop.

- Three farms (alpha, beta, delta) were identified during surveys carried out in 2005. The rest of the land is considered to belong to a single farm ("unknown").

- The landscape is enclosed by woods and a quarry on the northern and eastern sites.

- OSR covers 21% of the land. The mean area of the fields not permanently left fallow is 1.96 ha (minimum 0.17 ha, maximum 7.30 ha, median 1.52 ha).

The open frontiers of the landscape on the western and southern sides may lead to the results being slight underestimations.



Figure 2: Field map used for simulations (visualisation of polygons); each colour corresponds to a farm

² Original data kindly supplied by the Office National Interprofessionnel des Céréales, ONIC (National Office for Cereal Producers)

	Ai	nnual area	a	Cum	ulative area	(20 years)	
				Crop (including			
Farm	Crop*	Border	Total	OSR)	OSR	Border	Total
Alpha	21	1	22	420	142	20	440
Beta	37	1	38	740	150	20	760
Delta	47	1	48	940	210	20	960
Unknown	129	5	134	2580	480	100	2680
DDE**	0	1	1	0	0	20	20
TOTAL	234	9	243	4680	982	180	4860

Table 2: Areas used for simulation

* including permanent fallow (10 ha) ** Direction Départementale de l'Equipement

The preparation of input files for GENESYS from the data collected involved landscape simulation. The data collected did not cover the entire territory (the information available for some fields was not precise) or the entire duration of the simulation (information available only for provisional rotations over several years). The aim was to use this fragmented information about the cropping systems of a region at a given moment in time to generate an exhaustive simulation of the entire landscape at some time in the future. Casagrande (2005) described in detail the rules of extrapolation for filling in gaps due to a lack of information for particular fields during surveys (appendix). This work led to the creation of an initial landscape ("landscape A"). Two variants of this landscape were generated to increase variability in the distribution of OSR fields:

- Variant 1 (landscape B): rotations for the "unknown" farm displaced by one year.
- Variant 2 (landscape C): rotations for the "unknown" farm displaced by two years.

The duration of the simulation was fixed at 20 years, which was considered a suitable compromise between the possibility of demonstrating cumulative effects, calculation time and realism for predictions. Changes in seed stocks were simulated over a 20-year period, to generate an initial state. A variant of each landscape was generated for these "presimulations", to ensure that the rotations coincided, avoiding, for example, situations in which there were two successive OSR crops on a given field corresponding to the last year of the presimulation and the first year of the simulation.

Marolles is one of the worst possible cases in terms of gene flow, due to field fragmentation and size and the duration of rotations. OSR covers 21% of the land in this area, a proportion greater than that for the Beauce region as a whole. Other cropping practices are representative of the region.

The area simulated corresponds to an extreme case, and cannot be used to draw conclusions for the entire Beauce region. However, due to the critical nature of the field pattern, it is possible to establish a maximalist scenario in which no measure is excluded as may happen if less difficult zones were used.

2.2.2 Choice of dissemination pattern

We simplified the evaluation by distinguishing between spatial coexistence (GM and conventional OSR in the same area in the same year) and temporal coexistence (a given field used for conventional OSR after GM OSR).

The following table summarises the various examples considered, with the biological phenomena responsible for contamination:

Next landscape	GMO and conventional mixed	Conventional only
Previous landscape		
GMO and conventional mixed	Pollen and volunteers	Volunteers
GMO only	Pollen and volunteers	Volunteers
Conventional only	Pollen and volunteers	/

Only the cells shown in grey are considered and simulated in our analysis. In cases of **spatial coexistence** of GM and conventional OSR, the fields containing GM and conventional OSR are separated: the two types of OSR may not both be grown on the same field during the duration of the simulation. Temporal coexistence (i.e. the cultivation of conventional OSR on a field previously planted with GM OSR) is dealt with from the point of view of definitive varietal **reconversion** over all or part of the area under OSR.

The specifications of this study included provision for two types of introduction of GM OSR: by farm or by field, in a random, farm-independent manner. Given the small size of the territory studied and the small number of farms, the patterns of dissemination for the rates of GM OSR introduction given relate to field level only. Dissemination by farms concerned all but one of the farms, making the situation equivalent to studying the sensitivity of non-GM farms in a GM environment.

2.3 Method for processing and analysing the results of the simulations

The simulation work provides new information at the scale of the area covered by the simulation, making it possible to refine the identification of critical points relating to cropping system management. This process involves two steps:

- Overall evaluation: starting from the landscape, without changing the usual practices of farmers, we determined overall performance, by quantifying gene flow.

- Evaluation of the contribution of certain key practices to overall performance: this involved a targeted sensitivity study. Several critical points were identified from the results of previous studies, and the aim was to determine the extent to which the landscape was sensitive to modifications to these critical points. The aim is not to quantify total gene flow, but to classify the cropping practices currently used making the greatest contribution. Two methods can be used, depending on the type of critical point considered:

- Crop allocation and rotation: the landscape generator was not available at the time of this study for the generation of alternative landscapes. We therefore analysed the risk factors at field level, based on the results of the initial evaluation, comparing the results obtained with the various characteristics of the fields (post hoc analyses). This type of analysis makes it possible, for example, to determine the effects of the distance between fields and the time period between two successive OSR crops in the rotation.

- Cropping practices: the results obtained for the variants of the initial landscape are compared with those obtained for the initial evaluation. These variants differ in terms of cropping practices and field border management.

Indicators and rules for interpreting simulation results

The results are presented at the scale of the production area considered: the field; and for certain analysis, at the level of the individual farm. The indicators used were:

- Production area scale: overall GM OSR levels in harvested conventional OSR, for each year of the simulation. This scale is equivalent to country elevator level.

- Field scale: the percentage of the land under conventional OSR with a GM OSR content exceeding a certain threshold, for each year of the simulation. For most analyses, we used a threshold of 0.9%, corresponding to EU regulations. This threshold is the only known threshold for OSR production as buyers make no particular demands, by contrast with the situation for maize. This indicator is calculated as follows: the level of GM OSR present in the harvest from each field is calculated. If this level exceeds the threshold, the harvest of the entire field is rejected. No calculations for intrafield areas are possible, whereas such calculations can be made for maize with MAPOD.

- Farm scale: the level of GM OSR in the total conventional OSR harvest from a particular farm.

The GM variety introduced is a variety of OSR tolerant to glyphosate and homozygous for the dominant allele A. The percentage of GM OSR is calculated as follows:

 $GMO = (number of AA seeds + number of aA seeds) \times 100/ total number of seeds.$

This percentage corresponds to the phenotype of the seeds and is not comparable to the results of quantitative PCR. However, we feel that it accurately represents the GM OSR levels in the sense intended by EU legislation, the precise meaning of this level being unclear at the moment.

GENESYS underestimates spatial gene flow. We therefore introduced a correction to facilitate interpretation of the results. Based on a proposal put forward by the designers of the model (Colbach 2004), a value of 0.4% was considered to correspond to the EU norm of 0.9%, and was used as the threshold value in most studies. We took into account the effects of seed impurity, given the additive nature of these effects (COEX2), by using a threshold of 0.266%, corresponding to 0.3% seed impurity, for some results. Similarly, a rate of GM seed detection of 0.01% was corrected to 0.0044% for certain analyses.

3 Simulation results

3.1 Evaluation of the initial situation, with no change in practices other than the introduction of GM OSR

We simulated the introduction of GM OSR, using different proportions of the total area under OSR: 10%, 50% or 75%. No particular measures were taken to ensure coexistence. The output variables were:

- The proportion of the total area under conventional OSR (sown with non-GM OSR) for which contamination with GM OSR exceeded a certain threshold. This variable was calculated by summing the areas of fields considered contaminated on the basis of this threshold.
- The mean level of GM OSR in the total harvest of conventional OSR. This variable was obtained by calculating the mean level of contamination per unit area for all the fields of conventional OSR in the landscape studied.

3.1.1 Spatial coexistence: random dissemination at field level

For each GM introduction rate, we carried out 30 repetitions. This required the generation of 30 landscapes, by randomly distributing the GM OSR fields among the OSR fields (10 variants of landscape A, 10 of landscape B and 10 of landscape C). By "OSR field" we mean a field carrying a cropping system including OSR. By "GM OSR field", we mean a field carrying a cropping system including GM OSR. In these simulations, GM and conventional OSR could not be grown on the same field. Figures 4 to 6 show the results in the form of a boxplot, and the rules for interpreting this boxplot are shown in figure 3.



Figure 3: How to read the boxplots



Figure 4: Areas not satisfying the threshold and GM content in conventional OSR batches when 10% of the OSR grown is GM (30 simulations; 0.9% threshold, corresponding to 0.4% for GENESYS outputs)



Figure 5: Areas not satisfying the threshold and GM content in conventional OSR batches when 50% of the OSR grown is GM (30 simulations; 0.9% threshold, corresponding to 0.4% for GENESYS outputs)



Figure 6: Areas not satisfying the threshold and GM content in conventional OSR batches when 75% of the OSR grown is GM (30 simulations; 0.9% threshold, corresponding to 0.4% for GENESYS outputs)

These graphs illustrate the dilution effect that occurs when passing from the scale of the field to that of the landscape or production area. This effect is fully expressed for a rate of GM OSR introduction of 50%: at this rate, there is a large risk of the threshold being exceeded for an individual field, whereas the risk is almost non-existent at the level of the production area (1 case in 600). Similar results were obtained with a GMO introduction rate of 10%, but with smaller risks at field level. By contrast, when the frequency of GM OSR in the landscape exceeds 75%, there is a real risk of the threshold being exceeded at the level of the production area. The distributions tend to become wider as the proportion of GM OSR increases, due to the small number of fields. At 75% GM OSR, the mean number of conventional OSR fields is only 6 (range: 1-13). Simulations on larger landscapes would probably have given narrower distributions, with greater grouping around the median value.

Furthermore, for the threshold considered, no cumulative effect was observed over the duration of the simulation: risks were similar at the start and end of the simulation.

The proportion of the area for which harvests are rejected depends on the threshold used, as shown in figure 7. The three thresholds, corrected as required for the interpretation of the results, correspond to the legal threshold (0.9%, corrected to 0.4%), the legal threshold taking into account 0.3% seed impurity (or 0.6%, corrected to 0.4%) and the detection threshold (0.01%, corrected to 0.0044%). The extent to which cumulative effects are apparent seems to depend on the threshold considered, as shown in figure 8. If a threshold of 0.01% is used, the proportion of the area for which harvests are rejected tends to increase over time.



Figure 7: Proportion of the area for which harvests were rejected, as a function of threshold (30 simulations for different frequencies of GMO introduction into the landscape, with results shown for year 20.



Figure 8: Proportion of the area for which harvests were rejected, using a threshold of 0.01% (30 simulations, with 10% GM OSR in the landscape)

Simulating the introduction of GM OSR on all neighbouring farms resulted in similar levels of GM contamination for each of the conventional OSR farms, except for farm alpha. This farm was the smallest, and the small number of conventional OSR fields in this case rendered the results highly variable.



Figure 9: GM OSR levels in conventional OSR harvests from each farm (four simulations, each corresponding to one of the four farms growing conventional OSR, all the neighbouring farms growing GM OSR).

3.1.2 Return to conventional OSR

We have simulated a complete return to conventional OSR after 20 years of growing GM OSR over the entire production area. Three simulations were carried out, one for each variant (landscapes A, B and C).



Figure 10: Proportion of the area under conventional OSR exceeding the threshold of 0.04% in the case of a complete return to conventional OSR (3 simulations)



Figure 11: Proportion of GM OSR in the harvest of conventional OSR at the scale of the entire production area in the case of a complete return to conventional OSR (3 simulations)

During the first three years after a complete return to conventional OSR, the previous OSR crop was a GM OSR crop for all the conventional OSR fields considered, because the duration of the rotation was at least three years. Pollen dispersal had little influence on the configuration tested, with GM OSR levels markedly similar during the first three years, and decreasing thereafter. However, the results obtained seem to indicate a slight increase in risk in year 2, followed by a decrease in year 3. This discrepancy may be a consequence of the small sample size. Indeed, the area under OSR on the farms surveyed varied considerably from year to year (see figure 7; we are not dealing here with the areas measured in the survey, but with the areas calculated on the basis of crop allocation in 2004 and the rotations used). Farms beta and delta had large areas under OSR in years 1 and 2, whereas the "unknown" farm had the largest areas under OSR in year 3. This result corresponds to the data collected for the parts of the farms used for simulation (at the scale of the entire farm, the results obtained would have been markedly different, with a more even distribution of OSR over the years). The rates of GM OSR use were much higher on farms beta and delta (figure 12): the high rates recorded for years 1 and 2 may therefore result from a major contribution from these farms.



Figure 12: Contribution of the various farms in the simulation to the area under OSR (cumulative area over the three simulations for a complete return to conventional OSR).



Figure 13: Mean GMO levels in the harvest of conventional OSR, per farm, in the case of a complete return to conventional OSR (3 simulations)

Given these considerations, the risk period of two years identified for the whole production area should be revised upwards to three years. This period is linked to the short duration of the rotations practiced. At the field level, the risk period is four years, with a return to normal values in the fifth year.

3.1.3 Conclusion

The results of the different dissemination patterns can be summarised in the following table:

Table 3: Risk of the conventional OSR harvest exceeding the threshold of 0.4% for the area used for simulations

Dissemination pattern	Spatial coexistence of GM and conventional OSR	Complete return to conventional OSR
Scale		
Field	75 and 50 % GM OSR: high risk (f 52% of the area at most exceeding thresholds for 50% GM OSR) <u>10% GM OSR</u> : moderate risk (15% at most)	Risk for 4 years
Landscape	75% GM OSR: real risk(threshold exceeded in 6.67%of cases)50% GM OSR: very low risk(threshold exceeded in only 1case in 60010% GM OSR: risk negligible	Risk for 3 years

Few cumulative effects were noted. In the case of a complete return to conventional OSR, GM OSR remains in the landscape at very low levels, even after 20 years. However, note that these results are

highly dependent on the characteristics of the simulated area. There is, for example, a direct link between the risk period following a return to conventional OSR and the duration of the rotations used.

3.4 Contribution of basic practices

3.4.1 Critical points for pollen dispersal

Risks associated with the spatial arrangement of crops: post hoc analysis

The following data concern polygons rather than fields (a field is composed of 1 to n four-sided polygons and the field map simulated contains 354 polygons, comprising 321 fields). Thirty simulations involving 50% GM OSR show the relationship between GM OSR levels in the conventional OSR polygons and distance to the nearest GM OSR polygon. Results are presented for the start (year 1) and end (year 20) of the simulation.



Figure 14: Relationship between GM OSR levels and distance to the nearest GM field (triangles: polygons < 0.5 ha; circles: polygons > 0.5 ha)

A threshold phenomenon was observed for small distances, with fields either in contact or more than 10 m apart, corresponding to the width of a path and its borders.

If the nearest GM OSR polygon was at least 50 m away, contamination levels did not exceed 0.034%. In the first year, all the polygons sown to conventional OSR with contamination levels exceeding 0.4% were directly adjacent to GM OSR fields. In year 20, a few nearby polygons exceeded this threshold, indicating a cumulative effect. Small polygons were most at risk of contamination. Such polygons were included in the fields studied here, undoubtedly resulting in an overestimation of the risks of contamination.

Cropping and border management practices: comparison of variants

We carried out a simulation with 50% GM OSR, comparing different border management methods with the initial situation:

- No border management
- Glyphosate treatment
- Double cutting at the start of April and end of May

We also assessed the effects of manipulating OSR sowing date (conventional OSR sowed two weeks after GM OSR) and of lower levels of herbicide efficacy in cereal crops (80% versus 95%).

Border management practices had no marked effect on contamination levels if a threshold of 0.4% was used. By contrast, sowing date manipulation was found to improve the situation at field level, although the final result was not very evident at the level of the entire production area (figures 15 and 16). Indeed, improvements at field level were discrete (a single field below the threshold had a strong impact), whereas those for the production area as a whole were continuous.



Figure 15: Comparison of different cropping practices at the level of the entire production area (1 simulation with 50% GM OSR in the landscape)



Figure 16: Comparison of different cropping practices at field level, using a threshold of 0.4% (1 simulation, with 50% GM OSR in the landscape)

3.4.2 Critical points relating to the deleterious impact of volunteers

Cropping system management: post hoc analysis

OSR cropping systems differ widely from farm to farm. Thus, an analysis of the results of simulations involving a complete return to conventional OSR by farm (the three known farms + the unknown farm) can identify critical points, relating these results to the type of cropping system management. We expressed the results for each farm using the indicator "% GM OSR in the conventional harvest from each farm" in years 1 to 3 (total number of GM seeds harvested in years 1 to 3/ total number of seeds harvested. Three simulations were carried out for each landscape (A, B and C).

Table 4: GM OSR levels in the cumulative conventional OSR harvest from each farm in years 1 to 3, in the case of a complete return to conventional OSR (3 simulations)

		Landscape			
Farm	OSR cropping system	Α	В	С	Total
Alpha	<u>3-year rotation</u> : deep ploughing \rightarrow OSR \rightarrow chisel				
	ploughing \rightarrow wheat \rightarrow chisel ploughing \rightarrow wheat				
		0.26%	0.26%	0.25%	0.26%
Beta	<u>3-year rotation</u> : deep ploughing \rightarrow OSR \rightarrow shallow				
	ploughing → wheat → deep ploughing → wheat				
	(spring barley)	0.73%	0.71%	0.69%	0.71%
Delta	<u>4-year rotation</u> : deep ploughing \rightarrow OSR \rightarrow deep				
	ploughing → wheat → deep ploughing → wheat				
	→ deep ploughing → winter barley	0.61%	0.61%	0.58%	0.60%
Unknown	<u>3-year rotation</u> : chisel ploughing \rightarrow OSR \rightarrow chisel				
	ploughing \rightarrow wheat \rightarrow deep ploughing \rightarrow wheat				
	(18% of the total area) or				
	<u>4-year rotation</u> : chisel ploughing \rightarrow OSR \rightarrow chisel				
	ploughing \rightarrow wheat \rightarrow deep ploughing \rightarrow wheat				
	\rightarrow deep ploughing \rightarrow wheat (winter barley) (72%)				
	of the total area)	0.15%	0.15%	0.16%	0.15%

These results show that there are large differences between farms. These differences may be related to elements of the cropping system (alternation of ploughing with minimal tillage techniques).

We explored this "cropping system effect" linked to soil tillage, all other factors being equal, by setting up a virtual experiment in a simplified landscape of fields 1 km apart, rendering pollen flow negligible. On these fields, all sequences of soil tillage (deep or chisel ploughing) were tested in a situation in which there was a complete return to conventional OSR after 20 years of GP OSR use in three- and four-year rotations. The reference cropping system was considered to be that used on the unknown farm. The following tables classify the combinations of soil tillage sequences for several variants of the cropping systems tested; the aim is to illustrate the sensitivity of the model to differences in soil tillage.

Table 5: Levels of GM OSR in conventional OSR harvests in a situation of a complete return to conventional OSR in an OSR-wheat-wheat-wheat rotation (conventional OSR grown on a given field four years after the last GM OSR crop)

Soil tillage*	Deep (D) or chisel (C) ploughing before wheat on 16/09	Deep (D) or chisel (C) ploughing before wheat on 01/10	Last wheat crop in the rotation replaced by spring barley
DCCC	0.02%	0.03%	0.02%
CCCC	0.04%	0.04%	0.03%
DCCD	0.05%	0.06%	0.05%
CCCD	0.07%	0.08%	0.07%
CDCD	0.07%	0.08%	0.06%
DCDC	0.10%	0.10%	0.09%
CDCC	0.13%	0.13%	0.10%
DCDD	0.15%	0.15%	0.14%
CCDD	0.19%	0.19%	0.19%
CDDC	0.45%	0.40%	0.28%
DDDC	0.80%	0.63%	0.55%
DDCD	1.02%	0.79%	0.78%
DDDD	1.08%	0.82%	0.81%
CDCD	1.24%	0.96%	0.96%
CDDD	1.30%	1.00%	1.00%
DDCC	1.46%	1.10%	1.08%

*D indicates deep ploughing and C indicates chisel ploughing. A soil tillage sequence for the crops of the rotation, in order, of CDDD indicates chisel ploughing before OSR and then deep ploughing before each of the subsequent three wheat crops

Table 6: GM OSR levels in the conventional OSR harvest in the case of a complete return to conventional OSR in the context of an OSR-wheat-wheat rotation

Soil tillage *	Deep (D) or chisel (C) ploughing before wheat on 16/09	Last wheat crop in the rotation replaced by spring barley
DCC	0.13%	0.11%
CCC	0.21%	0.17%
DCD	0.30%	0.27%
CCD	0.40%	0.38%
CDC	0.68%	0.46%
DDC	1.54%	1.41%
DDD	1.65%	1.65%
CDD	1.96%	1.98%

The best results seem to be obtained as follows:

- In general, less deep ploughing is associated with better results
- Deep ploughing should be avoided after the OSR crop. If this is not possible, the risks of contamination can be minimised by not ploughing before the next crop in four-year rotations (CDCC rotation)
- If deep ploughing is carried out after the OSR crops, the risks are smaller if ploughing is carried out later
- The replacement of the last wheat crop by spring barley does not greatly improve the situation.

These results are largely consistent with those obtained for the Marolles landscape: farm alpha (DCC) had GM OSR levels of 0.26% (table 4), versus 0.13% for the same sequence in this test (table 5); farm delta (DDDD) had GM OSR levels of 0.60%, versus 0.82% in this test; the unknown farm (mostly CCDD) had GM OSR levels of 0.15%, versus 0.19% in this test. By contrast, farm beta had GM OSR levels of 0.71%, versus only 0.27% in this test. This discrepancy may be due to technical differences in cropping practices. Indeed, the no-tillage practices employed by this farmer did not involve chisel ploughing in autumn. By replacing chisel ploughing with additional stubble breaking in summer in the simulation, we obtained a level of 0.59% GM OSR, consistent with the 0.71% obtained for Marolles. This result demonstrates that simplified cultivation practices in summer may prove highly risky.

The differences in technical practices between farms were accentuated in the simulations presented. The clear differences in soil tillage practices simulated (typical technical schedules) may be attenuated by wet summers, after which most farmers resort to deep ploughing. Weather conditions in the summer may therefore have a major impact on GM OSR levels in non-GM OSR harvests.

Cropping practices: comparison of the variants

Following a complete return to conventional OSR after 20 years of GM OSR use, we evaluated the effects of the following variants during both the duration of the simulation and the prior constitution of seed stocks:

- Changes to soil tillage practices: deep ploughing before but not after OSR. This strategy for dealing with volunteers aims to bury the youngest seeds just before the OSR crop and to avoid their burial after harvest. This modification to practices would affect only farm delta and the unknown farm.
- Reducing seed losses from 7% to 2%. This would involve the use of a "special OSR" cutting bar by all the farms in the zone (Sausse *et al.*, 2006), whereas none of these farms currently have such equipment.
- Changes to stubble management: a stubble-breaking operation no longer carried out before wheat and OSR. As stubble breaking is generally carried out twice before sowing, this change amounts to delaying the first stubble breaking operation after harvest.
- Decrease in the efficacy with which herbicides kill volunteer OSR in cereal crops: 80% rather than 95%

For each of these options, a single situation of a complete return to conventional OSR was simulated, for comparison with the original practices.



Figure 17: Comparison of different practices on the proportion of GM OSR in the conventional OSR harvest after a complete return to conventional OSR (1 simulation for each modification).



Figure 18: Comparison of different practices on the proportion of the land under conventional OSR exceeding the threshold of 0.4% (1 simulation for each modification).

The risks of contamination were greatly reduced if grain loss was reduced, particularly at field level. However, seed losses at harvest probably vary with climatic conditions, whereas these simulations take into account only mean losses. Changes in soil tillage practices were less effective at field level, but made it possible to achieve quality objectives for the landscape as a whole. This result confirms those of the previous analysis (post hoc analysis on the management of cropping systems) and the importance of soil tillage in the management of volunteers. Delaying the first stubble breaking operation gave a slight improvement in contamination levels for the entire production area. By contrast, changes to the efficacy of herbicides against OSR volunteers had no effect.

Soil tillage practices are, in reality, adapted to the conditions each year. This "year" effect, linked to climate, is indirect, as it concerns the input variables of the model. The model is particularly sensitive to the sequence of soil tillage operations in the rotation and to the loss of OSR seed at harvest. All the other input variables, except field and varietal characteristics, are also subject to climatic fluctuations, but the model is less sensitive to these fluctuations. Consequently, the results presented here can only provide indications as to the possible room for manoeuvre, but cannot predict the efficacy of management strategies for which the results obtained depend on the climatic context.

3.4.3 Critical points relating to seed use

Impurities in certified seed lots have an essentially additive effect (COEX1), making it possible to reason in terms of modifications to the threshold for harvest contamination. The dotted line on the graph indicates the new threshold for the harvest corresponding to a seed impurity level of 0.266%, allowing evaluation of the associated increase in risk.

For farm-saved seed, spatial coexistence was simulated based on the use of such seeds on the unknown farm, corresponding to about 50% of the land under OSR.



Figure 19: Effect of the use of farm-saved seed on the proportion of GM OSR in the conventional OSR harvest, at the level of the entire production area (1 simulation)



Figure 20: Effect of the use of farm-saved seeds on the proportion of the area under conventional OSR not meeting the threshold of 0.4% maximal contamination (1 simulation)

The results show a strong cumulative effect from year 10 onwards, confirming the conclusions of the COEX2 study.

3.4.3 Conclusion

As far as contamination through pollen is concerned, only adjoining fields are at high risk. This result was unexpected, as the high risk linked to field pattern and the high frequency of OSR might have generated much higher levels of contamination. Good volunteer management within rotations may

have contributed to this result (no bare fallow and intercropping practices including multiple passages), by limiting cumulative effects. By contrast, border management seems to have a limited effect.

Volunteers may be responsible for high levels of contamination in the case of a return to the use of conventional OSR. We identified the following critical points in this context:

- Short rotations
- High seed losses during harvest
- High-risk soil tillage sequences: the sequence of tillage operations in the rotation may have a major effect on the results obtained.

These critical points vary considerably between farms (duration of the rotation and soil tillage strategy) and as a function of climate (losses at harvest and soil tillage strategy). The simulations carried out did not take the effects of climate fluctuations into account and the results obtained depend strongly on the hypotheses concerning the management of cropping systems. Large differences were observed between farms, so caution is required when extrapolating the results obtained for the small areas used for simulations.

The use of farm-saved seed proved to be extremely risky, due to cumulative effects. The situation of the case study in terms of the use of farm-saved seed is difficult to assess. The survey carried out in 2005 indicated that the use of farm-saved seeds was marginal, at odds with estimates obtained at national level.

3.5 The case of organic OSR production

No organic OSR crops have been identified in the study zone or the area used for simulation. We investigated possible organic/GM OSR coexistence, by transforming farm delta into an organic farm, based on the advice of experts and published references (ENITA, 2003). The organic cropping system had the following characteristics:

- Adoption of a single rotation for the whole of the area considered: alfalfa/alfalfa/wheat/OSR/wheat/sunflower.
- Herbicide treatments replaced by mechanical weeding.
- Systematic deep ploughing before each crop.
- Modifications to cropping practices for OSR: sowing advanced by two weeks; deep ploughing after alfalfa and use of a power harrow for preparation of the seedbed.

The question of effects on organic farms arises when GM OSR is introduced by neighbouring farmers. It does not concern the conversion of fields previously planted with GM OSR into organic fields, at least in the medium and short term. The results presented above show that, over a 20-year period, GM OSR is not completely eliminated.

We carried out three simulations, with 50% GM OSR in the neighbourhood of the organic farm. Mean GM OSR levels in the organic harvest were compared with those for the conventional OSR harvest of the unknown farm.



Figure 20: Comparison between an organic farm and a conventional OSR farm in a landscape containing 50% GM OSR

There seems to be slightly less GM OSR in the organic harvest. As contamination occurs principally through pollen, this result may be a consequence of a statistical bias (too few situations simulated, local effects of field arrangement). There seem to be no cumulative effects, consistent with a cropping system in which OSR is grown only once every six years, with effective volunteer control achieved through the cultivation of alfalfa. These results should be interpreted with caution, as they are highly dependent on the local configuration, and on account of the limited size of the area used for simulations.
4 Identification of critical points: summary

The simulations carried out for landscapes at risk showed that, using a threshold of 0.9%, the possibility of GM and conventional OSR coexisting with no change in current practices depends on two factors: the rate of introduction of GM OSR and the unit of analysis considered (field or elevator). For 10% GM OSR in the landscape, the risk of exceeding the threshold at the elevator is negligible, but this risk is real if results are analysed at field level. Similarly, simulations showed that, with the introduction of 50% GM OSR into the landscape, management at elevator level would be possible, but there would be a very high risk of contamination thresholds being exceeded at field level. With the introduction of 75% GM OSR, there would be a high risk of contamination at both elevator and field levels.

A review of the literature and simulations identified several critical points at the level of the cropping system for the "Beauce" region.

Spatial coexistence may be compromised by:

- Spatial proximity. Conventional OSR fields adjacent to GM OSR fields are at risk of contamination. This risk is linked to the high proportion of OSR in the landscape and is particularly high for certain types of field patterns (small and dispersed fields).
- Seed purity: the use of farm-saved seed may have cumulative effects, gradually decreasing performance over a number of years. Impure seed lots have a directly visible additive effect.

The possibility of returning GM OSR fields to conventional OSR depends on a number of critical points:

- The interval between successive OSR crops in the rotation: three- to four-year rotations are the most at risk.
- Soil tillage practices: deep ploughing after OSR has a deleterious effect on GM OSR contamination levels.
- Seed purity (see above).
- Seed losses: the seeds lost at harvest increase seed stocks. These losses depend on a number of factors, which may (cropping practices and harvesting material) or may not (weather) be controllable.

The degree of heterogeneity of the spatial distribution of these critical factors varies:

- There is a tendency for there to be small homogeneous zones within the field pattern of the region, due to local programmes aiming to combine the fields of individual farms. A single farm may overlap several of these zones, with or without continuity (in the case of farms dispersed into several blocks). The distribution and number of farms of different type is not currently available to use. We also lack information concerning the frequency of adjoining OSR fields.
- Rotations depend on the technical and economic orientation of the farm, but also tend to be homogeneous within small zones within the region in the case of specialised production systems (for seed). Deliverable 7.1 provides information about the rotations and their duration, but not their distribution within the region.
- The use of risky cropping practices (use of farm-saved seed, tillage) depends largely on the farm, with no pattern over the production area as a whole. Data concerning these practices are provided in deliverable 7.1.

These characteristics also tend to change over time:

- Soil tillage practices depend on the climatic context and, thus, on the year. However, they also depend on more long-term strategic decisions (choice of equipment).
- Rotations are likely to change rapidly, depending on the price obtained for particular crops and regulations. They also depend on the technical and economic orientation of the farm.
- The types of field patterns are likely to change much more slowly.

Table 7 provides a summary showing the relationship between critical points and their principal determinants.

Other critical points have been identified outside the strict framework of the cropping system:

- The cleaning of material, principally harvesting equipment, in cases of mixed use (coexistence on a single farm or for the farms using a given agricultural work contractor)
- -Transport from field to elevator.
- Storage on the farm and sale to multiple buyers.

Time scale	Determinant	Туре	Spatial distribution	Change over time	Type of risk	Critical points	Consequence	Vector
Invariant	Useful	Weak to strong	Grouping into small	None	Low: few alternatives	Amount of OSR	High probability of	Pollen
	reserves		homogeneous zones		to OSE	and interval	adjoining GM and	
Centuries,	Field pattern	3 types according	Grouping into small	Tendency towards	Types A	between	conventional OSR	
decades		to Casagrande	homogeneous zones	combining fields on		successive OSR	fields	
				a given farm		crops		
Several	Crop	Possibility of seed	Grouping into small	Traditional	No contracts: few	Type of field	OSR frequent in	Pollen
years	diversification	contracts	homogeneous zones	production zone	alternatives to OSR	patterns	rotation	
Several	Choice of	Irrigation	Intra- and interfarm	Strong increase	No irrigation: few			
years	equipment		differences	since the 1990s	alternatives to OSE			
Several	Choice of	Deep ploughing or	Intra- and interfarm	Limited tillage	Risk associated with	Soil tillage	Increase in seed	Volunteers
years	equipment	limited tillage	differences	systems becoming	ploughing generally		stocks	
-		systems		increasingly	and with ploughing			
				common	after OSR in particular			
Several	Choice of	Early or classic	Farm-specific	Specific cutting	Classic cutting			
years	equipment	cutting time		equipment for OSR				
				becoming more				
				common				
Before	Crop	Amount of land	Mean of 13%, but with	Tendency for the	Large proportion of the	Seed purity	Multiplication of seed	Volunteers
sowing	allocation	under OSR	differences, organised into	amount of land	land under OSR		stocks	and pollen
			small homogeneous zones	under OSR to				
			(associated with structural	increase				
			factors)					
Before	Choice of	Farm-saved or	Intra- and interfarm	Relative stability	Impure farm-saved or			
sowing	seed	certified seed, pure	differences		certified seed			
		or impure seed						
		batches						
Current	Weather	Conditions before	Unknown	Unknown	Hail	Seed losses		
growing		harvest						
season								
Current	Weather	Summer conditions	Unknown	Unknown	Ploughing			
growing								
season				1			1	

Table 7: Proposed link between the critical points concerning cropping system and their principal determinants

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APPENDIX 1 : rotations

num(nom de la rotation

duré ensuite, pour chaque année de la rotation num nom d'itinéraire technique

1	agri1C/BD/BD	3	3 Vcolza	4 Vble	4 Vble		
2	agri1CR/BD/BD	3	2 VcolzaGM	4 Vble	4 Vble		
3	agri2C/B/B	3	3 Xcolza	4 Xblecolza	4 Xbleble		
4	agri2CR/B/B	3	2 XcolzaGM	4 Xblecolza	4 Xbleble		
5	agri2C/B/OP	3	3 Xcolza	4 Xblecolza	5 Xorgep		
6	agri2CR/B/OP	3	2 XcolzaGM	4 Xblecolza	5 Xorgep		
7	agri2PH/B/OH	3	4 Xpoish	4 Xblepoish	4 Xorgeh		
8	agri2Gelfixe	1	9 gelfixe				
9	agri2T/B/OH	3	5 Xtournesol	4 Xbletournes	4 Xorgeh		
10	InconnuC/BD/BD	3	3 icolza	4 ibledcolzal	4 ibledble		
11	InconnuCR/BD/BD	3	2 icolzaGM	4 ibledcolzal	4 ibledble		
12	InconnuC/BD/BD/BT	4	3 icolza	4 ibledcolzal	4 ibledble	4 ibletble	
13	InconnuCR/BD/BT	4	2 icolzaGM	4 ibledcolzal	4 ibledble	4 ibletble	
14	InconnuC/BD/BD/OH	4	3 icolza	4 ibledcolzal	4 ibletble	4 iorgeh	
15	InconnuCR/BD/BD/OH	4	2 icolzaGM	4 ibledcolzal	4 ibletble	4 iorgeh	
16	InconnuC/BD/BT/BT	4	3 icolza	4 ibledcolzal	4 ibletble	4 ibletble	
17	InconnuCR/BD/BT/BT	4	2 icolzaGM	4 ibledcolzal	4 ibletble	4 ibletble	
18	InconnuC/BT/BT	3	3 icolza	4 ibletcolzal	4 ibletble		
19	InconnuCR/BT/BT	3	2 icolzaGM	4 ibletcolzal	4 ibletble		
20	InconnuC/BT/BT/OH	5	3 icolza	4 ibletcolzal	4 ibletble	4 iorgeh	4 iorgeh
21	InconnuCR/BT/BT/OH	5	2 icolzaGM	4 ibletcolzal	4 ibletble	4 iorgeh	4 iorgeh
22	InconnuC/BT/OP	3	3 icolza	4 ibletcolzal	5 iorgep		
23	InconnuCR/BT/OP	3	2 icolzaGM	4 ibletcolzal	5 iorgep		
24	InconnuGelfixe	1	9 gelfixe				
25	InconnuMD/BD/BD	3	5 imais	4 ibledmais	4 ibledble		
26	InconnuPH/BD/BD	3	4 ipoish	4 ibledpoish	4 ibledble		
27	InconnuPH/BD/BD/BT	4	4 ipoish	4 ibledpoish	4 ibledble	4 ibletble	
28	InconnuT/BD/BD	3	5 itournesol	4 ibledtournes	4 ibledble		
29	InconnuT/BD/BD/BT	4	5 itournesol	4 ibledtournes	4 ibledble	4 ibletble	
30	InconnuT/BD/BD/OH	4	5 itournesol	4 ibledtournes	4 ibledble	4 iorgeh	
31	agri3C/BD/BD/BT	4	3 XIIcolza	4 XIIbled	4 XIIbled	4 XIIblet	
32	agri3CR/BD/BD/BT	4	2 XIIcolzaGM	4 XIIbled	4 XIIbled	4 XIIblet	
33	agri3C/BT/BT/OH/OH	5	3 XIIcolza	4 XIIblet	4 XIIblet	4 iorgeh	4 iorgeh
34	agri3CR/BT/BT/OH/OH	5	2 XIIcolzaGM	4 XIIblet	4 XIIblet	4 iorgeh	4 iorgeh
35	agri3PH/BD/BD/BT	4	4 XIIpoish	4 XIIbled	4 XIIbled	4 XIIblet	
36	bordureagri1	1	0 bordureagri1				
37	bordureagri2	1	0 bordureagri2				
38	bordureagri3	1	0 bordureagri3				
39	bordureinconnu	1	0 bordureinconnu	ı			
40	bordureDDE	1	0 bordureDDE				

culture	nom de la c	u nom de l'itk	variété cola	za variété2 colz	part variété 1 mode d	e re
	0 bordure	bordureboul	a∖x	х	-1	-
	0 bordure	bordureadar	n x	x	-1	-
	0 bordure	borduresalm	io x	x	-1	-
	0 bordure	bordureinco	nrx	x	-1	-
	0 bordure	bordureDDE	x	x	-1	-
	2 colza	VcolzaGM	RU	non	1 NON	
	2 colza	XcolzaGM	RU	non	1 NON	
	2 colza	XIIcolzaGM	RU	non	1 NON	
	2 colza	icolzaGM	RU	non	1 NON	
	3 colza	Vcolza	classique	non	1 NON	
	3 colza	Xcolza	classique	non	1 NON	
	3 colza	XIIcolza	classique	non	1 NON	
	3 colza	icolza	classique	non	1 NON	
	4 hlé	Vhle	v v	Y	-1	_
	4 blódur	Xblecolza	× ×	x	-1	
	4 blótondro	Xhlahla	×	×	-1	-
	4 blédur	Xhlenoish	×	×	-1	-
	4 blódur	Xblotournos		~	-1	-
	4 bleuui 4 bló	XIIblot	01X	×	-1	-
	4 DIE 4 bló	XIIbled	×	×	-1	-
	4 blódur	iblodcolzal	~	~	-1	-
	4 blédur	ibledcolzall	×	×	-1	-
	4 blétondro	ibletcolzal	~	x	-1	-
	4 blétendre	ibletcolzall	x	X	-1	-
	4 bletenure 4 blédur	ibledble	x	X	- 1	-
	4 blétandra	ibletble	x	X	- 1	-
	4 bletenure 4 blédur	ibledneich	*	x	-1	-
	4 bledur 4 blédur	ibleapoisn	X	X	-1	-
	4 bledur 4 blédur	ibledtournes	SOLX	x	-1	-
	4 piedur 4 maiabiwan	Diedmais	x	X	-1	-
	4 poisniver	Xpoisn	x	x	-1	-
	4 orgeniver	Xorgen	x	x	-1	-
	4 poisniver	XIIpoisn	x	x	-1	-
	4 orgeniver	ipoisn	x	X	-1	-
	4 orgeniver	lorgen	х	х	-1	-
	5 orgeprintem	p: xorgep	х	х	-1	-
	5 tournesol	Xtourneso	х	х	-1	-
	5 orgeprintem	p: Xllorgep	х	х	-1	-
	5 poisprintem	osipoisp	х	х	-1	-
	5 tournesol	itournesol	х	х	-1	-
	5 maïsdoux	imais	х	х	-1	-
	5 orgeprintem	p: iorgep	х	х	-1	-
	9 gelfixe	gelfixe	Х	Х	-1	-

APPENDIX 2 : agricultural practices

culture	nom de la cu types de semprop de MS ∈nature du co⊦type date						
			de	nsité max broya	age 1		
	0 bordure	-1	-1	-1	1	999	
	0 bordure	-1	-1	-1	1	15/06/1900	
	0 bordure	-1	-1	-1	1	15/06/1900	
	0 bordure	-1	-1	-1	1	15/06/1900	
	0 bordure	-1	-1	-1	1	999	
	2 colza	0	-1	-1	0	-1	
	2 colza	0	-1	-1	0	-1	
	2 colza	0	-1	-1	0	-1	
	2 colza	0	-1	-1	0	-1	
	3 colza	0	-1	-1	0	-1	
	3 colza	0	-1	-1	0	-1	
	3 colza	0	-1	-1	0	-1	
	3 colza	0	-1	-1	0	-1	
	4 blé	-1	-1	400	0	-1	
	4 blédur	-1	-1	400	0	-1	
	4 blétendre	-1	-1	400	0	-1	
	4 blédur	-1	-1	400	0	-1	
	4 blédur	-1	-1	400	0	-1	
	4 blé	-1	-1	400	0	-1	
	4 blé	-1	-1	400	0	-1	
	4 blédur	-1	-1	400	0	-1	
	4 blédur	-1	-1	400	0	-1	
	4 blétendre	-1	-1	400	0	-1	
	4 blétendre	-1	-1	400	0	-1	
	4 blédur	-1	-1	400	0	-1	
	4 blétendre	-1	-1	400	0	-1	
	4 blédur	-1	-1	400	0	-1	
	4 blédur	-1	-1	400	0	-1	
	4 blédur	-1	-1	400	0	-1	
	4 poishiver	-1	-1	128	0	-1	
	4 orgehiver	-1	-1	400	0	-1	
	4 poishiver	-1	-1	165	0	-1	
	4 orgehiver	-1	-1	400	0	-1	
	4 orgehiver	-1	-1	400	0	-1	
	5 orgeprintemps	-1	-1	400	0	-1	
	5 tournesol	-1	-1	11	0	-1	
	5 orgeprintemps	-1	-1	400	0	-1	
	5 poisprintemps	-1	-1	135	0	-1	
	5 tournesol	-1	-1	11	0	-1	
	5 maïsdoux	-1	-1	11	0	-1	
	5 orgeprintemps	-1	-1	400	0	-1	
	9 gelfixe	-1	-1	-1	1	15	

culture	nom de la cu type	e date /age 2	da	ate de semi den	sité de s∉pert	te à la réc
	0 bordure	1	999	-1	-1	-1
	0 bordure	1	999	-1	-1	-1
	0 bordure	1	999	-1	-1	-1
	0 bordure	1	999	-1	-1	-1
	0 bordure	1	999	-1	-1	-1
	2 colza	0	-1	27-août	65	0.07
	2 colza	0	-1	31-août	41	0.07
	2 colza	0	-1	2-sept	42	0.07
	2 colza	0	-1	1-sept	50	0.07
	3 colza	Ő	-1	27-août	65	0.07
	3 colza	0	-1	31-août	41	0.07
	3 colza	Ő	-1	2-sent	42	0.07
	3 colza	Ő	-1	1-sept	50	0.07
	4 blé	0	-1	27-oct	300	-1
	4 blédur	Ő	-1	7-nov	265	-1
	4 blétendre	0	-1	7-nov	265	-1
	4 blédur	0	-1	20-oct	230	-1
	4 blédur	0	-1	7-nov	265	-1
	4 blé	0	-1	22-oct	275	-1
	4 blé	0	-1	27-oct	300	-1
	4 blédur	0	-1	1-nov	300	-1
	4 blédur	0	-1	1-nov.	300	-1
	4 blétendre	0	-1	20-oct	280	-1
	4 blétendre	0	-1	20-oct	280	-1
	4 blédur	0	-1	1-nov.	300	-1
	4 blétendre	0	-1	20-oct.	280	-1
	4 blédur	0	-1	1-nov	300	-1
	4 blédur	0	-1	1-nov	300	-1
	4 blédur	0	-1	1-nov	300	-1
	4 poishiver	0	-1	20-nov.	85	-1
	4 orgehiver	0	-1	10-oct.	210	-1
	4 poishiver	0	-1	17-nov.	110	-1
	4 orgehiver	0	-1	10-nov.	90	-1
	4 orgehiver	0	-1	15-oct.	220	-1
	5 orgeprintemps	0	-1	20-févr.	220	-1
	5 tournesol	0	-1	7-avr.	7	-1
	5 orgeprintemps	0	-1	20-févr.	220	-1
	5 poisprintemps	0	-1	1-mars	90	-1
	5 tournesol	0	-1	15-avr.	7	-1
	5 maïsdoux	0	-1	1-mai	7	-1
	5 orgeprintemps	0	-1	20-févr.	220	-1
	9 gelfixe	1	999	-1	-1	-1
	3	-		-		

culture	nom de la cu var	iété T1 vari	iété T2 var	iété T 3 clas	siques/s vari	été T 1
	mo	rtalité herbici	de appliqué a	u stade plantu	lle (semismor	talité her
	0 bordure	0	0	0	0	0
	0 bordure	0	0	0	0	0
	0 bordure	0	0	0	0	0
	0 bordure	0	0	0	0	0
	0 bordure	0	0	0	0	0
	2 colza	0	0	0	0.9	0
	2 colza	0	0	0	0.9	0
	2 colza	0	0	0	0.9	0
	2 colza	0	0	0	0.9	0
	3 colza	-1	-1	-1	-1	-1
	3 colza	-1	-1	-1	-1	-1
	3 colza	-1	-1	-1	-1	-1
	3 colza	-1	-1	-1	-1	-1
	4 blé	0.95	0.95	0.95	0.95	0.7
	4 blédur	0.95	0.95	0.95	0.95	0
	4 blétendre	0.95	0.95	0.95	0.95	0
	4 blédur	0.95	0.95	0.95	0.95	0.9
	4 blédur	0.95	0.95	0.95	0.95	0
	4 blé	0.95	0.95	0.95	0.95	0
	4 blé	0.95	0.95	0.95	0.95	0
	4 blédur	0.95	0.95	0.95	0.95	0
	4 blédur	0.95	0.95	0.95	0.95	0
	4 blétendre	0.95	0.95	0.95	0.95	0
	4 blétendre	0.95	0.95	0.95	0.95	0
	4 blédur	0	0	0	0	0.9
	4 blétendre	0.95	0.95	0.95	0.95	0
	4 blédur	0.95	0.95	0.95	0.95	0
	4 blédur	0.95	0.95	0.95	0.95	0
	4 blédur	0.95	0.95	0.95	0.95	0
	4 poishiver	0.9	0.9	0.9	0.9	0
	4 orgehiver	0.95	0.95	0.95	0.95	0
	4 poishiver	0.9	0.9	0.9	0.9	0
	4 orgehiver	0.9	0.9	0.9	0.9	0
	4 orgehiver	0.95	0.95	0.95	0.95	0
	5 orgeprintemps	0	0	0	0	0.95
	5 tournesol	0	0	0	0	0.9
	5 orgeprintemps	0	0	0	0	0.95
	5 poisprintemps	0	0	0	0	0.9
	5 tournesol	0	0	0	0	0.9
	5 maïsdoux	0	0	0	0	0.9
	5 orgeprintemps	0	0	0	0	0.8
	9 gelfixe	0	0	0	0	0

culture	nom de la cu var	iété T2 var	iété T3 clas	siques/s bina	ge nbd	e travau:
	bici	de appliqué a	u printemps	prop	o surface biné	e
	0 bordure	0	0	0	-1	-1
	0 bordure	0	0	0	-1	-1
	0 bordure	0	0	0.8	-1	-1
	0 bordure	0	0	0	-1	-1
	0 bordure	0	0	0.8	-1	-1
	2 colza	0	0	0.9	0	5
	2 colza	0	0	0.9	0	4
	2 colza	0	0	0.9	0	3
	2 colza	0	0	0.9	0	4
	3 colza	-1	-1	-1	0	5
	3 colza	-1	-1	-1	0	4
	3 colza	-1	-1	-1	0	3
	3 colza	-1	-1	-1	0	4
	4 blé	0.7	0.7	0.7	0	4
	4 blédur	0	0	0	0	5
	4 blétendre	0	0	0	0	5
	4 blédur	0.9	0.9	0.9	0	3
	4 blédur	0	0	0	0	3
	4 blé	0	0	0	0	4
	4 blé	0	0	0	0	4
	4 blédur	0	0	0	0	4
	4 blédur	0	0	0	0	4
	4 blétendre	0	0	0	0	4
	4 blétendre	0	0	0	0	4
	4 blédur	0.9	0.9	0.9	0	4
	4 blétendre	0	0	0	0	4
	4 blédur	0	0	0	0	4
	4 blédur	0	0	0	0	2
	4 blédur	0	0	0	0	2
	4 poishiver	0	0	0	0	4
	4 orgehiver	0	0	0	0	4
	4 poishiver	0	0	0	0	4
	4 orgehiver	0	0	0	0	3
	4 orgehiver	0	0	0	0	3
	5 orgeprintemps	0.95	0.95	0.95	0	4
	5 tournesol	0.9	0.9	0.9	0	4
	5 orgeprintemps	0.95	0.95	0.95	0	4
	5 poisprintemps	0.9	0.9	0.9	0	4
	5 tournesol	0.9	0.9	0.9	0	4
	5 maïsdoux	0.9	0.9	0.9	0	4
	5 orgeprintemps	0.8	0.8	0.8	0	4
	9 gelfixe	0	0	0	-1	-1

culture	nom de la cu pour chaque travail, le type et la date	
---------	--	--

0 bordure			
0 bordure			
2 colza	MELANGE5	30-juil. MELANGE5	15-août LABOURRAS
2 colza	MELANGE5	30-juil. LABOURRAS	15-août MELANGE10
2 colza	MELANGE5	20-juil. LABOURRAS	1-août HROT
2 colza	MELANGE5	30-juil. CHISEL	15-août MELANGE10
3 colza	MELANGE5	30-juil. MELANGE5	15-août LABOURRAS
3 colza	MELANGE5	30-juil. LABOURRAS	15-août MELANGE10
3 colza	MELANGE5	20-juil. LABOURRAS	1-août HROT
3 colza	MELANGE5	30-juil. CHISEL	15-août MELANGE10
4 blé	MELANGE5	30-juil. MELANGE5	15-sept. CHISEL
4 blédur	MELANGE5	30-juil. MELANGE5	15-août MELANGE10
4 blétendre	MELANGE5	30-juil. MELANGE5	15-août LABOURRAS
4 blédur	MELANGE5	30-juil. MELANGE5	15-août HROT
4 blédur	MELANGE5	15-sept. LABOURRAS	6-nov. HROT
4 blé	MELANGE5	25-juil. MELANGE5	15-août LABOURRAS
4 blé	MELANGE5	25-juil. MELANGE5	15-août LABOURRAS
4 blédur	MELANGE5	30-juil. MELANGE5	15-août CHISEL
4 blédur	MELANGE5	30-juil. MELANGE5	15-août LABOURRAS
4 blétendre	MELANGE5	30-juil. MELANGE5	15-août CHISEL
4 blétendre	MELANGE5	30-juil. MELANGE5	15-août LABOURRAS
4 blédur	MELANGE5	30-juil. MELANGE5	15-août LABOURRAS
4 blétendre	MELANGE5	30-juil. MELANGE5	15-août LABOURRAS
4 blédur	MELANGE5	30-juil. MELANGE5	15-août LABOURRAS
4 blédur	LABOURRAS	15-sept. HROT	1-nov.
4 blédur	LABOURRAS	15-sept. HROT	1-nov.
4 poishiver	MELANGE5	30-juil. MELANGE5	30-août LABOURRAS
4 orgehiver	MELANGE5	30-juil. LABOURRAS	1-sept. MELANGE10
4 poishiver	MELANGE5	25-juil. MELANGE5	15-août LABOURRAS
4 orgehiver	MELANGE5	30-juil. LABOURRAS	15-oct. HROT
4 orgehiver	MELANGE5	30-juil. LABOURRAS	15-sept. HROT
5 orgeprintem	p: MELANGE5	1-août LABOURRAS	15-nov. MELANGE10
5 tournesol	MELANGE5	30-juil. LABOURRAS	15-nov. MELANGE10
5 orgeprintem	p: MELANGE5	1-août LABOURRAS	15-nov. MELANGE10
5 poisprintem	ps MELANGE5	5-août LABOURRAS	15-nov. MELANGE10
5 tournesol	MELANGE5	1-août LABOURRAS	15-nov. MELANGE10
5 maïsdoux	MELANGE5	30-août LABOURRAS	15-nov. MELANGE10
5 orgeprintem	p: MELANGE5	1-août LABOURRAS	15-nov. MELANGE10
9 gelfixe			

culture nom de la cu

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0	bordure				
0	bordure				
0	bordure				
0	bordure				
0	bordure				
2	colza	26-août	MELANGE10	26-août HROT	27-août
2	colza	29-août	HROT	30-août	
2	colza	2-sept.			
2	colza	29-août	HROT	1-sept.	
3	colza	26-août	MELANGE10	26-août HROT	27-août
3	colza	29-août	HROT	30-août	
3	colza	2-sept.			
3	colza	29-août	HROT	1-sept.	
4	blé	25-oct.	MELANGE10	26-oct.	
4	blédur	15-août	MELANGE10	6-nov. HROT	7-nov.
4	blétendre	1-oct.	MELANGE10	6-nov. HROT	7-nov.
4	blédur	20-oct.			
4	blédur	7-nov			
4	blé	20-oct	HROT	22-oct	
4	blé	20-oct	HROT	27-oct	
4	blédur	15-sept	HROT	20-oct	
4	blédur	15-sept	HROT	20-oct	
4	blétendre	15-sept.	HROT	20-oct.	
4	blétendre	15-sept	HROT	20-oct	
4	blédur	15-sept.	HROT	1-nov.	
4	blétendre	15-sept	HROT	20-oct	
4	blédur	15-sept	HROT	20-oct	
4	blédur	ro oopt.		20 000	
4	blédur				
4	poishiver	19-nov	HROT	20-nov	
4	orgehiver	2-sept	HROT	10-oct	
4	poishiver	10-oct	HROT	17-nov	
4	orgehiver	10-nov			
4	orgehiver	15-oct			
. 5	orgeprintemps	19-févr	HROT	20-févr	
5	tournesol	6-avr	HROT	7-avr	
5	orgeprintemps	19-févr	HROT	20-févr	
5	poisprintemps	15-févr	HROT	1-mars	
5	tournesol	5-avr	HROT	15-avr	
5	maïsdoux	15-avr	MELANGE10	1-mai	
5	orgenrintemos	5-févr	HROT	20-févr	
9	aelfixe	0 10 11.		201011.	
	J				

D 7.2 Identification of critical points from validated results - Effect of the structural variables on GM admixture in non GM harvest based on simulation results -

Fife case study - oilseed rape

C. Sausse - CETIOM

Introduction

This report aims at identifying critical points for gene flow on cropping systems and farm levels concerning the SIGMEA Fife case study, with the help of GENESYS-OSR. The whole methodology is described in detail in the Beauce case study report. The conclusion emphasizes the comparison between both case studies.

Simulations implemented

The case study area is described in detail in deliverable 7.1. The table 1 gives the main characteristics of the simulation spot in comparison with Beauce. Data were provided by SCRI on 5 farms, but simulations were carried out only on the biggest one (figure 1), for some technical reasons. Real practices and rotations on this farm are not typical for the region (no plough, and crop gathered by blocks). In consequence, we used typical practices and rotations from the other farms (table 2). Rotations were randomly allocated, and we checked the landscapes with the Rothamsted landscape generator in order to avoid discrepancies between years with the following objectives: 5% (+-2%) set aside and 11% (+-4%) OSR each year. Field margins were not taken into account, as we did not see any effect of field margin management in Beauce. Seeds were assumed to be pure. Agricultural practices are described in detail in annex 1 and 2.

Spot	Beauce	Fife
area (ha)	243	768
field area (ha)	1.97 ± 1.46	7.0 ± 6.0
OSR area / total area (20 years)	21%	11%
cropping systems	OSR followed by 2 or 3 cereals; in some cases : spring crops; no set aside	diversified rotations possibly including set aside, temporary grass (3 years), potatoes
delay for return of OSR in rotation	3 or 4 years	4 years or more
Soil tillage	4 to 6 use of plough depends on the farmer, the previous crop	Plough + rotary harrow in all situations

Table 2: rotations used

Rotation	Frequency
Wheat / Beans / Wheat / Spring barley / OSR	2/10
Wheat / Spring barley / Winter barley / OSR	2/10
Potatoe / Wheat / Spring barley / Winter barley / OSR / winter wheat	1/10
Wheat / Spring barley / Set aside / Wheat / OSR	4/10
Wheat / Spring barley / Grass (4 years) /OSR	1/10

Simulations were carried out with the GENESYS version of 20/02/06. Two kinds of simulations were carried out: (1) co-existence during 20 years between cropping systems supporting GM or conventional non-GM OSR, focusing mainly on spatial aspects where a conventional OSR cannot be grown after a GM OSR. Cropping systems with GM OSR were randomly allocated to field aiming at introduction rates of 10 and 50% of total OSR area (30 replications); (2) return to conventional OSR

after 20 years of GM OSR cultivation on three similar landscapes, focusing on temporal aspects. These simulations did not take into account any specific measures to avoid gene flow. All simulations were carried out with pure seeds, and varieties were considered isogenic. The threshold of 0.4%, more severe than 0.9% enacted by European regulation, was chosen in the calculation in order to take the underestimation of geneflow by GENESYS into account.

We used 2 main indicators for gene flow assessment:

Landscape level: for each year, we mixed the harvests of all the fields in conventional OSR, and we calculated the GM content.

Field level: the proportion of conventional area in OSR above the 0.9% threshold (incremented by field)



figure 1: the farm chosen for simulation in 2005

Results

<u>Coexistence in space</u>: GM OSR and conventional OSR were grown in separate cropping systems for 20 years. GM OSR was followed by another GM OSR in the rotation. GM content in conventional harvest was mainly due to pollen dispersal. The figure 2 and 3 give the results concerning this dissemination scheme for 10 and 50% OSR. We will notice that the low field number makes it impossible to perform simulations with 75% GM OSR as done in Beauce. Risks appeared to be very low on both landscape and field levels. As observed in Beauce, the non GM fields above the threshold were close to GM fields (figure 4). These fields seemed to have a very special geometry: they were used for agronomic tests by the farmer and thus were long and quite small in comparison with other fields.

As in Beauce, no cumulative effect appeared: the results of the last years were similar to those of the first years. We performed complementary simulations with bad management practices of volunteers in order to see the main factor possibly introducing cumulative effect: when set asides were not sown and not cut, the GM content on the whole landscape level increased over the year (figure 5). On the contrary, the results were the same when we decreased herbicide efficiency on cereals from 95 to 80%, or when we increased seed loss from 7 to 10%.



figure 2: GM and non GM side by side. 10% of OSR is GM.



figure 3: GM and non GM side by side. 50% of OSR is GM. Number of fields with conventional OSR is low (0 to 11, median = 5) It explains the point in year 17 with 100% of the area above the threshold (only 1 field).





figure 4: relation between distance and GM presence in conventional OSR fields.

figure 5: comparison between reference cropping systems and cropping systems with unsown set aside. Results are expressed at the landscape level. 1 simulation; 50% GM OSR

<u>Case of return from GM to conventional OSR</u>: GM OSR was grown for 20 years, and then only conventional OSR was grown for 20 years (3 simulations. GM content in conventional OSR was mainly due to volunteers. The figure 6 gives the results on field and landscape levels. Risky period is 4 years on both levels.



figure 5: downgraded area and GM content in conventional OSR harvest at the landscape level in case of return to conventional OSR after 20 years cultivation of GM OSR. Three similar landscapes were simulated for each case.

Conclusion for Fife

Pollen dispersal is not a problem for the tested dissemination schemes and threshold, if we consider the landscape level. But risks are possible on the field level with low frequency. Nevertheless, in our dissemination scheme, fields are GM or not GM, independently of each other, as in a more realistic landscape it would not be the case: if we make the assumption that decision to grow GM or conventional OSR is taken on the farm level, field aggregation by farm which is typical from this region (clustered "holdings") would lead to lower risks than those identified in this study. On the other hand, OSR share in the landscape will possibly increase in the future due to better prices, with a rate around 20% leading to higher risks. Concerning gene flow in time, volunteer management is critical in case of return to conventional for both landscape and field levels, with a delay corresponding to the duration of rotations including OSR (4 years for 8/10 of the simulated cropping systems). This work based on simulations does not take into account other critical points after harvest: storage on farm in case of coexistence inside the farm and shared machinery on small holdings.

Attempt of comparison between Beauce an Fife

The table 3 shows a comparison with Beauce based on the probability to see a single field or a whole silo downgraded. The Beauce area is far more sensitive to gene flow than the Fife area under spatial dissemination schemes. This result is due to differences in landscape structural variables shown in deliverable 7.1: field size and geometry, and higher OSR area.

Nevertheless, the data were obtained on small spots chosen for their high risks, and we have to consider the variability of these structural variables towards space and time in both regions. Beauce seems more heterogeneous than Fife. Indeed, land consolidation in Beauce is still going on leading to various field patterns, whereas already done in Fife. Moreover, Beauce shows a greater number of cropping systems than in Fife for various reasons. Lastly, OSR share in the landscape over the last fifteen years shows a greater flexibility in Beauce (figure 6). Nevertheless, this figure should be confirmed with the new context of development of biofuel.

The landscapes in Beauce are on average less favourable to coexistence, but other characteristics of the cropping systems are more favourable. This is the case for soil tillage. Fields were systematically ploughed in Fife with little exceptions, but in Beauce, ploughing was only an option depending on the farm, the climate of the year or other agronomic decision rules. In this context, some flexibility could be proposed to manage volunteers. Moreover, storage on farm practiced in Fife but not in Beauce was less favourable in case of coexistence on the same farm.

These conclusions show that the design of coexistence measures should take into account variability of the cropping systems and landscapes between regions, but also inside each of them.

Spot	% GM OSR	Probability to see a single field downgraded (%)	Probability to see the whole non GM harvest downgraded (%)
Beauce	10%	2.6	0
	50%	14.7	0.17 (1/600)
	75%	22.1	6.67
Fife	10%	0.5	0
	50%	1.7	0.17 (1/600)

Table 3 : simulation results for various dissemination schemes for the 0.9% threshold



figure 6 : comparison of winter OSR areas in Beauce (department of Loir et Cher) and Scotland (base 100 in 1989, sources : SCEES and Scottish Executive)

APPENDIX 1 : hypothesis for set aside, grass and land management contract

1. set aside

Place in the rotation wheat / spring barley / set aside / wheat / OSR

Species : perennial ryegrass

Management	
Plough	1 sept
Rotary harrow / drill	15 oct
Sowing (1400 seed/m ²)	15 oct
Cut	15 july

2. Temporary grass

Place in the rotation Wheat / spring barley / grass / grass / grass / grass / wheat / OSR

Species : perennial ryegrass

Management : first year	
Plough	1 sept
Rotary harrow / drill	15 oct
Sowing (1400 seed/m ²)	15 oct
Cutting	1 may and 15 july
Grazing	yes
Management : other years	
Cutting rank patches	1 may and 15 july
Grazing	yes

Note : GENESYS cannot simulate more than 2 cuts by year

3. Permanent grass

ManagementCuttting rank patches15 mayGrazingyes

Note : no cut for silage or hay

4. Land management contract

No management

APPENDIX 2 : agricultural practices (only p_crop used; p for plough)

nom de la ci	u nom de l'Itk	type broyage 2	date	date de sem	il densité de s	seperte à la ré	d variété T 1 mortalité he	variété T 2 rbicide appliq	variété T 3 jué au stade p	classiques/se plantule (semis
bordure	bordure	1	999	-1	-1	-1	0	0	0	0
colza	m_GMOSR	-1	-1	05/09/1900	100	0.07	0	0	0	0.9
colza	p_GMOSR	-1	-1	05/09/1900	100	0.07	0	0	0	0.9
colza	m_OSR	-1	-1	05/09/1900	100	0.07	-1	-1	-1	-1
colza	p_OSR	-1	-1	05/09/1900	100	0.07	-1	-1	-1	-1
blé	m wheat	-1	-1	15/10/1900	460	-1	0.95	0.95	0.95	0.95
blé	m wheat	-1	-1	01/11/1900	460	-1	0.95	0.95	0.95	0.95
blé	p wheat	-1	-1	15/10/1900	460	-1	0.95	0.95	0.95	0.95
blé	p_wheat	-1	-1	01/11/1900	460	-1	0.95	0.95	0.95	0.95
avoine	m oats	-1	-1	20/09/1900	400	-1	0.95	0.95	0.95	0.95
avoine	p oats	-1	-1	20/09/1900	400	-1	0.95	0.95	0.95	0.95
orge printem	rm_sbarley	-1	-1	25/03/1900	420	-1	0	0	0	0
orge_printem	tp sbarley	-1	-1	25/03/1900	420	-1	0	0	0	0
orge hiver	m wbarley	-1	-1	20/09/1900	420	-1	0.95	0.95	0.95	0.95
orge hiver	p wbarley	-1	-1	20/09/1900	420	-1	0.95	0.95	0.95	0.95
patate	potatoe	-1	-1	01/05/1900	7	-1	0	0	0	0
pols	p beans	-1	-1	07/03/1900	90	-1	0	0	0	0
pols	m beans	-1	-1	07/03/1900	90	-1	0	0	0	0
Jachére	set aside	1	999	15/10/1900	1400	-1	0	0	0	0
herbe	grass1	1	999	15/10/1900	1400	-1	0	0	0	0
herbe	grass	1	999	-1	-1	-1	0	0	0	0
land_manage	e ÊMC	1	999	-1	-1	-1	0	0	0	0

nom de la cu	i nom de l'Itk	variété T 1	variété T 2	variété T 3	classiques/s	binage	nb de travau	:: pour chaque travall, le typ	e et la date
		mortalité her	bicide appilq	ué au printem	рв	prop surface	binée		
bordure	bordure	0	0	0	0	-1	-1		
colza	m_GMOSR	0	0	0	0.9	0	2	ROTAVATOR 02/09/1900	ROTAVATOR
colza	p_GMOSR	0	0	0	0.9	0	2	LABOURRAS 02/09/1900	HROT
colza	m OSR	-1	-1	-1	-1	0	2	ROTAVATOR 02/09/1900	ROTAVATOR
colza	p_OSR	-1	-1	-1	-1	0	2	LABOURRAS 02/09/1900	HROT
blé	m_wheat	0.7	0.7	0.7	0.7	0	2	ROTAVATOR 20/08/1900	ROTAVATOR
blé	m wheat	0.9	0.9	0.9	0.9	0	2	ROTAVATOR 20/09/1900	ROTAVATOR
blé	p wheat	0.7	0.7	0.7	0.7	0	2	LABOURRAS 20/08/1900	HROT
blé	p_wheat	0.9	0.9	0.9	0.9	0	2	LABOURRAS 20/09/1900	HROT
avoine	m_oats	0.7	0.7	0.7	0.7	0	2	ROTAVATOR 20/09/1900	ROTAVATOR
avoine	p oats	0.7	0.7	0.7	0.7	0	2	LABOUR/RAS 20/08/1900	HROT
orge printemi	rm_sbarley	0.95	0.95	0.95	0.95	0	2	ROTAVATOR 15/03/1900	ROTAVATOR
orge_printem	p_sbarley	0.95	0.95	0.95	0.95	0	2	LABOURRAS 15/03/1900	HROT
orge_hiver	m_wbarley	0.7	0.7	0.7	0.7	0	2	ROTAVATOR 01/09/1900	ROTAVATOR
orge hiver	p wbarley	0.7	0.7	0.7	0.7	0	2	LABOUR/RAS 01/09/1900	HROT
patate	potatoe	0.95	0.95	0.95	0.95	0	2	LABOURRAS 15/03/1900	HROT
pols	p beans	0.9	0.9	0.9	0.9	0	2	ROTAVATOR 01/03/1900	ROTAVATOR
pols	m beans	0.9	0.9	0.9	0.9	0	2	LABOURRAS 01/03/1900	HROT
Jachére	set aside	0	0	0	0	0	2	LABOUR/RAS 02/09/1900	HROT
herbe	grass1	0	0	0	0	0	2	LABOURRAS 02/09/1900	HROT
herbe	grass	0	0	0	0	-1	-1		
land manage	EMC .	0	0	0	0	-1	-1		

nom de la c	u nom de l'Itk	variété T 1	variété T 2	variété T 3	classiques/s	binage	nb de trava	u: pour chaque travall, le typ	oe et la date
		mortalité he	rbicide appilq	ué au printen	прв	prop surface	e binée		
bordure	bordure	0	0	0	0	-1	-1		
colza	m_GMOSR	0	0	0	0.9	0	2	ROTAVATOR02/09/1900	ROTAVATOR
colza	p_GMOSR	0	0	0	0.9	0	2	LABOURRAS 02/09/1900	HROT
colza	m OSR	-1	-1	-1	-1	0	2	ROTAVATOR 02/09/1900	ROTAVATOR
colza	p_OSR	-1	-1	-1	-1	0	2	LABOURRAS 02/09/1900	HROT
blé	m wheat	0.7	0.7	0.7	0.7	0	2	ROTAVATOR 20/08/1900	ROTAVATOR
blé	m_wheat	0.9	0.9	0.9	0.9	0	2	ROTAVATOR 20/09/1900	ROTAVATOR
blé	p wheat	0.7	0.7	0.7	0.7	0	2	LABOURRAS 20/08/1900	HROT
ble	p_wheat	0.9	0.9	0.9	0.9	0	2	LABOURRAS 20/09/1900	HROT
avoine	m oats	0.7	0.7	0.7	0.7	0	2	ROTAVATOR 20/09/1900	ROTAVATOR
avoine	p oats	0.7	0.7	0.7	0.7	0	2	LABOURRAS 20/06/1900	HROT
orge printen	nrim_sbarley	0.95	0.95	0.95	0.95	0	2	ROTAVATOR 15/03/1900	ROTAVATOR
orge_printen	tp_sbarley	0.95	0.95	0.95	0.95	0	2	LABOURRAS 15/03/1900	HROT
orge hiver	m wbarley	0.7	0.7	0.7	0.7	0	2	ROTAVATOR 01/09/1900	ROTAVATOR
orge hiver	p wbarley	0.7	0.7	0.7	0.7	0	2	LABOURRAS 01/09/1900	HROT
patate	potatoe	0.95	0.95	0.95	0.95	0	2	LABOURRAS 15/03/1900	HROT
pols	p beans	0.9	0.9	0.9	0.9	0	2	ROTAVATOR 01/03/1900	ROTAVATOR
pols	m beans	0.9	0.9	0.9	0.9	0	2	LABOURRAS 01/03/1900	HROT
jachére	set aside	0	0	0	0	0	2	LABOURRAS 02/09/1900	HROT
herbe	grass1	0	0	0	0	0	2	LABOURRAS 02/09/1900	HROT
herbe	grass	0	0	0	0	-1	-1		
land manao	e LMC	0	0	0	0	-1	-1		

nom de la cu	u nom de l'Itk		paturage	date de récol détourage		
bordure	bordure		-1	-1	-1	
colza	m_GMOSR	05/09/1900	-1	05/08/1900	0	
colza	p_GMOSR	05/09/1900	-1	05/08/1900	0	
colza	m OSR	05/09/1900	-1	05/08/1900	0	
colza	p_OSR	05/09/1900	-1	05/08/1900	0	
blé	m wheat	15/10/1900	-1	01/09/1900	-1	
blé	m wheat	01/11/1900	-1	01/09/1900	-1	
blé	p wheat	15/10/1900	-1	01/09/1900	-1	
blé	p_wheat	01/11/1900	-1	01/09/1900	-1	
avoine	m oats	20/09/1900	-1	20/08/1900	-1	
avoine	p oats	20/09/1900	-1	20/08/1900	-1	
orge printem	rm_sbarley	25/03/1900	-1	20/08/1900	-1	
orge printem	p sbarley	25/03/1900	-1	20/08/1900	-1	
orge hiver	m wbarley	20/09/1900	-1	20/08/1900	-1	
orge hiver	p wbarley	20/09/1900	-1	20/08/1900	-1	
patate	potatoe	01/05/1900	-1	01/10/1900	-1	
pols	p beans	07/03/1900	-1	05/09/1900	-1	
pols	m beans	07/03/1900	-1	05/09/1900	-1	
lachére	set aside	15/10/1900	0	-1	-1	
herbe	grass1	15/10/1900	1	-1	-1	
herbe	grass		1	-1	-1	
land manage	e ÉMC		0	-1	-1	

D 7.2 Identification of critical points from validated results - Effect of the structural variables on GM admixture in non GM harvest based on simulation results -

Schleswig Holstein case study - oilseed rape

C. Sausse - CETIOM

Introduction

Simulations carried out on the Schleswig Holstein case study aims at 1) assessing the sensitivity of the landscape to gene flow 2) focussing on the dilution process between the field and the silo, in a simulated landscape more large than in Beauce and Fife case studies. Detailed methodology is given in the Beauce report.

1. Material and methods

Data were provided by University of Kiel¹ on two spots (description in appendix). We carried out simulations on one of them (Schleswig Flensburg). The spot is much larger than Beauce and Fife, because we worked with a version of GENESYS allowing direct treatment of complex polygons, with subsequent low calculation duration. The following table shows the main characteristics of the spot:

Area	Schleswig Flensburg	Beauce	Fife
Area (ha)	2536	243	768
Field area (ha)	2.62 ± 3.33	1.97 ± 1.46	7.0 ± 6.0
OSR area / total	17%	21%	11%
area			
total OSR volume by year (t) (yield = 3 t/ha)	1292	153	253
Cropping systems	diversified rotations possibly including set-aside, temporary grass (3 years), potatoes; permanent pastures	OSR followed by 2 or 3 cereals; in some cases: spring crops; no set aside	diversified rotations possibly including set-aside, temporary grass (3 years), potatoes; permanent pastures

Table 1 : description of the simulation spot in comparison with Beauce and Fife

We started from a real landscape (i.e. : the occupation of each field was recorded during 10 years). Field occupations were divided into 4 categories: OSR, set aside, other crops, fodder crop. The two last categories were splitted in order to describe crop diversity with more accuracy. Starting from data provided by Kiel, we transformed field occupations in realistic crop successions, taking into account basic agronomic rules, and the global proportion of each crop in the landscape.

Hypothesis and methods for simulations were equivalent than those chosen in Beauce and Fife. The threshold of 0.4%, more severe than 0.9% enacted by European regulation, has been chosen in the calculation in order to take the underestimation of geneflow by GENESYS into account. The versions of GENESYS were 26012007 (spatial.exe) and 15022007 (GENESYS.exe). We carried out only simulations concerning spatial coexistence, where non GM and GM OSR were grown on different fields side by side, but where a GM OSR could not come after a non GM in the rotation and vice versa.

2. Results and discussion

2.1 Sensitivity to gene flow

The figures 1a to 1c show the sensitivity to gene flow according to two indicators: the average GM content in the whole non GM harvest (landscape level), and proportion of area of non GM OSR above the 0.9 threshold (field level).

¹ Kiel University, Ecology Centre, Department of Ecosystem Research; contact : Wilhelm Windhorst and Ulrike Middelhoff



Figure 1a : Areas not satisfying the threshold ("field level") and GM content in conventional OSR batches ("landscape level") when 10% of the OSR grown is GM (30 simulations; the 0.4% threshold corresponds to the 0.9% labelling threshold after correction.)



Figure 1b : Areas not satisfying the threshold ("field level") and GM content in conventional OSR batches ("landscape level") when 50% of the OSR grown is GM (30 simulations; the 0.4% threshold corresponds to the 0.9% labelling threshold after correction.)



Figure 1c : Areas not satisfying the threshold ("field level") and GM content in conventional OSR batches ("landscape level") when 75% of the OSR grown is GM (30 simulations; the 0.4% threshold corresponds to the 0.9% labelling threshold after correction.)

The dilution effect is so high, that in the worst case (75% GM introduction), risks are nil at the landscape level, whereas they are significant in the same time at the field level.

The version of the model was not the same as in Fife and Beauce, leading to higher long distance pollen dispersal. However, harvest pollutions seem similar. The new thing, in comparison with other cases studies, came from variations over years for both indicators. Volunteers were well controlled, and we tried to identify an effect of the spatial distribution of the OSR in the landscape. The following table shows yearly values of total OSR area and of fields' areas.

year	average OSR field area (ha)	standard error	total OSR area (ha)	% of OSR area > 0.9% (75% GM OSR)
1	3.04	3.64	322	10
2	3.09	4.61	399	13
3	2.70	2.61	337	15
4	3.02	2.85	341	15
5	2.79	2.89	307	14
6	3.07	3.71	224	10
7	3.47	4.98	378	8
8	3.05	3.66	360	16
9	2.77	3.21	349	11
10	2.89	3.42	361	13

Table 2: landscape characteristics possibly affecting gene flow

The variation of the fields' area can partially explain variations of harvest pollutions: the little the fields are, the more they are sensitive to gene flow and vice versa. However, this variation could seem unusual. Taking into account the large area of the whole spot, distribution should have been similar over years. On the contrary, a chi square test shows the year 7 having a higher rate of large fields than the other years.

Moreover, the total OSR area varied over years more than expected (if we assume farmers are prone to diversify economic risks with regular temporal crop allocation). As data reflect the real landscape occupation, this information is interesting for management issues. The choice between the different pre-scenarios described in Deliverable 7.3 should take into account this variability. Indeed, for a same GM introduction rate, sensitivity to gene flow can vary over years. If one cannot perform simulations in diverse situations, risks deducted from mere landscape characteristics could lead to underestimations in certain conditions.

2.2. Dilution effect

The figures 2a to 2c show a comparison of the distribution of GM admixture at the field level and at the landscape level (effectives are not the same).



Figure 2a : GM content in the fields ("field level") and GM content in conventional OSR batches ("landscape level") when 10% of the OSR grown is GM (30 simulations; the 0.4% threshold corresponds to the 0.9% labelling threshold after correction.)



Figure 2b : GM content in the fields ("field level") and GM content in conventional OSR batches ("landscape level") when 50% of the OSR grown is GM (30 simulations; the 0.4% threshold corresponds to the 0.9% labelling threshold after correction.)



Figure 2c : GM content in the fields ("field level") and GM content in conventional OSR batches ("landscape level") when 75% of the OSR grown is GM (30 simulations; the 0.4% threshold corresponds to the 0.9% labelling threshold after correction.)

The figures show the dilution effect (or smoothing effect), when harvest pollution on the field level are aggregated on the landscape level. The table 3 shows how this dilution varies the first year, according to the percentage of GM OSR in the landscape. Dilution is here defined with three indicators.

%OGM	indicator 1*	indicator 2**	indicator 3***
10%	1	88	1.550%
50%	9	22	1.446%
75%	6	10	1.351%

Table 3: dilution according to the percentage of GM OSR

*indicator 1 = height of the box at the field level / height of the box at the landscape level

**indicator 2 = (maxi - mini at the field) / (maxi - mini at the landscape)

***indicator 3 = maximum pollution at the field - maximum pollution at the landscape level

Results depend on the indicator. Whatever the percentage of OSR in the landscape is, the worst fields show more or less the same harvest pollutions. The "smoothing" effect is thus more important when the percentage of OSR in the landscape is low (indicator 2). On the other hand, if we do not take into account atypic values (indicator 1), results are not so clear.

An interesting result comes out from these outputs: the distribution of harvest pollution does not seem to follow a gaussian distribution at the field level, whereas it is the case at the landscape level. GM admixture at the field level is asymetric, with few very high values.

Logically, the dilution effect should grow with the size of the landscape. Thinking as a manager liable for grain quality, the landscape corresponds to the silo. Considering the present case (table 1), the "silo" is here particularly large, more or less twice than those usually used.

APPENDIX : description of the case study provided by University of Kiel

Case study Region Schleswig-Holstein, Germany within Project "Sustainable Introduction of GMOs into European Agriculture" (SIGMEA)

Date: 21 Oct 2005

Ref: Case study Schleswig-Holstein, contribution to WP7 by University of Kiel, contracted by CETIOM, contact Christophe Sausse

Summary

1.) Selection of case study areas

The selection of the study area was based on community statistics. In order to reach a target area of about 50 km² it was necessary to identify 3-5 neighbouring communities with a similar structure. Fig. 1 gives the locations of two selected areas in the districts of Schleswig-Flensburg (area 1) and Herzogtum Lauenburg (area 2).



Fig. 1: Location of case study areas in Schleswig-Holstein, Germany. Area 1 is located in the district Schleswig-Flensburg. Area 2 is located in the district Herzogtum Lauenburg.

The two areas consist of 5 and 6 communities each. Tab. 1 and 2 list main characteristics, number of farms as well as mean size of farms and fields on community level. Mean farm and field sizes are higher in area 2 compared to area 1. Tab. 3 and 4 list main land use characteristics: area and share of cropping area, permanent grassland, forest area, sealed area, water bodies and other in the communities. The fraction of permanent grassland is slightly higher in area 1 compared to area2. In total, area 1 and 2 cover the area of 29 km² and 49 km², respectively.

Community code	Name	number of	Mean size of farms (ha)	Mean size of fields (ha)				
"gkz_gem"		farms						
59021	Dollrottfeld	12	38,074	2,64				
59063	Norderbrarup	7	49,245	2,07				
59072	Saustrup	15	48,653	2,52				
59074	Scheggerott	14	40,735	2,80				
59095	Wagersrott	13	37,809	2,62				

Tab. 1: Area1, selected communities in the	district Schleswig-Flensburg,	farm numbers, mean farm	n
size and mean field size.			

Tab. 2: Area 2, selected communities in the district Herzogtum Lauenburg, farm numbers, mean farm size and mean field size.

Community code	Name	number of	Mean size of farms (ha)	Mean size of fields (ha)
"gkz_gem"		farms		
53014	Breitenfelde	19	57,895	3,50
53095	Niendorf/Stecknitz	14	45,347	3,51
53113	Schretstaken	13	45,631	3,28
53125	Talkau	7	50,537	3,28
53126	Tramm	9	44,715	2,67
53134	Woltersdorf	13	44,167	2,75

Tab. 3: Area1, land use characteristics of the selected communities in the district Schleswig-Flensburg.

	comn	nunity	community		comr	community community		nunity	community	
	59	021	5	9063	59072		59074		59095	
land use	ha	share	ha	share	ha	share	ha	share	ha	share
		(%)		(%)		(%)		(%)		(%)
cropping area	394	79,0	257	64,3	664	81,8	539	85,3	423	75,1
permanent grassland	63	12,6	88	22,0	66	8,1	31	4,9	69	12,3
Forest	13	2,6	18	4,5	51	6,3	21	3,3	42	7,5
sealead area	25	5,0	33	8,3	28	3,4	37	5,9	26	4,6
water bodies	4	0,8	1	0,3	2	0,2	3	0,5	2	0,4
other	0	0,0	3	0,8	1	0,1	1	0,2	1	0,2
total	499	100,0	400	100,0	812	100,0	632	100,0	563	100,0
total area 1	29 km	2								

Tab. 4: Area2,	land use ch	aracteristics (of the sele	cted comr	nunities in	n the district	t Herzogtum
Lauenburg.							

	comm 530	nunity)14	com 53	munity 3095	com 53	munity 3113	comi 53	nunity 125	comi 53	munity 126	comm 531	nunity 134
land use	ha	share	ha	share	ha	share	ha	share	ha	share	ha	share
		(%)		(%)		(%)		(%)		(%)		(%)
cropping area	985	78,5	555	66,0	521	61,7	322	66,0	350	52,0	537	67,9
Permanent grassland	115	9,2	80	9,5	72	8,5	32	6,6	53	7,9	38	4,8
Forest	52	4,1	164	19,5	219	25,9	101	20,7	238	35,4	139	17,6
sealed area	82	6,5	39	4,6	31	3,7	31	6,4	22	3,3	32	4,0
water bodies	1	0,1	1	0,1	0	0,0	0	0,0	2	0,3	1	0,1
other	20	1,6	2	0,2	2	0,2	2	0,4	8	1,2		5,6
total	1.255	100,0	841	100,0	845	100,0	488	100,0	673	100,0	791	100,0
total area 2	49 km ²											

2.) Overview on delivered data sets

The delivered data sets consist of shape-files and tables in Annex1_geodata.zip and a list of management specifications for different crop types. The geo-datasets for area 1 and area 2 are

specified by "fall1" and "fall2", respectively. Each of the geo-datasets consist of several shape files. For area 1 these are fall1.shp (poygones), fall1str.shp (linear elements roads) and fall1flu.shp (linear elements of waterbodies). The field "Objektart1" specifies land use options of polygons which are described in Fig. 2. In addition the specifications given in field "BETNR" (= farm number) and field "BETTYP" (= farm type, with 1 = cash crop farm and 2 = farm with animals) are of interest for the model runs.



Fig. 2: The geo-datasets for area 1 consist of several shape files: fall1.shp (poygones), fall1str.shp (linear elements roads) and fall1flu.shp (linear elements of waterbodies). The field "Objektart1" specifies land use options of polygons.

For each polygon that has been specified as cropping area, a series of crops has been specified describing the crop rotation over a time span of 10 years. The data are listed in the fields "J1", "J2" to "J10" in table "ffall1V.dbf" for area 1 and table "ffall2V.dbf" for area 2. The "real" data given are the specifications for oilseed rape which have been obtained via satellite images. All other crops have been specified according to statistical data giving an idea of the relations between different crops. According to these data other crops are winter cereals (winter wheat 50%, winter barley >40% and winter rye <10%), fodder plants (typically grass and maize). The fractions of fodder grass : maize are in area 1 about 80% : 20% and in area 2 about 30% : 70%. A third important land use of cropping areas has been identified as set-aside which is be further specified. We know that 3 to 5 % can be typical set-aside where virtually no management occurs. In addition on up to 3 % of the cropping area potatoes or beets are grown which is not specified in the data but can be implemented in cropping scenarios. Tab. 5 lists the mean shares of the crops listed above as they have been specified in the data of area 1 and area 2 for a period of 10 years. In Fig. 3 the spatial distribution of the specified crops is given for area 1 in year five. The legend in Fig. 3 also gives some information on how the crop data are linked to the shape files. The crop specifications given in the fields "J1", "J2" to "J10" are as follows: 1 = winter cereal, 2 = fodder plants, 4 = set-aside and values > 10 = oilseed rape.

Tab. 5: Shares of different crops as sp	pecified in the data of area 1 and area 2
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	area 1	area 2
specified crops	mean share in 10	years (% of total)
winter cereals	0.64	0.73
fodder plants	0.13	0.07
set-aside	0.06	0.06
oilseed rape	0.17	0.21

Fig. 3: Spatial distribution of specified crops in area 1 in year five. The legend also gives some information on how the crop data are linked to the shape files. The crop specifications as specified in the legend are given in the fields "J1", "J2" to "J10" of table ffall1V.dbf.

crops on fields of area 1 in year 5

