PROCEEDINGS

The 1st European Conference on the Co-existence of Genetically Modified Crops with Conventional and Organic Crops

GMCC-03

GM Crops and Co-existence

Edited by

Birte Boelt

Published by the Danish Institute of Agricultural Sciences, Research Centre Flakkebjerg
DK-4200 Slagelse, Denmark

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Preface

This proceedings contains invited papers, offered oral presentations and poster abstracts presented at the 1st European Conference on the Co-existence of Genetically Modified Crops with Conventional and Organic Crops, GMCC-03, held in Snekkersten, Denmark, on 13-14 November, 2003.

The production of genetically modified crops outside the EU is increasing, and the European Commission has initiated discussions to clarify the need, and possible options, for agronomic and other measures to ensure the viability of conventional and organic farming and their sustainable co-existence with genetically modified crops. The conference is organised in the recognition that, until now, the scientific discussions on co-existence strategies and the implications of these for European agriculture have been scarce. Therefore, co-existence is the key word for the conference.

The programme consists of seven invited papers focussing on aspects of gene dispersal and management measures, 2 x 3 parallel sessions with offered oral presentations focussing on specific crops, post harvest management and monitoring, strategies and economic assessments, modelling and on-farm stewardships. Members of the programme committee have reviewed each paper in draft, and only papers focusing on co-existence has been accepted. Our intention was to put together a scientific programme, which encourages to open and constructive discussions. It is the hope of the programme committee that during these two days the more than 200 registered participants will share results, experiences and ideas on co-existence and related subjects.

The Danish Institute of Agricultural Sciences (DIAS) organises the conference in cooperation with the Danish Plant Directorate, the Danish Research Institute of Food Economics, The Royal Veterinary and Agricultural University, Risø National Laboratory, The National Environmental Research Institute, Federal Biological Research Centre for Agriculture and Forestry (D), National Institute for Agricultural Research (FR), NIAB (UK), and the University of Manitoba (CA). We would like to thank the representatives from these institutes for their participation in the programme committee.

We would also like to thank the Danish Ministry of Food, Agriculture and Fisheries for economic support.

Birte Boelt
Chairman of the programme committee

Invited papers

The role of research on ensuring co-existence between GM and non-GM crops and seeds: what are the needs?

K. Lheureux & E. Rodríguez-Cerezo
European Commission, Directorate-General Joint Research Centre (JRC), Institute for Prospective Technological Studies (IPTS), Edificio Expo, E-41092 Sevilla, Spain
E-mail: karine.lheureux@jrc.es; emilio.rodriguez@jrc.es

The decision by the EU of introducing labelling thresholds for the adventitious presence of GM crops in non-GM crops is at the origin of the co-existence debate. It is essential to regard labelling thresholds as what they are: tools to ensure consumer choice after GM release. Co-existence measures are not environmental or health risks management measures. What is then the main role for scientist/researchers in addressing co-existence measures? A multidisciplinary effort by scientists (agronomists, agricultural economists, social scientists…) is needed to address the dimension of the co-existence issue, directed to answering the following main questions:

(1) What would be the % of adventitious presence of GM crops in non GM crops in Europe in a situation where GM crops are introduced and current farming practices continue without significant changes?
(2) If there are cases where estimations for adventitious presence are fairly above the 0.9% threshold, what are the agronomical measures needed to meet the threshold and at what cost?

We shall review in this talk different studies in the EU and elsewhere directed to answer these questions. In 2000, the European Commission's (EC) Joint Research Centre (JRC) through its Institute for Prospective Technological Studies (IPTS) received the request from EC's Directorate General Agriculture to launch a prospective study on agronomic and economic aspects of co-existence of GM and non-GM crops at European level. IPTS undertook the coordination of a number of European scientific institutes that carried out the studies summarised in the IPTS 2002 report (Bock *et al*., 2002). For this conference we have elaborated an exhaustive list of publications/reports (2000-2003) that in a way or another deal with the issue of co-existence as defined above. The results of this search, with few lines of description of each report are available at the JRC's Institute for Prospective and Technological Studies (IPTS) web page[1].

[1] http://lifesciences.jrc.es (Selected links)

Adventitious presence of GM crops in non GM crops: few real world data

The direct approach to answer this question would be to perform yearly detailed sampling of non-GM plots at harvest in areas where GM crops and non GM crops are grown commercially. Quantitative analysis (quantitative PCR and/or phenotype screening) of the samples would yield estimations for adventitious presence in a given crop in a given region in a given season. Such data are unfortunately missing in the EU and elsewhere.

Perhaps the piece of work addressing most directly this question is research performed in Australia on the adventitious presence of Herbicide Tolerant (HT) canola grown in commercial fields on harvests of non-HT canola (Rieger *et al*., 2002). Admixture levels, once averaged by field, did not surpass 0.07% and the vast majority were below 0.03%. These levels are much lower than those suggested by previous studies, and are well below the 0.9% level agreed by the EU.

One important conclusion from this research is that *gene flow at a large scale cannot necessarily be predicated from small scale, experimental plot studies*. But the second important conclusion is that canola gene flow in commercial conditions can go a long way and remain relatively independent of the distance of GM plots (up to 3 km), even at very low levels. Therefore absolute zero tolerance crops are unlikely to be achieved, a conclusion supported by many other studies listed in our review.

Recently, the reports released by DEFRA provide a set of new field data. The DEFRA report on oilseed rape (Ramsey *et al*., 2003) confirmed that *pollination of one large field to the next is likely to be less than 0.1% averaged over the field,* specifying also that *greater than 1% crossing might occur in crops with impaired male sterility over hundreds of metres a*nd that *low level of cross-pollination was detectable over very long distances* (up to 26 km).

Co-existence studies based on modelling, expert opinion and field trials: overview of conclusions

In the absence of real world data**,** the various EU reports on co-existence might be grouped into three categories: i) reports based on opinions and expert panel discussions (e.g. EC Scientific Committee on Plants, 2001; Tolstrup *et al.,* 2003), ii) reports based on computer modeling and expert estimations (e.g. Bock *et al*., 2002; Meynard *et al*., 2001), iii) surveys of farmers' compliance with agreed guidelines large field trials (ADAS 2003).

More research to generate new empirical field data is needed to validate models and help expert opinion.

Different types of models are used: i) models predicting the probability of gene flow as a function of a distance (dispersion curve) and ii) decision models which integrate both gene flow dispersion curve and agronomical practices. The last ones allow comparing the effects of changing farming practices (Bock *et al*., 2002; Meynard *et al*., 2001). Both models are needed and more research should be encouraged.

- A conclusion from all studies is that **co-existence depends on the farm type/crop considered**, and on the specific farming practices including **all steps** from seed production to processing, including cultivation, harvest, transport and storage. Nevertheless, while all studies mentioned the importance of post-harvest management, not all co-existence studies analyse in depth this source of adventitious admixture.
- Of the main GM crops studied, (maize, oilseed and potato), oilseed rape is considered as the most problematic for co-existence due to cross pollination, and persistence of seeds in the soil. Maize is placed in an intermediate position, while other crops looked at, such as potatoes, are considered to present little problems of co-existence. Reviews of one of the sources of impurities (pollen flow) for many crops have been published (Estham & Sweet, 2002) and are useful to classify crops according to their risk of gene-flow. However, as stated, many other factors such as the purity of starting material (seeds), the size and form of the plot, and the farming practices, all influence the final adventitious presence level. Others, based on expert panels, have considered all sources of impurity and classified crops according to their co-existence problems for a given country, such as Denmark (Tolstrup *et al.*, 2003). Future studies on co-existence should target in detail crops such as cotton or sugar-beet that are in the pipeline for release in EU (Lheureux *et al*., 2003).
- **Co-existence may be trait/dependent**. Depending on the genetic modification introduced, co-existence of GM and non-GM crops might be easier. For example, GM crops with Bt traits are grown with mandatory protocols requesting planting of non-GM refuges, what surely has an impact on the final levels of admixture.
- **Adventitious presence** of GM crops in non GM crops may increase with time (all other factors fixed) for some crops but not for others, depending of the biology of the crop.
- Comparing the results obtained for a specific crop (levels of adventitious admixture) from different studies is very difficult due to the different methodologies and inherent variability of farming practices. Efficient and cost-effective measures for co-existence are crop-farm specific. Continuous research to identify efficient/potential, un-expensive measures to reduce adventitious mixture at farm level (including development of biological tools to minimise gene flow) is needed.

The measures to ensure co-existence and their economic costs

Current research on the economic costs of co-existence for the EU again suffers from the lack of data from real life situations.

In the JRC-IPTS report (Bock *et al*., 2002) there is a detailed analysis of possible costs in a large number of farm/crop/measures combinations, and for compliance with different thresholds. The expert panel review produced by Denmark (Tolstrup *et al*., 2003) concluded, as the JRC study did, that co-existence at the 0.9% level for certain crops is possible but with different costs and needs for changing farming practices. In their report, most crops need little additional changes to meet the 0.9% threshold, with the exception of oilseed rape.

Monitoring costs are an important component of the total additional costs to ensure co-existence as demonstrated in the JRC study. The JRC study concluded that organics farms, as they already operate rules of segregation and traceability of produce, will support less additional costs than "conventional" non-GM farms to achieve the 0.9% threshold.

Another conclusion of the JRC study is that (apart from the inherent biological inability to obtain 100% pure crops) *zero tolerance thresholds, when technically achievable, would be very expensive*.

Co-existence in the seed production business, and the impact of seed purity on co-existence

The case of *seed production needs a separate treatment and specific studies on co-existence*. This has been done only for seed production of oilseed rape, in the JRC-IPTS study (Bock *et al*., 2002). Specific studies focusing on seed production of maize are a priority. Other studies should focus on co-existence on seed production of those crops for which GM varieties exist, as described below.

A draft Directive from the European Commission on the marketing of seeds provides a tolerance labelling thresholds for adventitious presence of GM seeds in seed lots of non-GM varieties of 0.3% for oilseed rape, 0.5% for maize, cotton, beet, potato, chicory, tomato and 0.7% for soybean (European Commission, 2003b).

There is a need to study, based on available knowledge and existing models, which crops are more affected by starting seed impurities. As said above, the crops more sensitive to the effect of purity of starting material *should be identified and the effect of varying levels of seed purity simulated to quantify final admixture levels in the crop.*

Final perspectives

To ensure efficient research on co-existence at EU level we need to move from considering co-existence as a general issue to understand that it may or may not be a problem depending on many factors. At least, current studies have established firmly that co-existence is a crop and farm type dependent issue. But we know little about the influence of landscape patterns and the influence of global rate of GM crop share in a region.

We could go farther in our "forecasting" ability by analyzing the probabilities of uptake (adoption) of GM crops in different EU farming areas. Economic theory provides a sound background to analyse the spread of new technologies. The rate of adoption and spatial distribution of GM crop uptake in the EU will not be uniform. Econometric models could be used to predict when and where the technology will spread. This ex ante studies are difficult to perform but some work has been done already, for example in forecasting the levels of adoption of GM HT rape crops by French farmers, or GM sugar beet by UK farmers.

A significant effort along this line will yield the results most useful for policy makers: we will be able to predict which crops will be adopted, by what type of farmers, and consequently "map" hotspots for co-existence, moving therefore the discussion to more precise grounds and redirecting our research and monitoring efforts to those hotspots.

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Pollen dispersal and cross pollination

J. Sweet
NIAB, Huntingdon Road, Cambridge
United Kingdom
E-mail: jeremy.sweet@niab.com

Summary

This paper considers the significance of pollen-mediated gene flow in the major crop types that have been genetically modified and have been commercialised or are close to commercial release in the European Union, specifically in relation to issues of co-existence. Each crop type has its own distinctive characteristics of pollen production, dispersal and potential outcrossing, giving varying levels of gene flow. Measures to restrict gene flow need to take full account of both the general dispersal characteristics of the pollen from each species and the biological factors which govern cross compatibility, pollination competition and seed set.

Background

Genetic modification can potentially improve crop quality and productivity. The molecular techniques employed to do this essentially involve the insertion and integration of a short segment of DNA from a wide variety of novel genes from unrelated plants, microbes and animals into the genome of a plant. Genetic modification has the advantage of allowing the addition of a single character to breeding lines and varieties without the need for backcrossing to remove unwanted genetic linkages (DoE, 1994). Genetically modified (GM) crops were first released commercially in 1992. Their global area covered 11.0 million hectares in 1997 increased to 27.8 million hectares in 1998 and estimates for 2002/3 suggest that approximately 56 million hectares of GM crops are now grown. The five main GM crops grown are, in order of the largest area, soybean (*Glycine max*), maize, cotton (*Gossypium hirsutum*), oilseed rape and potato, with herbicide tolerance and insect resistance the most utilised genetic traits.

In 1998 the first commercialised GM crop was grown in the European Union (EU). Estimates suggest that introductory quantities of insect resistant maize were grown primarily in Spain (20,000 hectares) and France (2,000 hectares). Other crops being developed for commercial application in the EU include sugar beet, oilseed rape (herbicide tolerance) and potatoes (modified starch) (Dale, 1999). There is no commercial growing of GM crops in most European countries including the UK. However certain imported products have been approved for food use: slow ripening tomatoes, soya that is resistant to a broad-spectrum herbicide (glyphosate), insect-resistant maize and herbicide tolerant rapeseed for oil (Lheureux *et al*., 2003).

Despite the potential benefits of GM crops, there is also concern over the possible environmental and agronomic impacts if the transgenes 'escape' and become established in natural or agricultural ecosystems. From an agronomic point of view, the transfer of novel genes from one crop to another could have a number of implications, including depletions in the quality of conventional and organic crop seed leading to a change in their performance and marketability. Maize, for example, will cross-pollinate with other cultivated maize and sweetcorn (*Zea mays* ssp. *saccharata*), directly affecting the quality and acceptability of the marketed product. Concerns over the ecological impacts of GM crops lie with whether or not a crop has wild relatives and the ability to cross-pollinate them. If crops hybridise with wild relatives and gene introgression occurs wild populations could incorporate transgenes that change their behaviour and they could present an economic threat as weeds or an environmental threat as competitors in natural communities. Oilseed rape, beet, grasses and several vegetable and fruit crops have varying degrees of sexual compatibility with a number of wild relatives found in Europe, and introgression of novel crop genes into some of these relatives is likely. Other crops, for example maize, have no wild relatives with which they could potentially cross-pollinate in Europe.

This paper draws on the recent reviews by Eastham and Sweet (2002), Bartsch *et al*. (2003), Gepts and Papa (2003) and several others.

Factors affecting pollen dispersal and cross-pollination

Distance between source and sink

Pollen concentrations decline rapidly with distance from source in classic exponentional decline or leptokurtic curves which often have long "tails" showing that low levels of pollen can disperse over long distances. Dispersal and distribution patterns will depend on the environmental and geographical factors and insect vectors described below. Levels of outcrossing are usually directly proportional to the concentration of pollen if there is no competitive pollen. However most receptor crops will be producing their own pollen which will dilute incoming pollen and compete for pollination sites on stigmas. Thus outcrossing levels will decline very rapidly from the edge towards the centre of the recipient crop. This factor is already exploited in seed crops, where the outer rows are used as a barrier for reducing levels of outcrossing and are discarded at harvest to maintain the high purity of the crop.

Size of pollen source and sink: The extent of cross-pollination between fields of crops or between crops and wild plant populations is largely dependent on the scale of pollen emission and dispersal (Raybould & Gray, 1993). A theoretical study by Crawford *et al.* (1999) examined the effect of increasing pollen source size on resulting levels of cross-pollination.

He concluded that a square 400 m^2 crop would emit ¾ the amount of pollen that a 4ha (40,000 m^2) crop would emit, but suggested that the effectiveness of pollen dispersal would decline significantly in crop areas of less than 400 m^2. Walklate *et al.* (in press) use models to estimate the dispersal patterns of pollen from various sources. The cross-pollination rate from one field to another was shown to depend on the sizes of both fields. As pollen disperses from larger source areas it develops a leptokurtic distribution curve with larger measurable amounts at greater distances. Evidence indicates that most airborne pollen from small to moderate sized fields contributes to the local component in this way (Treu & Emberlin, 2000). Due to conclusions of this kind many believe that small-scale field trials have done little to remove uncertainty over the scale of pollen emission and dispersal likely to emanate from genetically modified crops.

In addition there is no doubt that field shape, the juxta position of fields and the area of field exposed to the influx of pollen in relation to concentrations of local pollen are all additional factors contributing to levels of out crossing. (Thompson *et al.,* 1999) There is a need for good data from studies carried out on an agricultural scale in order to develop conclusions on potential outcrossing.

Pollination vectors: Pollen is effectively distributed by insects and is the main mechanism for hybridisation in some species (e.g. legumes). Pollen produced by some crops, for example oilseed rape, can be dispersed over considerable distances by both wind and insects. The weather can affect the behaviour of pollinating insects on the crop and the occurrence of airborne pollen movement so the amount of cross-pollination can vary significantly from crop to crop and day to day. The numbers and even species of natural pollinating insects can vary considerably in their contribution to successful pollination (Williams, 1987). The bumblebee (*Bombus* sp.) is an example of a pollinator, which moves only short distances between flowers so the majority of pollen is deposited in the immediate surroundings of the pollen source. By contrast, the foraging habits of the pollen beetle (*Meligethes aeneus*) mean that they emigrate from a crop in large numbers and often fly over long distances (Skogsmyr, 1994).

Environmental factors: Air currents and wind are important for the dispersal of pollen for a wide range of species. Pollen can be lifted high in the atmosphere and distributed over long distances. Dispersal patterns are a product of the nature of the pollen (shape, size, density, nature of surface) and the air currents occurring over the crop at pollen dehiscence. (McCartney & Lacey, 1990). Pollen released on the airflow can settle by gravity, can be removed by precipitation, be absorbed into water droplets, or can impact onto surfaces including vegetation, buildings, soil and water bodies. The relative importance of these sinks and the impacts they might have will vary with factors such as the terminal velocities of the pollen grains, climate, local vegetation and topography (Treu & Emberlin, 2000).

Weather: Pollen dispersal can be heavily influenced by the weather and changes in temperature, humidity and light, as well as wind and rain. For example, studies on pollen dispersal by Scott (1970) over several years revealed that the average concentration of oilseed rape pollen during one day of one year measured 1.4% of that on the same day the following year. This was due to heavy rain and high humidity on the first day compared with sunshine and low humidity on that day a year later. Wind strength can also have an important role in distributing pollen grains significant distances within their viability periods.

Local environment: Patterns of pollen dispersal can be heavily influenced by variable factors in the immediate local environment such as the nature of the plant canopy, surrounding vegetation and topography. Wind velocity and airflow are affected by topography, potentially influencing pollen movement from a pollen source to receptor plants.

Physical barriers: Woods and hedges can serve as barriers to air flow, having dual effects of depleting some pollen from the air flow by impaction and filtering and also creating a sheltered zone in the lee. Dense stands of shrubs, herb covers and tree-sized vegetation with full foliage act as catchments for airborne particulates, including pollen (Treu & Emberlin, 2000). Jones & Brooks (1950; 1952) conducted experiments with tree barriers adjacent to a crop of maize. The results indicated that a single row of trees with underbush were effective in reducing the amount of outcrossing by 50% in the plants situated immediately behind the barrier, but was much less effective at greater distances from the barrier. The authors concluded that the tree barrier was less effective in reducing outcrossing than an area of barrier crop occupying an area of equal size to the trees.

Pollen viability and competitive ability: Biological factors influencing successful pollination begin with the ability of the donor plant to produce viable pollen, and the length of time the pollen grain retains its potential for pollination. If the competitive ability of the pollen grain is poor its capacity to compete with fresher pollen produced in the vicinity of the receptor plant will be poor. Pollen viability can vary greatly between species but is also dependent on environmental variables such as temperature and humidity (Treu & Emberlin, 2000).

Levels of outbreeding in the crop: The amount of outbreeding in the crop is an important aspect to consider, and there is a significant positive correlation between outcrossing rates (largely determined by pollination mode) and gene flow variables, reflected in the different isolation requirements for various crops. Wheat, for example, is typically self-pollinated, with cross-pollination under field conditions usually involving less than 2% of all florets. Oilseed rape is known to be mainly self-fertilising and/or insect pollinated although pollen can become airborne and travel several kilometres downwind. Floral morphology and pollen characteristics are also important as the morphology and terminal velocity of pollen grains influence dispersal patterns.

Degree of synchrony in flowering times: There must be some overlap in flowering times between the pollen donor and the receptor plant so that ripe pollen and receptive stigmas are produced at the same time, in which case a higher degree of cross-pollination might occur than if partial self-pollination had begun in one of the plants.

Homogeneity of the GM crop: GM crop plants of maize, beet and many other species are often the product of a hybridisation between parent lines, of which only one may be GM. These heterozygous GM plants will therefore produce pollen, which segregates for the GM character so that only a proportion of the pollen is GM. Thus levels of outcrossing will often be higher than the actual rate at which the transgene appears in the recipient crop.

Hybridisation, gene flow and introgression

In its broad sense 'hybridisation' can be defined as the cross-breeding of genetically dissimilar individuals. Such individuals may differ by one or a few genes (the pure lines of plant geneticists), by several genes (e.g. hybrid maize) or be very different genetically (as in most hybridisations between members of different genera). Hybridisation is common within species but can also occur between species and occasionally with species in different genera. Hybridisation between different species can be described as 'interspecific' hybridisation or, where species belong to a different genus, 'intergeneric' hybridisation (DoE, 1994). The incidence of natural interspecific and intergeneric hybridisation varies substantially among plant genera and families.

Hybridisation is a frequent and important component of plant evolution and speciation, although the resulting F_1 plants are often sterile and relatively few populations persist, except where the parents remain in contact or where they are able to spread vegetatively (Raybould & Gray, 1993). Table 1 demonstrates the many factors that determine the production and establishment of viable hybrids. The frequent occurrence of fertile hybrids increases the chances of introgression, the incorporation of alleles from one taxon to another, mediated through repeated backcrossing of hybrid individuals to one of the parents.

Gene flow can be defined as "the incorporation of genes into the gene pool of one population from one or more populations" (Futuyma, 1998). Such gene movement is a major determinant of genetic structure in natural populations. Gene flow is strongly influenced by the biology of the species and is likely to vary with different breeding systems, life histories and modes of pollination. Assuming sexual compatibility between a crop and wild relative, the entry and subsequent spread of a transgene into natural populations will be determined to some extent by pollen movement. Different crop species have different pollination mechanisms (insect and wind) and different seed dispersal patterns. Both may act as vectors for transgenes from crops, but the subsequent dispersal of the genes through pollen and seeds may be completely different (DoE, 1995), depending on the reproductive characteristics of the species.
Gene flow is measured in various ways. The most common direct method for plants is the observation of seed and pollen movement, which gives an estimate of potential gene flow

(dispersal). Other methods use genetic and transgenic markers to estimate actual gene flow. A simple method is to introduce or identify a plant in a population with a unique genetic marker (e.g. an isozyme allele or herbicide tolerance) and to follow the appearance of the marker in the next generation. (Eastham & Sweet, 2002).

Table 1. Factors determining the likelihood of hybrids, between crop plants and related species, becoming established in agricultural or natural habitats. From Dale (1994).

The production of viable hybrid seeds

1. Compatibility of the two parental genomes (mitotic and genetic stability)
2. Ability of the endosperm to support hybrid embryo development
3. Direction of the cross: one parent may support embryo and seed development better than the other
4. Number and viability of hybrid seeds

Establishment of hybrid plants from seeds in soil

5. Seed dormancy
6. Vigour of the hybrid plant
7. Direction of cross: maternal effects influencing seedling vigour
8. Nature of habitat: wild, semi-wild or agricultural
9. Nature of competition from other plants
10. Influence of pest, disease and animal predators

Ability of the hybrid to propagate vegetatively and sexually

11. Method of vegetative propagation
12. Persistence of vegetative propagules in agricultural habitats
13. Dissemination of vegetative propagules
14. Invasiveness of vegetative propagules in natural habitats
15. Sexual breeding system: cross-compatible, self-compatible, ability to cross to either parental species
16. Male and female fertility: meiotic stability and chromosome pairing
17. Seed number and viability
18. Seed dormancy
19. Nature of habitat: wild, semi-wild or agricultural
20. Nature of competition from other plants
21. Influence of pest, disease and animal predators

The frequency and occurrence of genetic movement between different plants forms the basis of practical decisions about the isolation requirements of crops where varietal purity of the seed is essential. Some crops have sexually compatible relatives that are found as wild plants and arable weeds. Sugar beet, for example, can be accompanied by related wild beet, and there is well-documented evidence of gene transfer between the two (Boudry *et al.*, 1993).

Crops

Oilseed rape can be described as a high-risk crop for crop-to-crop gene flow and from crop to wild relatives. At the farm scale low levels of gene flow will occur at long distances and thus complete genetic isolation will be difficult to maintain. This particularly applies to varieties and lines containing male sterile components, which will outcross with neighbouring fully fertile GM oilseed rape at higher frequencies and at greater distances than traditional varieties. Gene stacking in *B. napus* has been observed in crops and it is predicted that plants carrying multiple resistance genes will become common post-GM release and consequently GM volunteers may require different herbicide management. Oilseed rape is cross-compatible with some wild relatives and thus some of these (especially *B. rapa*) can also act as sources of GM admixture for non-GM crops.

Sugar beet can be described as medium to high risk for gene flow from crop to crop especially in relation to seed production. Pollen from sugar beet has been recorded at distances of more than 1km at relatively high frequencies. Cross-pollination in root crops is not usually considered an issue since the crop is harvested before flowering. However a small proportion of plants in a crop will bolt and transgene movement between crops may occur in this way. Hybridisation and introgression between cultivated beet and wild sea beet has been shown to occur and thus wild beets can act as a source of impurity as well as stimulating the production of flowering beets in beet crops.

Potatoes can be described as a low risk crop for gene flow from crop to crop and from crop to wild relatives. Cross-pollination between production crops is not usually considered an issue since the harvested tuber is not affected by incoming pollen. In true seed production areas, however, the likelihood of cross-pollination between adjacent crops leading to contamination is higher. The risk of gene flow exists if volunteers are allowed to persist in a field from one crop to the next. Naturally occurring hybridisation and introgression between potato and its related wild species in Europe is unlikely.

Maize can be described as a medium to high-risk crop for gene flow from crop to crop. Evidence suggests that GM maize plants would cross-pollinate non-GM maize plants up to and beyond their recommended isolation distance of 200 m. Thus specific measures such as removal of outside rows at harvest may be necessary to achieve crop thresholds. There are no known wild relatives in Europe with which maize can hybridise.

Wheat and triticale can be described as a low risk crop for gene flow from crop to crop and from crop to wild relatives. Cross-pollination under field conditions normally involves less than 2% of all florets so any outcrossing usually occurs with adjacent plants. Hybrids formed between wheat and several wild barley and grass species generally appear to be restricted to the first generation with little evidence for subsequent introgression due to sterility.

Table 2. Frequency of pollen mediated gene flow in different crop types.

Crop	**Frequency of Gene Flow from Outcrossing**	
	Crop to Crop	**To Wild Relatives**
Oilseed rape	High	High
Sugar beet	High (seed). Low (crop)	High (seed), Medium (crop)
Maize	Medium to high	No known Wild Relatives
Potatoes	Low	Low
Wheat, Barley, Oats	Low	Medium (Oats). Low (W & B)
Rye & Rice	Medium	Low (rye) High (Rice)
Grain/forage Legumes	Low	Low
Grasses	Medium to High	High to medium
Vegetables	High (seed). Low (crop)	High (seed) Medium (crop)
Fruits – Strawberry, Apples, Grapevines and Plums	Medium to high	Medium to high
Raspberries, Blackberries, Blackcurrant	Medium to high	Medium to high

Barley and oats can also be described as a low risk crop for gene flow from crop to crop and from crop to wild relatives. They reproduce almost entirely by self-fertilisation, producing small amounts of pollen so that most outcrossing occurs between closely adjacent plants. There are no records of naturally occurring hybrids between barley and any wild relatives in Europe but oat will hybridise with wild oat at low frequencies.

By contrast rye and rice are outcrossing species and will require isolation from GM crops and possibly rye also from GM triticale depending on the compatibility of specific varieties. Rice has weedy relatives often occurring in or around crops, which can hybridise and hence become sources of transgenes.

Leguminous crops such as peas, beans, soya, lupin etc and forage legumes such as clover and lucerne are mostly self pollinating with low levels of hybridisation. However many of them are attractive to pollinating insects and so appropriate measures for isolation are needed for these crops that take account of hybridisation frequencies and the movement of bees between crops.

Some fruit crops, such as strawberry, apple, grapevine and plum have outcrossing and hybridisation tendencies, which suggest that gene flow from GM crops to other crops and to wild relatives is likely to occur. For raspberry, blackberry and blackcurrant the likelihood of gene flow is less easy to predict, partly due to lack of available information.

Managing gene flow

At present none of these crops has pollen, which can be completely contained. This means that the movement of seed and pollen will have to be measured and managed much more in the future. Management systems such as spatial and temporal isolation can be used to minimise direct gene flow between crops, and to minimise seed bank and volunteer populations. The use of isolation zones, crop barrier rows and other vegetation barriers between pollen source and recipient crops can reduce pollen dispersal, although changing weather and environmental conditions mean that some long distance pollen dispersal will occur (Champolivier *et al*., 1999). Biological containment measures are being developed that require research in order to determine whether plant reproduction can be controlled to inhibit gene flow through pollen and/or seed.

The possible implications of hybridisation and introgression between crops and wild plant species are so far unclear because it is difficult to predict how the genetically engineered genes will survive in a related wild species. While it is important to determine frequencies of hybridisation between crops and wild relatives, it is more important to determine whether genes will be introgressed into wild populations and establish at levels, which will have a significant ecological and agricultural impact as sources of transgenes for crops. This information is needed to determine management of related wild species in crop areas.

Seeds may be distributed in time through their dormancy mechanisms as well as in space. Together with cross pollination, they play a significant part in the movement of transgenes. Thus managing gene flow will also involve careful management of seed, seed banks and volunteers since the latter will provide sources of pollen.

Recommendations for further research

Better management systems and stewardship schemes to minimise GM contamination and gene flow require good scientific information on both seed and pollen mediated gene flow. Further research priorities are:

- More data on the effects of the scaling up of GM crop production on GM pollen emissions in agricultural regions and the impact on non-GM crops in the region. These data need to be modelled building on studies such those by Squire *et al.* (1999) and using models such as those developed by Colbach *et al*. (1999).

- Both temporal as well as spatial gene flow also arises through seed persistence and dispersal. More information is needed on the role of seed banks and dispersed seed of GM crops on contamination of subsequent crops.
- Gene transfer through cross pollination can be limited by effective biological and physical barriers. More research is needed to examine the options for these in the light of recommendations from the EU on thresholds for contamination of non-GM crops.
- Future monitoring of experimental and commercial releases of GM crops must be based on good scientific knowledge of the behaviour and ecology of the GM crop and its wild relatives. Understanding gene flow and introgression is a key part of this requirement.

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Co-existence of conventional, organic and GM crops – role of temporal and spatial behaviour of seeds

P.J.W. Lutman
Rothamsted Research, Harpenden, Herts AL5 2JQ, United Kingdom
E-mail: peter.lutman@bbsrc.ac.uk

Abstract

Seeds pose a threat to the co-existence of GM and non GM crops (including organic ones). No harvesting operations remove all crop seeds from the fields and these seeds will persist but will eventually germinate, carrying transgenes into the same crops sown in subsequent years. The longevity of these seeds is crucial, because it will determine the frequency of re-cropping with the same crop but with different GM/non GM characteristics. Seeds transferred out of the field on/in machinery can establish feral populations in semi-natural habitats such as roadside verges. Most of such populations appear transient but a minority can persist for some years. For the current northern European crops the main concern resides with oilseed rape as copious quantities of seeds (5-10,000 seeds/m^2) can be left in the field and with inappropriate management considerable numbers can remain 4-5 years later (evidence of persistence for up to 10 years). It is therefore critical that as few rape seeds as possible are incorporated into the seedbank. Rape seeds on the soil surface tend to germinate and/or are eaten by predators. Cereal crop seeds can also persist but they are not as persistent as oilseed rape. If sugar beet is allowed to flower, it too will produce a long-lived seedbank, as will the true seeds of potatoes.

Introduction

There are two main elements to the spatial and temporal co-existence of conventional, organic and GM crops, the behaviour of pollen and the behaviour of seeds. Much research has focused on the distribution of pollen and the potential for gene flow between crops, and between crops and related wild species. Much less attention has been paid to the impact of seeds, which have both a spatial and temporal component to their behaviour in this context. Pollen flow is an issue for all crops that flower but the behaviour of seeds is only of major concern for crops that have the potential to establish long-lived seedbanks. The current species of most concern is oilseed rape, but other crop species, such as cereals can form persistent seedbanks. There is a third aspect – the potential for vegetative propagules to persist. This is particularly relevant to 'groundkeeper' potatoes but also to sugar beet.

No harvesting operation removes every crop seed from a field. Precisely 25 years ago Cussans (1978) reviewed the potential for crops to be weeds. The paper focused on cereals and potatoes but also mentioned ryegrasses and the then minor crop, oilseed rape. The review concluded that approximately 5% of the crop seeds/tubers would be left in the field after harvest. Moreover, seeds can also be spilt from harvesting and transport vehicles and thus be moved away from the original field. This is particularly an issue with the small round seeds of oilseed rape but also readily occurs with other small seeds such as cereals.

Oilseed rape (*Brassica napus*)

This section focuses on oilseed rape but is also relevant to turnip rape (*B. rapa*). Many oilseed rape seeds can remain in the field after harvest; 5-10,000 seeds/m^2 are not unusual. This applies to winter and spring rapes both in Europe and N. America (Price *et al*., 1996; Lutman *et al*., 1998; Hobson & Bruce, 2002; Gulden *et al*., 2003). These seeds have no primary dormancy and have the potential to germinate immediately, if given adequate moisture (and appropriate temperatures). However, they can become secondarily dormant if exposed to stress and darkness. The stress could be lack of moisture or low temperatures, depending on the climate of the rape growing areas. Once the seeds have been incorporated into the seedbank and have become dormant, they can persist for some years (see below).

Seed losses

Before considering seed persistence, one has to address the issue of how many seeds become incorporated into the seedbank. This can be influenced by initial seed losses and the timing and method of post-harvest cultivation. Obviously, the more seeds that germinate, the fewer the seeds that can form the long-term seedbank and the fewer the seedlings that can emerge in subsequent crops.

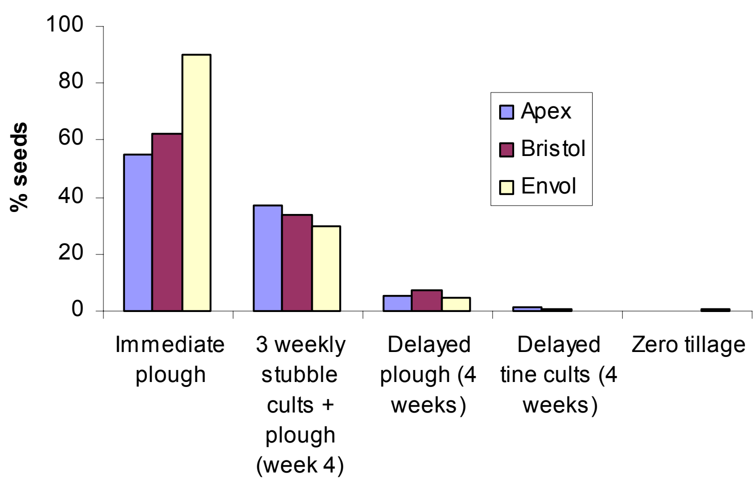

Figure 1. Effect of five different post-harvest cultivations on % of seed numbers c.6 months after seeds was broadcast onto plots. (Experiment (Stackyard field) started in August 1995, c. 10000 seeds/m^2 broadcast onto plots) (from Pekrun *et al.*, 1998).

Work at Rothamsted indicates that delaying post-harvest cultivation can substantially reduce seed numbers entering the seedbank (Figure 1). This is particularly important when the weather is dry over the harvest period (July and August in UK), such as in 1995 and 2003. Best advice is to delay cultivation until rain has stimulated many surface seeds to germinate. Approximately 10mm seems adequate and even 4mm may be sufficient in cool conditions (Lutman *et al.,* 1998). Conversely, immediate ploughing, burying seeds into dry soils, appears to maximise the seedbank in the soil. Studies in Canada do not support this, as some experiments have shown no major differences in seedbanks between plots uncultivated and those cultivated with non-inversion tillage techniques (Gulden, 2003). This difference between UK and Canada probably reflects climatic differences. In Canada, soil can be dry in late summer and additionally temperatures tend to be low. This contrasts with UK conditions, where temperatures remain relatively high from August – October and intermittent rain is common. Thus, one cannot generalise as to optimum conditions to minimise the initial seedbank. Studies in Germany on post-harvest cultivation (Pekrun *et al*., 2000; Roller *et al*., 2002, 2003) tend to support the UK conclusions that immediate post harvest cultivation, especially ploughing, decreases seed losses. Pekrun's work also emphasises that annual variation in late summer and autumn weather can have a greater impact on the numbers of seeds generating a persistent seedbank, than cultivation practice.

Temporal survival of the seedbank

Having established a seedbank the key question in relation to the co-existence of different types of oilseed rape is 'how long do the seeds remain in the soil'? This is not easy to answer. Anecdotal evidence from Europe and Canada suggests that persistence can be appreciable, 5-10 years (Sauermann, 1993; Pessel *et al.,* 2001; Simard *et al.,* 2002) but detailed research information derived from realistic field situations is limited.

Persistence appears to be influenced by both biotic and abiotic factors. Firstly, laboratory studies suggest that some cultivars are intrinsically more persistent than others (Pekrun *et al.,* 1997; Momoh *et al.,* 2002; Gulden, 2003). Secondly, cultivation will increase seed decline rates, as undisturbed seeds appear to persist longer than those in disturbed environments. This applies to seeds of most weed species (see Thompson *et al*., 1997). Thirdly, it appears likely that persistence is somewhat greater on heavy soil than on lighter ones (Gulden, 2003; Lutman *et al*., 2003).

Seed loss in the first 6 months following harvest of the crop and subsequent cultivation is high and can reach 90% (Figure 2). However once the seedbank is established the seeds will persist for some years. Following light tine cultivation and delayed ploughing, 95% of the seeds were lost in 20 months (Figure 2) but where immediate ploughing was used instead of the light tine cultivation it took 39 months to lose the same percentage (data not shown). Data from Germany (Roller *et al*., 2002) also confirm survival of some glufosinate tolerant rape seeds for up to 5 years after the crops were harvested. In contrast, their more recent work demonstrated only minimal survival of two glufosinate cultivars two years after the incorporation of the seeds into the soil (Roller *et al.,* 2003). Interestingly the conventional cultivars, included for comparison, persisted somewhat longer. Clearly there is variation in long-term persistence, and under some conditions it can be appreciable.

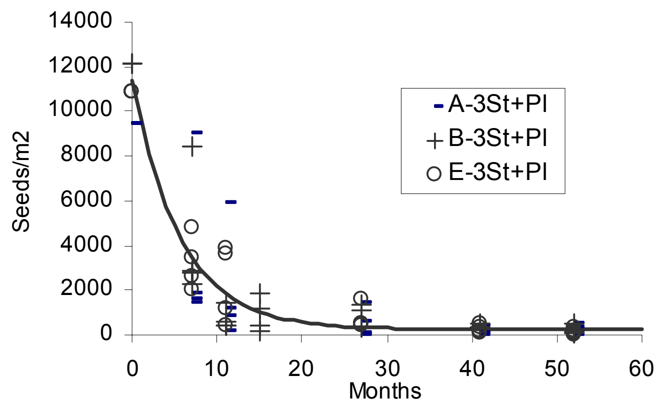

Figure 2. **Persistence of seeds of three cultivars on Stackyard field following tine cultivation weekly for 3 weeks and ploughing in week 4 (3St + Pl) in 1995, and normal arable cropping thereafter (A = Apex, E = Envol, B = Bristol) (from Lutman *et al.,* 2003).**

If it is assumed that the acceptable limit for GM seed in seed from a non GM crop is 0.9% (as required under proposed EU regulations) then one can estimate the impact of persisting volunteers (in field feral plants) on the basis that 1 GM plant/m^2 will exceed this threshold in a commercial crop established at 100 plants/m^2 (which approximates to normal establishment). Assuming the following:

a) seed loss at harvest 5,000 seeds/m^2 (a reasonable average figure (see above))
b) 2% of seeds surviving after 5 years (derived from Fig. 2 and other data, following conventional post harvest cultivation)
c) 2% of seedbank emerges as seedlings each year (this is probably a conservative estimate: P.J.W. Lutman unpubl.data)

- there would still be 2 GM plants/m^2 present in the field, even five years after the previous rape crop. Consequently, there would be a need for greater temporal separation. How long this should be is debate able as: a) we have very limited data from long-term persistence studies in arable fields and b) what data there is suggests that the decline rates of long-term buried seeds, even with annual soil cultivation, is slow.

The above scenario assumes that there is no segregation of the GM trait in subsequent generations. This is not the case as far as glufosinate resistant rape is concerned as segregation does occur in the F_2 generation and work has shown that only 86% of progeny are resistant (Beismann *et al.*, 2003). Consequently, the previous calculation would over-estimate the presence of resistant phenotypes.

Conclusions from a desk study reported by Bock *et al.* (2002) indicated that co-existence of commercial crops would be possible (within the 1% admixture guideline), though it would be much more difficult to meet the 0.3% limit proposed for seed crops. If gene flow is suspected and/or volunteers for a previous crop are present, then it would be difficult to meet the requirements for a seed crop. A recently published report on the potential of volunteer oilseed rape to cause impurities in later rape crops in UK also concludes that it would be difficult to achieve a 1% threshold (Squire *et al*., 2003), especially if the crops were established using 'home-saved' seeds. Lax controls over seed production can result in the farmer using home-saved seeds planting some GM seeds with his 'non' GM crops.

To minimise persistence it seems to be absolutely vital that rape seeds are not incorporated into the seedbank. Rape stubbles should be left uncultivated until rainfall has caused germination of most shed seeds. Such a strategy seems to 'work' in the mild damp autumns experienced in the UK but may not be appropriate elsewhere in the world as has been shown by Gulden's work in Canada, where dry and cold late summer weather prevented germination. The same may apply in continental Europe but Pekrun's work in Austria and Germany tends to show the same responses as seen in the UK (Pekrun *et al.*, 2000). In all situations absence of rain will reduce the success of delaying cultivation in reducing seed numbers. However, if no rain occurs seeds on the soil surface can be predated and although anecdotal evidence of the effects of pigeons suggests that these and other birds can have a marked effect there are little data to quantify their impact. Recently completed studies of seed decline on undisturbed oilseed rape stubbles in UK showed appreciable survival into August but with only 15% of seeds remaining at the end of August (E.J.P. Marshall pers. comm.) In contrast, in Canada, predation of seeds in the autumn seems minimal (Gulden, 2003).

Seed movement off fields

Seeds can be moved off fields in harvesting machinery and then be returned to the soil when the next field is harvested. A requirement of GM field trials with rape in the UK is to thoroughly clean harvesters. Several kilograms of seeds can be removed by cleaning. Anecdotal evidence suggests that perhaps 2 kg of seed could be held inside the combine (drum and augers) and a further 2 kg could be lying on the header. Seeds on the header could be removed by cleaning but not those inside the machine (unless it is dismantled). A study by Norris & Sweet (2002) found 3.9 kg of seed in a combine leaving a GM rape field. Assuming 1000 seed wt of 4 g, even only 2 kg would contain 500,000 seeds. This seed may fall off in transit or can be shed from the combine when the next field is harvested. A study of the presence of volunteer rape plants in the stubble of a winter barley field harvested immediately after a rape field, identified a 'spike' of rape seedlings 52m down the field, with few before or after (P.J.W. Lutman & K. Berry; unpubl. data). Norris and Sweet (2002) reported a similar incident where rape seedlings were found in the stubble of a winter barley field for the first 200 m from the gate. This machine had not been cleaned post rape harvest and had travelled 2 km to reach the barley field. Thus, it is likely that some GM rape seeds will be transported from field to field. This can be minimised by cleaning the combine but not eliminated.

Losses from trailers and trucks are discussed in relation to feral rape. This problem could be minimised by farmers sowing a small patch of conventional rape on one edge of the GM rape field, and then harvesting it last. However, agronomic management of such a 'trap crop' would not be very easy and would increase growers' costs.

Soil movement by cultivators can also move seeds from field to field. The amount of soil moved depends on the machine and the wetness of the soil. Experiments by Mayer *et al.* (2002) concluded that ploughs carried the most soil, especially when wet, and that harrows carried less. Their work showed that ploughs could carry 35 kg of soil in wet conditions. This soil could be transported 1-2 km from the source. From studies of the volunteer rape seedbank (e.g. Lutman *et al.*, 2003) it can be calculated that there could be 2-13 seeds/kg of soil in the first winter after harvesting a rape crop. Thus 35 kg of soil would move a maximum of 455 seeds. This is clearly a less serious issue than the seeds carried in the combine.

Feral populations – persistence and role in geneflow

Feral populations arise from seeds that fall from farm machinery. These may be on field margins, roadside verges or waste ground near to processing plants and depots of agricultural merchants. Some populations would arise within the farm and some on distant sites, as a result of transportation. A detailed survey of the road margins of the M25 London orbital motorway in 1993 and 1994 (Crawley & Brown, 1995) has shown that 33-48% of the margin contained volunteer rape. The populations appeared transient, as many sites occupied in year 1 were not in year 2. However, other work indicates that some feral populations can survive for some years (Pessel *et al*., 2001; Squire *et al*., 1999). These plants could become the source of low level gene flow into neighbouring crops, but as their pollen production is likely to be low in relation to that produced by neighbouring crops, their impact would be small. In reality they would be more vulnerable to incoming pollen from the crop and so if that crop was genetically modified they could become a new source of genes in later years and could also become a source of seeds exhibiting gene-stacking. However, except if cross-pollination thresholds were zero, or if any gene-stacking conferred a clear selective advantage, which as far as herbicide tolerance (in the absence of the herbicides) seems not to be the situation, their impact would still be small.

Sugar beet

Sugar beet is a biennial crop and the roots are produced in year one. But some plants can become vernalised at the seedling stage and produce seeds in the first year. This potential to 'bolt' is affected by weather conditions and varies between cultivars. If GM beet plants are permitted to flower, the seeds produced could survive for at least 10 years (MJ May, pers. comm.). Thus, it is vital that bolting is prevented in any commercialised GM beet crops. These needs are exacerbated by the presence of annual beet in many European sugar beet fields (Longden, 1993) and if cross pollination between these weedy beet species and the GM crop were to occur, especially if it was transformed to be herbicide resistant, then much of the benefit of the GM technology to control weed beet would be lost. A recent paper by Arnaud *et al.,* (2003) identified the risk of moving soil containing beet seeds out of beet fields, as this soil with its beet seeds could be moved to a new site. As some soil movement is inevitable when the roots are taken to the sugar factory it re-emphasises the importance of preventing seed production in GM beet.

Cereals

Persistence of volunteer cereal seeds is not as important an issue as persistence of rape seeds. In general cereal seeds have less dormancy than rape, and tend to germinate readily in the autumn, post harvest. However, it is clear that some seeds can survive for longer than a year, once buried in the soil. Pickett (1993) concluded that wheat could have a 'lifespan in the soil of up to 5 years'. This conclusion is supported by more recent data from Canada that showed low level survival of spring wheat for up to 5 years (Beckie, 2001). However, a recent review (Anderson & Soper, 2003) concluded that wheat rarely survived more that 1-2 years. Barley seeds can also persist as volunteer barley plants can pose problems in subsequent crops of wheat. Thus, if GM cereals were to be commercialised in Europe, some temporal separation of cereal crops would be needed to minimise any possible cross-fertilisation with subsequent crops.

Potatoes

Although the primary problem of potatoes as weeds stems from the persistence of tubers, some varieties produce copious quantities of true seed, which can persist. Potato tubers tend to survive for only one year and depend for longer-term survival on new tubers being produced each year. True seed survives longer. Askew (1993) suggests that seedbanks of potato seed can reach 25,000/m^2 and that these seeds can survive for at least 7 years. Thus, if GM potatoes are to be commercialised the cultivars used should not be prolific producers of flowers. Genetically transformed true potato seedlings would not pose a great threat to other crops, as the plants tend not to be very vigorous, at least in the first season. If it was considered important to avoid the continued presence of transgenes in the environment, true seed production needs to be avoided, unless expression of the transgene in the seeds could be prevented.

Conclusions

There are clear co-existence issues associated with the sequential growing of GM and non GM crops in the same field, as a result of the presence of seeds and volunteer plants from earlier crops. This is most acute in N. Europe for oilseed rape, where large numbers of seeds are inevitably left in the field post-harvest, an appreciable proportion of which can remain dormant in the soil for some years. Cereal seeds can also persist and although Canadian and some UK work shows survival for several years, in practice most seeds seem to disappear in 12 months. If sugar beet is allowed to set seed, this too would create a very persistent seedbank, but the key issue here is the prevention of bolting by GM beet crops. In warmer climates both maize and sunflower can also form persistent seedbanks, but this is not an issue in more temperate conditions, as the volunteer plants are very sensitive to frost. Global warming and the consequent increased planting of these crops in more northerly latitudes would increase the risk of co-existence problems associated with volunteers.

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GMO and GMO free products in Europe: problems of organization in the agricultural sector

E. Valceschini
INRA 16, rue Claude Bernard 75231 Paris Cedex 05, France
E-mail: valcesch@inapg.inra.fr

Abstract

The separation between GMOs and GMOs free product is highly demanding from an organizational point of view. This paper analyses the characteristics of that organization and identifies the conditions of its effectiveness, its cost factors and its limits.

This paper is based on a case study[1] undertaken in 2000 in France. The study consisted of a survey on 21 operators in the agri-food sector, from seed companies to distributors, producers and users of maize, soy and products or ingredients containing them. We analysed five branches, which have the objective of supplying guaranteed "non-transgenic" products.

GMO/non-GMO separation: what is the problem?

The only economic agents who can identify products with GMO characteristics are their direct users in agriculture and seed producers. However, the farmer who does not use GMO seeds has no way of knowing whether his/her seeds are actually GMOs or not. It stands to reason that an individual consumer cannot either assess whether a product contains GMOs or not. When it comes to processed products, the multiple circuits used and the complexity of aggregating operations of material make the GMO or non-GMO characteristic of a product very difficult to determine. For all the economic operators at the various levels (manufacturers, retailers), checking whether production methods include some form of GMO or not is a difficult and complex process.

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[1] More generally, it draws on knowledge acquired during a research programme that I coordinated between 1999 and 2001 "Economic relevance and feasibility of a non-GMO sector". This programme consisted of four parts:
Ruffieux, B. (2001) *Analyse de la propension des consommateurs à acheter des produits garantis "sans OGM"*. ENSGI, Université de Grenoble;
Bertheau, Y. (2001) *Caractérisation scientifique et technique des produits garantis "sans OGM"*. INRA Versailles.
Meynard, J. M., Le Bail, M. (2001). *Isolement des collectes et maîtrise des disséminations au champ*. INRA, INA P-G, 57 p.
Valceschini, E., Avelange, I. (2001). *Analyse économique et réglementaire de l'organisation d'une filière " sans OGM "*. INRA, March, 137 p.

The GMO/non-GMO characteristic of a product thus remains hidden at all stages of the chain. Economists[2] talk of "confidence characteristics", i.e. characteristics not directly identifiable, neither before purchase nor after use.

In view of this problem, consumers can demand information in the form of labelling. In a customer-supplier relationship economic agents can use a certificate of guarantee. However, on their own they cannot assess the accuracy of the information. The management of information concerning the transgenic nature of a product or production process involves considerable risks related to asymmetric information between sellers and buyers, to potential opportunistic behaviours, to possible failures in coordination between the different production stages, and to difficulties of control.

The question is then, what economic organization is capable of maintaining the credibility of information on the separation between GMO and GMO free? In economic terms, credibility relates to suppliers' ability to guarantee their commitments. What commitments must farmers and economic agents at the different levels of the agri-food chain make individually and collectively to ensure that this information is reliable? What systems of guarantee and control have to be implemented?

The key problem is managing the risk of products being mixed[3]. This type of problem is already well known in the agri-food sector. Two other types of case involve a similar problem. The first concerns agricultural production for specific industrial use (e.g. waxy maize for starch manufacturing; oil-rich maize for animal fodder; vegetables for industrial processing). The production of certified seeds is another example, although it is distinguished by the requirement of a higher level of purity. The second type of case corresponds to products guaranteed by an official quality label (e.g. the *Agriculture Biologique* -organic farming- label) or products identified by a collective quality label.

In all cases, the problem turns around the organization established to separate production and product flows, and an information system to control and support that separation. From an economic point of view, the aim is to minimize risks of mixing, at the lowest organizational costs. In this respect, the organization of the GMO/GMO free separation has many points in common with the cases cited. But it also has specific characteristics that are particularly important.

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[2] In these cases economists distinguish three types of characteristic, depending on the time at which the buyer is informed of these characteristics: "search characteristics" that the buyer identifies before the purchase; "experience characteristics" that are identifiable only after purchase; and "confidence characteristics" that are never identified.

[3] The risk considered here concerns only the objective of purity of a product. It does not concern possible consequences regarding the environment or human health.

Separation and guarantee: what forms of economic organization?

Complete separation

Technically, control of the absence/presence of GMOs cannot be conceived of without a complete separation between the transgenic and non-transgenic sectors[4]. It is necessary to ensure that the separation of ingredients can be maintained throughout the entire process, "from seed to fork". Pollution of fields, accidental contamination during storage or transport, and mixing of batches during production or distribution are obviously all possible and raise questions of organization, traceability and control.

From the agricultural production stage, it is necessary to organize the identification and separation of cultivated fields and of the batches harvested and stored. It is then necessary to establish procedures for separating and identifying batches of products – something which is complicated by the fact that most food processing activities today involve the breaking up of raw products (grinding of wheat flour, for example) and then the combination of different products (e.g. cooked dishes).

A dedicated supply chain

The solution that minimizes risks of mixing consists in setting up an entirely dedicated separate supply chain[5]. Risk is controlled more effectively when the entire chain uses specialized tools. This organization has a high cost in terms of equipment, especially because it allows less flexibility. It requires a collective agreement by all parties in the supply chain, and delegation of steering of the organization to a centralized body[6] with some authority over the various individual partners. Its effectiveness requires it to control all supplies from the first raw materials (and even to certify their production processes via a set of requirements).

A regional farmers' organization

Seed and agricultural production are critical stages that have the potential to block or facilitate dissemination and mixtures. In this respect commercial enterprises that collect and store products play a key part. They have management capacities in three main areas: dissemination of seeds; location of production (choice of areas for cropping, farms and fields); and management of facilities for collecting, storing and commercialising products.

The first type of organization aims at avoiding any GMOs whatsoever in a supply area, on the scale of a region or even of a continent (e.g. Europe, Brazil). In terms of an agreement between all the economic players in the area under consideration, any sale, planting or collection of GMO seeds in a "naturally" isolated production area is banned[7]. The aim is total

[4] Note, however, that guaranteeing "zero GMO" products is a scientifically inconceivable and technically impossible goal.
[5] Either a GMO or a non-GMO speciality.
[6] This may be a private promoter (in France, the retailer Carrefour, for example) or a collective or inter-professional body that federates agricultural and industrial actors (e.g. in France the label *"Soja de pays"*).
[7] For example, the Alsace region is "protected" by the Rhine and the Vosges mountains from possible contamination from neighbouring areas.

geographic isolation. In this way separation costs – basically, costs of eliminating GMOs – can be avoided. Guarantee costs concern *ex ante* control of the seed distribution stage and *ex post* control after the constitution of batches (from samples).

Another type of organization is based on planning, with centralized coordination of farmers' individual production plans. On the basis of contracts with farmers – either individually or as groups – the aim is to create "islands" of protected production intended for dedicated facilities, for a particular industrial use. It is often the commercialisation or processing firm that takes care of the planning. This type of organization is more flexible than the preceding one, especially if storage is delegated to farmers themselves, who nevertheless remain under the central authority. On the other hand, risks of mixing are higher, and consequently so are control costs.

Control by certification

Considering the "hidden" nature of GMO characteristics, certification is the most effective method for guaranteeing reliability of information throughout the agri-food chain. Certification bases the credibility of information on control by an independent outside organization. The guarantee is based on controls, both *ex ante* (producers' accreditation) and *ex post* (verification of products).

A complex system of traceability

Certification is a demanding control system in terms of informational and organizational techniques. The now-widely accepted term traceability refers to this system. In the case of GMOs, the traceability system is particularly complex.

Identifying and then guaranteeing the stability of the GMO free characteristic requires a combination of three conditions. The first is traceability of origin, the aim of which is to define the original characteristics of products and to monitor and control their movements throughout the agri-food chain. The second condition is traceability of processes[8] aimed above all at finding the source of a defect revealed by *a posterior* control (withdrawal of product if it does not meet the tolerance threshold). The third condition completes *a posterior* control of products on the basis of a sampling plan, by analyses to detect the presence/absence of GMOs.

Additional costs of separation and guarantee

In the case of GMO/non-GMO separation we distinguish two main types of additional costs: those associated with separation *per se* and those related to guarantees of purity of products. Separation costs concern all the mechanisms needed to control risks of mixing and to eliminate batches containing mixtures. They consist primarily of additional expenditure on the purchase of raw material due to identification, costs of operations for maintaining separation,

[8] This distinction is not necessarily found in the physical and organizational system. Both types of traceability are articulated within an information, detection and production management system.

and hidden costs that essentially cover the loss of flexibility (e.g. equipment is no longer multi-purpose). Guarantee costs are inherent in information management procedures, which ensure that the non-GMO identity is preserved, i.e. that separation is both reliable and effective. They involve traceability costs, detection costs, certification costs, and insurance costs (e.g. against commercial risks).

The level of costs is determined by the mechanisms set up to eliminate or control sources of mixing. The control procedures and separation devices required depend both on the probability of mixtures and on the accepted level of risk of mixture. Not all sectors have identical costs. For example, in the current situation the non-GMO maize "commodities" sector in Europe is under little pressure from GMOs[9]. In this case, additional costs are only guarantee costs. The situation of specialized "non-GMO soy" is different since it accumulates additional costs of separation and guarantee.

Specific characteristics of GMO/non-GMO separation

A very high performance constraint

At the production and commercial stages of agricultural produce, the "non-GMO" requirement has introduced strong specific constraints due to the severity of the required purity levels, to which collection and storage firms are not familiar with. The 0.9% threshold under which it is not compulsory to indicate the presence of GMOs on a label has no equivalent today except in the most tightly controlled micro-branches (e.g. seeds). This order of magnitude is unknown to actors in the cereal sector whose quantified evaluation of risk remains highly empirical and therefore based on "usual" thresholds (between 2 and 4%, depending on the branch). Furthermore, recognition of the presence of GMOs, per field or per batch, is more difficult than for classical quality criteria whose evaluation often involves the identification of variety (often based on visual criteria) or simple chemical tests (content of certain elements).

A complex control system

In addition to this strict obligation of performance, the GMO/non-GMO separation is subjected to an obligation of means. This implies the establishment of a very complete traceability mechanism that combines traceability of origin and traceability of processes. Until now in the agri-food sector, the two systems of traceability were not implemented together. For example, in the case of products with public labels, only traceability of origin was systematic, whereas in food processing it was essentially traceability of processes that was implemented. Thus, until now the maize and soy supply chains used systems of traceability of processes but never of origin. Likewise, separation was not based on the origin of the batches that were often large, so that different origins were mixed.

[9] Apart from 20,000 ha of maize in Spain and a few dozen hectares of experimental fields, Europe is free of GMO crops.

Risks of mixing: considerable externalities
Another specific characteristic is related to externalities. The case mentioned above concerns chains that wish to produce a speciality or to identify a particular quality that is subject to the risk of mixtures. In the case of GMOs the opposite is true: it is GMO producers who subject other producers (conventional or organic) to the risk of mixture. In case of mixture, it is the non-qualified product that suffers the consequences (possible downgrading and loss of value). Unlike the above-mentioned cases, the GMO innovation is not only likely to create value for those who adopt it; it also creates major externalities for the other producers. Considering the level of hostility of European consumers to GMOs, these are strong negative externalities[10].
This problem of externalities is further complicated by the existence of two types of risk. First, endogenous risks in agricultural production, related to farmers' strategic decisions concerning GMOs ("I grow GMOs" or "I don't grow them") can be controlled at macroeconomic levels (a field, area of crops). For example, the main endogenous risk is related to the plant species under consideration: colza is more sensitive to risks of mixture than maize, itself more sensitive than soy.

On the other hand, certain so-called "exogenous" risk factors totally escape individual farmers' decisions. Their control depends on collective or public management. I have aggregated their exogenous factors into a synthetic indicator called "GMO pressure". GMO pressure is does make sense in relation to a particular geographic area, as far as Europe is concerned. It is an increasing function of four main factors: the size of areas with GMO crops; the number of GMO plants authorized by regulation in the European Union; the proportion of agricultural raw materials (authorized for commercialisation) incorporated into the production process of products intended for mass consumption; and, lastly, the quantities of seeds imported.

Conclusion

Externalities are certainly the key issue in separation, related to two problems that as yet have not been solved: who should be liable for the dissemination of GMOs? Who should be responsible for the organization of separation: users of GMOs or conventional and organic farmers?
Development of GMO pressure in Europe will be the decisive factor determining the effectiveness and cost of separation. There is a chance that very soon no organization will be able to enforce the legal tolerance threshold.

[10] Noussair., C., Robin., S., Ruffieux, B. (2004) have shown that in France the consumer's propensity to pay dropped by 40% on average for products labelled GMO.

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Certified seed production

H. Klein
Section for Biotechnology and Genetic Engineering
German Plant Breeders' Association
Kaufmannstraße 71-73
53115 Bonn
Germany
E-mail: hklein@bdp-online.de

Abstract

Certified seed is genetically stable quality seed produced in tested multiplication conditions. Strict obligations concerning maintenance breeding and the production of pre-basic and basic seed ensure that the value-added properties of a variety as concerns quality, resistance and yield remain on the same high level.

Certified seed is officially tested and approved seed. The certifying agencies ensure that the multiplication of certified seed was done in conformity with the seed marketing requirements. In particular, they inspect the field plots, taking into account volunteer plants, plant health and minimum distances as well as the minimum requirements of the seed lot with respect to germination, moisture, sorting quality, technical purity and health conditions.
Certified seed is available to the farmer in the quantities needed, wherever it is needed and when it is needed.

Certified seed has guaranteed quality. Legal certainty for the farmer is ensured on the basis of the seed marketing law as well as the "General Conditions for the Trade and the Distribution of Seeds".

Certified seed is subject to continuous official inspection. The official control of legal minimum standards in form of supervision of the seed marketing as well as the control of certified seeds for varietal purity, health and the other requirements mentioned is a means to ensure customer protection.

Certified seed is the basis for a successful cultivation and market-oriented processing of the harvest.

Co-existence of different farming systems with or without genetically modified varieties is manageable, provided workable and feasible thresholds will be laid down for seed as well as for food and feed for all purposes. This enables seed suppliers, farmers and other participants of the entire food and feed chain to meet the requirements of the markets they choose.

High quality certified seed is one of the measures for the management of co-existence.

Quality seed is an asset of outstanding importance with decisive influence on the success in agricultural plant production. In the future, its role in shaping a plant production adapted to market needs will even get more decisive.

Seed production – faithful to the original

The entire food chain profits from the progress achieved by breeding. The certified seed production system ensures that only seed of highest quality will be produced and marketed. The multiplication and marketing of quality seeds are set forth in the Seed Marketing Act and the regulations thereto. The seed industry goes even beyond the already strict legal requirements in form of voluntary supplementary efforts in order to achieve even further improvement of the already high quality level.

The seed production takes place in selected farms, the so-called seed multiplication companies. They produce basic seed from pre-basic seed, or certified seed from basic seed for the breeders. Distribution is done by specialized distribution companies, mostly agricultural co-operatives or private companies trading in agricultural goods.

They determine the quantities of seeds needed in their catchment area and select quality seed of the varieties, which are of particular interest for the cultivation in this region, in co-operation with the official advisors.

Seed certification as a means of customer protection

Seed production is carried out according to the official certification guidelines. This is to ensure that the legally guaranteed quality standards for certified seed must conform to a wide range of requirements and pass numerous tests. It may only be descended from listed seed, normally basic seed. In field inspections and quality testings, this basic seed is subject to even stricter standards than certified seed. The seed multiplication farm and the fields used for seed multiplication also must fulfil well-defined requirements. The seed producer must have the technical equipment and the skills necessary for the production of quality seed. The state of cultivation on the production area must be properly treated and prepared.

The official certification agencies carry out field inspections in order to test the seed multiplication crops for varietal purity, for presence of volunteer plants and seed borne diseases as well as minimum distances.

Experienced seed certifiers inspect each individual seed multiplication field plot for conformity with the legal minimum standards and maximum tolerances. Only a small number of plants of other species or deviating from the variety properties may be found in the samples taken. Very strict standards apply to seed-borne diseases. In the case of cross-pollinating crops, minimum distances to neighbouring field plots cultivated with the same crop are to be respected.

An important task of the seed multiplier is therefore not only to carefully select the multiplication field plot, but also to keep these plots free of plants of other crops or weeds, continuous inspection of the seed multiplying crop, their protection against diseases and rouging of undesirable plants.

After the harvest, the seed will be treated and an official sample will be taken for testing at the official regional testing agencies. This is to ensure that the legal requirements for germination rates, health, varietal purity and presence of volunteer plants are fulfilled. If the samples conform to the standards established by the seed marketing regulations, the seed certification agency grants official certification. Only then may the seed lot be marketed in closed or defined containers marked with the blue label for certified seed.

The seed industry surpasses the legal standards for seed quality in crucial points as e.g. technical purity, volunteer plants and germination rates. For customer protection purposes, the rules and regulations on seed marketing only allow seed to be marketed in closed packaging as e.g. bags, containers, cardboard boxes or small packages. The regulations do not only provide for the nature and the closing of the packaging, but also for the labelling of the seed. It may only be marketed with the appropriate labels. In certain cases an additional inside slip is necessary.

The label gives information to the seed consumer on the category of seed. A white label is for basic seed, a blue label for certified seed.
Label and inside slip contain important information for the seed consumer.

Regarding certified seeds, information is provided as follows: applicable EC-Standard, certifying country, reference to the certifying agency, crop, variety denomination, seed category, certification number, closing date (month and year), country of origin, package weight or nominal number of grains as well as additional information.

The additional information in the case of cereals may be e.g. the thousand seed weight, the germination rate and information on seed treatment. They enable the farmer to calculate exactly the amount of seed to be sown and give data on protection of the seeds against diseases and pests.

And finally, legal requirements as to the closing of the package are in place for the protection of the seed consumer. In general, they must be sealed with official lead seals, bands, sealing labels or other forms of adhesive labels. The farmer can refuse to accept opened or damaged packages.

The farmer buying certified seed enjoys a high level of customer protection. The following is guaranteed to him:

- crop and varietal purity
- legal standards for minimum germination rates and technical purity
- legal standards for maximum contents of seeds of other species
- maximum moisture content

Certified seed of high quality - one of the measures for management of co-existence

High quality certified seed is one of the measures for the management of co-existence.
Co-existence of different farming systems with or without genetically modified varieties is manageable, provided workable and feasible thresholds will be laid down for seed as well as for food and feed for all purposes. This enables seed suppliers, farmers and other participants of the entire food and feed chain to meet the requirements of the markets they choose.

Co-existence of different farming systems with or without genetically modified varieties is manageable, provided workable and feasible thresholds will be laid down for seed as well as for food and feed for all purposes. This enables seed suppliers, farmers and other participants of the entire food and feed chain to meet the requirements of the markets they choose.
High quality certified seed is one of the measures for the management of co-existence.

In **crop production**, farmers make use of a lot of measures in conformity with the rules of integrated crop management. There is a complex interaction between the selection of a variety on the one hand and climatic conditions, soil preparation, plant protection, fertilisation, harvesting and crop rotation on the other hand (genotype-environment-interaction). Depending on the intended quality, farmers may adopt additional practices like optimised harvesting, field inspections, keeping field records. Every introduction of an innovative new variety into crop production results in changes of this interplay of rules, measures, conditions and objectives. An example is the introduction of oil seed rape hybrids in comparison to open-pollinating varieties.

In **seed production**, additionally some legal obligations like e. g. minimum distances and regulations on crop rotation are applied to meet general quality criteria. Other measures traditionally used like special field inspections and elimination of off-types, taking care of the ratio of male and female plants in hybrid production, barrier crops and many others are voluntary requirements set up for the special purpose of producing seeds for very specific market demands which could define the seed production as a „premium market“ in comparison to the commodity market of crop production.

All participants of the supply chain have to observe the rules of good practice and to take all reasonable measures to meet the given thresholds. Even today, conformity with the principles of good practice is a genuine interest of all market players and is a responsibility of each individual company. Examples for codes of good practice are the Quality Management (QM) systems of the different processing stages and the descriptions of good agricultural practice.

Co-existence: the seed industry's expertise

Today, the European seed industry operates under a wide range of legislative as well as voluntary rules of production. These rules and regulations are foremost designed to assure the purity levels set by the European Seed Marketing Directives. To do so, specific requirements as regards isolation distances from other seed or crop production areas, rotation cycles and many other requirements are established at national level and in conformity with existing European legislation.

When required by specific market segments and possible under the natural conditions, product qualities even beyond the legal requirements can sometimes be produced, of course with adequate supplementary compensation for premium quality. At the same time, the quality requirements set forth in the Seed Marketing Directives are frequently lowered on demand of individual Member States in cases in which the usual product qualities cannot be supplied in sufficient quantities due to bad harvest conditions or other natural or unforeseeable factors.

Plant breeders and seed producers have also a proven track record of supplying all crop production with the seed of their choice, be it conventional, organic or GM, provided the legislative and market conditions allow for an economic production.

The seed industry is convinced that the existing European and national legislative provisions in connection with the expertise and experience of seed producers in their respective production environment have successfully guaranteed to all European farmers the high quality seed necessary for competitive crop production in all forms of production and markets.

In connection with the taking on of the new technology of GM in seed and crop production, seed producers therefore require solely the additional setting of practical thresholds for all possible sources of adventitious presence of GMOs in conventional seed.

The setting of such thresholds alone will be sufficient to continue to supply the different markets and consumers with the seed of their choice.

Co-existence: Recommendations from the breeders' point of view

In comparison to crop production, seed production obviously is carried out on quite limited areas and under very strict quality assurance policies of companies and seed producers.

Following the existing approach of seed and crop production achieving the highest possible purity levels right at the beginning of the crop production cycle and under conditions which cannot be transferred to the field, i.e. to general crop production, the approach to co-existence must be proportionate to the objective and economically viable if one is to prevent that particular forms of agriculture and especially particular farm or enterprise sizes are placed at an impossible competitive disadvantage.

The seed industry therefore recommends to

- set thresholds for the adventitious presence of GMOs covering all possible sources of events. Such thresholds are preconditions for any further measures to be considered.
- consider the foreseeable future development of GM production in Europe even beyond the setting of such thresholds in order to ensure that they will be economically viable and technically sustainable
- always make use of the least costly and least demanding measure in view of on-farm management measures for co-existence (principle of proportionality)
- develop and exchange best practices and stewardship protocols on such measures
- renounce all attempts to place one form of production at a competitive disadvantage to another. This includes in particular renouncing all attempts to ban any form of production from large scale areas in the European Union.

Conclusions

1. Co-existence is **not a new phenomenon**. Co-existence of varieties of the same crop for different purposes and markets is an existing reality.
2. Co-existence is possible, **provided workable and appropriate thresholds** will be laid down for food, feed and seed, for all products (be it conventional or organic) and for all sources of adventitious presence.
3. In **seed production**, integrated crop management applies to different measures interacting both with each other and with the genotype (variety) and environment. The existing rules for seed production are sufficient to resolve the challenge of co-existence between seed production and crop production.
4. **Farmers** use their traditional experience with respect to specific local natural, biological and technical conditions. Therefore, no uniform measures of good agricultural practice can be defined by legislation. These measures being taken by the farmers have to be flexible and adapted to individual crops and farms as well as cost-effective.

5. Co-existence as a general concept will affect **the entire food and feed chain**, including seed and crop production. But already today differentiated measures are applied by crop producers, particularly by seed producers, to meet the demands for specific product qualities and standards, not all of which are reflected in the labelling.
6. Research, producers (e.g. seed suppliers and farmers), agricultural advisory offices and federal and governmental bodies should work out a set of guidelines for meeting the quality standards concerning adventitious presence. They should develop product **stewardship concepts including management principles for co-existence**. Plant breeders and seed industry will contribute with their expertise.

Management of varietal purity in seed production in France: organisation and cost

G. Sicard
Fédération Nationale des Agriculteurs Multiplicateurs de Semences (FNAMS)
Impasse du Verger, F – 49800 Brain sur l'Authion
E-mail: g.sicard.fnams@wanadoo.fr

Abstract

An important challenge for seed growers is to achieve the varietal purity standards required by the regulation or the market. For many species, mainly allogamous seed crops, varietal purity depends on the management of the surroundings of the farm. Therefore, a collective organisation, on a large scale, is needed. To help growers to resolve co existence between seed production of different cultivars and other commercial crops, several "protected areas" were created in France, according to the law set up in December 1972. The management of seed crops within this protected areas and the organisation evolution during the past years, due to the use of new tools, are described in this paper.

Introduction

The seed grower is responsible for the varietal purity of the seed lot he produces on his farm. For most of the species, varietal purity requirements are defined by the certification standards and vary from 90 to 99.7% (for agricultural crops). Concerning vegetable seeds, for which certification is not compulsory, very high varietal purity levels are often required by the seed companies (99.9 or 100%), to satisfy the needs of the professional market. Insuring the right varietal purity has always been a real challenge for the growers. Many factors affecting varietal purity are linked to good management of the seed production crop itself and good practices on the farm: accurate rotation, control of volunteers, good concordance of flowering (in case of hybrid production), management of pollinator insects, machineries and storage facilities cleaning. For many allogamous species, the risk of undesirable cross-pollination is coming from both cultivated and wild plants growing in the vicinity and must also be controlled. This risk, linked to the environment, varies a lot according to the biology of the species. It is always much more important in hybrid productions than in populations. Different isolation distances have been edicted by National or interprofessional rulings, mainly based on practical experience. Those distances can be revised by the actors of seed sector, like in 1998 for vegetables when the new "standard convention" was agreed. When the isolation distances involve a scale or area much bigger than an average farm, an individual farmer management is not sufficient. Collective management of isolation has therefore been organised for a long time by the growers in different seed production areas in France.

The 22 December law on "Protected Area"

During the sixties, the difficulties to manage co-existence with non-growers in seed production areas raised the need of reglementation. Its aim was to give higher priority to seed crops in specific areas. In December 1972, a law was voted by the French parliament, which gives the profession the opportunity of applying for the creation of "protected areas for seed production". Since 1972, more than one hundred protected areas were created for maize, sunflower, beets, various vegetable and recently also for hybrid rye. In these areas, all the crops of the designated species must be declared to the Public Authority. The same crop for grain production (and not for seed) is not allowed in the area, except if it is in accordance with isolation requirement defined for the seed production fields. Public Authorities can destroy a crop if the isolation policy is not respected. Legal penalties can be inflicted to farmers who do not respect the law. New protected areas are created at profession request. Public inquiries are opened at a regional level and the agricultural organisations are involved. If everybody agrees, the new protected area is officially created by order of the Ministry of Agriculture. The administration of the seed plots in the protected area is directly managed by the seed sector, organised within the GNIS (Groupement National Interprofessionnel des Semences), and local public authority.

Management of isolation in protected areas

The management of isolation distances within a protected area is based on a compulsory declaration of seed plots location several months before sowing date. Formerly drawn on paper maps, this "cartography" is now more and more computerized. After recording all proposed seed production plots, the computer lists automatically the fields, which are not well isolated, according to the standard or particular isolation, distances. Meeting with representatives of seed companies and seed growers are organised to find a solution to each problem. Once accepted by all parts, the cartography become official and all the crops can be sown. Consultation of the cartography is possible at any time on an Internet site.

Management costs

This management has a cost, mainly in terms of time spent by different actors. A rough estimate of that time was made on 2 different protected areas in Anjou (Maine et Loire): the protected area for maize seed production and the protected area for several vegetable seed productions. In maize, an approximate 210 days were devoted to that management, which means for about 4000 hectares an average of 25 minutes per hectare. 2/3 of that time is spent by the farmers themselves (in fact, benevolent representatives) to prepare the isolation at a very local level. For vegetable seed, an approximate 200 days were spent for 2800 hectares

(35 minutes per hectare). In that case, seed companies, GNIS and public administration spend the biggest part of that time.

Conclusion

Management of varietal purity can be considered as efficient in seed production in France, due to several factors.

- It is an activity of general interest and done on limited acreage;
- There is an attractive added value for the seed growers;
- There is a good organisation of the profession;
- Public Authority is involved in the system.

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GM/non-GM wheat co-existence in Canada: Roundup Ready® wheat as a case study

R.C. Van Acker, A.L. Brûlé-Babel, L.F. Friesen & M.H. Entz
Department of Plant Science, Faculty of Agricultural and Food Sciences
University of Manitoba, Winnipeg, MB, R3T 2N2, Canada
E-mail: rene_van_acker@umanitoba.ca

Abstract

The pending approval of the unconfined release of Roundup Ready wheat in North America has brought to the attention of regulators and scientists in North America the issue of co-existence of genetically engineered (GM) and non-GM crops. In Canada, there has been great adoption by farmers of Roundup Ready and other GM canola varieties. At the time of commercial release of GM canola in Canada no specific GM, non-GM co-existence plans were considered or implemented. GM traits (transgenes) have since become ubiquitous in canola crops in Canada and this has led to some problems. For example, although Roundup Ready canola provides direct operational value to adopting farmers, controlling volunteer Roundup Ready canola in low disturbance direct seeding systems adds cost for farmers. The spread of the Roundup Ready trait in canola means that this added cost is borne by both adopters and non-adopters of the technology. The factors and conditions which led to the spread of the Roundup Ready transgene in canola in Canada appear to be similar for wheat if Roundup Ready wheat were to be released in Canada in the same way that Roundup Ready canola has been. To minimize potential negative impacts from movement of the gene conferring glyphosate (Roundup) tolerance among volunteer wheat populations after the release of Roundup Ready wheat, a co-existence plan must be created and implemented. The plan must be species and trait specific and based on knowledge of biology, ecology and agronomy. The plan must be made functional by legislation and regulation and must provide formal routes of recourse for non-adopters affected by transgene movement. Co-existence plans are progressive and will facilitate the introduction of new traits into crops both by GM and non-GM means.

Introduction

In Canada and the United States, approvals for the unconfined release of Roundup Ready spring wheat (*Triticum aestivum* L.) are pending. Roundup Ready wheat is modified through recombinant DNA technology to be herbicide-tolerant and is considered genetically engineered wheat (GM wheat). There has been some debate and controversy in North America surrounding the pending approval of Roundup Ready wheat. Proponents of this product suggest that it will simplify weed control in spring wheat, reduce herbicide injury to wheat, improve control of current herbicide-resistant weed biotypes, eliminate off-type wheat within a given wheat crop, and increase in-crop opportunities for the control or suppression of perennial weeds (Harker *et al*., 2003; Van Acker & Entz, 2002). Those concerned about the unconfined release of Roundup Ready wheat suggest a number of risks associated with its release including difficulty and cost of controlling volunteer Roundup Ready wheat in low-disturbance direct seeding (no tillage prior to seeding) cropping systems, the evolution of glyphosate resistant weeds in glyphosate dependent cropping systems, the loss of farm saved seed for wheat (Ogg & Jackson, 2001; Van Acker *et al*., 2003), and the adventitious presence of GM-wheat in non-GM wheat segregated for sale to satisfy domestic and export customers of North American wheat who are unwilling to purchase GM wheat (Rosher, 2003). At the core of most of the concerns are questions about movement of the transgene conferring Roundup tolerance from GM to non-GM wheat crops and whether co-existence of Roundup Ready and non-Roundup Ready wheat is possible. In western Canada, Roundup Ready canola (*Brassica napus* L.) has been grown commercially for 8 years on large acreages. Experiences with Roundup Ready canola can be used as a valuable reference for consideration of possible intraspecific transgene movement in wheat and the creation of a functional co-existence plan for Roundup Ready and non-Roundup Ready wheat.

Intraspecific transgene movement in canola in western Canada

GM canola is very popular with western Canadian farmers. In 2003, 48% of the canola grown in western Canada (2.25 of approximately 4.7 million ha in 2003) was Roundup Ready, and since it's commercial release in 1996, more than 8 million ha of Roundup Ready canola have been grown by western Canadian farmers (M. Lawton, Monsanto Canada Inc, pers. comm.). Farmers value the operational benefits of this product citing simplicity and effective weed control as key values they capture by growing Roundup Ready canola.

At the time of unconfined commercial release of Roundup Ready canola in Canada, it was known that there was significant potential for out-crossing within the canola (*Brassica napus* L.) genome and that transgene movement from canola crop to canola crop would occur (Canadian Food Inspection Agency 1995). Work after the release and massive adoption of GM-canola in western Canada has shown that pollen mediated gene flow in canola can be an effective cause of transgene movement. Beckie *et al*. (2001) and Rieger *et al.* (2002) found that out-crossing in canola (*B. napus*) occurred to a distance of 800 and 2500 m, respectively. These studies helped in part to explain why Friesen *et al*. (2003) and Downey and Beckie (2002) found that a majority of the western Canadian grown pedigreed non-GM canola (*B. napus*) seedlots they tested contained genetically engineered herbicide tolerance traits. This adventitious presence of transgenes was not caused by pollen flow alone. Thirty-three percent of the seedlots (9 of 27) tested by Friesen *et al.* (2003), and 18% of the seedlots tested by Downey and Beckie (2002) (13 of 70) had the Roundup Ready transgene present at levels above 0.25%. Given current knowledge of pollen mediated gene flow in *B. napus*, it is unlikely that pollen flow would cause greater than 0.1% presence in a single generation of pedigreed seed production given strict seed production protocols. Adventitious presence levels above 0.25% were likely the result of inadvertent mechanical mixing of certified seedlots during harvest or handling, or contamination occurring in earlier generations of pedigreed seed production (i.e., Breeder or Foundation seed).

The general spread of transgenes among canola crops within a region such as western Canada is a function of canola biology and ecology and the environmental and agronomic conditions under which it is grown. The species characteristics and agronomic conditions interact to create opportunities for genes to move from crop to crop. The characteristics and conditions which have combined to create effective transgene movement for the Roundup Ready trait in canola in western Canada include:

- A very large number of acres of Roundup Ready (2.25 million ha in 2003) and non-Roundup Ready canola (2.45 million ha in 2003) grown in fields across western Canada in a temporal and spatial randomly stratified fashion.
- The relatively high frequency of canola in crop rotations in western Canada (e.g. on average 1 in 4 years on any given field in Manitoba) (Thomas *et al*., 1999).
- The large volunteer canola population in fields in western Canada (Leeson *et al*. 2002 a,b; Thomas *et al*. 1996; Thomas *et al*. 1998 a,b; Gulden *et al*., 2003).
- Volunteer canola commonly survives to flowering at significant occurrence densities in a significant proportion of fields in western Canada (Leeson *et al*., 2002a,b; Thomas *et al*., 1996).
- In western Canada, glyphosate use is extensive and farmers who practice low-disturbance direct seeding use glyphosate every spring for pre-seeding weed control. Low-disturbance direct seeding is currently practiced on 25-30% of the annually cropped acres in western Canada, and that percentage is rising (Statistics Canada 2002). A tremendous selection pressure is created for the Roundup Ready trait in volunteer canola populations. In this situation Roundup Ready volunteer canola has a very large positive fitness advantage over non-Roundup Ready volunteer canola and, according to population genetics theory and experience with herbicide resistant weed populations, the frequency of the Roundup Ready trait will rise rapidly in the volunteer canola populations (Brûlé-Babel *et al*., 2003; Gealy *et al*., 2003; Jaseniuk *et al*., 1996).
- Volunteer canola can persist until, emerge in, and flower in subsequent canola crops (Simard *et al*., 2002; Légère *et al*., 2001; Leeson *et al.*, 2002 a,b).
- Plant to plant out-crossing rates in canola is relatively high (Cuthbert & McVetty, 2001).
- The current canola pedigreed seed production system was designed to maintain varietal purity standards related to performance and end-use function. The system was not designed to prevent gene flow at the level required to prevent problematic appearance of the Roundup Ready trait in non-Roundup Ready canola varieties.

Sometimes gene movement matters

The result of Roundup Ready transgene movement in western Canada is that essentially all volunteer canola populations in western Canada contain some proportion of Roundup Ready volunteers. This is true even if Roundup Ready canola cultivars have never been intentionally planted in a given field. Farmers now cannot be certain of the herbicide tolerance status of their volunteer canola population. When Roundup Ready volunteer canola is present in a field, pre-seeding weed control in low-disturbance direct seeding systems requires the addition of another herbicide as well as glyphosate, adding cost and complication in the crop rotation because of the pre-seeding residue left by some herbicides (Van Acker *et al*., 2003). Farmers who choose to grow Roundup Ready canola balance the added costs and complications against the measurable benefits they receive from this technology. However, because of the ubiquitous presence of the Roundup Ready trait in volunteer canola populations, the added costs and complications in rotation are also borne by farmers who choose not to grow Roundup Ready canola (non-adopters).

The impact of gene movement depends upon the crop. For example, controlling Roundup Ready wheat volunteers in a low-disturbance direct seeding system would cost more than controlling Roundup Ready canola volunteers (Harker *et al*., 2003; Van Acker *et al*., 2003). If the transgene conferring glyphosate tolerance became ubiquitous in volunteer wheat populations in a manner similar to what we have witnessed in canola, then the cost associated with low-disturbance direct seeding systems in western Canada would rise significantly. This would threaten the economic viability of these systems and in turn threaten Canadian farmers' ability to capture the environmental, resource conservation and economic value of low-disturbance direct seeding (McRae *et al*., 2000). In this manner, a production economics issue related to the movement of one trait within a crop species can become an environmental issue.

The potential for damage resulting from gene movement also depends upon the gene (trait) that is moving (Gealy *et al*., 2003). Other novel herbicide tolerance traits in canola (glufosinate tolerance and imidazolinone tolerance in the Liberty Link and Clearfield canola systems, respectively) also move into conventional non-herbicide resistant canola varieties in western Canada (Hall *et al*., 2000). The movement of these traits does not create problems for non-adopting farmers in western Canada because they do not currently depend on glufosinate or imidazolinone herbicides for pre-seeding weed control to replace pre-seeding tillage in low-disturbance direct seeding systems. However, if there were a segregated market for GM and non-GM canola for Canadian farmers, then the movement of transgenes conferring either glufosinate or glyphosate tolerance would matter because these are both GM traits. It should be noted as well, that gene (trait) movement can be a problem whether or not the trait is considered GM. For example, if the Roundup Ready trait had been incorporated into wheat using conventional breeding means, the movement of this trait among volunteer wheat populations would still pose a threat to the economics of low-disturbance direct-seeding systems in western Canada because of the dependence of these systems on Roundup herbicide for inexpensive, non-selective, non-residual, pre-seeding weed control.

The potential for intraspecific transgene movement in wheat

Pollen movement in wheat is facilitated by wind and gravity. In wheat, anthers normally open within the floret, followed by filament elongation and extrusion of the anthers outside of the floret. A small amount of pollen is shed on the stigma within the floret, while 80% of the pollen is shed outside of the floret. Florets that have not been successfully self-pollinated will remain open and be receptive to pollen from other sources for up to 13 days after flowering (de Vries, 1971). Estimates of out-crossing rates in wheat are dependent on synchrony of flowering between pollen donors (males) and pollen receptors (females), the presence of receptive females, and the availability of single dominant nuclear marker genes to facilitate detection of out-crossing. Waines and Hegde (2003) stated that "...there is enough evidence to show that cross-pollination [in wheat] regularly occurs and the reproductive biology of wheat is favourable to facilitate varying degrees of gene flow in a variety of situations."

The factors and conditions that facilitated movement of the Roundup Ready transgene in canola in western Canada appear to be similar for wheat. These include:

- A large number of acres of wheat grown in all agricultural regions of western Canada (up to 10 million ha annually).
- The relatively high frequency of wheat in crop rotations in western Canada (e.g. on average, 2 in 5 years in any given field in Manitoba) (Thomas *et al*., 1999).
- The high population levels of volunteer wheat in average fields in western Canada (Leeson *et al*., 2002 a,b; Thomas *et al*., 1996).
- Volunteer wheat commonly survives to flowering at significant occurrence densities in a significant proportion of fields in western Canada (Leeson *et al*., 2002a,b; Thomas *et al*., 1996).
- In low disturbance direct-seeding systems, Roundup Ready volunteer wheat would be selected for within the volunteer wheat population and, according to population genetics theory and experience with herbicide resistant weed populations (Jaseniuk *et al*., 1996), this would cause the glyphosate tolerance gene frequency to rapidly rise in the volunteer wheat population.
- Empirical evidence shows that in practice wheat is as persistent as canola both in terms of quantity (density) and frequency (% of fields) and it can persist to a measurable level for up to five years (Beckie *et al*., 2001).
- Volunteer wheat can persist until, emerge in, and flower in subsequent wheat crops (Beckie *et al*., 2001).
- Out-crossing rates in wheat are relatively high from plant to plant within a commercial crop (Brûlé-Babel *et al*., 2003; Hucl & Matus-Cádiz, 2001; Waines & Hegde, 2003).

- The current wheat pedigreed seed production system was designed to maintain varietal purity standards related to performance and end-use function. The system was not designed to prevent gene flow at levels required to prevent problematic appearance of the Roundup Ready trait in non-Roundup Ready varieties.

Co-existence planning for Roundup Ready wheat and non-Roundup Ready wheat

The appearance of the Roundup Ready trait in non-Roundup Ready pedigreed canola seedlots in Canada was arguably predictable (Warwick *et al*., 1999). In Canada, the experience with canola can be used as the basis for planning for Roundup Ready and non-Roundup Ready wheat co-existence. Currently, industry led stewardship plans are being proposed to prevent potential negative impacts resulting from transgene movement after the release of Roundup Ready wheat in western Canada. These plans are functionally problematic because industry has limited ability to demand, monitor, or enforce adherence to such plans. In the case of non-adopters, industry may have no ability to demand adherence to these plans. This is especially problematic for the containment of the Roundup Ready trait because prevention of transgene movement via pollen flow in wheat relies critically on management of receptor wheat crops (Waines & Hegde, 2003), and in many (and perhaps most) cases receptor wheat crops will be grown by non-adopters of the Roundup Ready technology. To be effective, co-existence plans for Roundup Ready and non-Roundup Ready wheat need to have certain characteristics. The co-existence plan must:

- Be based on realistic, science-based, robust, tested models of transgene movement in wheat in western Canada.
- Specifically recognize that the Roundup Ready trait is particularly difficult to contain because glyphosate is used extensively for pre-seeding weed (and volunteer wheat) control in western Canada and this gives Roundup Ready volunteers a selective advantage within volunteer wheat populations in western Canada.
- Represent the reality of the biology of pollen mediated gene flow in wheat with specific recognition of the fact that in the absence of genetic technology preventing pollen mediated gene flow, transgene flow has to be controlled at the receptor wheat crop. This poses a particular challenge for transgene containment when receptor crops are grown by non-adopters of the technology.
- Represent a realistic expectation of commitment from farmers to implement the plan given the reality of the vast acreages, the short cropping season in western Canada, and the almost total reliance of current cropping systems on herbicides for weed control.
- Incorporate a mechanism for dealing with non-compliance, and recognition of the jurisdiction and responsibilities of the various stakeholders. Issues of liability and compensation will also need to be addressed.
- Incorporate a mechanism for recourse for those affected by gene movement. With Roundup Ready canola in western Canada mitigation of gene movement impacts were *ad*

hoc and left to the technology developer even though the technology developer was not bound by law or regulation to provide such mitigation.

- Be made functional and enforceable via regulation arising from legislation.
- Be regional and systemic. The experience with movement of the Roundup Ready trait in canola showed that in western Canada volunteer canola existed as a metapopulation with respect to the Roundup Ready transgene. Therefore, containment will require co-existence plan application and adherence throughout the entire cropping system and across the entire region of western Canada. Management for containment within a given field and for a given crop alone will be insufficient to achieve co-existence.

Conclusion

The need for co-existence plans and the stringency of a given plan is a function of the crop and the genes (trait). In some cases gene (trait) movement matters. Whether the movement matters is not necessarily a function of whether the trait is considered GM or not. For those traits for which movement matters, co-existence plans must be created which are based on biology, ecology, agronomy, and competitive advantage of the trait (selection pressure). These plans must be made functional and enforceable through regulation arising from legislation and they must include a formal route of recourse for those affected by gene movement. Effective and functional co-existence plans will protect choice for farmers and consumers. For GM and non-GM crops, or more generally, for crops, which contain traits that must be contained, co-existence planning is progressive. It will facilitate the introduction of new traits in crops using either recombinant DNA (GM) or non-GM means.

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Maize and cereals

Introduction to the co-existence of GM and non-GM maize

E. Alcalde
Syngenta
Ribera del Loira 8-10, 28047 Madrid, Spain
E-mail: esteban.alcalde@syngenta.com

Co-existence, a summary of the current legislative status

Co-existence refers to the ability of farmers to provide consumers with a choice between conventional, organic and GM products that comply with European labelling and adventitious presence standards. Co-existence is not about environmental or health risks because only GM crops that have been authorised as safe for the environment and for human and animal health can be cultivated in the EU.

The EU legislative framework establishes a labelling threshold of 0.9%. This means that crops grown from non-genetically modified seed should have a GMO content not exceeding 0.9%. The 0.9% threshold, which has been adopted by the Council and the European Parliament and provided for by Regulation on genetically modified food and feed and by Regulation on traceability and labelling of GMOs, was an administrative compromise reached without reference to plant biology.

The proposals for co-existence guidelines of GM crops must be assessed on a case-by-case basis. Different crops need different measures that correspond to their propensity for cross-pollination and other crop-specific characteristics. More importantly, co-existence measures must be specific to regional characteristics and farming practices.

Present status of co-existence in maize

Maize is widely grown in the EU for food, animal feed and industrial products. The question of ensuring co-existence between different production lines is not new to maize. Commodity yellow field maize (the predominant type, essentially grown for animal feed) coexists in European agriculture with several types of "speciality maize" such as sweet maize (the type sold for human consumption, as fresh produce and in cans) and waxy maize (a type grown for the starch industry).

Maize alone accounts for 29% of all the seed value in the world (12-15 bn US), being by far the main crop for the seed industry. The establishment of seed purity standards has obliged the industry to develop procedures to guarantee the maintenance of these standards. Operating procedures, which allow crops designated for different production chains to be grown in the same region, and deliver them to their respective processing stream with a high level of confidence, are routine in the agricultural industry.

In Spain, since 1998, 20-25000 ha of GM maize is cultivated by farmers annually in areas where conventional and organic maize are also produced. Under conditions given in this country (one GM variety grown on 10 to 13% of the maize acreage), co-existence between GM and non-GM products has been made possible. However, a zero-threshold is not achievable in all situations (two incidents have been reported) but these are still less than the 0.9% threshold. Prior to the publication of the Commission Guidelines for Co-existence, Denmark had already advanced guidelines for co-existence, which established an isolation distance of 200m and considered the seed purity and cleaning of machinery as the most critical factors in ensuring co-existence. The recommended separation distances between GM and non-GM crops given in the UK Supply Chain Initiative on Modified Agricultural Crops (SCIMAC) guidelines for growing GMHT crops are 200 m for sweet corn and 80 m for forage maize.

Scientific basis for co-existence

It is important to understand the scientific basis of the co-existence practices in maize in order to put the issue of GM gene flow into the wider context of how important GM gene flow is relative to other practices that affect seed purity, such as harvesting and storage practices. Maize is an open pollinating crop. Male and female flowers are separated on the plant and most of the varieties currently used display protandry (e.g. male flowering begins before female flowering). Pollen is spread from plant to plant between neighboring plants and by wind. Although bees and other insects may visit male tassels, they have a small role in pollination because female flowers are not attractive to pollinating insects. Most of the pollen released remains within several meters of the emitting plant, and the quantity of pollen dispersed diminishes with distance.

The following biological characteristics are major factors for crop-to-crop pollination. The flowering time of the pollen source and of the receiving population are important, and the degree of overlap of the respective flowering periods. The duration of pollen viability, which depends on environmental conditions, such as the humidity of the air and the temperature. The competition among pollen, which is influenced by the production of pollen in the receiving population and the pollen pressure generated by the pollen source and the relative sizes of emitting and receiving fields (in hybrid seed production the larger plant number corresponds to “female” parents that do not produce pollen on their own, and this makes them more vulnerable to pollen pressure from outside sources).

There are other important maize characteristics and practices that could influence maize co-existence. Although single seeds or ears can remain on the ground after harvesting and germination has been observed, volunteer plants are rare and are easily controlled by agricultural techniques or destroyed by frost. Out-crossing to wild relatives is not an issue for maize as no wild relatives (e.g. teosinte) are established in Europe. Finally, the farming systems themselves, for example fodder versus grain production (silage and grain maize), produce differences which can affect co-existence practices.

Several studies on co-existence between GM and non-GM crops, based on existing data, expertise and models, have been performed (INRA, 2001; JRC, 2002; Danish analysis, 2003). The study undertaken for the EU Commission's Joint Research Council (JRC) in 2002 on co-existence is probably the most detailed piece of research on the subject in the EU to date. A key point drawn from the work indicates that all farm types would be able to meet thresholds of 1% for maize, but that some farms would have to initiate changes and that co-operation with adjacent farms might also be required. The data and models have shown that pollen dispersal and out-crossing decline exponentially with distance but never have an end point so that very low levels (below threshold levels) could occur at long distances.

At the Brussels Round Table on Research in Co-existence of GM and Non-GM Crops, in April 2003, three points regarding maize crop were noted as still open. First, measurements and impacts of pollen flow at large distances and over fragmented landscapes. Second, models are a key tool in co-existence, however they have been largely based on small scale or single point sources of GM pollen. Field data obtained at a larger scale (either from trials or from commercial fields) would be useful to provide more evidence of model prediction and evaluation. Third, how should cross-border GM adventitious presence be addressed.

The oral presentations of Dr. Brunet on*"Evidence for long-range transport of viable maize pollen*; Dr. Meier-Bethke on "*Effect of varying distances and intervening maize fields on outcrossing rates of transgenic maize*" and "*Crop-to crop gene flow in Maize: a challenge to co-existence in England?"* of Dr. Weekes will provide relevant data on the co-existence issue.

Evidence for long-range transport of viable maize pollen

Y. Brunet[1], X. Foueillassar[2], A. Audran[2], D. Garrigou[1], S. Dayau[1] & L. Tardieu[2]
[1]INRA-Bioclimatologie, BP 81, 33883 Villenave d'Ornon Cedex, France
[2]ARVALIS-Institut du Végétal, 21 chemin de Pau, 64121 Montardon, France
E-mail: brunet@bordeaux.inra.fr

Abstract

The possibility for long-range transport of viable maize pollen was studied by sampling the atmospheric boundary layer up to 1800 m from a light aircraft, on several days during the pollination period in 2002 and 2003. Viable pollen was found every day at all heights, with a vertical variation in concentration typical of what could be expected in such climatic conditions. These results have profound implications on the co-existence of genetically modified crops with conventional crops in cropping systems.

Introduction

Most experiments on the dispersal of maize pollen have been performed using ground-based systems with pollen traps located at various distances downwind from a source. Such measurements can only provide information on short-range dispersal in the surface boundary layer (*i.e.*, within horizontal distances not exceeding a few hundred metres). However during the pollination period there is frequent convective activity in the atmospheric boundary layer (ABL, *i.e.*, the first 1-2 km of the atmosphere) that may result in mass transport to occur over much longer distances. In order to investigate whether this applies to maize pollen, we study here the presence and viability of pollen within the ABL.

Material and methods

A series of ten flights spanning six days was performed with a light aircraft (Cessna 180) over a 4000 ha set of maize fields embedded in the Landes pine forest (South-West France) in July 2002 and 2003, during the pollination period. Each flight consisted in 12 km long legs made at various altitudes between 150 m and 1800 m, i.e. within and above the ABL. A sampling device was built to collect pollen grains by impaction onto a set of 8 Petri dishes located around the main axis of a plastic tube set up under the wing and facing forward. A Pitot tube was used to monitor the air flow rate in the tube. Total and germinated pollen grains were counted under a microscope after each flight was completed. Corrections for non-isokinetic sampling and untrapped pollen were performed to estimate pollen concentration. Radio-soundings were made simultaneously to characterize the structure of the ABL.

Results and discussion

The radio-soundings performed each day reveal a range of atmospheric conditions from moderate to strong convective activity (ABL height from 800 to 2000 m, respectively). During each flight maize pollen grains were found at all heights, with average concentrations across the ABL ranging from 0.2 to 1.1 grains m^{-3}. Such concentrations are of the same order as those found near the ground at distances as short as 40 m downwind from a source (*e.g.*, Raynor *et al.*, 1972). On the days when several flights were performed the time variation of the vertical profiles in pollen concentration are as expected: as the boundary layer grows it gets progressively loaded with pollen. The average vertical variation in concentration is typical of the structure of a convective ABL (Figure 1): the pollen concentration decreases up to $z/h \approx 0.2$ (where z is the altitude and h the ABL height), then remains approximately constant throughout the mixed layer; above the top of the ABL it decreases again.

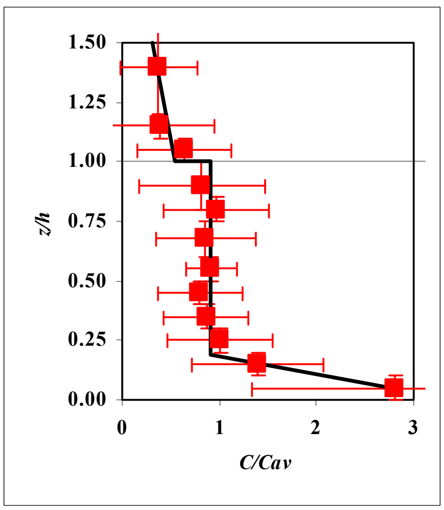

Figure 1. Vertical variation in pollen concentration *C*, normalised by the mean concentration found for each flight C_{av}, and averaged over all complete flights.

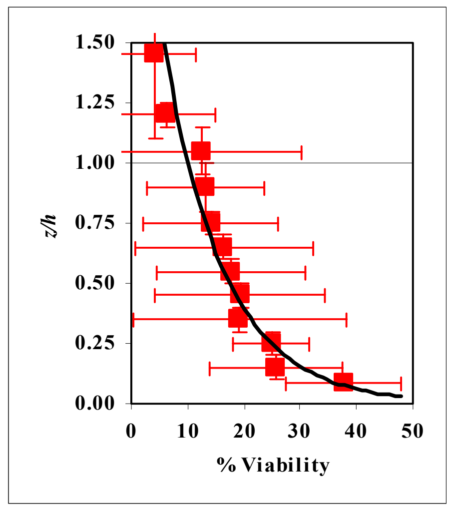

Figure 2. Vertical variation in viability, averaged over all complete flights.

Pollen viability (Figure 2) shows a smooth decrease from about 40-50% close to the ground to about 5-10% above the ABL. At the top of the ABL it is still significant (about 15%).

Perspectives

These results have profound implications on the possibility of long-range gene dissemination, because they show that in such climatic conditions as those encountered during the pollination period viable pollen grains can be transported over considerable distances (dozens of km) before they settle (which is likely to occur during the evening and night time). They should be confirmed by setting up sampling systems at the ground at both small and large distances from maize fields. We also plan to use a model of ABL flow, in order to estimate the dispersal pattern of pollen at a regional scale.

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Effect of varying distances and intervening maize fields on outcrossing rates of transgenic maize

S. Meier-Bethke & J. Schiemann
Federal Biological Research Centre for Agriculture and Forestry, Institute for Plant Virology, Microbiology and Biosafety, Messeweg 11-12, D-38104 Braunschweig, Germany
E-mail: s.meier-bethke@bba.de; j.schiemann@bba.de

Introduction

Within the current discussion of large scale, commercial cultivation of transgenic crops in Europe referring to the co-existence of GM and non-GM cultivation systems, the probability and extent of outcrossing of transgenic traits is one of the main topics. One of several measurements to reduce the extent of outcrossing in commercial cultivation of crops is the employment of mechanical barriers. We examined the effect of distance and mechanical barriers on outcrossing rates of transgenic maize (T25).

Materials and methods

In a field experiment in 2001, downwind of a glufosinate tolerant transgenic maize field four non-transgenic maize recipient fields were grown in parallel at varying distances (50 m, 75 m, 100 m and 120 m = first line). Two further parallel non-transgenic maize recipient fields were grown behind these fields in 50 m and 75 m distance (in 150 m and 200 m distance to the transgenic pollen donor maize field = second line). As a reference for outcrossing rates within an adjacent maize field, another transgenic pollen donor maize field and a non-transgenic maize recipient field were grown. Outcrossing rates were determined by an herbicide germination test with harvested kernels from the field trial in the greenhouse.

Outcrossing rates at sampling points at the field edges (first line) towards the transgenic maize pollen donor field were significantly higher than outcrossing rates at sampling points within the reference maize field, but were also decreasing with increasing distance. At the edges of non-transgenic maize recipient fields in the second line, outcrossing rates were higher than at the ends of the fields in first line, although the sampling points at the ends of the first line fields were more than 50 m or 75 m nearer to the transgenic pollen donor maize field. Within the non-transgenic maize recipient fields in the second line the decline of outcrossing rates was steeper than in the fields of the first line and within the reference field. Within the fields in first and second line outcrossing rates were detected on a very low level.

Results

This field study indicates that mechanical barrier zones will not prevent or reduce far field pollen dispersal and outcrossing, because far field pollen dispersal is mediated first through vertical transport due to upward air currents (Digiovanni F. & Kevan P.G., 1991). Viable maize pollen was found in 1800 m height (Brunet, Y. *et al.,* 2003). Pollen dispersal in higher air layers is not hindered by mechanical barriers near the ground. Pollen deposition rates were higher at mechanical barriers due to local eddies (Digiovanni F. & Kevan P.G., 1991). A maize field influences strongly the horizontal and vertical distribution of wind like a windbreak (Du M. *et al.,* 2001). Hence, outcrossing rates were higher at the field edges than within a maize field in comparable distance. Our findings indicate that a mechanical barrier zone will prevent or reduce outcrossing only effectively if it is established around the recipient field.

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Crop-to-crop gene flow in maize: a challenge to co-existence in England?

R. Weekes[1], C. Henry[1], D. Morgan[1], C. Boffey[2] & R. Daniels[2]
[1]Central Science Laboratory, Sand Hutton, York YO41 1LZ, United Kingdom
E-mail: r.weekes@csl.gov.uk
[2]Centre for Ecology and Hydrology, Winfrith Technology Centre, Dorchester
Dorset DT2 8ZD, United Kingdom

Abstract

Gene flow was monitored at the farm-scale evaluation (FSE) sites of fodder maize, genetically modified to be herbicide tolerant (GMHT) and released under the authority of the Genetically Modified Organisms (Deliberate Release) Regulations. Maize samples were collected from the conventional crop halves of a total of 55 FSE sites in England, (during 2000 to 2003) at a range of distances from the GM crop. Cross hybridization between GM and non-GM crops was detected and quantified using molecular methods. Additional data on wind direction and landscape were also collected for each trial site. Overall the results showed that rates of cross-pollination decreased with distance. Evidence of cross-pollination was found up to 650 m away from the GM crop. There was significant variation in levels of cross-pollination between sites in each year ($p < 0.01$), although the variation between years across all sites was not significant ($p > 0.05$). The importance of isolation distances in contributing to reducing adventitious pollen intrusion will be discussed with respect to sustainable co-existence of GM, conventional and organic crops.

Introduction

No GM crops are currently grown for commercial purposes in England. A decision on whether or not to lift the current moratorium on GM crops may be made during late 2003. The decision process is being supported by a series of public debates (King *et al*., 2003) along with the publication of results from the FSE study and reviews of the costs and benefits of GM crops (Strategy Unit of the Cabinet Office, 2003). The UK government has stressed its commitment to ensuring the co-existence of GM, organic and conventional farming should GM become commercialized (Defra, 2002).

Agriculture accounts for about 70% of land use in England and Wales and in 2001 1.1% of this land was used for growing maize (Environment Agency, 2003). Most of the maize is fodder and is grown and harvested for silage, although some sweetcorn is also grown. No maize is currently grown for seed production in the UK. The recommended separation distances between GM and non-GM crops given in the Supply Chain Initiative on Modified Agricultural Crops (SCIMAC) guidelines for growing GMHT crops are 200 m for sweetcorn and 80 m for forage maize (this distance was increased from 50 m in 2001). The current threshold value (for adventitious GM presence) for food and feed agreed by the EU Council is 0.9%.

The FSE study was initiated in 1999 to assess the effects of the agricultural management of field-scale releases of GMHT crops on farmland wildlife abundance and diversity compared with conventional (non-GM) crops. In conjunction with these trials, a study of gene flow from GM to conventional crops was also commissioned, using the FSE sites of GMHT winter and spring oilseed rape and GMHT fodder maize. This paper reports the results of the fodder maize trials.

Methods

The FSE sites were well distributed across England, covering 15 counties from Dorset to North Yorkshire and from Shropshire to Lincolnshire. Sites consisted of a split field design; half planted with Liberty Link™, line T25 (containing the *pat* gene, conferring tolerance to the herbicide glufosinate ammonium) and the other half with an equivalent conventional maize variety. Samples were taken from 55 sites and at each site samples were collected from 3 transects in the conventional crop, perpendicular to the GM: non-GM divide and evenly spaced across the field. Along each transect, cob samples were collected at the following distances: 2 m, 5, 10, 20 (or 25 m), 50 and 150 m away from the junction with the GM crop. At each site additional information on landscape features and wind direction during the flowering period was also collected. Maize grains from each sample were tested for GM content using real-time (TaqMan) polymerase chain reaction (PCR). The GM content was quantified by comparing the amount of *pat* gene to an endogenous control gene found in all maize cells to produce a GM: non-GM ratio. T25 maize seed kindly provided by Bayer CropScience was used as positive reference material.

Results and discussion

Results from individual fields in this study showed that the extent of gene flow is very variable between fields. High levels of gene flow could be linked to the prevailing wind direction (GM to conventional) during the overlapping flowering period, whereas low levels of gene flow were linked to large isolation distances and also to a lack of synchrony in the flowering times of the two crops.

Overall the level of gene flow decreased with distance. There was a rapid decline in the first 20 m from the GM crop and thereafter the rate of decrease was greatly reduced. A regression equation was used to describe the trend in the results and from this it was predicted that an isolation distance of 24.4 m would be required to meet the 0.9% threshold and that an isolation distance of 80 m would be sufficient to ensure that GM contamination was below 0.3%. The results also indicate that the 200m-separation distance (recommended by SCIMAC for sweetcorn and organic crops) would be sufficient although elimination of the 'edge effect' by removal of the first few GM-facing rows prior to harvest should be considered.

It is evident from this work that successful co-existence will be dependent not only on the threshold levels and isolation distances set down in the guidelines but also on factors such as regional cropping practices, environmental and landscape conditions and flowering synchrony. This study is unique both in the number and range of the trial sites and in the molecular approach to quantification of gene flow. It has not, however, addressed the problem of how multiple fields planted with GM crops would interact in a particular landscape. It is essential that our knowledge in this area be improved to support any future co-existence strategy in England.

Acknowledgements

We would like to thank Defra for funding this work (project EPG 1/5/138), also Bayer CropScience for kindly providing the positive control T25 maize seed and finally we would like to thank all those involved with sample collection and laboratory work at CEH, IACR, SCRI and CSL.

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Introduction to co-existence perspectives of GM wheat and cereals with conventional production

M.C. Chueca
Departamento de Protección Vegetal
Instituto Nacional de Investigación y Tecnología Agraria y Alimentaria. (INIA)
Spain
E-mail: chueca@inia.es

Wheat is the mean food grain consumed directly by humans and it is a staple for about 40% of the world's population (Eastham & Sweet, 2002). In the European Union, wheat is cultivated around large areas and some wheat fields will be grown adjacent to each other. In this circumstance gene flow is considered to be a major concern because of the potential risk associated to commercial release of transgenic plants.

Despite the fact that wheat may be described as a low risk genetically modified crop under normal conditions in crop-to-crop, crop-to-weed and crop-to-wild gene flow, biological and environmental conditions can bring variation the extend of gene flow. Gene flow may vary among species, population and individual plants, with the size of the pollen source and season (Ellstrand, 1992; Waines & Hegde, 2003).

Cross-pollination between wheat cultivars has been reported to occur in field at rates between 0.3 to 5.6% (Griffin, 1987; Martin, 1990; Hucl 1996). Notwithstanding that wheat pollen has short life of 20 to 30 minutes (de Vries, 1971) an hour in some conditions (Fritz & Lukaszewski, 1989), and that a minimum isolation of 10 meters is required for production of select seed, data of many authors show that outcrossing is possible at distances of up to more than 10 meters with a 2 x 2 meters pollen source (Loureiro *et al*., 2001), 27 meters (Hucl & Matus-Cadiz, 2001), and 80 meters with emasculated ears (Zhao H. *et al*., 2000).

Rice is another staple food for world's population. GM rice provide an important tool in red rice control, the main problem in rice culture, but herbicide tolerant rice present the risk of the escape of genes from engineered plants to other cultivars and to weeds. Despite the autogamous conditions of rice hybridization between rice and red rice is possible at distances of 1 and 5 meters and 10 meters is considered a distance of isolation (Messeguer *et al.,* 2001)

This variability leads us to determine that gene flow levels in wheat and rice may be sufficiently high that it will not be possible to guarantee 0% GM trait in non GM. Recently Waines and Hegde (2003) have published a review stating that scientific literature lacks sufficient rigorous and systematic gene flow studies in wheat. Rice and other cereals share the same problem.

This is the situation of knowledge in which GM wheat (and rice) is ready for commercial release in North America some years after the first GM crops were commercialized, with little resistance of the market to adopt these products. However, in recent years there has been a rejection of GM crops in many markets all over the world, mainly in EU were legislation mandates labeling rules enabling European consumers to identify GM content in their products and especially in their food, which is the principal destination of wheat.

Surveys by the Canadian Wheat Board indicate that 82% of Canadian wheat buyers would reject GM wheat. In America US Wheat Associates surveys of Asian and European buyers gathered similar responses (Anonymous, 2003a). The release of GM wheat has now been interrupted until five conditions are fulfilled, among them, segregation systems must be in place and there must be sufficient research into gene flow, weediness and management of weedy herbicide resistant wheat (Anonymous, 2003b). Gene flow data has dealt with crop-to-wild transfer, however, gene transfer from GM to non GM cultivars of the same crop may occur more frequently because of sexual compatibility and physical proximity. Therefore, prior to releasing GM crops it is of the outmost importance to establish standards for tolerance levels of GM by nonGM crops for both conventionally and organically produced crops.

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Pollen dispersal and potential hybridization between wheat cultivars

I. Loureiro, M.C. Escorial, J.M. García-Baudín & M.C. Chueca
Dpto. Protección Vegetal. INIA. Ctra. La Coruña Km 7.5, 28040 Madrid, Spain
E-mail: chueca@inia.es

Abstract

Risk assessment is now recognized as being an important step towards the release and application of novel transgenic crops. In assessing the value of transgenic wheat gene dispersal data it is important to know whether there has been the opportunity for cross-pollination and what factors have influenced the results. How far wheat pollen can travel is important for assessing the risks of future genetic pollution, with attendant safety and economic implications.

Introduction

Wheat is the world's most important food crop species, grown on an area of over 200 million hectares and now yielding almost 600 million tons annually (FAO, 2002). Wheat is predominantly a self-pollinating crop that has variable outcrossing rates depending on populations, genotypes and different environmental conditions (Jain, 1975).

Genetically modified (GM) wheat lines with herbicide or disease resistance, improved yield or quality have been or will be developed. Numerous field trials have already been conducted. Transgenic herbicide-resistant wheat varieties are being developed and field-tested and probably in the next few years certified cultivars of transgenic wheat will be commercially available. Many of the GM wheat cultivars will be grown next to non-GM wheat, thus, how far wheat pollen can travel is important for assessing the risks of future genetic pollution by GM crops to non-GM crops, with attendant safety and economic implications. It seems practical to develop a set of isolation distances based on acceptable levels of trait presence in cultivars and the amount of gene flow risk involved

Data on potential hybridization events are useful in the development of crop management practices, as desired to maintain crop purity for both GM and non GM-cultivars. In wheat, several studies on the effect of pollinator distance on seed set have been conducted in the field using male-sterile bread wheat (*Triticum aestivum* L., AABBDD, hexaploid) with great variations in the outcrossing rates estimated among studies, due to weather conditions at blooming time, day and night temperatures, light, moisture and wind direction and speed, that have a large influence on gene flow in male-sterile wheat lines (Khan *et al.*, 1973). The use of sterile receptor plants is useful in order to measure the maximum pollen dispersal and hybridization rates. Seed set percentages ranged from 14-73% around the pollen source, 17-70% at 1.5 m, 8-54% at 3 m, 9-33% at 12 m distance (see the review of Waines, 2003)

Bread wheat can also coexist in the field with the second major cultivated wheat species, the durum wheat (*Triticum turgidum* var *durum,* tetraploid, AABB) usually grown for bread, beer and pasta. Crossability between *T. aestivum* and *T. turgidum* has been studied in view of the fact that this specie is a source of genetic variability for wheat improvement. There are no data on the literature about the variation of the hybridization rate with the distance in the field between *T. aestivum* and *T. turgidum*, and information is necessary for the risk assessment study in the case of co-existence between these species.

Materials and methods

Pollen flow and hybridization rates between two *T. aestivum* cultivars and between *T. aestivum* and *T. turgidum* have been estimated studying the pollen dispersal in a field plot assay during three years, using emasculated plants were used as pollen traps in order to establish the maximum potential for hybridization.

Results and discussion

The average hybridization rate between *T. aestivum* cultivars average a 63, 20 and 7% at inside the pollen source, 1 and 3 m respectively. At the distance of 14 m a 0.65% hybridization rate was obtained. Hybrid progeny was also obtained with *T. turgidum* as recipient plants at a rate lower than *T. aestivum*. Data show that environmental conditions, wind, temperature and humidity, which varied from year to year during the crossing periods bear a great influence on the pollen dispersal and the hybridization rates. Humid weather makes the pollen heavy, so pollen may not disperse far from the parent plant, whereas dry weather causes desiccation and loss of viability, reducing the chance of effective gene flow. Extreme cold or hot temperatures are also unfavorable for pollination and fertilization.

Given the importance of the wheat crop grown world wide, it is necessary to develop more studies about of the potential risk of hybridization and pollen dissemination from transgenic wheat to other cultivars and wild relatives under various environmental conditions.

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A field study of pollen-mediated gene flow from Mediterranean GM rice to conventional rice and the red rice weed

J. Messeguer[1], V. Marfà[1], M.M. Català[2], E. Guiderdoni[3] & E. Melé[1]
[1] Departament de Genètica Vegetal. Centre de Cabrils. IRTA. Carretera de Cabrils s/n. Cabrils, 08348, Barcelona, Spain
E-mail: joaquima.messeguer@irta.es
[2] Estació Experimental del Delta de l'Ebre. Carretera de Balada s/n. Amposta 43870 Tarragona, Spain
E-mail: mar.catala@irta.es
[3] Biotrop program, Cirad-Amis, TA40/03, F-34398 Montpellier Cedex 5, France
E-mail: guiderdoni@cirad.fr

Abstract

The objective of this study was to assess the frequency of pollen-mediated gene flow from a transgenic rice line, harbouring the *gusA* and the *bar* genes, to the red rice weed and conventional rice in the Spanish *japonica* cultivar Senia. A circular field trial design was set up to investigate the influence of the wind on the frequency of pollination of red rice and conventional rice recipient plants with the transgenic pollen. Frequencies of gene flow based on detection of herbicide resistant, GUS positive seedlings among seed progenies of recipient plants averaged over all wind directions were 0.036 ± 0.006% and 0.086 ± 0.007 for red rice and conventional rice respectively. However, for both red rice and conventional rice, a clear asymmetric distribution was observed with pollination frequency favoured in plants placed under the local prevailing winds. Gene flow detected in conventional planted at 1, 2, 5 and 10 m distance revealed a clear decrease with increasing distance which was less dramatic under the prevailing wind direction.

Introduction

Cultivated rice (*Oryza sativa* L.) is often associated with various weedy forms that are genetically related. Recently, new forms of weedy rice - often called red rice due to the frequently occurring red coloration of the seed pericarp - have emerged in direct seeded, irrigated paddy fields of temperate rice growing areas where related wild species are not found. Red rice strains usually exhibit many traits related to weediness, which facilitate seed dispersal and persistence in paddy fields. Given the ecological importance of the rice crop (grown on 147 million hectares world wide (FAO, 2003)), it is important to determine the extent of pollen dissemination from transgenic rice wild relatives and also to other cultivars and in various environmental conditions. The objective of the present study is to use a circular design to assess the frequency of gene flow in field conditions from transgenic plants to recipient red rice and non-transgenic plants of the Mediterranean variety Senia. The influence of the distance and of the wind on pollen-mediated gene flow will be also studied.

Material and methods

Transgenic homozygous seeds (containing the *gus*A and *bar* genes) non-transgenic seeds of Senia and red rice seeds were transplanted in the paddy field when they reached the 4-5 leaf stage. A circular design was used: seedlings were transplanted to the field in concentric circles. The inner circle (3 m diameter) was planted with non-transgenic plants and surrounded by one circular row of red rice plants, in turn surrounded by two successive circular rows of transgenic plants, a circular row of red rice plants and then a last circular row of non-transgenic plants. To evaluate the influence of the distance on gene flow frequency, 8 plots of 8 non-transgenic plants were planted at 1 m, 2 m 5 m and 10 m distances from the rings of transgenic plants, following the 8 directions of the compass card. Panicles from all plants were manually and individually harvested, and their location in relation to the geographic orientation was recorded in the field. Wind speed and direction were measured during the heading-flowering period. Samples of seeds harvested from each non-transgenic Senia and red rice plants were sown in greenhouse conditions and seedlings at the 3-4-leaf stage were treated with commercial herbicide. After 3-4 weeks, all surviving seedlings were transferred to individual pots for GUS histochemical assay, and progeny analysis. A total of about 165,000 seedlings harvested from non-transgenic Senia plants and 125,000 seedlings harvested from red rice plants were processed.

Results

Only 41 seedlings among all the seeds analysed from red rice recipient plants clearly survived to the herbicide treatment and were positive for the GUS assay with Mendelenian segregation of their progeny, demonstrating that all individuals resulted from pollination by the transgenic pollen. Average gene flow obtained considering all the wind directions was 0.036 ± 0.006% but a clear asymmetric distribution of the results was observed analysing separately the inner and the outer circles demonstrating the influence of the prevailing wind in cross-pollination process. In the case of Senia progeny, 52 seedlings (from a total of about 60,000 analysed) were identified among progenies of recipient Senia plants placed in concentric circles contiguous to red rice. Values of the gene flow obtained, grouping the results by main compass card directions, were averaged (0.086 ± 0.007) to estimate the total gene flow obtained in plants at a 50 cm distance from the transgenic Senia ring. In the discontinuous outer Senia circles, only 13 resistant seedlings were recovered from 88,000 plantlets tested. Most (11) of them were from plants grown in the NW position (5, 4, 1 and 1 at a 1 m, 2 m, 5 m and 10 m distance, respectively).

Discussion

The influence of the wind is clear as the transgenic seedlings found in the two red rice circles have a complementary pattern. N, NW and W directions have the higher values in the outer circle and the lowest in the inner one. The opposite pattern can be found in the S, SE and E direction. The null values encountered in the inner circle at N, NW and W direction have special significance in this assay as the red rice plants in this area could have received the pollen from the transgenic plants of the opposite part of the circle, situated at 6 m distance. This fact suggests that gene flow decreases quickly with the distance. Results obtained here suggest that within a commercial transgenic rice field, the influence of the wind appears a less determinant factor than in our circular design, because red rice plants usually will grow isolated or in patches surrounded by transgenic plants and consequently can be pollinated by all of them. On the other hand, the wind influence on cross-pollination has to be taken into account for the plants growing in the borders. This is a very essential question to consider because the real introgression of the genes will be minimized inside the field by the usual control practices tending to destroy the red rice, but the wild plants in the borders can act as reservoirs of the transgenic characters. Moreover, although the gene flow values are relatively low, the shattering and dormancy of the red rice seeds, which ensure their persistence in the field, lead into an undesirable effect of durability of the transferred genes. Therefore, whether one wants to avoid gene flow to the red rice, crop management has to be considered. In the case of gene flow from transgenic to non-transgenic Senia plants, the wind effect is also very clear producing this asymmetric distribution that matches perfectly the wind data recorded by the field sensors. This strong dependence of the wind suggests that security isolation distances can be totally different according the compass card direction and a correct decision-making will require preliminary, precise meteorological study of the wind at the blooming period.

Oil seed rape and beets

Gene flow in rapeseed: a major concern for co-existence

F. Angevin[1] & A. Messéan[1,2]
[1] INRA, Unité Eco-innov, BP 01, F-78850 Thiverval-Grignon, France
E-mail: angevin@jouy.inra.fr
[2] CETIOM, BP 04, F- 78850 Thiverval-Grignon, France
E-mail: messean@grignon.inra.fr

Although gene flow is a common phenomenon for crop species, its implications for Genetically Modified Plants have raised new concerns. Undesirable effects related to gene flow in rapeseed have ecological or agronomic consequences (persistence of resistant volunteers; creation of new weeds; multiple resistance) and commercial consequences (unintended presence of GMOs in conventional rapeseed production would affect its market value).

Gene flow occurs through pollen flow as well as through seed dispersal along the field-to-food chain. Seeds are dispersed within the field at harvesting time, during transportation and storage. Admixture also occurs in the downstream chain (crushing, etc) as well as in the upstream chain (seed production). Several factors affect gene flow along the on-farm routes: crop biology, environmental conditions, crop management and post-harvesting practices.

Adventitious presence of GM seeds in non-GM rapeseed harvest could have several causes:

- Crop-to-crop pollination between neighbouring fields;
- Presence of volunteers in conventional fields resulting from former GM rapeseed cultivation in the field;
- Pollination from feral GM rapeseed plants occurring in field borders and resulting from seed dispersal during transportation;
- GM impurities in seed lots (cross-pollination during seed production or admixture in the post-harvest process).

Results on these topics are now available. In this session, we will focus on results concerning crop to crop pollen flow, as well as seed persistence and dispersal – especially volunteers in subsequent crops and feral population in road edges – which have a major impact on adventitious presence and highly depends on the landscape and on farmer practices.

However, the data and models have shown that pollen dispersal and out-crossing decline exponentially with distance but never have an end point so that very low levels can occur at long distances. Research is needed on long distance gene flow. As we will see it in the “modelling “ session, models can then help in designing proper protocols for such experiments.

From the available results, it can be stressed that herbicide tolerant rapeseed cannot be cultivated without applying specific guidelines for crop management. Those crop management guidelines should achieve three main objectives:

1) the development or extension of practices aiming at reducing, in time and space, the persistence of undesirable plants (volunteers and hybrids with wild relatives);
2) the avoidance of selection pressure on these undesirable plants (a major issue for herbicide tolerance);
3) as gene flow occurs both in time and in space, its impacts highly depend on farming practices and regional variability and co-existence rules should be designed on a local and dynamic basis.

Mitigation measures have been defined and their relevance has been assessed through experiments (Farm scale evaluation in the United Kingdom, Inter-institutes platforms in France and through models simulations (see interventions in the “modelling session”). They should now be assessed in terms of feasibility and acceptability by the stakeholders.

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Gene flow from transplastomic oilseed rape into *Brassica rapa*

M.J. Johannessen & R.B. Jørgensen
Plant Research Department, Risø National Laboratory, DK-4000 Roskilde, Denmark
E-mail: marina.johannessen@risoe.dk; rikke.bagger.jorgensen@risoe.dk

Abstract

The hybrid production in a winter variety of oilseed rape co-cultivated with *B. rapa* was estimated. Under the assumption that plastids are inherited uniparental maternally, hybrid production on oilseed rape is the only transgene escape route when cultivating transplastomic oilseed rape. Therefore transplastomic lines are assumed to reduce admixtures of transgenes in the harvest from non-GM fields. Field trials were made with two different proportions between the species and three different plant densities. The paternity of 3000 progenies was assessed. The relative production of hybrids was lowest when *B. napus* was cultivated at intermediate to high densities and the abundance of *B. rapa* in the field area was kept low. The numerical number of hybrids produced per mother plant was lowest at intermediate density and still at a low abundance of *B. rapa*.

Introduction

Reducing the spread of crop-transgenes, from crop-species who coexist with wild and weedy pollen recipients, is of course desirable. In most angiosperms inheritance of plastids are mainly uniparental maternal, thus transformation of plastids is one approach of transgene biocontainment. But the containment is not complete in that hybrid production still occurs if the wild and/or weedy species sire the crop-species.

We have estimated the hybrid production on a winter variety of oilseed rape, co-cultivated with *B. rapa*.

Materials and methods

The oilseed rape variety 'Capitol' (winter type) and *B. rapa* (wild) were co-cultivated in the field in two different proportions (3:1 and 1:1 (*B. napus*: *B. rapa*)) and three different densities (16 plants/m^2, 44,5 plants/m^2 and 100 plants/m^2). Fifty progenies from each of ten mother-plants from each of the six plot-types, giving 3000 individuals, were screened to assess their paternity. *B. rapa* specific molecular markers, obtained from the PCR-based fingerprinting techniques Inter-SSR and SSR, were used to identify the hybrids.

Results and discussion

At low plant densities the relative production of hybrids on average was the same irrespective of the abundance of *B. rapa* in the population, thus the contribution of *B. rapa* pollen to the pollen-cloud seemed to be the same. On average the relative production of hybrids at intermediate and high densities was, not surprisingly, lower when the abundance of *B. rapa* was lowest. Thus at these densities a lower abundance of *B. rapa* probably was reflected in the pollen-cloud contribution from *B. rapa*. When *B. napus* and *B. rapa* were equally represented the relative production of hybrids on average was the same at all densities. Thus a plant-density-effect did not exist at this proportion. When *B. napus* was three-times more abundant than *B. rapa* the relative production of hybrids on average was lower at intermediate and high densities than at low density. So a density-effect existed at this proportion, probably caused by the reduction of resources for *B. rapa*, causing smaller plants with less pollen production. Therefore these results are in consistence with immediate expectations, namely, that if *B. napus* is cultivated at intermediate to high densities and the abundance of *B. rapa* in the field area is kept low, the relative production of hybrids will be lowest.

The relative production of hybrids per mother plant was converted into the actual production, since the numerical number of hybrids produced in the population is highly dependent on the amount of the seed set on the mother-plants. At the high plant density the production of hybrids was generally low, and at low density the production was in general comparatively higher. Thus the hybrid production was apparently independent of the species composition (3:1 and 1:1) at these specific densities. The hybrid production at intermediate density would be expected to be intermediate, and this was true at the 1:1 proportion. But at the 3:1 proportion the hybrid production was the lowest observed over all. One explanation may be that exactly this combination of density and plant-type composition favours oilseed rape, and intensifies the competition against *B. rapa*, resulting in smaller *B. rapa* plants with less pollen production.

Perspectives

We investigated the hybrid production. The next step in the introgression process of transgenes from *B. napus* to *B. rapa* depends on the fitness of the hybrids, plus the frequency whereby it is pollinated by *B. rapa* again and self-pollinated. Therefore we have made field trials with F_1-hybrids coexisting with *B. napus* and *B. rapa* in different proportions and the same densities as in the above mentioned experiment. We are currently making the paternity analysis, and the results will provide further knowledge about gene dispersal from transplastomic lines of oilseed rape to the environment.

Pollen and seed dispersal during the large scale cultivation of transgenic oilseed rape

A. Dietz-Pfeilstetter & P. Zwerger
Federal Biological Research Centre for Agriculture and Forestry
Messeweg 11-12, D-38104 Braunschweig, Germany
E-mail: a.dietz@bba.de

Introduction

Pollen-mediated gene flow between different oilseed rape varieties is influenced by flowering times, environmental conditions, field sizes and the distance between fields. Few data have been available on the consequences of outcrossing during agricultural cultivation. Transfer of an herbicide resistance transgene, for instance, not only causes the adventitious presence of transgenic seeds in conventional seed products, but can also give rise to volunteer plants with an unexpected herbicide resistance pattern. Outcrossing of herbicide resistance genes was shown to be the cause of multiple-resistant *Brassica napus* volunteers in Canada (Hall *et al.*, 2000).

Materials and methods

In the years 1999/2000 and 2000/2001, we carried out a large scale field release experiment with Glufosinate resistant (LibertyLink®, LL) and Glyphosate resistant (RoundupReady®, RR) oilseed rape. Each of the 4 square transgenic plots was about 0.5 ha in size, surrounded by 8 ha non-transgenic oilseed rape. The different transgenic plots were either in direct contact with each other and with the non-transgenic field, or they were separated by 10 m fallow. In order to determine the outcrossing frequencies of the herbicide resistance genes to neighbouring fields, seed samples were collected in the transgenic plots and in the surrounding non-transgenic field at different distances (Figure 1). The samples were screened for resistant seedlings using herbicide germination tests and PCR. For the detection of volunteers with an herbicide resistance gene originating from outcrossing, oilseed rape plants emerging after harvest were treated with one of the complementary herbicides (see Figure 1), followed by PCR analysis of individual resistant plants.

Results and discussion

In both years it was found, that an isolation distance of 10 m reduced the outcrossing frequencies to plants at the inner border of the surrounding non-transgenic field. However, at greater distances, the isolation distance had no influence on the rate of transgenic seeds in samples from the non-transgenic field. Outcrossing was found to decrease gradually within the field, and at 50 m it was well below 0.1%. The effect of a greater isolation distance was less pronounced for the outcrossing between the neighbouring transgenic plots, which may be due to a greater influence of competing pollen from the surrounding non-transgenic field. Double resistant volunteers could be detected in each of the transgenic plots, but were mostly limited to the inner field borders. Herbicide resistant volunteers were mechanically destroyed during the shallow soil cultivation preceding the sowing of the next crop. No double resistant volunteers were detected in the following years. However, single herbicide resistant volunteers were found in the second crop of the rotation of the 1999/2000 field experiment (spring barley) at locations of the LL and RR plots, respectively. Therefore we conclude, that shallow soil cultivation after the harvest of oilseed rape does not always guarantee, that all the seeds from seed losses germinate and emerge in the harvest year.

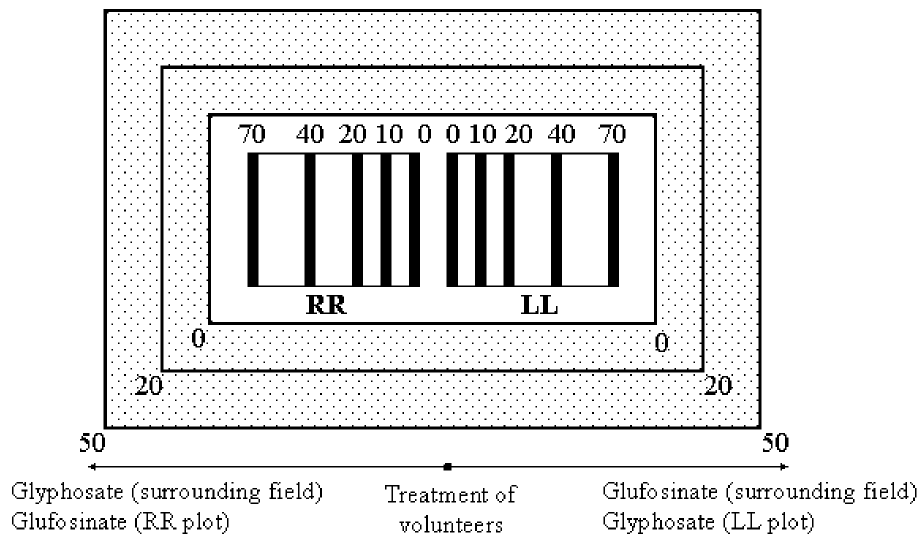

Figure 1. Outline of one half of the field experiment, showing the location of sampling strips (distance in meters). The scheme for the selection of single and of double resistant volunteers in the surrounding non-transgenic field (dotted) and in the transgenic RR and LL plots, respectively, is shown below.

Gene flow from crop fields can also be mediated by seeds, as was demonstrated in a recent report (Arnaud *et al.*, 2003). The experimental design of our field release experiment enabled us to detect not only outcrossing events, but also to estimate the unintentional spreading of seeds through harvesting machines. Despite extensive cleaning of the combine between the different transgenic plots, in the first harvest year about 16% of the seeds in the sample taken immediately after changing from the LL plot to the RR plot (outer sampling strip) were only Glufosinate resistant and turned out to be carry-overs from the LL plot. At the end of the 70 m long outer sampling strip the frequency of seeds carried over from the LL plot was reduced to

0.1%. For comparison, the average rate of double resistant seeds resulting from outcrossing was well below 0.1% in this sampling strip, which was located at 80 m distance from the LL plot.

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Origin and dynamics of feral oilseed rape populations

A. Deville[1], A. Garnier[1], J. Lecomte[1], K. Adamczyk[2], S. Huet[2], A. Merrien[3] & A. Messéan[4]
[1]UMR 8079-CNRS, Ecologie, Systématique et Evolution, Université Paris XI-Centre d'Orsay, F - 91405 Orsay Cedex, France
E-mail: alexandra.deville@ese.u-psud.fr
[2]UR341 Biométrie et intelligence artificielle, INRA Domaine de Vilvert, F - 78352 Jouy-en-Josas Cedex, France
[3]CETIOM, Laboratoire d'analyses d'Ardon, BP 90635, 45166 Olivet Cedex, France
[4]CETIOM, Centre de Grignon, BP 4, F – 78850 Thiverval-Grignon, France

Abstract

We developed concurrently a study of oilseed rape (OSR) populations in a production basin and a stage structured model of population dynamics to assess the potentialities of gene flow in an agricultural landscape from feral populations of OSR and to determine an optimal management of these populations. These studies show that feral populations of oilseed rape can persist several years in spite of road verges management and confirm that such populations can be a source of (trans)genes dispersal through space and time.

Introduction

Within the framework of the introduction of Genetically Modified Oilseed Rape (GM-OSR) into the environment, the OSR populations localized in field margins (i. e. feral populations) are likely to rise problems of management such as (1) a source of corridors for genetic pollution between fields, (2) a risk of increasing weediness in the case of herbicide tolerant OSR and (3) an irreversible dissemination of a GM-variety if its cultivation is stopped. The estimation of the risks associated with the persistence and the spread of a transgene through space and time is conditioned by a good knowledge of the origin and the dynamics of these populations. Our preliminary study on an agricultural landscape reveals a persistence of OSR at least 8 years after their setting in culture (Pessel *et al., 2001*) via secondary dormancy, local recruitment and/or farm seeds. In the continuity of this study, we have developed a survey at a landscape level and a model of dynamics of feral populations of OSR.

Material and methods

Field surveys at landscape level

This study aims to assess the respective importance of the different events of foundation of new feral populations (seed losses during harvest or sowing transportation or local colonization from neighboring fields) and to estimate the impact of the seed bank and the self-recruitment on their persistence. Since 1999, every feral and cultivated populations of winter

OSR are geographically localized with inframetric precision (GPS measures) in the Selommes area (Loir-et-Cher, France), that is an agricultural landscape of approximately 100km^2 surrounding a grain silo. In addition, surveys provide data concerning crop rotations, management of the field edges and ways followed by seed trucks from fields to crops. All this information is gathered in a database which enables us to study the origin of the feral OSR populations and their possibilities of persistence in a year and between years.

Feral populations dynamics model
The stage-structured model of the local dynamics of a feral population takes into account road side management (cuttings and chemical treatments), external seed flows (from crops or from seed transport), density-dependence and stochastic factors. As seeds can develop a secondary dormancy if buried (Pekrun & Lutman, 1998), a seed bank is also included in the model.

Results

Field studies show that feral populations are not only funded by fields neighbouring previous years and suggest there is a strong influence of seed losses during transportation. Their maintenance lays on seed recruitment and on the presence of a seed bank. Moreover, the amount of feral populations found in the studied area and their flowering period indicate that there is a high ability of exchanges of (trans)genes with OSR crops.

The model underlines the capacity of persistence of feral populations during at least 10 years, even with management by cutting or by herbicide treatment. Mean extinctiontime and elasticity analyses show the pronounced effects of management parameters, seed bank and seed flows on the dynamics and the persistence of the feral populations.

Perspectives

Projections of several scenarios of treatments and seed flows associated to an increased knowledge of the origin and of the dynamics of feral populations will provide a useful tool for the management of feral populations to limit gene escape.

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Understanding and predicting landscape-scale gene flow in oilseed rape

G. Ramsay, G.R. Squire, C.E. Thompson, D. Cullen, J.N. Anderson & S.C. Gordon
Scottish Crop Research Institute, Invergowrie, Dundee DD2 5DA
United Kingdom
E-mail: g.ramsay@scri.sari.ac.uk

Abstract

A study of the patterns of cross-pollination on the landscape-scale in oilseed rape has, using male-sterile plants, revealed that the decline in cross-pollination with distance is initially rapid but falls to a long tail, which persists to all distances tested, including 26 km from the nearest known source. The majority of the cross-pollination was shown to be due to insect activity rather than wind-borne pollen. Experiments were conducted to determine the likely levels of cross-pollination to similar small groups of male-fertile plants and to larger plots of male-fertile plots. Predictions from the data have been made to indicate possible cross-pollination rates across wide areas of landscape to recipient populations of different types.

Introduction

Determining means of achieving co-existence of GM and conventional cropping depends on an understanding of the various routes by which impurity can arise in a farmer's harvested crop. Cross-pollination is particularly contentious, given that other farms in the area may be affected. The patterns of decline across landscapes are crucial to prediction of the conditions required to meet certain thresholds, and this paper will present a synthesis of work designed to assist such prediction.

Materials and methods

Groups of 10 male-sterile plants of oilseed rape were placed around a typical arable landscape in E Scotland to determine the patterns of the decline in cross-pollination with distance. Results are presented as the proportion of seed set expected from the receptive ovules present during the 14-day periods of the experiments. In one experiment, a non-GM herbicide-tolerant field was used as a defined source so that comparisons of rates of gene flow into 10-plant groups of male-sterile and male-fertile plants and plots of 10 x 10m and 30 x 30m male-fertile rape could be compared. Fuller details of these experiments, the molecular verification of progeny, and experiments on pollen vectors are described in Ramsay *et al.* (2003).

Results and discussion

In three seasons rates of cross-pollination to male-sterile plants were found to be very variable between sites (Figure 1). In each season there was a high variability within each distance class (e.g. 500-2000 m from the nearest field, a mean of 11.1% ovules fertilised with 95% confidence limits 8.1 to 14.7%). In the second season, an experimental period dominated by poor weather, the values dropped very rapidly with increasing distance from the source, yet verified pollination events were still found 5 and 26 km from the nearest source. In one experiment using a non-GM herbicide tolerant source field of 7 ha, cross-pollination to male-steriles, male-fertiles and larger blocks of male-fertiles were compared. Estimates of the reduction in gene flow when moving to male-fertile recipients and to larger blocks of recipient were approximately one order of magnitude in each case. A combination of insect-exclusion cages and continuous monitoring of wind throws at the source field has confirmed that cross-pollination was largely due to insect activity. Over ranges of several km bees are likely to be the main vectors but the furthest site was beyond bee foraging range.

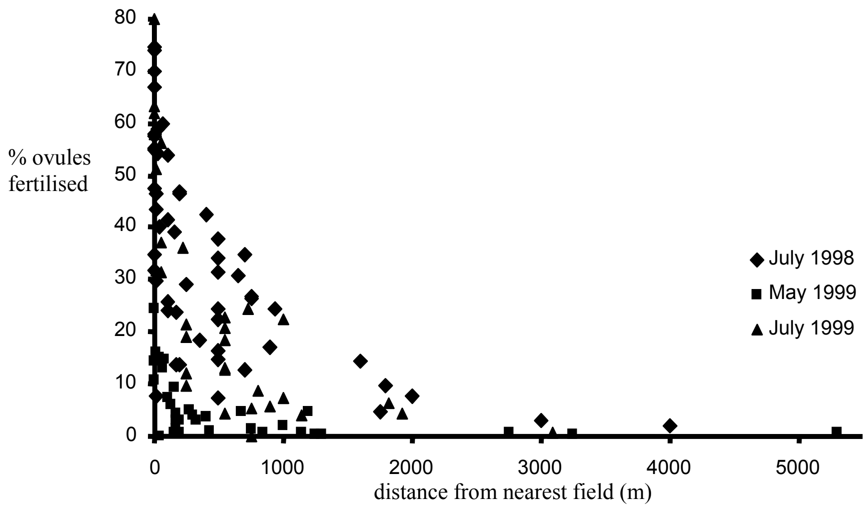

Figure 1. Cross-pollination (as the percentage of ovules fertilised) of male-sterile oilseed rape over three seasons. One result at 26 km (0.15%) not shown for clarity.

The data generated here have been used produce charts designed to aid the prediction of gene flow in different situations (Figure 2, and further detail in Ramsay *at al.* 2003). This synthesis will enable predictions to be made of the situations when the 0.9% GM threshold and other possibly more stringent thresholds may be breached.

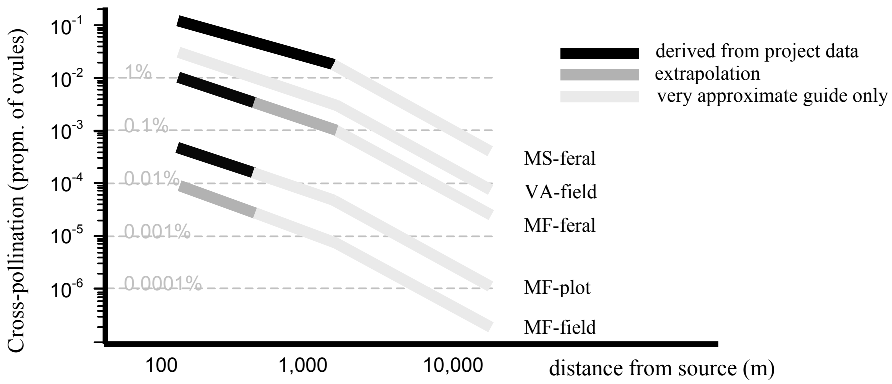

Figure 2. Predicted cross-pollination where about 25% of the OSR field area donates pollen of interest. (VA – varietal association types).

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Introduction to field trial data of crop to weed beet gene flow

D. Bartsch[1], U. Wehres[2], U. Göddecke[2] & A. Gathmann[2]
[1]Center for Genetechnology, Robert Koch Institut, Wollankstrasse 15-17, D-13187 Berlin, Germany
E-mail: bartschd@rki.de
[2]Chair of Biology V (Ecology, Ecotoxicology, Ecochemistry), RWTH Aachen, D-52056 Aachen, Germany

Abstract

Pollen-mediated gene flow is hard to control in wind-pollinated plants like beet (*Beta vulgaris*). Unintended products of cultivated beets pollinated by wild beets are weed beets that bolt and flower during their first year of planting. Weed beets cause yield losses, delay harvest, and may lead to adventitious presence of genetically modified (GM) weed beets in non-GM sugar beet crop. Field data suggest that crop-to-weed gene flow is of minor implication for co-existence questions between GM and non-GM sugar beet cultivation. The key elements for minimizing gene escape are 1) isolation measures in seed production areas to prevent wild-to-crop and crop-to-crop gene flow, and 2) bolter control in crop production areas to prevent the establishment of GM weed beet reservoirs.

Introduction

Beta vulgaris is a good example for a wild/cultivar complex, in which gene flow and hybridization can be observed (Mücher *et al.,* 2000**)**. Besides cultivated beet, a weed beet form is also known to grow in sugar beet fields, where, due to its genetically based first year flowering, it bolts late in the season. Producing lignescent inflorescence and large amounts of seed instead of commercially desirable fleshy beet roots, weed beets can be a serious problem for sugar beet farmers (Soukup *et al*., 2002; Viard *et al*., 2002). Due to the similarity of their seedling morphology and physiology to sugar beet, conventional methods do not control flowering weed beets. Gene flow from GM sugar beet to wild and weedy relatives is very likely to occur. Hybrids formed by spontaneous crosses between cultivated beets and south European wild types are typically flowering in their first year of growth, since this life cycle trait is genetically dominant and common in southern wild populations.

Field data on beet gene flow

In Germany, a research program was carried out addressing gene flow consequences of GM sugar beet to wild and weed beet. The GM beet studied had a single copy of the beet necrotic yellow vein virus (BNYVV) coat protein (*cp*), and neomycin-phospho-transferase (*nptII*) genes. We found that the virus-tolerant phenotype of hybrids with wild sea beet were not significantly different to naturally virus-tolerant genotypes, but isolation measures in seed production areas are crucial for limiting crop-to-wild gene flow (Bartsch *et al*., 2003). In one field study carried out in 2001, we measured gene flow from a source plot [656 hemizygous GM weed beets planted on 800 m²] into non-GM beet bolters found naturally in beet crop field within 500 m radius of the source field (Bartsch, unpublished data). We harvested all beet bolter mother plants from an area of 3.6 ha sugar beet fields, and transferred seeds into the greenhouse for GM offspring testing. In total, 31 mother plants gave 1068 beet seedlings, of which 7 were tested positive for GM (= 0.7% hybridization rate). Similar rates (0.8%) were reported by Vigouroux *et al*. (1999) for a French field trial. These data can be used for calculating the adventitious presence of GM beet biomass within the next crop rotation. On a per plant basis, 7 GM weed beets on 3.6 ha will contribute to 0.002% GM biomass proportion, if 80,000 non-GM beets are planted per ha in the next sugar beet rotation. But this low number will increase dramatically, if no bolter control measures are taken. We calculate, that in the worst case population increase could lead to 70,000 GM weed beet on 3.6 ha within 12 years (in a three year beet crop rotation, if the finite rate of population increase is 10). Therefore, bolter control is the key element for minimizing gene flow and adventitious GM presence in beet crop production areas.

Conclusion

The beet example is a well documented case how unwanted economic effects arise by uncontrolled gene flow in conventional breeding (Soukup *et al.,* 2002). The primary question related to GM crops is whether the GM will get fixed in recipient plant populations (Desplanque *et al*., 2002; Arnaud *et al*., 2003). The secondary more ecologically and economically important question is whether the GM cause effects that are fundamentally different from conventional gene flow consequences.

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Modelling the influence of cropping systems on gene flow from herbicide resistant sugar beet. Adapting the genesys model to sugar beet

M. Sester, N. Colbach & H. Darmency
UMR Biologie et Gestion des Adventices, INRA, 17 rue Sully, BP 86510, 21065 Dijon Cedex France
E-mail: colbach@dijon.inra.fr

Abstract

A model quantifying the effect of cropping systems on gene escape from sugar beet to weed beet was developed. Field trials were carried out to estimate the parameter values of the model. The model will be used to develop new cropping systems limiting gene flow.

Introduction

The transfer of genes from sugar beet to weed beet, which could lead to the apparition of a more persistent weed, is probably the greatest risk of planting genetically modified beet varieties resistant to non-selective herbicides. A model is developed to quantify the processes involved and to rank cropping systems according to their risk of transgene escape from GM sugar beet.

Model structure

The input variables are the regional field pattern, the crop successions, the cultivation techniques of each crop and climate. Output variables are, for each field and year, the density and genotype proportions of the seed bank, adult plants and newly produced seeds.

The model is based on the life-cycle of sugar and weed beet (Figure 1). Cultivated sugar beet is biennial and accumulates sucrose during the first year. Annual plants are either weed beets or prematurely bolting cultivated sugar beet. Groundkeepers are small sugar beet roots lost during harvest which flower in the following crop. Each day the density and genotypes are calculated for every life-stage in each field, depending on cultivation techniques and crop environment. Herbicide resistant and sensitive plants only differ in their response to the herbicide against which the transgene confers resistance. During flowering, pollen is dispersed between fields, depending on field areas, shapes and distances and flowering dates.

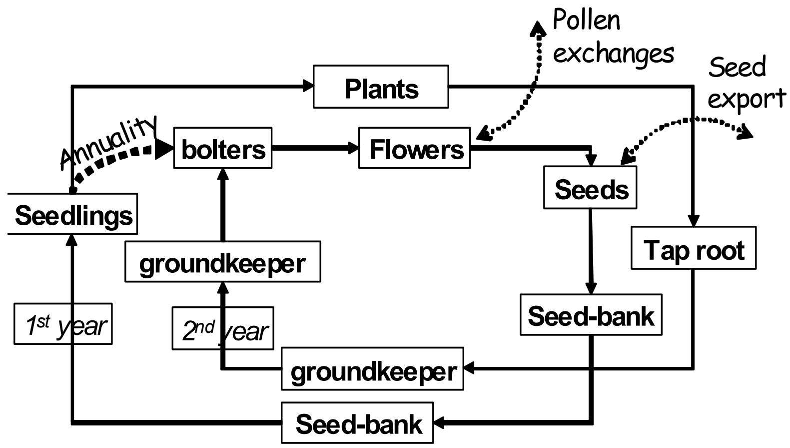

Figure 1. Life-cycle for annual (weed beet or prematurely bolting cultivated sugar beet) and biennial (cultivated sugar beet).

Choice of parameter values

Parameters describing cultivated sugar beet are found in literature. Processes specific to weed beet are often unknown, *e.g.* seed survival in soil, the evolution of germination ability with seed age, or the competitive effects of crops on flowering and seed production of weed beet and of groundkeepers. Field experiments were set up to study these processes and their results will be used to estimate model parameter values.

Simulations

The programming of the model is presently being terminated. Simulations carried out with the model will then allow an *a priori* evaluation of advantages and disadvantages of innovating cropping systems. Simulations on individual fields are already possible. Figure 2 shows the dates and intensity of weed beet flowering in different crops and therefore, the possibility and importance of gene flow via pollen.

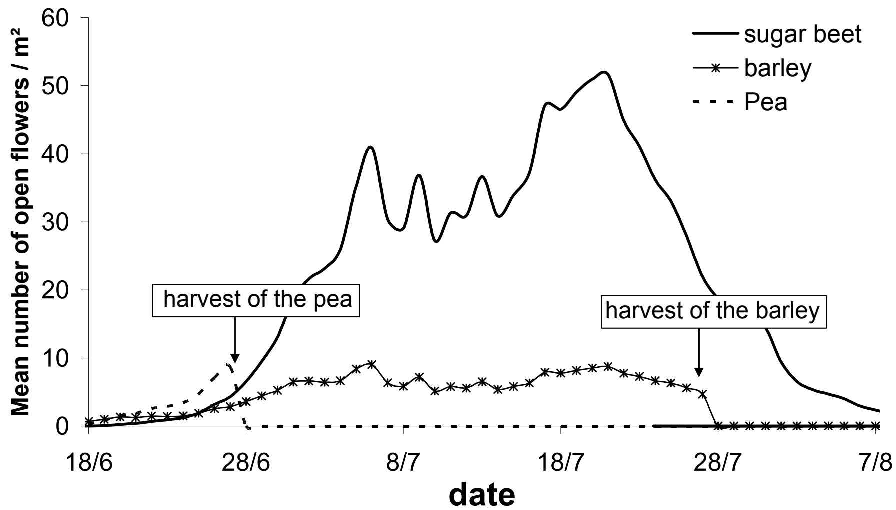

Figure 2. **Simulation of the impact of the crop (spring barley, pea and sugar beet) on the flowering dynamic of weed beet in fields originally infested by 0.5 weed beet seedlings· m^{-2}.**

Conclusion

Both the beet and the rapeseed versions (Colbach *et al.*, 2003) of GENESYS are based on the same principle: a life-cycle describing seed and plant evolution under the influence of cropping systems, with pollen and seed exchanges between fields. This example shows the possibility of adapting GENESYS to other species. However, it also shows that the difficult part of the adaption is to obtain information of the volunteer or feral counterpart of the crop.

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Definition of specific rules for GM sugar beet seed growing

G. Sicard & L. Nardi
Fédération Nationale des Agriculteurs Multiplicateurs de Semences (FNAMS)
F-49800 Brain sur l'Authion, France
E-mail: fnams.brain@wanadoo.fr

Abstract

In 1998, in the prospect of GM varieties registration and marketing, the French Committee for GMO gave a group of experts the responsibility for defining specific rules for GM sugar beet seed growing. The aim was to avoid dispersal of GM genes and preserve the ability of producing conventional seeds in the traditional seed production areas. During 3 years (1999, 2000, 2001) the pertinence and practicability of the proposed rules were tested on 3 conventional fields each year, as if they were GM seed crops. The result of this work shows that most of the proposed measures for GM seed production are in fact, although not statutory, already in use for conventional seed production, because seed companies require high levels of both varietal and sanitary purities. Nevertheless, further efforts are needed such as traceability measures, a complete destruction of excess plants, volunteers and re-growth after harvest, and a careful cleaning of all machinery, especially combine harvesters.

Introduction

In France, more than 60% of the sugar beet seeds are produced in the traditional and very favourable area of the South West, around the city of Agen, where main European sugar beet seed companies are operating. In the prospect of GM varieties marketing, several studies were carried out over the period 1998-2001. Their objective was to assess the ability of producing GM beet seeds without any risk of undermining the conventional production, by uncontrolled dissemination of GM genes, through pollen, seeds or plant regrowth. 3 complementary studies were carried out by the different partners of French seed production (FNAMS, GNIS, seed companies) with the collaboration of ENSAT – INP University of Toulouse and a financial support of the French Ministry of Agriculture:

- A 3 year study on pollen and gene flow from sugar beet to wild relatives (Alibert *et al*., 2003);
- Several trials to ensure the control of volunteers in the following crop and environment;
- Elaboration of specific rules for GM seed production and determination of their feasibility and acceptability by seed growers and the seed companies. This is the subject of the present paper.

Materials and methods

In 1998, a group of 16 experts designated by the French Official Committee "Commission du Génie Biomoléculaire" proposed a series of specific rules for growing GM sugar beet seeds, to be considered as additional to those already in application for conventional seed production. From 1999 to 2001, those specific rules were tested on conventional plots. 3 farmers, each year, growing for 3 different companies (ADVANTA, NOVARTIS, KWS) were asked to manage their crops according to these specific rules. The seed companies' technicians regularly visited the growers to record their practices and check the actual crop management and the plot environment.

Results

Table 1 summarizes the most important specific rules proposed and the way they were applied and discerned by the professionals.

Table 1. Main specific rules for GM sugar beet seed growing and their practicability.

Specific rules	Comments on practicability
On the nursery plot	
➢ Very careful cleaning of seed drill after each sowing	➢ Time costly
➢ Preparation and conditioning of the plants on the nursery plot	➢ Needs a specific organization
➢ Storage and transportation into hermetically sealed containers	➢ No difficulty
➢ Complete destruction of the excess plants on the plot	➢ Needs chemical and mechanical control
➢ Control of the potential re-growths for several years	➢ Needs regular visits to the following crops
On the growing plot	
➢ The plots used for GM seed production must be localised on a cadastral map.	➢ No difficulty
➢ Isolation distances are enlarged to 1000 meters every time the GM character is carried by the male.	➢ No problem with other fields, but needs to control feral beets on a large area
➢ Careful cleaning of all machinery leaving the crop from flowering time to harvest.	➢ Needs a water wash and a period of drying
➢ No contractors intervention without the agreement of the seed company.	➢ No difficulty
➢ Complete destruction of male rows after pollination	➢ No difficulty
➢ The combine must be cleaned before leaving each field and carefully cleaned on the farm	➢ Very time costly
➢ The seed company organises the transport of the seeds lots with traceability measures and insures the cleaning of containers	➢ No difficulty
➢ The seed grower has to check his plots and to destroy GM re-growth or volunteers.	➢ Needs regular visits to the following crops

Conclusion

This 3-year work demonstrates that all the proposed measures are technically feasible by the actors of seed production. Most of the specific rules are not basically different from the management already used by the seed companies and seed growers to insure the good purity and quality of the seed lots.

There are two important points, which need a specific attention and are time-consuming

- a complete cleaning of all the equipment used in the seed growing plots, which may require spending many additional hours
- a careful monitoring of the plots during the following years.

A complete traceability of all the crop management must of course also be required.

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Post harvest management and monitoring

Introduction to the post harvest management and monitoring

J.M. Puumalainen
European Commission - DG Joint Research Centre
Biotechnology and GMOs Unit – IHCP
TP331 via Fermi
I – 21020 Ispra (Va), Italy
E-mail: janna.puumalainen@jrc.it

The management of the co-existence of GM, traditional and organic products does not end after the harvesting or at the farm gate. In contrast, the consumer should be free to make his choices on the market and place on his dinner table organic potatoes as well as smoke a transgenic cigar after the dinner should he wish to do so. Traceability, segregation and identity preservation systems can be applied to assure the end user that the product has maintained its unique identity from farm gate to end user. Such systems – and co-existence – are based on the tolerance of an admixture (i.e. thresholds). Certain traces of GMOs in products may be adventitious and technically unavoidable, and such presence should not trigger labelling and traceability requirements.

The recently introduced EU regulation concerning "the traceability and labelling of genetically modified organisms and the traceability of food and feed products produced from genetically modified organisms" provides the framework of implementing GM traceability systems in Europe. In principle, at the first stage of the placing on the market of a product consisting of or containing GMOs operators shall ensure that this information together with the "GMO identity" is transmitted to the next operator that receives the product. This information should then be passed further through the production and marketing chain. However, the details of the operations – both the traceability documentation and practical segregation – must still be tailored to the practices prevalent different countries, regions, products and food.

The prerequisite for the monitoring of the co-existence after harvesting – and the traceability - is the development of sampling and testing schemes, which can be applied at the different stages of the production chain and for instance for seeds, grains and processed food products. Proper sampling for seeds and grains is not trivial, and a generalized sampling approach for GMO analyses on food is difficult to define because of different variable factors such as the diverse nature of the target analyte in terms of treatment, concentration in the product, production process, packing and distribution channels.

The current scientific and technological knowledge does not allow the detection of all possible genetically modified organisms by using a single method. Thus, multiple tests need to be carried out. In addition, the regulation requires the identification of the GM while being “live” but that this is no longer required for derived products and that therefore the type of information and the eventual tests required may differ in nature between GMOs and derived products. There are several different testing approaches, which are likely to provide equally satisfactory results. These include, for instance, qualitative and/or (semi-)quantitative screening methods or methods specific for certain GMO(s).

The methods commonly based on protein or DNA detection, and some of them take the advantage of the use of plasmids. Some of the methods are more appropriate for the analysis of seed and grain samples (e.g. qualitative methods, ELISA-based methods or strip-tests) than for testing final food products (e.g. quantitative PCR methods).

The session “Post harvest managements and monitoring” will provide insights and examples, on one hand, to the management, implementation and costs of the traceability and segregations systems and, on the other hand, to the challenges of the monitoring – sampling and testing – the success of the co-existence.

GMOs analysis in large kernel lots: modelling sampling of non-randomly distributed contaminants

C. Paoletti[1], M. Donatelli[2], E. Grazioli[1] & G. Van den Eede[1]
[1]European Commission, DG JRC, IHCP, B&GMOs Unit, Via E. Fermi 1
21020 Ispra (VA)
Italy
E-mail: claudia.paoletti@jrc.it
[2]Research Institute for Industrial Crops (ISCI), Via di Corticella, 133, 40128 Bologna
Italy

Introduction

The definition of internationally harmonized strategies for the evaluation of GMO safety is a priority and there is a strong interest in sampling schemes to ensure accuracy and precision of GM surveys. Our work focuses on one critical aspect of GMO control: the definition of sampling protocols for GMO detection and/or quantification. Several guidelines defining sampling strategies for purity analyses are adopted for the detection of GM materials, while waiting for the *ad hoc* protocols.

Kernel lot sampling is a complex multi-stage procedure that should reduce a lot to an analytical sample, of suitable working size, representing the lot. Most kernel sampling plans are based upon the assumption of random distribution of GMOs so that the mean, the standard deviation and both the producer and consumer risks can be estimated according to the Binomial or the Poisson distributions. Given the high likelihood of non-detectable *strata* of GM material in kernel lots (Lischer, 2001), assuming randomness is very risky because even modest deviations from randomness have a strong effect on the accuracy (GMO %) and precision (variance of GMO %) of GMO estimates (Paoletti *et al.*, 2003).

Here we present a model to estimate the sampling error associated to different sampling protocols, in terms of both number and size of samples taken from the lot (primary samples), applicable to any consignment of particulate material with respect to any kind of contamination, including GMOs. The novelty of our approach is the freedom from any distribution constraints.

Results

Preliminary results from our simulations done with KeSTE (Kernel Sampling Technique Evaluation http://www.sipeaa.it/ASP/ASP2/KeSTE.asp; Paoletti *et al.*, 2003) indicated that the pattern of convergence to the “true” contamination value is similar for different contamination levels and lot heterogeneity scenarios. Specifically, the spread of the simulation results (*SD* - standard deviation of the estimates) for each sampling numerosity (number of primary samples taken from a lot) provides an indication of the possible sampling error (*SE*). When a given population is sampled multiple times for an increasing number of primary samples, the error associated to the contamination estimates in the bulk sample decreases. Figure 1 shows such decrease for 50 repeated samples at each sampling numerosity.

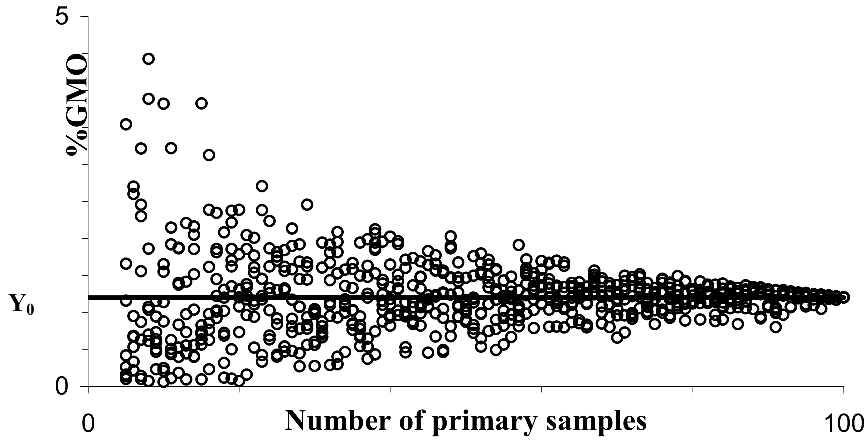

Figure 1. Decrease of contamination estimates error as function of primary samples.

The presence of a common convergence pattern allows the definition of a model to estimate the maximum possible *SE* associated to any sampling numerosity. We found that a negative exponential model $SD = h\, e^{(-sx)}$ (*SD* = standard deviation, *x* = number of primary samples) best describes the decreasing trend of *SD* as a function of the increasing number of primary samples (Figure 2). The parameter *h* is indicative of lot heterogeneity and *s* is associated to primary samples characteristics. The level of lot heterogeneity will determine how large is the error and how rapidly *SD* converges to 0.

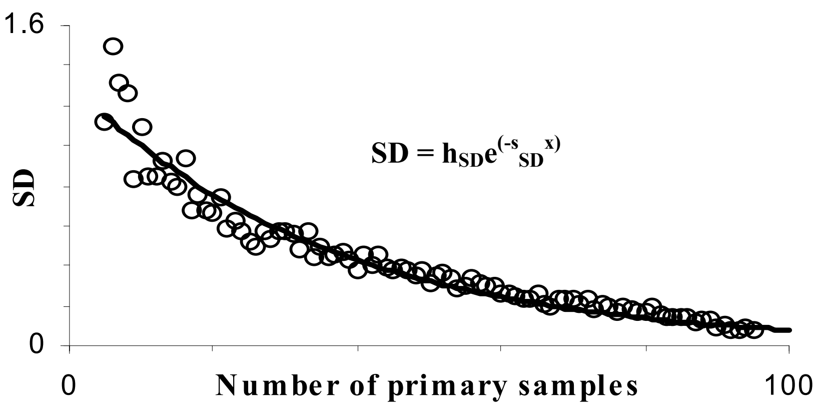

Figure 2. Decreasing trend of standard deviation (SD) as function of primary samples.

Similarly to *SD*, also the largest values of contamination estimates (dominant values) obtained performing repeated sampling at each sampling numerosity, show a constant pattern for different contamination and heterogeneity scenarios, converging asymptotically to the true lot contamination value. Such pattern is described by the exponential model $y = y_0 + he^{(-sx)}$, where y_0 = true lot contamination.

The exponential *SD* curve is more robust compared to the exponential contamination curve given that it converges to 0, implying independency from the y_0 parameter, and it does not require selection of dominant values at each sampling numerosity. In addition, provided that lot characteristics were *a priori* defined in our simulations, we could impose the known value of y_0 and estimate parameters *h* and *s* for both the *SD* and contamination curve, for a series of different contamination and heterogeneity scenarios.

In our simulation primary samples characteristics were maintained constant. As a result, we could use the average value of *s* (calculated over a broad range of heterogeneity and contamination conditions) to re-estimate *h*. This improves the precision of *h* estimates because *h* and *s* show correlation in both exponential models. Once the two series of *h* parameters are estimated, h_Y can be expressed as a function of h_{SD} (Figure 3) and, using the average value of s_Y, y_0 can be estimated with the greatest precision.

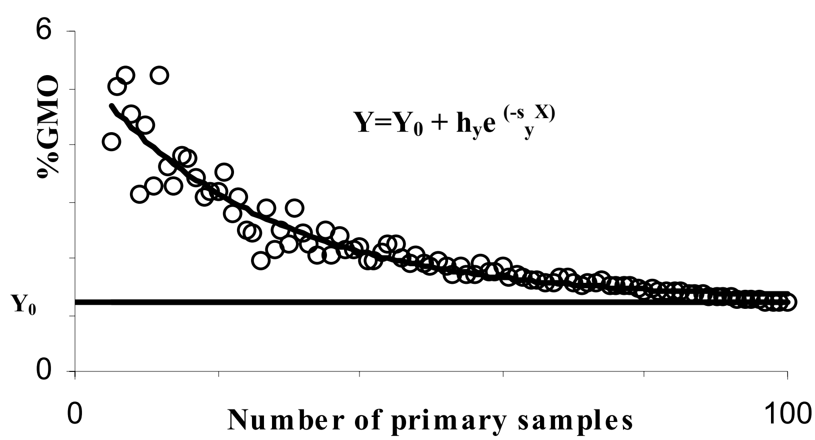

Figure 3. Convergence of dominant values to the true lot contamination level (Y_0) as function of primary samples.

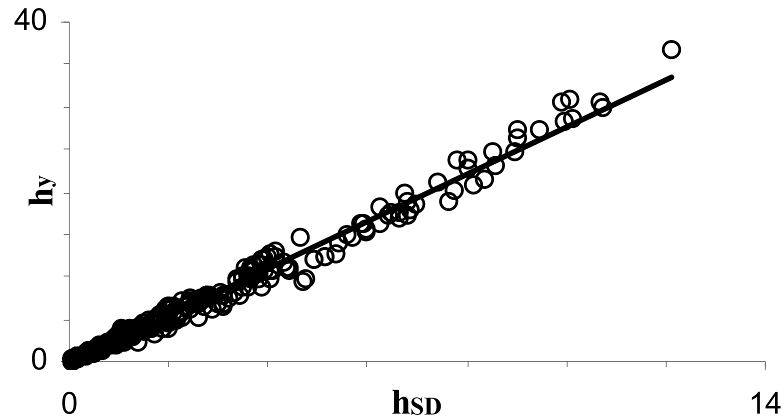

Figure 4. Correlation between h_Y vs h_{SD}.

Perspectives

In this paper we present the heterogeneity model and the preliminary results of our investigation on the stability of heterogeneity parameters. Our next goal is to evaluate, via a sensitivity study, the minimum number of primary samples necessary to maximize h_{SD} estimates robustness. The h_{SD} parameter will provide a proper description of lot heterogeneity and will allow to better estimate the lot contamination mean y_0. This will greatly impact the definition of sampling protocols, as it will ensure a proper sampling numerosity if non-random distribution of contaminants is observed or expected.

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Introduction to GMO detection methods, an essential tool for enforcing traceability and co-existence

M. De Loose[1], N. Papazova[1,2], I. De Grieck[1,3] & E. Van Bockstaele[1,3]
[1]Department for Plant Genetics and Breeding, CLO, Caritasstraat 21, 9090 Melle, Belgium
E-mail: m.deloose@clo.fgov.be
[2]AgroBioInstitute, bul. Dragan Tsankov 8, 1164 Sofia, Bulgaria
[3]Department for Plant Production, Ghent University, Coupure Links 653, 9000 Gent, Belgium

If a regulation such as co-existence and traceability has to be implemented, it is essential that the competent authority who is responsible for the implementation and enforcement has access to the necessary tools. On the one hand this can be based on documentation of the production chain, available in written form. But secondly it is absolutely required to test the trueness of this information at critical points.

Besides controls by the competent authorities it is absolutely necessary for the different stakeholders to have access to testing procedures and guidelines so that they can make the necessary evaluation of the production process and trade they are responsible for. It might be possible that for the different goals different testing procedures are used. This can be due to different knowledge available on the product, the legal context, the different legal responsibility, etc. However, the output of the experimental data has to be used in the framework of the European legislation.

All detection procedures are identifying compounds that differ between the GMO and the non-GMO or wild type. The genetic information for the difference is coded by the fragment that is inserted in the plant genome during the transformation process. This fragment can be transcribed in a mRNA, which lead to the translation in a protein and the production of metabolite(s). The interaction of all these compounds together with the endogenously produced compounds leads to a phenotype, which is different from the phenotype of the wild type. In other words, it is possible to detect a GMO by demonstrating one of the differences described above. However, different GMOs may contain the same fragment(s), express the same compound(s) and resulting in the same phenotype. This is a challenge for the detection to distinguish those – together with the hybrids that contain the transgene locus from both parents, the so called stacked gene constructs, which have to be interpreted as a different GMO. It should also be noted that by using different experimental approaches and determining different compounds this does not necessarily lead to the same quantitative raw data. It is therefore important that the data are interpreted in a correct way.

In the process of detection a step by step and a case by case procedure will be the approach to be followed in the future. In detection, one can distinguish strategies for screening for the presence of GMOs, quantification of the amount of GMOs and identification of the detected GMOs or a combination. A growing complexity for detection of GMOs can be expected in the future due to the fact that the number of transgenic events that is currently used in agriculture worldwide is constantly growing. Therefore in the near future it might be especially relevant in the context of controls to concentrate on screening for the presence of GMOs and in a second step identification of events. Where it is the responsibility of the companies to develop a quantitative event specific method for their product and to submit this method and the necessary control material for evaluation (validation of method) together with the application for authorisation, it will be a role of the authorities such as the members of the European Network of GMO Laboratories (ENGL) to develop tools and methods that can be used for general screening. These methods have to be reliable and economically feasible to be carried out and have to be able to guarantee the quality of the food production chain in respect to GMO content

Biological factors and the production process used will determine the precautions that have to be taken in function of co-existence. Knowledge can be based on the experience that is available today e.g. in seed production and in ecological farming. By using tools such as molecular markers it will be possible to generate lacking data. Finally the ultimate information will be obtained by experimental designs in which GMOs are grown and used in the production process at real scale production levels.

A case study in seed production will be discussed as an example on how to implement detection and quantification of GMOs and how to harmonize this in the context of traceability and co-existence.

GMO/non GMO segregation in supply zone of country elevators

M. Le Bail
Institut National Agronomique Paris Grignon - AGER UMR SADAPT
16 rue Claude Bernard 75231 Cedex 05 Paris, France
E-mail: lebail@inapg.inra.fr

The diagnosis of country elevators supply zone management in three corn and soy regions in France gave the opportunity to draw three GM crops/non GM crops segregation scenarios: "GMO-free region", "farm storage" and "contractual" strategies. They include field and farm management and technical and contractual coordination between farmers and firms.

To reduce the rate of adventitious presence of GM crops in non GM crops most of the agronomic analysis remained attached to the field scale. Few works are performed on the chain management impact on crop(s) segregation. In France collecting firms have a strategic role in segregation. They form a thick network providing concentration in the space and spreading over the time of most of the crops. They have invested in the past fifteen years in identity preservation and quality insurance strategies for specific EEC markets. The study of collection and storage processes from the farmer to the industrial firm (economic and technical planning and piloting) has been used to build agronomic management strategies for wheat quality (Le Bail & Makowski *in press*). Our hypothesis is that co-mingling GMO/"non GMO" risk management consists not only of improving cropping systems but also of evaluating and reducing risks during the various phases of crops collection and storage with technical devices (spatial and temporal organization of crops, transport, storage, drying and ventilation methods,...) and coordination (economic, technical and informational relationships) between farmers and country elevators.

Methods

Our aim (Meynard & Le Bail, 2001) was to study the contamination risks between GMO and non-GMO batches in corn and soy between the exit of the field and the exit of country elevators. By studying the organization of about twenty elevators firms in the south-west, Parisian Basin, and east of France where farming and collecting systems are different, we aimed to identify critical points for adventitious mixing. In fact, as no GMOs were produced in France we have made this diagnosis on particular types of corn or soy already existing in France for various markets (e.g. waxy and rich oil corn). Then, we analyzed the organizational devices set up by firms to manage these risks.

Results

If GM crops spread out in France, the elevators firms we have investigated would try to adapt, as cheaply as possible, their current systems. Three types of scenarios for "GMO"/"non GMO" segregation were identified.

A "GMO-free region" strategy: This logic aims to avoid all GMO in the supply zone of country elevators. By a coordination among all the chain actors everybody refrains himself from any sale, sowing, collecting of GMO crops in a "naturally" isolated region (current model for Alsace). System leans on three devices; a severe control of seeds; a pre-maturity control of the risks of dissemination in the fields by a hazard analysis device; a PCR control after corn dryer to test the level of purity of final batches (1000 to 6000 tons). The advantages of this procedure are to keep the same storage structures with few supplementary costs, particularly coordination costs (farmers advising, firms strategic alliances), seeds and statistic pre-maturity controls. The main drawbacks would appear particularly if GMO crops expand greatly in all regions around[1]. Indeed, in that scenario there is faint traceability before the dryer. It is enough that a field of 4 ha of not declared GMO corn so that a silo of 4000 tons is 1% polluted. After all, this system has to incite farmers to declare GMO sowing, to install severe controls of seed origin and to control particular crops compartments during the vegetative period.

A "farm storage" strategy: The farmers' dryers and storage materials are mobilized to avoid co-mingling between "non GMO" and GMO batches. The strategy result in a time delay segregation and a specialisation of farms in GMO or "non GMO" crops. The system leans on two devices: the farmers specialised in GMO crops (or non GMO) are the farmers with the most important storage capacities and a particular competence for drying their corn without quality damages (e.g. starch extractability); the PCR analysis is made at farm level to decide to include or not the farmer's corn in the final batch. The advantage of this strategy is to delete risks of inter-batches pollution at the country elevator level, because GMO and "non GMO" fluxes are separated and complete traceability is assumed. The first drawback here is a high cost if we consider an increasing of PCR analysis, but also the payment of farmers' storage service. A second drawback is a transfer of pollution responsibility to the only farmers who will try to cover themselves financially to compensate for these risks. In fact, for them it could be difficult (particularly in regions with small farms with dispersed fields) to avoid systematically gene dissemination from their neighbours. Finally, this strategy is possible only, if a significant number of farmers possess the equipments, which is far from reality in some regions.

A "contractual" strategy: Contracts with farmers are used to coordinate their practices in a supply zone and to optimize the use of infrastructures of the country elevator. If the country

[1] Refer to recent Brazilian withdrawal of "non GMO" policy.

elevator has a dedicated site for a particular type of corn with dedicated dryer and silos, the aim of the contract is to reduce risks at field and during transfer towards this silo: technical specifications could be associated to the contract on plot size, isolation distances, different flowering time varieties between GMO and "non GMO" corns... The contract must be signed with farmers closest to the dedicated site, creating an "island of GM corn (or non GM corn)" with particular isolation measures at borders. Particularly efficient to reduce risks of co-mixing this strategy suppose quite high transaction costs and a limitation of the volume produced at the level of the capacities of the dedicated silos. If the country elevator has no dedicated site the risks of co-mixing are maximum. In that case the elevator has to optimize the use of its dryer, by fixing in the contract different delivery dates for each type of corn and has to clean dryers between two types of corn. The advantage of this strategy is to reduce investments and to assure some traceability with the contract. The drawbacks are the risk of co-mingling by failure of differing flowering time for climatic reasons or delay in cleaning dryers, which can be too long for the collecting organization.

Conclusion

Strategies presented here are ideotypes to guide research on technical references on critical points of the systems. Besides, our results gave opportunities to the agronomists to simulate various organizations of segregation and to estimate the risks of adventitious presence of "GM grains" in "non GMO" batches by integrating into those organisational scenarios the last improvements of the biotechnical models (Angevin *et al*., 2002 and INRA current works).

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Segregation and quality control for the co-existence of GM and conventional maize

F. Benetrix[1] & C. Poeydomenge[2]
[1]ARVALIS – Institut du végétal, 64121 Montardon, France
E-mail: f.benetrix@arvalisinstitutduvegetal.fr
[2]MAIZ'EUROP', 64121 Montardon, France
E-mail: cedric.poeydomenge@agpm.com

Abstract

An operational Program for GM Crops Evaluation (POECB) was sep up to obtain operational information for segregation and traceability of GM crop from field to storage.
POECB is the first study led in France under real field conditions. Aware of civil society expectations, professionals were willing to build this program to study how to set up an organization and traceability system in order to allow the co-existence between GM and non-GM productions. How? By identifying critical points and controlling the batches segregation at each step of the chain.

Introduction

POECB has been conducted with a scientific committee composed of scientists and experts from INRA, IRTAC, technical institutes, industries, maize and seeds producers association. Four working groups included experts from national research, technical institutes and stake-holders of maize chain have been created to predict and evaluate the distribution and prediction of cross pollination, the identification of appropriate quality control and procedures for co-existence in real field conditions in France. So all the main subjects related to the co-existence of GM and conventional maize were covered.

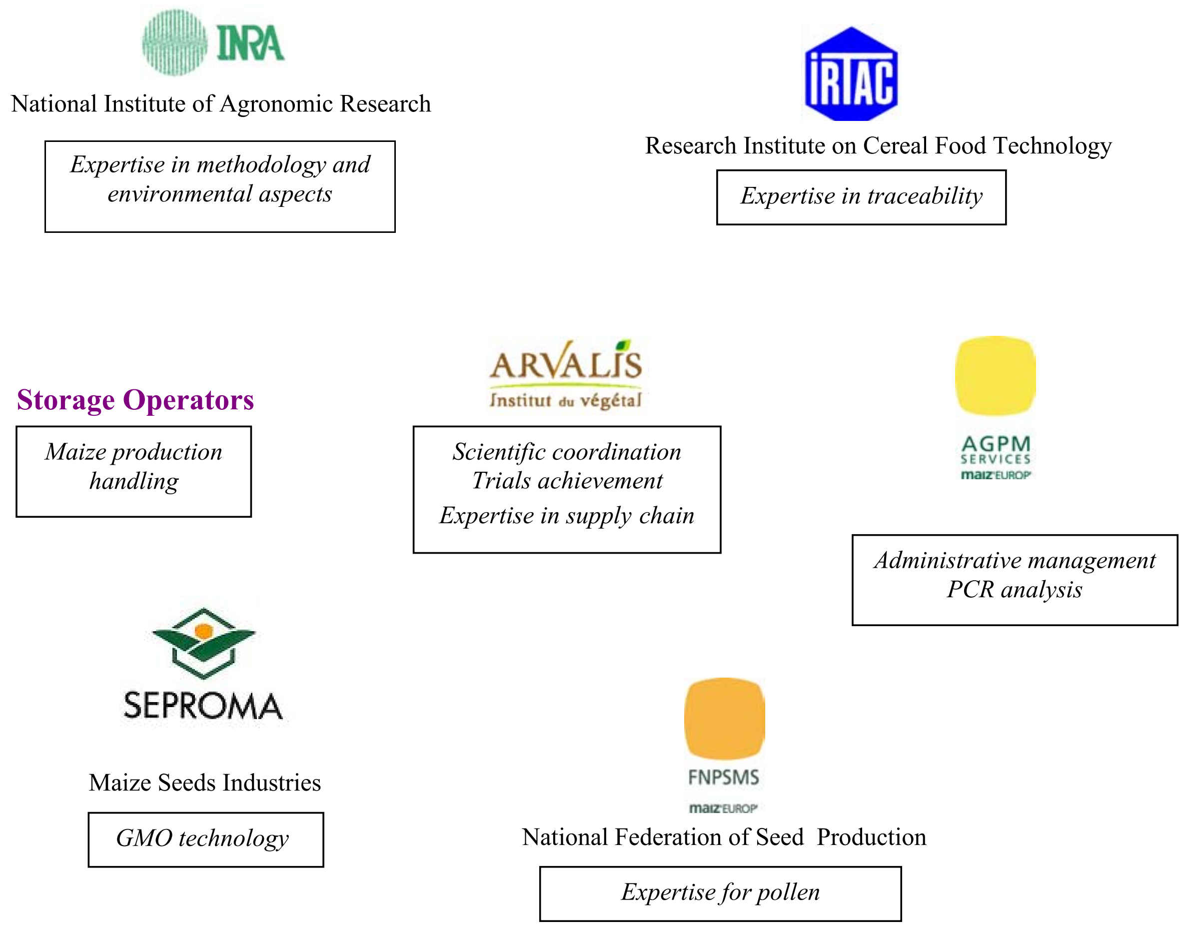

Figure 1. Composition of the Operational Committee.

Materials and methods

POECB has been conducted in three fields located in the South of France. 50 ha were dedicated for this program with 0.4 to 0.8 ha of Bt maize on each location.

Monitoring of pollen dispersion

Conventional and Bt maize were sown to be in a flowering concordance. Within the framework of the program, the pollination and dispersion of pollen from GM fields were studied: flowering cinetics, pollen quantification, pollen weight and viability. A statistical model of pollen dispersion has been tested: it is based on climatic data, crop situations, varietal characteristics.

Controls and monitoring from field to storage

At each step from seed to storage, quality controls and procedures have been used: field monitoring, agronomic notations, climatic registration, procedures for maize harvesting and drying, farm equipment cleaning, PCR controls.

Results and discussion

POECB has set up interesting results to make the co-existence possible.

A survey of cross pollination for harvest control
Within the framework of POECB, a survey about pollination and pollen dispersion from a GMO field was conducted. This program permitted to test the performance of a model of dispersion proposed by the National Institute of Agronomical Research (INRA). The level of cross fertilization at various distances from the original field can be predicted with the help of this model. Thanks to an accurate method of ear sampling in the receptor field, the analysis of cross pollination levels by PCR confirmed the forecasts.

Quality and traceability from field to storage
Methods, registration files and controls have been created and applied at every step of production, harvest, drying and storage in a quality assurance process. Harvest has been realized in a preset order from the GM contents results in field. The risks of mixing between a GM production and a non GM production have been evaluated at the harvesting stage in the combine harvester and in the dryer, used in continuous flow.

Co-existence of GM and conventional maize is achievable with more or less factors to take into account according to thresholds for GMO limits and field disposal.

Perspectives

An economical analysis is in progress. It will permit to evaluate the overcosts generated by the co-existence and separated management of both productions at each step of the chain and according to threshold values to be applied.

This survey provides data to set up a « good practice » program for the professionals of the agricultural sector. It will provide tangible elements concerning the realization of a GM – non-GM traceability procedure.

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A method to assess the costs of avoiding admixture in a combined GM and conventional processing system

S.M. Pedersen, M. Gylling & J. Søndergaard
Danish Research Institute of Food Economics
Rolighedsvej 25
DK-1958 Frederiksberg C, Denmark
E-mail: marcus@foi.dk

Abstract

This paper describes a method to assess the critical points and additional costs of avoiding admixture in a combined GM and non-GM processing system. To establish a realistic scenario we assume that GM crops will be processed at the same production facilities along with non-GM crops. This study examines the production chain from farm gate to products for further use in the food and feed industry. The costs of implementing different production systems differ significantly according to logistics, production methods, commodity type and possible production volumes. Some systems are highly complex and for other systems it is less costly to prevent admixture.

Introduction

The co-existence between non-GM and GM crops have for some years been of major concern among European consumers and crop producers. In this respect it is relevant to examine the critical points and additional costs of preventing admixture between non GM and GM crops.

Materials

This study focuses on the further processing of three well established vegetable commodities: Refined oil from rape seed, refined sugar from sugar beets and feed mixture compounds from phytase wheat. A number of Danish manufactures, supply companies and research institutes have participated in an interactive dialogue during the project period and provided information about the production structures and data for the cost analysis.

Methods

The additional costs in the various processing system are assessed by using traditional system and cost analysis. All critical points are depicted by adapting the principles from the HACCP analysis approach. This study emphasises on the production chain from field to products for further use in the food and feed industry. Focus is put on three production processes: Sugar beets for sugar production, rape seed for oil production and wheat for feed mixture compounds.

Results and discussion

Studies conducted by a Danish research group (Fødevareministeriet, 2003) indicates that co-existence between GM-crops, non GM crops and organic crops is possible given the necessary precautions. The difference in complexity between the systems implies that there are markedly cost differences depending on the crop variety, end product functionality and durability. For all crops it is possible to use existing production facilities given additional precaution, such as periodical cleaning and logistic optimisation. Additional costs of preventing admixture between GM sugar and non GM sugar are rather modest. The logistic facilities are well established with contractual agreements between farmers and the manufacturing industry, and as a commodity, sugar is easy to handle.

The cost of preventing admixture between GM and non GM rape seed seems relatively high. However, unlike sugar, rape seed oil is characterised as less durable than sugar and the manufacturing process gives higher cleaning costs and lower production volumes. Moreover the supply sources vary from many individual farmers to large external suppliers implying that each batch has to be analysed independently of the batch size and quantity. The costs of preventing admixture between GM wheat and non GM wheat for feed mixture compounds are relatively high due to low durability, high storage costs due to segregation, and high cleaning costs of the transport vehicles.

Perspectives

In general, it is possible to prevent admixture of GM and non GM crops given the necessary precautions. It is possible to use existing production facilities but the additional costs depend on production volumes, durability and end product functionality. The relative modest additional costs of co-existence are based on the fact that the individual companies already conducts control and documentation measures at a high level. A further structural change in capacities and supply of GM crops will have a direct impact on the additional costs.

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Strategies and economic assessments

Introduction to strategies and economic assessments

K. Menrad
University of Applied Sciences
Department of Horticulture and Food Technology
Am Staudengarten 10, D-85350 Freising, Germany
E-mail: klaus.menrad@fh-weihenstephan.de

In recent years there have been several studies targeting at least partly to analyse the cost effects and wider economic impacts of co-existence of GM crops with conventional and organic crops in Europe and overseas. These studies show that potential costs of co-existence schemes occur on different levels (e. g. on a single farm, a region, the agricultural sector or the agro-food chain of a specific country or internationally) and that different types of costs have to be distinguished when assessing the economic impacts of co-existence measures in agricultural production. In this context the most important cost positions are:

- Short-term financial losses in case of contamination with GM material (e. g. via gene flow, volunteers, mixture of GM and non-GM crops).
- Insurance and liability costs.
- In case a contamination with GM material has happened, mid-term costs for reducing or removal of the GM contamination.
- Costs of establishing and handling monitoring, segregation and labelling systems.
- Wider impacts of co-existence of GM crops on agricultural and food markets, production and processing structures as well as trade flows.

The estimation of cost effects or economic impacts of co-existence schemes or measures is further complicated by the fact that agricultural production systems and structures differ significantly between EU member countries and regions. This is one important reason why so far no generally accepted co-existence schemes and measures have been suggested by agronomists (and maybe never will) due to differing biologic characteristics of farm crops and regional production systems. Furthermore, the necessary co-existence measures highly depend on the adoption rate of GM crops in a specific region as well as the threshold levels of adventitious admixture, which are tolerated by existing regulations. In this sense all available assessments of costs or wider economic impacts of co-existence measures of GM crops with non-GM crops are based on case studies of specific plants (e. g. oilseed rape, maize, potatoes, soybean, sugar beet) and particular assumptions concerning e. g. agricultural practice, production systems, threshold levels, GM adoption rates or market reactions. So far studies are lacking which intend to assess the overall impacts of co-existence measures on agricultural production and the food processing chain (e. g. of a specific country). In addition, the costs of specific stewardship or training programmes for farmers in order to introduce and implement the suggested co-existence measures in agricultural practice are mostly not included in available studies.

Despite these specific challenges and existing uncertainties it could be shown in recent research that co-existence activities are almost impossible from a technical and cost-point of view if threshold levels of 0.1% of adventitious admixture with GM material are requested like e. g. from several organic farming associations. In most agricultural crops and production systems studied, thresholds of 0.5% in seed production and 0.9% in the food chain can be complied with through changes in farming practices and introduction of a monitoring system. In most cases their costs as well as likely insurance fees may result up to 10% of the current farmgate prices of major agricultural crops concerned (like e. g. oilseed rape, maize or sugar beet).

In case of GM contamination, organic farmers generally face higher costs (or at least short-term losses) per hectare (or per tonne produced crop) than conventional farmers due to the existing price premia of organic crops. In all cases, monitoring costs account for a large part of the additional cost effects on farm level. Cost reductions might be feasible in this area with segregation and identity preservation measures becoming a common part of agricultural practice and decreasing costs of test systems for GMOs due to economies of scale. Concerning potential insurance or liability costs of GM contamination, there is only limited empirical evidence for the EU in the available studies.

The papers presented at the session „Strategies and economic assessments“ of the Conference partly fill the research gaps existing in this field:

- Rodolphe de Borchgrave aims to provide a framework for co-existence economics and analyses the issues, frameworks and available results of the behaviour of single farmers and operators in the food and feed supply chain. In addition, welfare effects, market and trade balance considerations are also taken into account in the paper.
- G. Brookes & P. Barfoot present experiences from the six years cultivation of GM-maize in Spain. The paper examines the issue and covers current/recent plantings of conventional, GM and organic maize in Spain, the operation of conditions or recommendations for growing each crop and their relevance, possible future developments in crop plantings, possibilities of adventitious presence occurring and measures to minimise this.
- The paper of S. E. Frandsen & C. P. Nielsen aims to analyse the price, production and trade consequences of alternative consumer reactions to GM food. For this purpose an empirical model of the world economy is used which segregates the food processing chain in GM and non-GM lines of production. The results show the crucial role of the level of GM-rejection by consumers on the impacts of trade flows. In this sense this paper widens the view of recent research with respect to consumer reactions and the assessment of wider economic impacts of co-existence.

- J. Sondergaard, M. Gylling and S. M. Pedersen examine the possibilities and cost effects of co-existence between GM and non-GM products in the food processing chain. Therefore this paper is one of the first studies, which include food processing in co-existence economics. Using the example of a Danish producer of convenience food products, they analyse the modifications in the production structure and changes in costs when a non-GM process line is implemented parallel to a GM-line. In this example, they found an increase of total production costs of around 6.5% mainly due to higher labor, investments and control costs.
- K. Sinemus analyses the code of good agricultural practice, which have to be modified in order to achieve co-existence between GM and non-GM plants. Based on this analysis communication and consultation activities are outlined in order to implement co-existence schemes in practical agriculture. In this sense the paper widens the view to practical considerations and activities.

Co-existence from farm to fork

J. Søndergaard, M. Gylling & S.M. Pedersen
Danish Research Institute of Food Economics
Rolighedsvej 25
DK-1958 Frederiksberg C, Denmark
E-mail: janus@foi.dk

Abstract

The possibility of co-existence between GM and non-GM products in the last stage of the production chain is examined in this study. In order to ensure the consumers free choice preventing admixture in the production chain is a necessity. This study shows that co-existence is possible and that costs are not prohibitively high. The paper also supports the importance of looking at the coexisting issues in a chain perspective.

Using the proposed EU-labeling rules this paper examines the changes in production structure and costs of production related to implementation of a non-GM process line parallel to a GM-line in an all ready existing production system.

Introduction

The co-existence between non-GM, and GM crops have for some years been a major issue among agents in the production chain and among consumers. In this respect it is relevant to establish a method to assess the critical points and additional costs of preventing admixture between GM and non-GM crops. It's therefore relevant to move downstream in the production chain and also look at the processing industry because of the multi component handling problems facing the processing companies of GM and non-GM products in the future.

Materials

Based on the production set-up in one of the big Danish producers of convenience food products it is attempted to map the changes in the production structure and changes in costs of production when a non-GM process line is implemented parallel to a GM-line.

Methods

Mapping the production set-up based on Michaels Porters value system gives important information about where in the company's functions the changes might take place in connection with producing under co-existence in at processing company. Adaption of the production system will have an impact on all the company's functions. Some are direct and therefore easy to measure and some are indirect and therefore more difficult to measure. In relation to Porters value system it can be expected that the support functions contain the indirect difficult costs and the primary functions contain most of the direct cost in relation to producing GM and non-GM products. Based on the assumptions from above the focus point in the paper will be based at the changes in production and the costs at the "floor", the primary activities, in the processing company.

By using the HACCP concept as a risk analysis tool in relation to adventitious GM admixture in a non-GM processing system, the critical control points (CCP) are located and the costs of adopting the process line are estimated.

Results and discussion

The study shows that in practice there are relatively few problems in practicing co-existence in the production system. In relation to the costs of this co-existence it is found that there is a considerable increase in relative production costs. Labour, investment and control costs are the main sources for increased costs. The result from the study, regarding increased costs, is shown in Table 1.

Table 1. Overview of the % relative changes in costs and % increased total costs in the investigated processing company.

Cost categories	% relative changes in costs	% increased total costs
Labor (Adm. and prod.)	+14%	+4.04%
Machinery costs	+6%	+1.01%
Depreciation	+38%	+1.51%
Total		**+6.56%**

The costs of controlling and preventing the co-existing admixture between GM and non-GM are relatively high. The most important relates to the investments in new storage capacity to ensure the segregation of ingredients, which increases the depreciation of the production facilities with 38 percent. In relation to the increase in total costs the depreciation represent an increase of approx. 1.5 percent.

However, the study shows that the total percent increase in productions cost are at approx. 6,5 percent, which mostly is due to the increasing labour costs. A result of the co-existence is an increased focus on control, handling and cleaning of the products and the processing lines. These initiatives are labour intensive and count for approx. 60 percent of the increase in total costs.

Perspectives

This paper takes over from the first studies of the production chain and do not consider the problems with pre-handle product. The processing industry is the last product handler before the consumers and they are therefore the collector of pre-handled products containing GM and non-GM ingredients. It is assumed that the processor has all the right information available concerning the previous stages of the chain. This may give an asymmetric picture of the real cost structure; because the costs concerning information and contract negotiations in transactions between the different stages of the chain have not been taken into account.

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Co-existence of GM and non GM crops: case study of maize grown in Spain

G. Brookes & P. Barfoot
PG Economics Ltd, Dorchester, Dorset, United Kingdom
E-mail: graham.brookes@btinternet.com

Abstract

Given that GM (Bt) maize is in its sixth year of cultivation in Spain, this offers a unique (in the EU context) case study of whether approved GM, conventional and organic crops can coexist. This paper examines the issue and covers current/recent plantings of conventional, GM and organic maize in Spain, the operation of conditions or recommendations for growing each crop and their relevance, possible future developments in crop plantings, possibilities of adventitious presence occurring and measures to minimise this. It also examines whether the organic sector can coexist with GM maize production.

Summary

Current co-existence

Out of the current area planted to maize in Spain (about 460,000 hectares), about 32,000 ha (7%) is to GM insect resistant (Bt) varieties, 100-500 hectares (0.1%) is organic and the vast majority is 'conventionally produced'.

The evidence to date shows that these three types of maize production have coexisted without economic and commercial problems. This includes in regions such as Catalunya where GM (Bt) is concentrated[1]. Where non GM maize has been required in some markets, supplies have been relatively easily obtained, based on market-driven adherence to on/post farm segregation and by the purchase of maize from regions where there has been limited adoption of Bt maize (because the target pest of the Bt technology, the corn borer is not a significant problem for farmers in these regions). Isolated instances (two) of GMO adventitious presence in organic maize crops were reported in 2001.

Future co-existence

For the future, the likelihood of co-existence problems arising remains fairly limited, even if there is a significant expansion in both the areas planted to GMOs and to organic maize because:

[1] Bt maize accounts for about 15% of total maize plantings in this region

- GMO (Bt) maize is unlikely to dominate maize plantings in Spain, being concentrated in regions where there are significant (corn borer) pest problems. In other words there will continue to be regions of Spain where GM (Bt) maize will not be widely planted;
- The organic maize area is likely to continue to be a very small part of the total maize crop (even if there was a tenfold increase in plantings), with a very limited economic contribution relative to the rest of the Spanish maize crop;
- some changes to farming practices on some farms may be required. This will however, only apply where GMO maize crops are located near non GM or organic crops for which the non GM status of the crop is important (e.g., where buyers do not wish to label products as being GM or derived from GM according EU labelling regulations). These changes are likely to focus on the use of separation distances and buffer crops (of non GM maize) between the GM maize crop and the 'vulnerable' non GM/organic crop. GMO planting farmers are already made aware of these practices as part of recommendations for growing GM maize in Spain (co-existence and refuge requirements) provided by seed suppliers in their 'GMO stewardship programmes'. Few GM planting farmers have however, found themselves located near to 'vulnerable' non GM/organic crops to date and hence the need to apply these guidelines has been very limited. In the future, if for example, the areas planted to Bt maize and/or organic maize was to increase significantly, it is possible that more (GMO planting) farmers will need to apply these guidelines.

Co-existence economics: issues, frameworks and first results

R. de Borchgrave
Arcadia International E.E.I.G.
Avenue des Princes Brabançons, 35
B-1170 Brussels, Belgium
E-mail: rodolphe.de.borchgrave@arcadia-international.net

Introduction

On various occasions, Commissioner Franz Fischler publicly shared his views that "Co-existence is exclusively an economic problem"(Agra Europe, 2003). However, co-existence economics have been relatively little explored so far and only scattered results are available. A systematic and broad band approach is hence necessary to provide a practical perspective on managing co-existence scenarios. This paper aims at presenting a "state of the art" view of co-existence economics: issues, frameworks, available results and some agenda for research[1]. It focuses on the angle taken from entering the biotech agriculture route.

At the core is the individual farmer's decision making. Consolidated individual decisions will determine adoption rates for various segments of agriculture. From an economic perspective, farmer's decision will be influenced by his perceived benefits, costs and risks. Some of these costs will arise from management measures defined by local authorities. Some will be determined by operators' specifications in the supply chain, such as elevators. These might be in turn determined by anticipations on market prices resulting from the existence of several segments. Costs and benefits can be analysed from an individual perspective, but also from a collective viewpoint. This last perspective encompasses welfare effects such as consumer surplus or producer surplus. When we review these viewpoints, we will see that some first results are available but that there are also wide gaps and holes in our knowledge. This sets an agenda for research.

Farmer behaviour

According to economic wisdom, a rational farmer will decide on what to grow (biotech, conventional, organic) on the basis of his perceived net benefits, taking into account costs, risks and liabilities. What are exactly these perceived elements can be approached either by

[1] Co-existence economics will be systematically investigated as part of SIGMEA, a project submitted to 6th Framework Research. As a partner in the SIGMEA research consortium will participate in this effort. Although this paper was in part inspired by SIGMEA discussions, it reflects only Arcadia thinking or responsibility and does not commit other members of the SIGMEA project.

assessing *ex post* results from, say, biotech plantings and comparing these with actual decisions, or by obtaining from farmers an *ex ante* judgement on what their decision would be under pre-defined circumstances. The first approach is only feasible in areas where actual GM planting exists, that is in Spain. Two distinct pieces of research were recently conducted in Spain: a first one on biotech benefits (Brookes, 2002) and another one on the feasibility and costs of co-existence (IPTS-JRC, 2002). Allowing with all due reservations for putting together results from two independent studies, it looks like in the case of BT maize in Spain:

- average biotech net benefits: 146,50 €/Ha
- average co-existence cost, at 1% tolerance: ~ 100,00 €/Ha

On the basis of these preliminary results, it looks like implementing standard co-existence measures would severely reduce farmers' perceived benefits and therefore BT maize adoption rate in Spain. This remains of course to be confirmed and generalised.

Supply chain elements

Farmer decision will be influenced by specifications set market downstream by operators such as elevators, transport companies and processors as well as by contract terms such as premiums and penalties. Such specifications could result from:

- The cost of maintaining co-existence (identity preservation + traceability) along the food/feed supply chain;
- The influence of several market segments (organic, GM, non-GM, ..) on prices.

The first aspect was approached, among others, by Arcadia & partners (Borchgrave *et al.*, 2003) in previous research on "non-GM economics". Key results from this research could, to some extend and pending confirmation, be extrapolated to the case of co-existence, including:

- Identity preservation + traceability on-cost, compared with the value of the product, is more important in up stream activities of the feed/food chain;
- This on-cost tends to dilute in the value chain and to become minimal, in relative terms, as one reaches the consumer.

The potential impact of co-existence on prices is more difficult to assess, because it has to do not only with cost, but with willingness to pay. It is anticipated that:

- Consumer prices will be influenced by consumer willingness to pay for products from coexisting agriculture segments. This willingness to pay will be influenced by labelling and retail marketing. Most available results are not very informative, because they are based on opinion surveys rather than on purchasing behaviour;
- How consumer demand will be reflected in market equilibrium at up stream stages of the food chain, back to agriculture, will depend on price transmission into the supply chain, another element to be investigated.

Collective impact

From a government perspective, individual decision making are important mostly as a way to assess collective economic impacts of co-existence. These can be measured as welfare effects called consumer surplus or producer surplus. These measure what, as an aggregate, consumers (producers) would receive above their willingness to pay. This supposes that one can derive an estimate of aggregate demand/supply relationships for various agriculture segments. Literature provides little indication of this, which can be nonetheless addressed in several schemes of "experimental economics":

- Estimate of aggregate supply: from the analysis of a sample of farmers according to a relevant experimental design (e.g. Stated preference);
- Estimate of aggregate consumer demand: by a similar technique or by running an auction.

Market and trade balance

The situation will be very different for imported materials produced in countries where GM is a production standard, such as soja, and for products produced in the EU where co-existence will become a production standard. It can be anticipated that the cost of producing non-GM in the first group of countries and the cost of co-existence in the EU will act in a quite different way on production decisions, shifting supply relationships and on export/import positions. Impact of all this on trade balances also deserves to be investigated.

Conclusion

A multi-level framework is required to approach is co-existence economics. Results from partial analyses need to be fed back to each other. Short term and long term effects need be considered as well. Research efforts will be welcome in various directions in the times ahead.

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Genetically modified crops in Europe: co-existence or no existence?

S.E. Frandsen & C.P. Nielsen
Danish Research Institute of Food Economics
Rolighedsvej 25, DK-1958 Frederiksberg C, Denmark
E-mail: soren@foi.dk; chantal@foi.dk

Abstract

This paper analyses the price, production, and trade consequences of alternative consumer reactions to genetically modified (GM) foods in the context of co-existence with conventional foods. Farmers' choice to grow GM or non-GM crops depends not only on technical aspects related to the productivity gains and agronomic benefits to be had from adopting this technology, but also on consumers' preferences. Particularly in Europe, consumers continue to be concerned about the potentially adverse implications of widespread GM crop production for the environment and food safety. Their reactions to GM foods will determine the long-term viability of the GM and non-GM segments of the market, not only within Europe, but also in countries with which Europe trades.

Introduction

Co-existence between conventional and genetically modified (GM) crops is a reality. Sharp reactions against genetic modification by particularly European consumers has already resulted in the creation of differentiated marketing systems for e.g. GM and non-GM maize and soybeans in the United States. Growing GM crops allegedly provides farmers with a range of agronomic benefits, mainly in the form of lower input requirements. Yet the GM/non-GM choice is much more than simply a matter of different production cost levels. The long-term viability of markets for GM products depends crucially on consumer preferences. This paper analyses the price, production, and trade consequences of alternative consumer reactions to GM foods.

The economics of co-existence

Co-existence represents a market-based solution to a controversial technology. To the extent that markets are permitted to respond freely to changes in supply and demand, the relationship between producer and consumer interests will determine the long run shares of each segment of the market (Frandsen, 2003). On the supply side, the extent of GM adoption will be determined by the real or anticipated economic benefits of growing these crops. GM production allegedly requires fewer inputs, leading to lower production costs and thereby also lower product prices. Lower primary product prices will benefit the food-processing industries, more so the larger the share of the primary product in the overall cost structure. To date, GM technology has been more readily applicable to some crops (e.g. maize, soybeans, and cotton) than others (e.g. wheat). Hence, in the short to medium term, sectoral production structures will change in response to which crops are able to benefit from this technology-induced productivity gain. Depending on the technological developments in the longer run, such sectoral shifts may be dampened as the first-mover advantages wear off. In terms of international competitiveness, a country's initial production structure will determine the extent to which it benefits from GM technology. On the demand side, consumer reactions are a function of perceived risks and benefits concerning the potential effect of GM crops on the environment and food safety, the development of GM products that have more direct benefits for consumers, and finally, ethical considerations. Moreover, consumers' reactions depend on their trust in public authorities to correct potential negative externalities and information problems.

An empirical assessment

An empirical model of the world economy, in which the entire food processing chain is segregated into GM and non-GM lines of production (Nielsen *et al.*, forthcoming), is used to analyse the implications of widespread use of GM crops while consumers in Europe continue to adopt a critical attitude towards GM foods. Two alternative consumer responses are considered: (1) reduced sensitivity to a decline in the price of GM foods relative to non-GM varieties, and (2) a structural demand shift irrespective of the price differential. The results show that interpreting consumer dislike of GM foods as a reduced sensitivity to relative price changes dampens the impact of the productivity difference between the two varieties. Consumers are less inclined to buy GM products and so the positive production and export responses are less than in a situation where consumers are indifferent. If the consumer response is more a matter of rejection, the effects on prices, production, and trade flows are much more dramatic and the direction of effects reverses. Countries that initially are dependent on exporting GM-potential crops to GM-critical regions find themselves increasing exports and hence production of non-GM varieties, and reducing production of GM varieties in spite of the productivity benefit. Clearly, the results depend crucially on the extent on GM-rejection by consumers and the size of the productivity gain foregone in comparison with the relative price premium obtainable on non-GM varieties.

Future research

Due to the relative novelty of GM technology, data limitations have meant that the empirical analysis has had to rely on a number of simplifying assumptions, in particular those made about the size and nature of the productivity impact of adopting GM crops, and the interpretation of consumer responses. Furthermore, the analysis has not been able to take explicit account of the costs of preserving the identity of a crop or food product throughout the production and marketing chain, including labelling and related testing costs. Improved data availability would enable a richer empirical analysis. These caveats notwithstanding, this analysis has brought attention to the importance of consumer sentiment in determining the long-run viability of the co-existence of conventional and GM crops.

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Communication measures for establishing co-existence: technical, economic and social components

K. Sinemus
Genius GmbH, Robert-Bosch-Str. 7, 64293 Darmstadt, Germany
E-mail: ksinemus@genius-biotech.de

On 23 July 2003, the European Commission issued guidelines for the co-existence of different types of agriculture to serve as a basis for the development of national strategies and procedures. No form, whether conventional or organic, or making use of GMOs, should be excluded in the European Union. At the same time, the freedom of choice for consumers should be upheld. In some member states legislation processes have been set in motion to regulate co-existence measures. However, opinions range from no need whatever for national regulations to detailed crop-specific regulations.

In actual fact, the co-existence of different production types is not a new issue in agriculture. Conventional and organic crop production have coexisted for several years. Today about 3% of the agricultural area in Europe is organic land. The increasing number of contract growing and identity preservation systems, e.g. vegetables for baby food or barley for beer brewing, reflect the market demand for agricultural products with specific qualities. To this end, codes of good agricultural practice have already been established for quite some time.

However, the extensive growing of GMOs calls for substantial safeguards to minimize admixtures of GMO and non-GMO products at farm level. GMO and non-GMO farmers will have to take new and complex measures going beyond the current codes of good agricultural practice, e.g. isolation distances, pollination barriers, enhanced control of volunteers, adequate choice of varieties and separate facilities for storage and transport.

For this reason, farmers will have to adopt the necessary measures for calculating their economic impacts and coordinate their growing plans and measures with neighbours. The requirements will differ from region to region depending on different agricultural structures, marketing strategies and degrees of public acceptance. Farmers will remain at the centre of attention and must be prepared to deal with the requirements of members of the food chain, consumers, NGOs and neighbouring farmers. To enable them to implement co-existence measures and carry out an exchange of information, it will be necessary to provide them with an education and consultation program taking full account of specific regional characteristics.

This lecture will sum up the current status of the various types of agricultural growing systems and offer an analysis of the codes of good agricultural practice, which will have to be modified or additionally adopted. Starting from this situation, a suitable communication and consultation initiative at European level for farmers and related groups will be outlined.

Modelling

Introduction to gene flow modelling and co-existence

A. Messéan[1,2], F. Angevin[1], N. Colbach[1] & J.-M. Meynard[1]
[1]INRA, Dijon & Grignon, France
[2]CETIOM, Grignon, France
E-mail: messean@grignon.inra.fr

As already addressed in the sessions dealing with crops, gene flow is a common phenomenon for crop species and its implications for Genetically Modified Plants have raised new concerns. Undesirable effects related to gene flow may result in **ecological** or **agronomic** considerations (persistence of feral plants, creation of new weeds; impacts on biodiversity) as well as **in commercial** considerations (adventitious presence of GMOs in conventional crop production affecting its competitiveness in the marketplace). Consequently, the co-existence between different types of crops has become a major issue and has to be addressed *per se* whatever are the actual ecological, agronomic and safety impacts.

Adventitious presence of GM highly depends on crop biology, farming practices and regional environments

Adventitious presence of GM seeds in non-GM production could have several causes:

- Crop-to-crop pollination between neighboured fields;
- Presence of volunteers in conventional fields resulting from former GM crop cultivation in the field;
- Pollination from feral GM plants occurring in field borders and resulting from seed dispersal during transportation (or harvest combination);
- GM impurities in seed lots (cross-pollination during seed production or admixture in the post-harvest process).

Why models are necessary?

On-farm gene flow occurs both in time and in space, through pollen flow as well as through seed dispersal. Several factors are involved: crop biology, landscape fragmentation, environmental conditions, crop management and post-harvesting practices. For helping in the elaboration of co-existence rules, for assessing their feasibility and their consequences as well as for setting up monitoring and control schemes, one should be able to forecast the fate of GM plants at the landscape level in the wide range of agro-ecosystems. Specific field experiments are necessary for understanding the basic phenomena involved but are difficult to extrapolate for such a perspective even if several studies have been carried out in order to broaden the scope of the evaluation: the inter-institute platforms in France or the Farm Scale Evaluation programme in the UK.

For addressing such a challenge, modelling is a key element. Models reproduce the functioning of agro-systems and take into account the relevant factors and processes as well as their interactions. They thus allow simulating the behaviour of various agro-systems in non-observed situations and on a long term basis.

Several modelling strategies

There exists a wide range of modelling approaches: mechanistic or statistical, simple or complex, conceptual or analytical, knowledge oriented or decision-making oriented. The choice of a modelling approach depends on the specific objective we focus on. Within the co-existence framework, models can help in:

- structuring gene flow knowledge and thus identifying those research gaps that should be addressed through specific field experiments (e.g. long distance issue);
- implementing optimal design of experiments for testing specific hypothesis;
- ranking farming systems according to adventitious presence in non-GM production;
- forecasting the behaviour of transgenes in cultivated and non-cultivated lands;
- testing *a priori* the efficiency of mitigation measures or regulation schemes on adventitious presence of GMO in a wide range of agro-ecosystems;
- identifying relevant biological indicators for an early detection of undesirable adverse effects and implementing optimal sampling for monitoring schemes;

Models implement direct and interactive links between scientific knowledge and decision-making.

Issues related to modelling

The oral and poster sessions on modelling present some of the models under development and address issues related to modelling:

- Modelling feral oilseed rape dynamics (Begg *et al*.; Garnier *et al*.);
- Modelling pollen dispersal (Damgaard C. & Kjellsson G.; Walklate P. & Sweet J.B.);
- Using models for assessing impacts of cropping systems and agricultural practices (Colbach *et al*.; Angevin *et al*.) as well as effects of new biological characteristics (Fargue A. *et al*.);
- Design of long distance gene flow studies according to modelling results (Devaux C. *et al*.)

Results from these papers as well as from ongoing studies and modelling approaches have raised several issues that should be further addressed in the discussion:

- What does model validation mean? Models are never "true" but can always be used for designing new studies or for decision making. In fact, the model fitting process should determine the exact range of applications and domains models can be used for.
- Should we use specific co-existence models? Although co-existence and environmental safety are separate issues, gene flow is a common scientific knowledge and gene flow models could be designed for both purposes;
- Should we design crop-specific or more generic models?

- What are the factors that should be taken into account for forecasting adventitious presence? Crop biology, relative sizes of fields, agricultural practices are major factors but other factors related to landscape fragmentation should be considered as well;
- At what spatial scale models should be designed? Available models for gene flow and ecological impacts focus mainly on the field level or on a small region (group of fields). However, decisions on co-existence can be made at the farm, the local and the regional levels. Up-scaling of models at different biogeographical levels should thus be made possible and easy to handle.

Spatial distribution characteristics of out-crossing probability

P.J. Walklate[1] & J.B. Sweet[2]
[1]Silsoe Research Institute, Wrest Park, Bedfordshire, MK45 4HS, United Kingdom
E-mail: peter.walklate@bbsrc.ac.uk
[1]NIAB, Cambridge, CB3 0LE, United Kingdom
E-mail: jeremy.sweet@niab.com

Abstract

This paper compares field data with the output from a new pollen mediated gene flow model. The comparisons are made for a range of experiments with genetically modified (GM) source crops of different size (i.e. 0.0064 ha to 6 ha) and different levels of fertility in an adjacent non-GM target crop (i.e. self-fertile to 80% male-sterile). The deterministic part of the model uses a standardised set of conditions as the basis for predicting the effects of atmospheric wind and local-scale insect dispersal. It predicts the spatial distribution of out-crossing probability for a given field geometry and the variation due to wind and insect exposure during the flowering period of a field trial is represented as a specially constrained random exposure factor. Estimates of exposure factors vary in the range 0.3 to 1.0 for the range of field experiments presented here and with these the deterministic model accounts for 94% of the variability in the data throughout space. The current model predicts lower total levels of out-crossing than might be expected to occur in practice because it neglects other distributed sources of GM pollen admixture, such as from contaminant seeds and volunteers. Further model development and experimental verification is therefore needed to quantify these effects.

Introduction

The development of genetically modified (GM) crops has precipitated the need for risk assessment of pollen mediated gene flow. In response to this need we have developed a model to predict the spatial distribution of out-crossing between different progenitor populations of oilseed rape based on similarity distribution characteristics of transport by atmospheric wind and insects (Walklate *et al*., 2003). The model offers new insight into the factors that determine gene flow. In particular, it has been shown that the spatial distribution of out-crossing is rather insensitive to the assumptions that are made about the spreading of pollen by insects over distances below a local plant-scale of 7 m. For the purpose of the comparison presented here we have neglected insect transport at distances greater than a local plant-scale of 1 m.

Results

Model predictions based on standardised environmental conditions are compared with a range of different field experiments in Figure 1. We show the results from samples taken on two transects of the target crop area for each source of experimental data. These represent different orientation relative to the source and the boundaries of other surrounding crop structures. Therefore it seems reasonable to expect the derived estimates of exposure to represent different effects of wind and insect exposure. However, without further information about the full time history of wind direction during the flowering period it is not possible to separate these effects. The extremes of mean exposure are represented by the solid and dotted lines in Figure 1. Estimates of typical exposure factor range between 1 and 0.3 across the full range of these experiments even though these were performed with good synchronicity between the flowering periods of source and target crops.

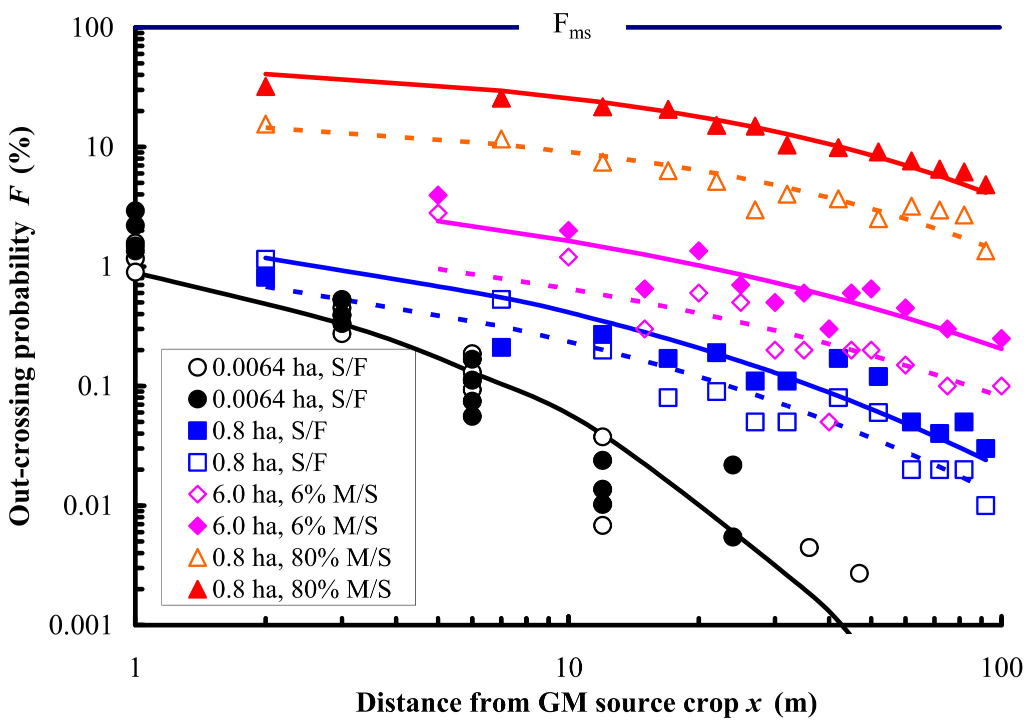

Figure 1. Comparison between measurements and predictions of out-crossing probability for a range of different field experiments based on GM source crops of different size and target crops of different fertility.

The results tend to represent high levels of competition between pollen from both GM and non-GM sources at the stigmas of target plants. The experiment for the smallest source and self-fertile targets produces the highest levels of competition. By contrast with this, very low competition can occur when the target crop contains a very high proportion of male-sterile plants or when male-sterile target plants are used in isolation (i.e. the asymptotic characteristic, represented by $F_{ms} \cong 100\,\%$ in Figure 1). This may be a condition worth avoiding because it is not capable of showing any decay with respect to space for levels of pollen above the detection limit of sampling.

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Pollen dispersal between fields of GM and non-GM oilseed rape: meta-analysis of available data and the possibilities for co-existence

C. Damgaard & G. Kjellsson
The National Environmental Research Institute (NERI), Department of Terrestrial Ecology,
Vejlsøvej 25, DK-8600 Silkeborg, Denmark
E-mail: cfd@.dmu.dk; gk@dmu.dk

Abstract

Results from model estimations based on existing data on gene dispersal in oilseed rape indicate that the GM dispersal by pollen to organic fields can primarily be limited by the use of isolation distances. The results also indicate that because of pollen dilution, large fields are better protected from GM pollen dispersal than smaller fields. For large fields (i.e. field width > 200 m), the risk of dispersal from GM fields by pollen will be limited (approx. 0.1%) at distances above 100 m. For small fields (i.e. width = 50 m), some GM pollen dispersal (up to approx. 0.3%) may be expected even with an isolation distance of 200 m. If the fields are very close, the use of additional protective buffer zones may be required.

Introduction

A meta-analysis of a number of field trials, where the gene flow between oilseed rape fields has been measured in e.g. England, France, Australia, Canada, USA, Denmark and Sweden, has been made in order to summarise and synthesise the obtained results. In most of these trials herbicide resistance was used as a genetic marker. The main questions, which were studied in the meta-analysis, are how the distance between fields and the field size of the pollen-receiving field affects the percentage of seeds containing GM.

Results and discussion

Results from the meta-analysis (Figure 1) show as expected that the GM-content of the seeds from a non-GM field will be reduced with increasing distance to the GM field. This is particularly evident for small fields. Furthermore the results indicate, that an increase of the size of the organic field (field width) has a relatively larger effect in reduction of the average GM-content than increasing the isolation distance. This effect is mainly caused by a dilution of the GM pollen by pollen from the pollen-receiving field. Other results from the study indicate that the use of a protective buffer zone (i.e., 5 m border, which is excluded from the harvest) will reduce the GM contents by approximately one third. Consequently, if the fields for some reasons cannot be separated by distance, then the use of a buffer zone may be a possible solution.

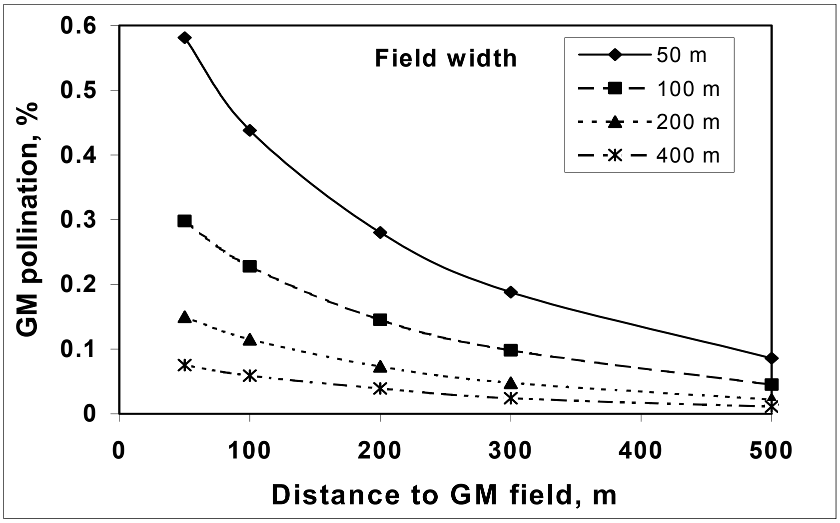

Figure 1. Total percentage of GM-containing oilseed rape seeds in an organic field in relation to isolation distance and the width of the field. Model results are shown for the upper 95% credibility level (in 5% of the fields, the average GM-content is expected to be higher than the shown value).

The results indicate that a critical level of 0.1% GM content in the organic crop of oilseed rape can be obtained by an isolation distance above 100 m if the field is at least 200 m wide (Figure 1). For small organic fields (width = 50 m), a low level of GM pollen dispersal (up to approx. 0.3%) may be expected even with an isolation distance of 200 m.

The results are based on the assumption that the GM field and the organic field are relatively equal in size. If the GM field is significantly larger than the organic field or if several GM fields are situated in the surroundings of an organic field, the extent of the GM dispersal will increase. It is also well known that the dispersal of pollen into a field may be irregularly distributed, and small pockets with higher concentration of GM-content will arise.

The assumptions for the model include that varieties with normal fertility are used. If hybrid varieties with male-sterile plants are cultivated in the organic field, the probability of GM pollination will increase depending on the percentage of male sterile plants. Furthermore, pollen dispersal from hybrids between GM oilseed rape and weedy relatives or from GM-volunteers in the surroundings has not been included in the model simulations.

When the relevant management measures are taken to reduce GM pollen dispersal, we expect that an isolation distance of 100 m will result in a GM content in the range of 0.1% to 0.3% of the total oilseed crop in the organic field (very small fields excluded). Single test samples collected from especially the field margins may however show a higher GM content.

Using the GeneSys model quantifying the effect of cropping systems on gene flow from GM rape varieties to rape volunteers for designing and evaluating scenarios for co-existence of GM, non-GM and organic crops

N. Colbach[1], F. Angevin[2], A. Fargue[3,4] & J.M. Meynard[3]
[1]UMR Biologie et Gestion des Adventices, INRA, BP 86510, 17 rue Sully, 21065 Dijon Cedex, France
E-mail: colbach@dijon.inra.fr
[2]Unité Eco-innov, INRA, BP 01, 78850 Thiverval-Grignon, France
[3]UMR d'Agronomie, INRA INA P-G, BP 01, 78850 Thiverval-Grignon, France
[4]CETIOM, BP 04, 78850 Thiverval-Grignon, France

Abstract

To quantify the effects of cropping systems on gene flow in time and in space, a model was built, evaluated and used to design new cropping systems that limit gene flow in case of coexisting GM, non-GM and organic rape crops.

Introduction

The GENESYS model (Colbach *et al.*, 2001) quantifies the effects of cropping systems on gene escape from rape crops to rape volunteers in neighbour plots and later crops. Its aim is to rank cropping systems according to their risk of gene flow and to propose low-risk systems.

Model presentation

The input variables of the model are the regional field pattern, the crop succession in each field, the management of each crop and border, and rape variety characteristics. The main output variables are, for each year and field/border, the adult rape plants, the newly produced seeds and the seed bank, both as densities and genotype proportions. The model is based on the annual life-cycle of the volunteer and cropped rape plants (seed bank, seedlings, adults...), which is simulated for each plot and year. For each stage, densities and genotype proportions are calculated. The relationships between the stages depend on crop type and management. Pollen and seed dispersal depends on plot areas, shapes and distances and on flowering dates.

Model evaluation

Four small regions were chosen. Their cropping history was recorded, based on the sensitivity analysis of the model (Colbach *et al.*, 2003). Rape volunteers (densities and genotypes) in crops and after crop harvests and the genotype proportions of the rape harvests were assessed. The comparison of simulated and observed variables shows that the model correctly ranks the situations according to their volunteer infestation and predicts volunteer densities accurately. The model is though very sensitive to several input variables that are difficult to estimate and usually based on literature (herbicide efficiency, seed loss during rape harvest, self-pollination rate, pollen emission). Moreover, volunteer populations in spring crops are sometimes and gene flow between fields frequently underestimated. Therefore, when using the model for simulations and advice, the user must not forget that actual overall gene flow would probably be larger than the simulated one, especially after rotations with frequent spring crops.

Model use for evaluating co-existence of varieties

GENESYS was used during a study for the European Commission to evaluate the consequences of coexisting GM, non-GM and organic crops (Angevin *et al.*, 2001). The proportion of GM seeds in conventional rape harvests and hybrid seed production was simulated depending on (a) the cropping system of the simulated farm and its neighbour farms and (b) the proportion of GM varieties in the neighbour farms. The simulated harvest contamination usually exceeded the studied thresholds (0.3 and 0.9%). It increased considerably in farms with small vs. large fields and slightly in organic vs. conventional farms. For each farm, changes in cropping practices were then simulated to identify techniques that minimise harvest pollution. Only time-consuming and expensive practices such as sowing set-aside would decrease harvest pollution below the acceptability threshold. Other practices such as clustering farm fields or permanently banning any rape crops next to fields used for hybrid seed production were also very efficient in reducing harvest contamination but are even more expensive and difficult to carry out.

Conclusion

GENESYS can be used to simulate cropping system effects on gene flow in agricultural regions, with a few precautions. It can be used to advise farmers, technical institutes and public decision-makers. Moreover, the mechanistic structure of the model makes it possible to integrate further varietal characteristics, to adapt the model to other conditions or species (see related papers in this book).

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Co-existence of GM, non-GM and organic maize crops in European agricultural landscapes: using MAPOD model to design necessary adjustments of farming practices

F. Angevin[1], C. Roturier[2], J.M. Meynard[3] & E.K. Klein[4]
[1] INRA, Unité Eco-innov, BP 01, F-78850 Thiverval-Grignon, France
E-mail: angevin@jouy.inra.fr
[2] INRA, DS PPV, 147 rue de l'Université, F-75338 Paris Cedex 07, France
E-mail: roturier@paris.inra.fr
[3] INRA, Département SAD, BP 01, F-78850 Thiverval-Grignon, France
E-mail: meynard@grignon.inra.fr
[4] UMR BIA 518, INRA/INA-PG, 16 rue Claude Bernard, F-75005 Paris, France
E-mail: klein@inapg.inra.fr

Abstract

MAPOD maize was conceived to quantify, on a spatial scale, cross pollination under real agronomic and climatic conditions. Numerous simulations were carried out to compare the effects of different cropping practices and certain varietal characteristics on gene flow as well as to determine the most efficient practices to ensure co-existence between GM, non-GM and organic maize crops.

Introduction

Adventitious presence of GM seeds in non-GM maize harvest results mainly in Europe from:

- Cross pollination between a GM crop and a neighbouring non GM maize field;
- GM impurities in seed lots.

Several research programs have been carried out and many results concerning pollen dispersal have been published now. Short distance pollen flow has been well assessed and pollen dispersal curves are now available (Klein *et al.*, 2003). Pollen flow occurs at long distances and is rather erratic. Climatic and agronomic conditions and field patterns have a major influence on cross pollination and therefore on rates of GM adventitious presence in harvests. In this context, field experiments are not sufficient to assess all existing variability in a wide range of agro-ecosystems. In order to forecast pollen dispersal and cross pollination as well as to design co-existence rules, a model comprising both theoretical knowledge and field trial results is required.

Model presentation

The MAPOD[1] model (Angevin *et al.*, 2001) can be used to predict, at the scale of the plot or a collection basin the rates of impurities for the harvest (or collection) due to cross-pollination under real agronomic and climatic conditions. It allows to the testing of the effects of isolation distances, cropping techniques, biological characteristics of different varieties on the rate of contamination by a transgene.

Studies presentation

In two studies (Le Bail & Meynard, 2001; Angevin *et al.*, 2002), we tested several scenarios for co-existence in representative maize production regions. Typical model farms were determined and the rates of impurities in maize harvests were estimated. We also simulated changes in cropping techniques in order to diminish rates of contamination in harvests to respect EC defined thresholds. Then, we went on to assess feasibility and costs of these changes.

Results and perspectives

These studies have highlighted several issues that need further research:

- The landscape fragmentation has a great influence on gene flow and its effect should be taken into account in modelling;
- Co-existence rules should involve at least three different decision levels: the field level with crop management practices, the “cropping system” within the farming systems strategy, the landscape and the regional levels.

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[1] MAPOD : Matricial Approach to Pollen Dispersal

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Modelling feral (volunteer) oilseed rape in relation to thresholds of impurity

G.S. Begg[1], M.W. Young[1], C. Hawes[1], J.W. McNicol[2] & G.R. Squire[1]
[1]Scottish Crop Research Institute, Invergowrie, Dundee DD2 5DA, United Kingdom
E-mail: gbegg@scri.sari.ac.uk
[2]Biomathematics and Statistics Scotland, Scottish Crop Research Institute, Invergowrie, Dundee DD2 5DA, United Kingdom

Abstract

Measurements of the arable seedbank have shown that feral (or volunteer) oilseed rape occurs in many arable fields at population densities around 100 m^{-2}. This is a small (usually <1%) proportion of the seedbank but a large enough density to cause impurities in crops of oilseed rape if seedbank seed germinates at the same time as the crop. A model of the life cycle dynamics of oilseed rape, which incorporated physiological (life-history) and environmental (management) variables, showed rates of decline and population densities consistent with field measurements. The model was used to explore combinations of management practices that reduced impurities below specified thresholds. The model indicated that an impurity threshold of 1% could be met within reasonable timescales (e.g. 5 years) but only with the rigorous application of measures to control ferals. Uncertainties in the understanding of feral population dynamics and emergence are considered.

Introduction

As seed pods of oilseed rape mature they weaken, leading to dehiscence and a loss of seeds when disturbed by wind, rain or harvesting. As a result, high densities of seed can be returned to the soil and incorporated into the seedbank. The prevailing environmental conditions can induce some of these seeds into dormancy to emerge later if subjected to appropriate germination triggers. As a result feral (volunteer) oilseed rape populations have become a common feature of arable rotations. The introduction of genetically modified (GM) varieties, and non-GM varieties with unique oil characteristics, means that the mixing of populations resulting from the presence of ferals could have important economic and environmental consequences if ferals persisted for long enough and at high enough densities. In the absence of definitive experimental information, a model was developed to explore the mechanisms linking feral dynamics to life-history processes and field management, to predict the persistence of GM or other notable ferals and to identify the management strategies necessary to restrict impurity below given thresholds.

Materials and methods

A population dynamics model was developed in which the density of individuals at three stages of the oilseed rape life-cycle was determined by a combination of life-history and management processes. The life-cycle stages considered are the seeds in the seedbank, emerged plants and mature seeds on the emerged plants. At each stage, the population is subject to mortality. Germination controls the flow of individuals from the seedbank into the pool of emerged plants. Seed dormancy, an important determinant of persistence in annual plant populations, is governed by temperature and depth dependence in the proportion of seeds that germinate. The per capita rate of seed production is determined by a density dependent function that incorporates the effect of intra- and inter-specific competition. The management events implemented within the model are sowing, cultivation, herbicide application and harvesting. At sowing, seeds are introduced to the depth strata of the seedbank at densities, which can be varied to represent different sowing practises. Cultivation redistributes seeds between the depth strata representing a range of cultivation types while removing emerged plants and seeds. Harvesting also acts to remove emerged plants and seeds although a proportion of seeds are returned to the seedbank representing the loss of seed at harvest. Finally, the application of herbicide is modelled by increasing the plant mortality rate over a predefined duration thereby determining the proportion of the emerged population that is killed (herbicide efficacy).

Results

A typical rotation was simulated in which winter oilseed rape was sown in the first year followed by winter wheat in the two subsequent years. This cycle was repeated six times. The first crop was considered to be the one introducing impurity into later crops and feral populations. The rate at which density declined was sensitive to basic life-history traits of fecundity, mortality and most acutely to dormancy, the fraction of a population germinating in a single flush. With no attempt to control the feral population the density of the contaminating population did not fall below 1% of the total oilseed rape density until 16 years after it was sown. Controlling feral populations with herbicide and preventing or minimising the loss of seed at harvest consistently reduced population persistence, although the model implied that individually these were unlikely to suppress feral populations sufficiently to meet the 1% threshold within a reasonable time. Using control measures in combination was effective, reducing seedbank density to 0.013 seeds m^{-2} and impurity to 0.12% within 3 years. However, the controls had to be consistently and stringently applied at levels that are outside the range in current practice.

Discussion

The control of dormancy in oilseed rape is a complex process that is affected by the genotype of the plants and the prevailing environmental conditions. Environmental effects and genotypic variation in dormancy traits may be responsible for much of the uncertainty in the persistence of feral populations. Current European legislation sets out 1% threshold above which conditions on labelling and traceability of GM food and feed must be imposed with proposals to impose lower thresholds in the future. The results of this study indicate that the current threshold could be met within a reasonable timeframe for oilseed rape but only with the rigorous application of feral control measures. The implication of this is that thresholds below 1% may prove very difficult to attain within, say, 5years of releasing the contaminating crop. The model will now be validated against feral populations left by GMHT crops.

Acknowledgements

This work was funded by the Department of Environment Food and Rural Affairs.

A first step for modelling pollen dispersal at the landscape level: determining the shape of dispersal functions at long-distance. The case of oilseed rape

C. Devaux[1], E.K. Klein[2] & C. Lavigne[1]
[1] Lab. ESE, bât. 360, Université Paris-Sud, CNRS UMR 8079, 91405 Orsay Cedex, France
[2] UR 546, INRA, Domaine Saint Paul – Site Agro Parc, 84914 Avignon Cedex 9, France
E-mail: celine.devaux@ese.u-psud.fr

Abstract

Simulation outputs from an oilseed rape pollen dispersal model compared to experimental data allowed us to show (1) that the individual pollen dispersal function of oilseed rape that is to be used at the landscape level to predict the consequences of the large scale cultivation of genetically modified plants has a fatter tail than an exponential curve and therefore (2) that pollen coming from different cultivars may be mixed at the landscape level.

Introduction

Modelling pollen dispersal at the landscape level is crucial for predicting the possibility of the co-existence of genetically modified (GM) and non-GM crops. Such modelling can be done on an individual plant basis by integrating the individual dispersal function of all pollen-emitting plants over the landscape. This individual dispersal function describes the probability that a pollen grain emitted at the (0,0) coordinates fertilizes a plant at any (x,y) coordinates (e.g. Lavigne *et al*., 1996; Klein *et al*., 2003). Determining the weight of the tail of that function (i.e. its shape for long distance dispersal events) is essential because it changes the composition of the pollen cloud over the landscape and thus may change the pattern of pollution of non-GM crops. However, pollen dispersal experiments are usually conducted on too small distances to enable researchers to discriminate between different types of function tails.

We therefore performed computer simulations and, from them, designed an experiment that allows us to determine the individual dispersal function family, according to the weight of its tail, in the case of oilseed rape. The choice of the function family is based on the comparison between the model and the experiment of the diversity and the differentiation of pollen clouds sampled randomly over the landscape.

Material and methods

Computer simulations

We built a landscape level pollen dispersal model that considers oilseed rape fields as point sources of pollen. Pollen is dispersed from every field with the same function but the quantity of pollen is adjusted from field to field according to its area. The landscape for this model was calibrated on a survey concerning the 2002 distribution of oilseed rape fields in a 10 * 10 km area surrounding the village of Selommes (Loir-et-Cher, France). Cultivars were determined for all fields. Dispersal functions were estimated from a prior experiment conducted on a one-ha field of oilseed rape (dispersal data in Lavigne *et al*., 1998). After pollen dispersal was simulated, the virtual pollen cloud was sampled randomly over the landscape and its composition in terms of oilseed rape cultivars was determined.

Field and laboratory experiments

Field experiments were conducted during the 2002 spring to sample real pollen clouds in the region of Selommes. We placed 50 male-sterile oilseed rape plants, divided into two temporal repetitions at 13 controlled scattered sites in the Selommes area during the flowering period. Mature seeds from each plant were sampled for genetic analyses using 4 microsatellites markers to determine the fertilising pollen genotype of each seed. These genotypes allow us to characterize the pollen cloud, in terms of cultivar composition, at each site and to compare it to that expected from the computer simulations.

Results

A geometric fat-tailed individual dispersal function generated pollen clouds that were diverse but little differentiated – clouds were composed of a mean of 6.5 (sd 3.2) cultivars - and therefore should produce high diversity between descents collected on male-sterile plants scattered across the landscape but little differentiation among the groups of these descents. In contrast, an exponential individual dispersal function would generate little diversity within seeds collected on traps but high differentiation among them – clouds were composed of a mean of 1.2 (sd 0.8) cultivars.

The 4 markers allowed us to obtain the genotypes of 1834 of the 1967 seeds sampled on 19 of the 38 surviving male-sterile plants from the two repetitions. The pollen clouds above these 19 plants were all diverse: pollen clouds were all composed of more than 2 cultivars.

Conclusion and perspectives

These preliminary results performed on a sample of the 2002 seeds suggest that the individual dispersal function has a fatter tail than an exponential curve. Consequently, exponential functions may be inadequate for modelling long-distance pollen dispersal because they severely underestimate long- distance events. Rigorous statistical analyses will be lead to formally assess this result and they could be obtained from an adaptation of classical methods of parentage analysis.

We reconducted the experiment on a larger scale in 2003 and in regards to the 2002 results.

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On-farm stewardship

Introduction to on-farm stewardship for co-existence of GM and non-GM crops

M.H. Entz & G. Martens
Department of Plant Science
Faculty of Agricultural and Food Sciences
University of Manitoba
Winnipeg, R3T 2N2
Canada
E-mail: m_entz@umanitoba.ca

Abstract

Co-existence of GM, conventional and organic crop production will require significant on-farm stewardship efforts. On-farm issues relevant to co-existence include field separation distance, which requires arrangements between neighbours; rotation management, which involves land tenure; purity of the seed supply; tillage system; manure management; volunteer plant control; other vegetation management; insect pollinator management; harvest, transportation and storage management; and straw management. These 13 issues are discussed in terms of the western Canadian experience of GM herbicide tolerant canola. It is suggested that the relative importance of these 13 on-farm stewardship issues is different when the GM trait confers a fitness advantage to the crop within the farming system compared with GM traits that do not confer a fitness advantage.

Introduction

On-farm stewardship issues are being discussed at this conference for one important reason - implementing a co-existence strategy begins on the farm. In order for co-existence to work, farmers must be aware of the major issues as well as all critical control points where problems could arise. The on-farm stewardship steps and critical control points are reviewed in this paper. On-farm stewardship issues are discussed in relation to the GM herbicide-tolerant (HT) canola experience in western Canada.

Framework for on-farm stewardship

Farming systems consists of a series of production steps including seed purchase, land preparation, crop production, pest management, harvest and crop storage. Production steps important in on-farm stewardship are identified in Table 1. Each production step provides an opportunity to practice on-farm stewardship.

It is important to recognize that the need for on-farm stewardship, and indeed the success of on-farm stewardship practices, will depend on whether the GM trait in question has a fitness advantage in the farming system. Traits that do not confer a fitness advantage over conventional types may be contained with an appropriate stewardship plan. On the other hand, GM traits that confer a fitness advantage to the crop within the farming system may require higher levels of on-farm stewardship.

Table 1. A framework for on-farm stewardship issues, practices and considerations.

Stewardship steps	Issues and considerations
Separation distance (field organization)	Management of separation distance requires co-operation between neighbouring farmers.
Land tenure	When a high proportion of land is rented or leased, agreements between farmers become more difficult.
Crop rotation planning	Crop rotation central to co-existence. Crop rotation interval depends on crop type and nature of the GM trait.
Seed supply	Properly managed certified seed production system should allow farmers to purchase uncontaminated seed.
Tillage system	Tillage system will affect seed bank persistence and opportunities for volunteer plant control.
Manure management	Manure, especially straw-based manure, often contains viable seeds. Composting and manure disposal systems must be adjusted accordingly.
Volunteer management	Volunteer plant management critical.
Vegetation management	Control of potential out-crossing plants important.
Insect pollinator management	Honey production or seed production may require alternative management.
Harvest management	Seed return to land during harvest creates future problems. Machinery sanitation critical.
Grain transportation	Extra care in grain transportation to avoid losses to the environment. Equipment sanitation.
Grain storage	Segregated storage and handling equipment required. Spillage during on-farm grain transfer common.
Straw management	Straw contains seeds. Movement of straw from farm to farm must be restricted or managed in new ways.

On-farm stewardship – the case of western Canada

M.H. Entz & G. Martens
Department of Plant Science
Faculty of Agricultural and Food Sciences
University of Manitoba
Winnipeg, R3T 2N2
Canada
E-mail: m_entz@umanitoba.ca

The prairie provinces of Western Canada (Manitoba, Saskatchewan and Alberta) contain 46 million ha of arable land and approximately 120,000 farms. Most farms are family farming units. There are 1,060 certified organic farmers in the region.

GM, HT canola was introduced into western Canada without a co-existence plan. Adoption of HT canola has been widespread and many benefits have been recognized. However, the lack of a co-existence plan has resulted in serious consequences for both commercial production as well as the seed production industry. Organic farmers have virtually stopped growing canola because of GM contamination.

The western Canadian experience can be used to examine what happens when GM, HT traits are introduced into broadacre agriculture without a co-existence plan. The experience also shows what happens when the GM trait in question has a fitness advantage within the farming system. Lessons from western Canada are invaluable in any future co-existence planning.

Each on-farm stewardship step is discussed separately:

Field separation

Western Canadian fields are separated into square mile "sections". Field size ranges from 80 acres (an 1/8 of a section) to 640 acres (a whole section). The only buffers between fields are narrow roadways (10 m) separating the sections, though wetlands and small forest patches also separate fields. Individual farms do not have contiguous fields. One farmer may cultivate many fields spread over a 20 km radius. Farmers do not typically co-ordinate cropping plans, though conventional farmers are increasingly aware of spray drift and genetic drift into organic fields. There are many examples of HT genes "drifting" into adjacent conventional canola fields. The main contamination occurs within the first 300 m of the field.

Land tenure

In western Canada 33,211,219 ha are cultivated by the actual land owners, while 13,164,889 ha are cultivated by farmers who rent the land from others, often on an annual basis (Census of Agriculture, 2001). Therefore, farmer-to-farmer land use arrangements on over 1/3 of the land area of western Canada are tenuous because of land tenure. The proportion of land rented or leased is increasing.

Crop rotation planning

Wheat dominates the prairie landscape. In 2001, wheat was grown on 10,272,805 ha (Census of Agriculture, 2001). The wheat production area is similar to the combined area of all other major crops (Canola: 3,740,666 ha; Barley: 4,316,401 ha; Peas: 1,340,431, Lentil: 700,000, Flax: 666,673; Sunflower: 70,000). Wheat is often grown every other year, while canola is grown every third or fourth year in the rotation.

Seed supply

Recent evidence has confirmed significant levels of GM contamination in pedigreed canola seedlots (Friesen *et al.*, 2003). Therefore, Canadian farmers cannot rely on the current pedigreed seed production system to ensure genetic purity.

Tillage system

Sixty percent of fields are managed using a no-till (25%) or minimum tillage (35%) system. The area under reduced tillage management is increasing. Adoption of reduced tillage systems is thought to reduce seed bank longevity in canola.

Reduced-tillage systems rely on broad spectrum herbicidal weed control in spring, mainly glyphosate. In an effort to maintain the use and efficacy of glyphosate in no-till systems, many no-till farmers avoid glyphosate tolerant canola and instead they use Liberty-link or Clearfield canola cultivars. However, cross contamination of seedlots has seriously compromised the strategy of avoiding glyphosate tolerant canola cultivars.

Manure management

Manure spreading is now recognized as a contributing factor to GM herbicide tolerant canola seed spread.

Volunteer plant management

HT canola was introduced without sufficient information on the potential for volunteers. After 7 years, "every system is in every field" (Kelner, pers. comm.). This statement refers to the fact that canola volunteers tolerant to all three herbicide systems (glyphosate, clearfield, liberty) are present as volunteers in most fields where canola has been grown. HT canola volunteer problems are growing and are especially severe in wet springs, when spring seeding is delayed.

Tactics for dealing with HT canola volunteers include spring tillage and herbicides. Glyphosate herbicide is widely used for preseeding non-selective weed control, especially in reduced tillage systems. The presence of glyphosate tolerant canola volunteers has forced farmers to use mixtures of glyphosate and phenoxy herbicides. However, this herbicide mixture is not appropriate in years when broadleaved crops are to be grown (e.g., canola, pea, lentil, bean, sunflower, etc.) since 2,4-D residues harm these crops. Therefore, glyphosate-tolerant volunteers are a particular concern for farmers using diversified, no-till or reduced-tillage cropping systems. This dilemma is complicated by the long seed bank life of canola seeds (up to four years in commercial fields).

Herbicide rotation (i.e., using herbicides with different modes of action to control the same plant species) could be an important form of volunteer management. However, extensive cross-contamination of canola seedlots has reduced this option.

Harvest losses of canola cause large seed bank inputs (Gulden *et al*., 2003). To minimize the effect of this seed bank, some farmers avoid fall tillage in an effort to increase seed germination in fall. These fall-germinating volunteer canola plants will freeze over the winter months and die. Some farmers in the tillage-dominant areas of western Canada now avoid deep tillage in autumn in an effort to minimize deep seed burial (Kelner, pers. comm.). The adoption of winter wheat, which is grown under no-till conditions to ensure snow insulation over winter, has further discouraged fall tillage of canola stubble.

Vegetation management

No special efforts have been made to eliminate weeds that might cross with HT canola since the risk of such crosses is thought to be low in western Canada.

Insect pollinator management

Bumble bees are common in western Canada and most flowering canola fields contain natural populations of bees. Bees are widely used for honey production, plus some are used in hybrid canola production. Leaf cutter bees are widely used in alfalfa seed production. To date, no regulations or recommendations have been developed regarding bee use in agriculture as it relates to co-existence.

Harvest management

Harvest losses form the basis of the volunteer crop problem, though some seed shattering also occurs. Optimum combine harvester setting is important. Some western Canadian farmers use a chaff collection system, which is highly effective in collecting volunteer seeds (Shirtliffe and Entz, 2003). The McLeod Harvest system is an innovative machine design that significantly reduces seed return to the land (http://www.mcleodharvest.com/). Approximately 20 such machines are now used on commercial farms in the region.

Transport from field to farm storage

Grain is transferred from the combine harvester directly into a truck or first to a grain cart and then to a truck. These trucks transport the grain to farm storage, a distance of up to 20 km. Some grain (less than 30% of harvest) is transported from the field directly to the silo. While it is recommended that farmers cover grain loads, this is rarely done when transporting grain from the field to farm storage. Road travel combined with windy conditions lead to grain blowing off trucks during transport from the field to farm storage.

Grain storage
Typically, well over 50% of the grain harvested on farms is stored on the farm. Transfer between granaries is common, especially where grain aeration or grain drying is practiced. Spillage is common during grain storage and granary transfer.

Straw management
Canola straw is rarely removed from the field; though canola straw was used as livestock feed the drought years of 2001, 2002 and 2003. Cereal straw is commonly removed from fields, and used as livestock feed, bedding as well as raw material for manufactured fiber products.

Critical points for on-farm stewardship

Whether or not a particular production step, as outlined in table 1, has a more important role than another depends on the crop type, and whether the GM trait confers a fitness advantage to the crop within the farming system.

GM trait does not confer fitness advantage
In cases where the GM trait does not confer a fitness advantage to the crop within the farming system, most of the 13 issues outlined in table 1 could be viewed as critical steps in on-farm stewardship. Purity of seed supply would still rank higher than other factors, however.

GM trait confers a fitness advantage
In cases where the GM trait confers a fitness advantage to the crop within the farming system some on-farm stewardship steps may be more critical than others. In the case of HT canola in western Canada, a contaminated seed supply created a major problem. Furthermore, insufficient separation distances as well as "tight" rotations also contributed to the problem. Therefore, the four most critical on-farm stewardship steps in the Canadian canola example are: seed supply; field separation; crop rotation; and land tenure (which contributes to problems in field separation and crop rotation). Other on-farm stewardship steps play a somewhat less critical role, but remain very important in the long-term.

Perspective and conclusion

On-farm stewardship approaches require a systematic analysis of at least 13 steps within the farming system. While all of the on-farm stewardship steps are important, some are more critical than others. In instances where the GM trait confers a fitness advantage to the crop in question, crop rotation; purity of seed supply; land tenure and field separation issues are of paramount importance. Where the GM trait does not confer a fitness advantage, all of the stewardship steps can play an equally important role in a co-existence system.

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Modelling the effect of management strategies on the seed bank dynamics of volunteer oilseed rape

I.A. Rasmussen[1], K.H. Madsen[1,2] & N. Holst[1]
[1] Danish Institute of Agricultural Sciences, Department of Crop Protection, Research Center Flakkebjerg, DK-4200 Slagelse, Denmark
[2] Center for Bioethics and Risk Assessment, KVL, Groennegaardsvej 8, DK-1870 Frederiksberg C, Denmark
E-mail: ilsea.rasmussen@agrsci.dk; khm@kvl.dk; niels.holst@agrsci.dk

Abstract

Seeds of genetically modified (GM) crops remaining in the field after harvest may later emerge as volunteers. These can affect the purity of subsequently harvested crops and cause an uncontrolled flow of GM pollen into the agro-ecosystem. Management strategies that reduce these long-term effects of GM crops include (1) minimizing the input of GM seeds into the soil, (2) preventing proliferation by volunteers and (3) fast depletion of the soil GM seed bank. Formulation of such strategies requires knowledge of the biology of the GM plant (seed loss at or before harvest, seed survival, germination and emergence rates, reproduction, etc.) and of the effect of management practices on the different life stages.

Of the GM crops considered for cultivation in Denmark, oilseed rape is one of the more problematic, since there is a large seed loss at harvest (5-10%, but up to 50% have been recorded Thomas *et al*., 1991), and the seeds are viable for 5 years or more. (Pekrun *et al*., 1998)

We extended an existing model ("FieldWeeds") of weed population dynamics under different management strategies (Rasmussen & Holst, 2003) to describe the population dynamics of oilseed rape volunteers. Based on already published data and expert knowledge, we derived a model to predict whether the seed bank of oilseed rape would increase or decrease under different management practices. Running the model with different scenarios we thus estimated how long time it will take to deplete the oilseed rape seed bank in different crop rotations and with different management practices.

With an alternative approach Madsen *et al.* (1999, 2002) developed a model to investigate the evolution of herbicide resistant weed populations and their effect on herbicide use. Results from these two simulation approaches will be described along with possible pitfalls, limitations and uncertainties.

The models

The objective of the FieldWeeds model is to estimate the time to deplete the oilseed rape seed bank in different scenarios. The model is designed with a user interface enabling the inclusion of all types of crops, agricultural interventions and of additional weed species. Growth and development of crops and weeds runs in thermal time, which makes the model generally applicable. There is no estimation of gene flow, either through pollen, or through seed movement; however, for the dynamics of GM oil-seed rape in a single field, these factors are expected to be of less importance. Effects of soil type or precipitation are not considered.

The objective of the Madsen model is to determine the amount and frequency of herbicide use, the development of the rape and hybrids and the resistance in the weeds. The crop rotation is restricted to one type (oil-seed rape and three years of cereals). The input variables can be varied according to new knowledge. There are no interactions with soil or climate.

What models can be used for is highly dependent on their objective. In this case both can be used to explore the population dynamics of GM oil-seed rape in a particular field, since both include the seed-bank as a component. FieldWeeds is the more universal of the models, since all parameter values can be changed according to the environment, and the growth process sub-models are dependent upon weather (ambient temperature). The germination of the weed is spread out in time, whereas in the Madsen model all weeds germinate at once. The Madsen model is a less universal model, since there is only one possible crop rotation, the changing scenarios are the use of herbicides, but this model includes more weed species, while FieldWeeds can only treat one species at a time.

For both models, the output of the model is shown without variation or confidence intervals. However, FieldWeeds is designed for sensitivity analysis, where all parameters can be tested automatically at different values to determine the sensitivity of the model to each parameter.

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The potential impact of volunteer rape as a link between previous and current rape crops – its relevance for managing HT-rape

C. Pekrun, S. Gruber, P.J.W. Lutman[1] & W. Claupein
Institute for Crop Production and Grassland Research
University of Hohenheim, 70599 Stuttgart, Germany
E-mail: pekrun@uni-hohenheim.de
[1]Rothamsted Research
Harpenden, Herts, AL5 2JQ
United Kingdom

Abstract

A whole set of management options exists that can be used to minimise the risk of HT-volunteer oilseed rape being present in subsequent crops. These include rotation, cultivar choice, harvesting methods, post-harvest tillage and effective volunteer control. Taking all management options together the number of volunteer oilseed rape plants acting as a pollen and seed source should be very low under European conditions.

Introduction

Oilseed rape seeds can persist in soil for several years and produce volunteer rape in succeeding crops. In Canada herbicide tolerant volunteer Canola is one of the most abundant weeds (Légère *et al.*, 2001). It can require additional herbicide applications and therefore may increase the costs for weed control in HT-cropping systems. In Europe this situation should be avoided. In this paper we present long-term research on persistence and population-dynamics of volunteer oilseed rape indicating possible means of control to facilitate the co-existence of HT-oilseed rape with conventional and organic oilseed rape production.

Management options

Rotation

Growing crops where control of volunteer oilseed rape is easy and cheap is the most effective means to avoid volunteer problems. In a rotation with a large proportion of cereals volunteer oilseed rape will play a minor role. As long as the type of oilseed rape cultivar is not altered and oilseed rape is grown every fourth or fifth year volunteers in rape crops will emerge at densities that can be tolerated.

Cultivar
Cultivars differ in their ability to persist (Gruber *et al.*, 2003). This finding implies a scope for breeding low-dormancy types. Alternatively, farmers may be supplied with information on dormancy of current rape cultivars and chose low dormancy types where necessary.

Harvesting
Harvesting losses are the origin for volunteer problems. Assessments of seed numbers on the stubble at various sites and in a number of years (10 assessments) generated a wide range of values: 1,300 - 14,500 with an average of 5,300 seeds m^{-2}. Seed losses can be minimised by careful harvesting using appropriate machinery and choosing the optimal date for combining.

Post-harvest operations
Some of the seeds that are left on the stubble may develop secondary dormancy and persist for a number of years. In field experiments in the UK, Austria and Germany between 0 - 10% of seeds broadcast on a stubble in July-August were still present in the soil the following spring (Pekrun, 2003; Gruber *et al.*, 2003). Persistence was significantly reduced when the first tillage operation post-harvest was delayed for two or four weeks. Few seeds persisted in no-tillage plots. However, in these plots up to 1 volunteer plant m^{-2} emerged within the following winter wheat, flowered and set seed and therefore could function as a pollen source for neighbouring rape crops or refill the seedbank. The reasons for these results are unknown but mirror the situation in arable farming in Canada where HT-rape is of considerable importance in no-tillage systems (Gulden, 2003) and show that some sort of tillage should be done to avoid volunteer problems in future crops.

Controlling volunteers within the following crops
Modelling runs showed that efficient control of volunteers in the rotation will be important to keep volunteer populations at a low level. Even a slight increase in control efficiency, e.g. from 85 to 90%, can increase the decline in volunteer populations significantly.

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On-farm monitoring and auditing of field scale genetically modified crops in the UK – a co-existence case study

J. Kilpatrick & S. Twining
ADAS Consulting Ltd, Woodthorne, Wergs Rd., Wolverhampton WV6 8TQ
United Kingdom
E-mail: john.kilpatrick@adas.co.uk

Abstract

In May 1999 SCIMAC (the Supply Chain Initiative on Modified Agricultural Crops) published a Code of Practice on the introduction of genetically modified (GM) crops and Guidelines *for* growing newly developed herbicide tolerant crops. In 1999 the UK Government established a three-year project called the Farm Scale Evaluations (FSEs) aimed at evaluating the comparative effect on farmland biodiversity of the management of GM Herbicide Tolerant (GM-HT) and equivalent non-GM crops. The GM crops within the trial were being grown on a field scale on commercial farms so SCIMAC Code of Practice and Guidelines were implemented. In order to monitor compliance with the SCIMAC Code of Practice and Guidelines ADAS Consulting Ltd were contracted to provide third party audits of all growers within the FSE Trial. Over the three-year reporting period there was a high level of compliance with the SCIMAC Code of Practice and Guidelines. No major non-conformances were found in the eight critical control points identified by SCIMAC. Information from the audits has been used effectively to ensure that high standards are maintained in the growing of GM-HT crops.

Introduction

In May 1999 SCIMAC (the Supply Chain Initiative on Modified Agricultural Crops) published a Code of Practice on the introduction of genetically modified (GM) crops and Guidelines for growing newly developed herbicide tolerant crops (SCIMAC, 1999 a & b). These publications were both formally endorsed by the UK Government. They establish a consistent, industry wide approach to information supply for GM crops from seed to primary end product, and promote best practice guidelines for the on-farm management and identity preservation of GM herbicide tolerant crops. In 1999 the UK Government established a three-year project called the Farm Scale Evaluations (FSEs) aimed at evaluating the comparative effect on farmland biodiversity of the management of GM Herbicide Tolerant (GM-HT) and equivalent non-GM crops (Firbank *et al*., 2003). The trials were established following an agreement between Government and SCIMAC that during the trial period no commercial introductions of GM crops would be made. The GM crops within the trial were being grown on a field scale on commercial farms so SCIMAC Code of Practice and Guidelines were implemented. In order to monitor compliance with the SCIMAC Code of Practice and Guidelines ADAS Consulting Ltd were contracted to provide third party audits of all growers within the FSE trial.

Method

The SCIMAC Code of Practice outlines the need for successive transfer of supplementary information at strategic points along the food chain. Because of the experimental nature of the FSE programme (i.e. none of the harvested produce entered the commercial food or feed chain), auditing of the growers has been limited to the farm only, but does include information on the supply of information from the seed supplier and the maintenance of the integrity of the crop post harvest.

In an adaptation of Hazard Analysis and Critical Control Point methodology (NAS, 1995), SCIMAC identified eight Critical Control Points at which non-conformance with the SCIMAC Code of Practice and Guidelines could result in failure to observe best agricultural practice or to maintain the identity of the GM crop and other non-GM crops. For the purposes of the audit, these Critical Control Points would be treated as major non-conformances if the Code of Practice and Guidelines were not followed.

1. Seed delivery, storage and handling
2. Drilling operations, including cleaning
3. Handling of surplus seed
4. Separation distances
5. Field operations, including harvest preparation
6. Harvesting operations
7. Transport and storage of GM bulk
8. Record keeping and post harvest monitoring

Auditing is a systematic examination to measure compliance with a pre-determined system. Any variation from procedure, practice or performance standard is classed as a non-conformance. Non-conformances can be of a major or a minor nature. Extrinsic audits, or third party audits, are carried out independently of the auditee or other interested party. The audits were carried out by a team of four ADAS or Scottish Agricultural College (SAC) consultants. All are agronomists with BASIS and FACT (Agrochemical and Fertiliser qualifications). They also received specific training in auditing procedures and the SCIMAC Code of Practice and Guidelines. Any potential for conflict of interest, such as the auditor being involved in the farm in another capacity, were checked at the start of the season and growers assigned to an auditor from outwith the area.

The audit comprised a two part process of telephone/postal interview with all growers and an on-farm verification audit of 20% of all crops. Two separate audits were done for each grower, during the growing season and again after harvest. The auditors assessed the answers given for each question and marked them as satisfactory, insufficient, or not satisfactory, according to agreed assessment criteria. Any events of suspected non-conformance under the eight critical control points identified by SCIMAC, were brought to the attention of SCIMAC for investigation.

Results

All farms within the FSE received a remote audit in each of the three years of the trial. A total of 257 crops were audited over the three year period, on 97 unique farms. Some farms were part of the FSE in more than one year and several farms had more than one trial site. The plan of achieving 20% of crops receiving an on-farm verification audit, was exceeded in all years and all crops, with around 30% of all crops being audited on farm. 40 out of the 97 unique farms received an on-farm verification audit at some stage in the three year reporting period. Most farms only received one on-farm verification audit. This level of follow-up ensures confidence in the results.

Overall there was a very high level of compliance and no CCP queries were upheld. During the remote or farm audits if any CCP question did not appear to be answered adequately the query was sent to SCIMAC.

A detailed analysis was completed on all the questions for each crop group each year. Main points from this analysis are:

Areas of good conformance

- Access to Codes of Practice
- Staff training
- Use of BASIS qualified agronomists
- Separation distances
- Information from the seed supplier
- Storage of seed before and after drilling
- Prevention of spillage at drilling and at harvest
- Harvest management
- Post harvest storage
- Post harvest field management
- Record keeping
- Security of records

Areas of poor conformance

- Documented weed control policy
- Documented policy to monitor fields post harvest
- Confusion over formal requirements to notify neighbours and/or maintain records of discussions

In many cases, these areas of poor performance were directly attributable to the experimental nature of the FSE process, with the requirements or responsibilities of individual growers addressed, according to consent conditions, by consent holders. In other cases, however, farmers mentioned their thought processes but did not necessarily have these formally documented.

The results from the on-farm verification audits were evaluated centrally, checking that appropriate information had been recorded. The results from this indicate that the telephone audit was a reliable and accurate means of determining conformance. On only two farms was it found that the information provided was poorer than expected.

Conclusions

Over the three year reporting period there was a high level of compliance with the SCIMAC Code of Practice and Guidelines. No major non-conformances were found in the eight critical control points identified by SCIMAC. Information from the audits has been used effectively to ensure that high standards are maintained in the growing of GM-HT crops.

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Genetically Modified Crops and Agricultural Landscapes: Spatial Constraints on Farmers' crop rotations

J. Abildtrup & M. Gylling
Danish Research Institute of Food Economics
Rolighedsvej 25
DK-1958 Frederiksberg C, Denmark
E-mail: jens@foi.dk

Abstract

Based on empirical data on farm and field structure in a case study area the implications of separation distances to ensure co-existence between genetically modified (GM) and non-GM crops for farm management are analysed. The results indicate that only a few fields can be managed independently of the adjacent farms' fields. However, the need for adjustments in crop rotations to comply with the separation distances is limited.

Introduction

Buffer zones are suggested as one of the measures in a protective scheme to ensure co-existence between GM and non-GM crops (Fødevareministeriet, 2003). Buffer zones imply that a non-GM crop may not be grown within a certain separation distance of a GM crop. For example, a separation distance of 300 metres has been proposed to avoid pollen influx from the GM oilseed rape to the non-GM variety. A farmer choosing to grow a GM oilseed rape will have to coordinate the crop rotation with neighbouring farms having fields located within the buffer zone of the field with the GM-crop. If a neighbouring farm is also growing the oilseed rape but a non-GM variety there may be a need for adjustment of the crop rotation at one or both of the farms to comply with the demanded separation distance. The need for coordination with neighbours and adjustment of rotations to comply with the separation distances implies costs for the farmers. The impact of buffer zones on farming depends on the farm size and the location of its fields as well as the field size and form. In the present study empirical data on farm and field structure is used to estimate the potential conflicts - or demand for coordination - for different scenarios for adaptation of GM-crops and separation distances.

Material and methods

The study is carried out in a case study area of 10x10 kilometres located in an agricultural landscape in the central part of the Danish peninsula, Jutland, using digital field map and a database linking fields with the farms managing the fields (Dalgaard and Nielsen 2002). A basic assumption in the analysis is that a farm cannot have a GM and a non-GM variety of the same crop. The demand for adjustments in crop rotations to comply with the separation distances is estimated using stochastic simulation, i.e. the farms growing a GM and non-GM crops are selected randomly and the number of violations of the separation distances is estimated as an average of the results of a number of simulations. In the case study area 66% of land is used for agriculture and 258 farms larger than 5 hectares are having fields within the case study area.

Results

Table 1 reports the number of transgressions of the separation distances in the case of oilseed rape in six scenarios deviating with respect to i) area cropped with oilseed rape, ii) the share of GM oilseed rape, and iii) the separation distances. It should be stressed that the results represents the number of transgression when farmers do not at all coordinate their crop rotation with the farms with adjacent fields.

Table 1. Transgression of separation distances in winter oilseed rape with separation distance of 100, 150 and 200 m.

Sepa-ration distance	Oilseed rape area of cultivated area	Share of area with GM oilseed rape of total oilseed rape area (%)	Number of oilseed rape fields	GM fields that do not comply with separation distances	GM fields that do not comply with separation distances
Metres	%	scenario	(number)	% of GM oilseed rape fields	% of all oilseed rape fields
150	2.5	10	32	10	1.1
150	2.5	50	32	6.2	2.7
150	5	10	68	15	1.9
150	5	50	72	12	5.7
100	5	50	72	8.2	4.1
200	5	50	72	15	7.5

In the study it was found that there is a high demand for contact between farms because only 4-8% of the farm's fields are located more than 100 meters from a neighbouring farm's field. However, the results above show that it will only be in a relatively few cases that adjustments of the rotations will be required to comply with the separation distances of 100 meter.

Discussion

There will be a relatively great need of contact between the neighbouring farms. However, it will only be in the relatively few cases where the neighbour intends to grow a similar non-GM crop that farm adjustments will be necessary. It should be stressed that the conclusions of the study are based on only one case area. As the area generally has smaller farms and fields than Denmark on average, the estimated need of adjustments would probably be higher than for Denmark as a whole.

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GMO-free areas, nature conservation and organic farming - results of a survey of experts' opinion

J. Hoppichler
Federal Institute for Less Favoured and Mountainous Areas, Marxergasse 2, 1030 Vienna Austria
E-mail: josef.hoppichler@babf.bmlfuw.gv.at

Abstract

The paper describes results of a survey among persons in administration and politics in Austria, who are confronted professionally with problems and possible environmental effects of releases of GMOs (experts in a broader sense). They were in majority of the opinion that the relationship between protected areas and the application of genetic engineering in agriculture needs to be clarified. They see great deficits in relation to the needs of organic farming and in GMO-free on-farm management of Plant Genetic Resources and prefer a concept of defining "large, GMO-free ecologically sensitive areas". As a result of this survey the paper provides some perspectives concerning GMO-free areas and reflects on the problem of thresholds and liability for GMO-pollution.

Introduction

Since in Austria organic farming is practised in a relatively high density (about 10% of farms and/or 10% of UUA is organic) and since the public discussion on releases and marketing of GMO intensified in the late 90ies, the adventitious presence of GM crops in organic agriculture was soon anticipated as one of the main future problems. In order to assess the chances for a social consensus on the idea of GMO free areas and to evaluate the problems concerning organic agriculture we conducted in 1999 a survey among persons (n=150), who are confronted professionally with problems and possible environmental effects of the release of GMOs (experts in a broader sense - administrative personnel, politicians, scientists, and NGOs in agriculture, nature and environmental protection) (Hoppichler, 1999).

Main results

- The relationship between protected areas and the application of genetic engineering in agriculture needs to be clarified. 75% of the responding experts think that the use of GMOs causes a significant disturbance in areas of nature protection.
- The concept of defining "large, GMO-free ecologically sensitive areas (e.g. the size of an Austrian Federal Province)" was supported by the majority of the experts (73%).

- There are great deficits in relation to the needs of organic farming. 89% of the respondents called for GMO-free areas for breeding and propagating organic seeds.
- As main strategies for assisting organic farming in coping with the problems of genetic engineering, the experts recommend to support GMO-free production through agricultural environmental programs (60%) and through regional food processing and marketing structures (60%), followed by defining GMO-free areas for seed breeding and multiplying (57%) and demarcation of "large, GMO-free ecologically sensitive areas" (also 57%). In response to the question as to who should bear the additional costs of analyses to ensure freedom from GMOs, 42% of the experts tend towards the "polluter pays" principle and claiming compensation from the seed industry (all percentages multiple responses).
- The great majority of experts are of the opinion that the in-situ conservation and on-farm management of Plant Genetic Resources should be GMO-free.

Perspectives

Despite the fact that the actual research on co-existence of GM, conventional and organic crops mainly focuses on small scale management solutions (e.g. isolation distances or buffer zones), there is a need to introduce GMO-free areas especially in regions of small-scale agriculture and/or in regions with higher densities of organic farming (Müller, 2003). All proposed technical solutions for co-existence and their applicability depend very much on the level of thresholds accepted in organic and GMO-free agriculture as well as on the regime of enforcement of measures including the rules of liability for direct and indirect economic loss (AEBC, 2003). The introduction of co-existence measures is also a question of economic incentives. As long as organic producers, but also conventional producers, are responding to consumer demand for as little GM material as possible in their food (freedom of choice), the threshold should not exceed the 0,1% level. This for sure would create big problems with separation distances and other on-farm management measures.

Additionally co-existence has also a social dimension, which includes social acceptability within the farming community and in relation to consumers. As a result GMO-free areas, without regard to whether introduced by voluntary or mandatory means, could be one of the main solutions to reach a certain kind of co-existence. As a counterpart some sort of GMO areas may be created. Depending on the EU-seed-thresholds there will also be a need to define closed areas for seed propagating including seeds for organic agriculture. And if the current results of the British Farm Scale Evaluation concerning impacts on biodiversity are taken serious, GMO-free environmentally sensitive areas for nature protection or even large GMO-free biosphere reserves could be a possible perspective (Hoppichler, 2000).

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Poster Session

Development of an adventitious pollen risk assessment model

R.W. Arritt, M.E. Westgate, C.A. Clark, A. Fonseca & J.M. Riese
Department of Agronomy, Iowa State University, Ames, Iowa 50011-1010
USA
E-mail: rwarritt@bruce.agron.iastate.edu

Controversy over use of genetically modified (GM) crops has led to requirements for evaluating and controlling the potential for inadvertent out crossing in open pollinated crops such as maize. In response we are developing an Adventitious Pollen Risk Assessment (APRA) model that couples physical and biological processes affecting maize pollination. The model is based on field and laboratory measurements of maize pollen transport and viability. The core of the APRA model is a Lagrangian model of pollen dispersion. The Lagrangian method is adopted because of its generality and flexibility: the method accommodates flow fields of arbitrary complexity; and each element of the material being transported can be identified by its source or other properties of interest. The Lagrangian approach thus allows one to trace the environmental conditions to which the pollen has been exposed in its travel from tassel to silk, so that the physical effects of wind and turbulence on pollen dispersion can be considered together with the biological aspects of pollen release and viability. Predictions of pollen dispersal by the Lagrangian model compare well both to observations and to results from a standard Gaussian plume model.

APRA includes evaluation of the biophysical factors that affect pollen viability. Preliminary results indicate that pollen viability can be quantified by an "aging function" that accounts for temperature, humidity, and time. As expected the reduction of viability is greatest for high temperature and low humidity. Decline of viability with time is nearly linear when elapsed time is multiplied by a factor that depends on temperature and vapor pressure deficit. A second biological component of the APRA model is specification of the diurnal trend of pollen shed, and especially the relation of pollen shed to ambient meteorology. We present results from a field experiment in the 2003 growing season that measured diurnal pollen shed and its relation to local temperature, humidity and wind.

A study to evaluate co-existence of GM and conventional maize on the same farm

F. Benetrix, D. Bloc, X. Foueillassar & B. Naibo
ARVALIS – Institut du végétal, 64121 Montardon
France
E-mail: f.benetrix@arvalisinstitutduvegetal.fr

Little operational information is available in Europe about real conditions of co-existence of GM and conventional maize on the same farm. In 2002, an Operational Program for GM Crops Evaluation (POECB) was set up to obtain data in order to ensure segregation and traceability of GM crops from field to storage.

This study has been conducted under control of a scientific committee with scientists and experts from INRA, IRTAC, technical institutes, industries, maize and seeds producers association. Four working groups included experts from national research, technical institutes and stake-holders of maize chain have been created to evaluate and predict the distribution of cross pollination, the identification of appropriate quality control and procedures for co-existence, and the impact of Bt technology on corn borer in real field conditions in France.

Perspectives of GM crops co-existence in Czech farming systems

M. Čeřovská & J. Soukup
Czech Agriculture University in Prague, AF-KOPRA, Kamýcká 129, 165 21, Praha 6
Czech Republic
E-mail: marie.cerovska@centrum.cz; soukup@af.czu.cz

This study applies to current status and possibilities of GM-crops co-existence with conventional and organic farming in the Czech Republic. It is focused on those crops most likely to be grown in the Czech Republic as GM-crops: oilseed rape, maize and beet.

The Act No.153/2000 „On the use of GMOs and their products“ regulates usage of GM-crops in the Czech Republic. Till now there are no registered GM varieties for planting. There is GM-maize MON810 (Monsanto) in approval procedure since January 2003. The only approved GM-crop is imported RR soybean (feed component, not for planting). The Czech GMO field research focuses on maize, oilseed rape, potatoes, fibre flax and plum tree (Ministry of Environment of the Czech Republic – www.env.cz).

Economic benefit by introducing GM-crops in the Czech Republic is expected by beet and oilseed rape in consequence of the reduced herbicide costs. Beet areas represent about 2.5% and rape areas about 12% of entire arable land in the Czech Republic (3.07 m.s ha). We expect also economic benefit by farmers planting maize in consequence of the reduced insecticide costs (Alcalde, 2003). The most of maize (7%) planting in the Czech Republic (9.5% of entire arable land) is used as fodder crop for silage production. To consider there are the costs by farmers producing seed and also influence of changes in EU labelling of GM products on the Czech legislature.

A key aspect by producing GM crops is consumer. Consumers behaviour, according to the rule of free choice, influences indirect the extent of particular production. A consumers study, carried out in the Czech Republic in 2001, have shown that the Czech consumers want to have free choice to buy the products of different origin – organic, conventional and GMO and most of them think that GM-crops are "future crops". For the Czech consumer it is more important labelling of Bio-products compared with products containing GMO (Čeřovská, 2002).

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Brassica napus is aerodynamically unsuited to cross-pollination by wind

J.E. Cresswell[1], T.W. Davies[2], M.A. Patrick[2], F. Russell[3], C. Pennel[3], M. Vicot[3] & M. Lahoubi[2]
[1]School of Biological Sciences, University of Exeter, Hatherly Laboratories, Prince of Wales Road, Exeter, EX4 4PS
United Kingdom
[2]School of Engineering and Computer Science, Harrison Building, University of Exeter, EX4 4QF, United Kingdom
[3]Institut Catholique d'Arts et Métiers (ICAM), 6 rue Auber, 59046 Lille Cedex
France

We used Computational Fluid Dynamics to study the aerodynamic interactions between a *B. napus* flower and a windborne suspension of its pollen. In combination with wind-tunnel experiments, our results demonstrate that the floral architecture of canola severely restricts its potential for cross-pollination by wind. Specifically, flowers captured pollen from the air stream in amounts that, at best, were commensurate with the ambient airborne pollen concentration and the stigma's small surface area, but only when they faced upwind because otherwise their petals sheltered the stigma completely. The radial deployment of flowers about the canola stem and their tendency to be blown temporarily into a downwind-facing position further restricts the capture of airborne pollen. The attenuation in the concentration of airborne pollen that occurs with distance from a source crop, coupled with the ineffectiveness of the flowers in capturing airborne pollen, implies that the wind is not the primary vector for gene dispersal over long distances in *B. napus*, which instead must be insects.

Outcrossing frequency at large distances from a GM oilseed rape field

I. Degrieck[1], E. Van Bockstaele[1,2] & M. De Loose[1]
[1] Ministry of the Flemish Community, Centre for Agricultural Research (CLO), Department for Plant Genetics and Breeding (DvP), Caritasstraat 21, 9090 Melle, Belgium
E-mail: i.degrieck@clo.fgov.be; e.vanbockstaele@clo.fgov.be; m.deloose@clo.fgov.be
[2] Ghent University, Faculty of Agricultural and Applied Biological Sciences, Department for Plant Production, Coupure Links 653. 9000 Gent, Belgium
E-mail: erik.vanbockstaele@rug.ac.be

Ever since the discussion started in Europe on the effects of GMO's on the environment, gene flow from GM plants to conventionally bred plants of the same species or to its close relatives has been one of the major concerns. This is particularly relevant for oilseed rape (*Brassica napus* L.) because this species is partially allogamous and presents several wild relatives growing nearby the cultivated areas. It is well known that crop-to-crop gene flow occurs in oilseed rape (Eastham & Sweet, 2002; Reboud, 2003). Among other things this raises the question whether growing GM and non-GM crops together in respect to co-existence is possible. This is because cross-pollination of the GM oilseed rape to other oilseed rape crops in the vicinity may contaminate the crop with a certain ratio of GM seeds, which could lead to organic crops losing their premiums or to conventional seed lots not meeting their high purity standards. Cross-pollination from non-GMO to GMO fields could lead to depreciation of the value of speciality GM crops.

While a large number of studies have reported on outcrossing rates within short distances from GM crops, fewer projects have addressed long distance gene flow from large pollen sources. It is however already known that oilseed rape pollen is carried over long distances by both wind and insects (Thompson *et al*., 1999). In this context outcrossing has been studied at large distances from a field trial with genetically modified and hybrid oilseed rape in Belgium. Frequencies of gene flow at various distances, ranging from 880 to 1280 meters, from the pollen source will be presented and discussed. The outcrossing frequencies fall well below any threshold proposed within the European legislation.

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Are rapeseed feral populations adapted to their environment?

A. Deville[1], H. Monod[2] & J. Lecomte[1]
[1] Laboratoire ESE, bât. 362, Centre universitaire d'Orsay, 91405 Orsay Cedex
France
E-mail: alexandra.deville@esv.u-psud.fr
[2] Laboratoire BIA, INRA - Domaine de Vilvert, 78352 Jouy-en-Josas Cedex
France

The persistence of oilseed rape outside fields via feral populations could influence gene flows in a landscape. It is then important to determine the origin of such populations i.e. if they are only funded by fields of previous years or if they can maintain by self-recruitment. We then carried out an experiment to compare the fitness of plants from 7 feral populations liable of self-recruitment with plants from 4 cultivated populations, both sampled the same year in the same region. If plants of feral populations have a better selective value than plants of cultivated populations in a road verge environment, and then if we show an interaction between genotype and environment, we could infer that plants of feral populations are able to adapt to their environment and are subjected to selection.

Seeds of each population were sown in two environments, corresponding to a road verge (soil packed down and inter-specific competition) or to a field environment (laboured and uncovered soil). This environmental factor is crossed with density of plants (high or low density, for field and for feral populations respectively). The persistence ability is assessed via measures of seed weight, germination rate and seedlings mortality, growth speed, height, date of beginning of flowering and plant mortality. The comparison of yield components lays on measures of number of spikes, number of pods per spike, number of seeds per pod, seed weight and germination rate. Preliminary results seem to indicate that there is a strong effect of the type of population on many components of selective value and that some interactions between genotype and environment are significant.

On-farm experiences during the first few years of glyphosate-tolerant canola production in Manitoba, Canada: canola volunteers

M.H. Entz & G. Martens
Department of Plant Science
University of Manitoba
Winnipeg, R3T 2N2
Canada
E-mail: m_entz@umanitoba.ca

Glyphosate-tolerant canola was introduced in western Canada in 1995. In general, farmers have embraced glyphosate-tolerant canola; in 2002, well over one-half of the canola produced in this region consisted of glyphosate-tolerant cultivars. However, a sizeable cohort of farmers have deliberately avoided growing glyphosate-tolerant canola cultivars on their farms; these farmers often use other HT canola systems. A major reason for avoiding glyphosate-tolerant cultivars is zero-tillage production which relies on glyphosate for pre-seeding non-selective weed control. Zero-tillage and minimum tillage (no spring tillage) are practiced on over one-half of the 40 million ha western Canadian production region. Other reasons for avoiding glyphosate-tolerant canola cultivars include concern about volunteers in crop rotations with a high proportion of non-cereal crops, where control of volunteers is more challenging, and concern with glyphosate overuse. Unfortunately, glyphosate-tolerant canola volunteers have appeared in fields where glyphosate-tolerant canola has never been grown, and even where canola was not produced for over 10 years. These situations were documented in a farm case studies in Manitoba in 1999. Results of this study indicated that: 1) farmers were poorly informed about how to manage glyphosate-tolerant canola volunteers; 2) canola can survive in the seedbank for longer than farmers expected; 3) containment of glyphosate-tolerant canola is very difficult due in large part to the high use of glyphosate in western Canadian farming systems (selection pressure), and the weedy nature of the canola plant.

Modelling the effect of genotype on gene flow between rapeseed volunteers and crops. Integration into the GENESYS model

A. Fargue[1, 2], N. Colbach[3], J.-M. Meynard[1] & M. Renard[2]
[1] UMR Agronomie, INRA-INA P-G, BP1, 78850 Thiverval-Grignon, France
[2] UMR Amélioration des Plantes et Biotechnologies Végétales, INRA-ENSAR, Domaine de la Motte, BP 35327, 35653 Le Rheu Cedex, France
[3] UMR Biologie et Gestion des Adventices, INRA, BP 86510, 17 rue Sully, 21065 Dijon Cedex, France
E-mail: colbach@dijon.inra.fr

The GENESYS model (Colbach *et al.*, 2001) was developed to quantify transgene dispersal in regional cropping systems. Its input variables are the regional field pattern, the crop successions, the cultivation techniques of each crop and rapeseed variety characteristics. New variables were added to describe the genotypic composition of the rapeseed varieties. This composition results from the combination of six genes: the transgene, modelled as a dominant allele A associated to a recessive allele a (already present in the previous version of the model); two genes H1/h1 and H2/h2 determining plant height; one gene C/c for flower morphology (closed, semi-open, open); a cytoplasmic gene M/m coding for male sterility and a nuclear gene R/r restoring male fertility.

The input variables influence the annual life cycle of cultivated and volunteer rape (seed bank, seedlings, adult plants, ...). The six genes influence various parts of the life-cycle. Herbicide tolerance depends on transgene presence. Flower and seed production increase with plant height (relative to neighbour plants). Seed production is larger for male-sterile plants but these produce no pollen. Pollen production and emission as well as self-pollination rates depend on flower morphology.

The parameters for pollen emission rates necessary for the associated model equations were measured in field experiments on conventional rapeseed, varietal association (80% male sterile, 20% male fertile), dwarf rapeseed and cleistogamous rapeseed (closed flowers). On the latter, self-pollination rates were also measured. Another series of experiments studied the effects of competition on seed and flower production of rape volunteers in rape crops of different varieties. The yield of individual plants decreased with total plant density and with the plant height relative to the height of the field cover.

The modified GENESYS was then used to simulate harvest pollution in case of coexisting rape varieties, depending on variety characteristics (self-pollination rate, plant height, ...) and distance.

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Distribution and prediction of cross fertilization level between two maize fields (GM and conventional)

X. Foueillassar & F. Benetrix
Arvalis, Institut du végétal, 64121 Montardon
France
E-mail: x.foueilassar@arvalisinstitutduvegetal.fr

In 2002, within the framework of the P.O.E.C.B. (Operational Program for GM. Crops Evaluation), a study on the pollination and dispersion of pollen from GMO fields was conducted in three different locations, in the South of France. In two of the locations we sampled ears windward at different distances from the source and analysed them by P.C.R. Under these conditions the results show that at 40 m from the GM source, the rate of GMO found in the conventional ears DNA is lower than 0.9%. In the third location, we tested an efficient model of pollen dispersion, elaborated by the National Institute Agronomical Research (I.N.R.A.), which allows predicting cross fertilization at different distances from the GMO source. Thanks to an intensive method of ear sampling in the conventional field, the analysis of cross fertilization levels by P.C.R. confirmed the model forecasts. This database model, after validation, will be implemented when both sectors (GMO and conventional) coexist.

Modelling the dynamics of feral oilseed rape populations

A. Garnier, A. Deville & J. Lecomte
Laboratoire Ecologie, Systématique et Evolution, bâtiment 360
Université Paris Sud
91405 Orsay Cedex
France
E-mail: aurelie.garnier@ese.u-psud.fr; alexandra.deville@ese.u-psud.fr;
jane.lecomte@ese.u-psud.fr

The risks of (trans)gene escape resulting from cultivation of genetically modified plants is particularly large in the case of crop species presenting wild populations or wild relatives. The widely cultivated winter oilseed rape (OSR) can remain outside fields in feral populations mainly located on road verges or in disturbed environments. They might act as a relay in gene dispersal in space and time *via* pollen and seed flow.

Our stage-structured model of the local dynamics of a feral OSR population gives a better understanding of the demography of those populations and enlightens on their potential impact on gene flow in an agricultural landscape. This model takes into account road side management (cuttings and chemical treatments), external seed flows (from crops and/or seed transport), density-dependence, and stochastic factors. The analysis shows that the key-parameters of the dynamics of the feral populations are management parameters, density-dependence, presence of a seed bank and external seed flows. Projections suggest that without seed bank, feral populations can persist at least two years, whereas they can persist from two years (for isolated populations) to more than ten years (for untreated populations) with a seed bank. This model can also provide data to determine treatments (herbicide strength, number and date of cuttings) required to limit gene escape from feral populations. Simulations show that the choice of an optimal management strongly depends on the magnitude and on the periodicity of external seed flows supplying feral populations. Model results are now integrated in metapopulation and invasion models to estimate gene dispersal *via* seeds in OSR populations at the landscape scale.

An experimental evaluation of the relative importance of pollination by insects vs. wind in oilseed rape (*Brassica napus*)

K.E. Hayter & J.E. Cresswell
Ecology and Evolution Group, School of Biological Sciences, Hatherly Laboratories,
University of Exeter, Exeter, Devon, EX4 4PS
United Kingdom
E-mail: k.e.hayter@exeter.ac.uk

Pollination mechanisms in oilseed rape (*Brassica napus*) were investigated in agricultural fields at two sites in the UK. The sites were characterised by their very different bee densities (high or low). Pollen accumulation rates of individual flowers in the field were quantified by checking the proportion of the stigma covered with pollen and the number of pollen grains deposited. Flowers were sampled from the time of flower opening to the time of senescence. At high bee densities stigmas appeared covered with pollen within 3 hours of flower opening. In contrast, at low bee density, most flowers were barely pollinated after 5 days. We conclude high rates of pollen deposition were associated with high bee densities. Experimentally manipulated flowers were used to clarify further the contribution of wind and insects as pollinating agents in oilseed rape. Some flowers had petals removed (w/o petals treatment) to limit pollen deposition by insects. Entire flowers (w/ petals treatment) were used as a reference to evaluate the contribution of insects to pollination. At high bee densities, flowers without petals accumulated many fewer pollen grains relative to intact flowers (w/o petals: median= 56 grains d^{-1}; w/petals: 546 grains d^{-1}). At low bee densities this pattern was maintained although the differential between treatments was rather less (w/o petals: median= 5 grains d^{-1}; w/ petals 19 grains d^{-1}). Some of the pollination in de-petalled flowers is doubtless due to mechanical contact between flowers and to insect visitation, albeit infrequent, which leaves only a small residual to be explained by wind pollination. The slow rate of pollen accumulation at low bee densities and the poor pollination of de-petalled flowers, even in the center of an agricultural field where airborne pollen densities are higher, leads us to conclude that the wind is not the primary pollen vector in oilseed rape, which must instead be insects.

Development of real-time PCR systems based on SYBR® Green I, Amplifluor™ and TaqMan® technologies for specific quantitative detection of the transgenic maize event GA21

M. Hernández, T. Esteve, S. Prat & M. Pla[§*]
Dpt. de Genètica Molecular, Institut de Biologia Molecular de Barcelona (IBMB), Centro de Investigación y Desarrollo- Consejo Superior de Investigaciones Científicas (CSIC), Jordi Girona 18-26, 08034 Barcelona, Spain
[§]Institute of Food and Agricultural Technology (INTEA), University of Girona, E-17071 Girona
Spain
[*]To whom correspondence should be addressed
E-mail: mpsgmp@ibmb.csic.es

Worldwide production and commercialisation of genetically modified organisms (GMO) is increasing and food labelling regulations have been established in several countries. Besides, measures intended to ensure the sustainable co-existence of GM crops with conventional farming are currently discussed in the EU. To evaluate the extent of adventitious presence of GMO in conventional productions and its possible sources, methods for detection, identification and quantification of GMO are required.

The *Zea mays* line GA21, exhibiting tolerance to the Roundup® glyphosate herbicide, is one of the GMO for which no specific assay has been developed to date. It integrated several tandemly repeated copies of the *r-act*-EPSPS-NOS cassette used for transformation. We amplified a nucleotide sequence corresponding to the polylinker plasmid vector flanked by NOS 3'- terminator and *r-act* promoter. Based on this transgenic sequence we developed a method for specific detection and quantification of Roundup Ready® transgenic maize line GA21 DNA using conventional and real-time PCR. GA21 specific primers and probe were designed targeting the vector-promoter junction region and amplifying a 67-bp DNA fragment. Quantification methods were optimized through three alternative real-time PCR chemistries i.e. SYBR® Green I, Amplifluor™ and TaqMan®. All three methods proved to be specific, highly sensitive and reliable for both identification and quantification of GA21 DNA.

Plasmid *pGAivr* containing single copies of the GA21 and *invertase* amplicons was constructed for use as external standard in calibration curves. Using *pGAivr*, a TaqMan® based real-time PCR assay was optimized in duplex format targeting the maize species-specific *ivr1* gene and the GA21 junction region. The detection limit of the method was 0.01% GA21, this method therefore being suitable for use in extensive GMO analyses.

Volunteer oil-seed crops in the Czech Republic and effective post harvest management for their control

J. Holec, J. Soukup & J. Mikulka
Department of General Plant Production and Agrometeorology, Faculty of Agronomy, Czech University of Agriculture in Prague, Kamýcká 957, 165 21 Prague 6
Czech Republic
E-mail: holec@af.czu.cz

Possible conflicts of GMO and non-GMO co-existence can be caused by emerging volunteers. In the Czech Republic, oil-seed rape and sunflower acreage have rapidly increased and their share on arable land is currently 12%. In field tests, different types of post-harvest management were used to investigate the influence of soil tillage timing and depth onto the sunflower soil seed bank formation and volunteer plants emergence in following crop (Table 1).

Table 1. The influence of post-harvest tillage onto volunteer sunflower emergence.

Date of testing/ Crop	Ploughing /18-20 cm/ [number of plants per m^2]	Shallow tillage /8-10 cm/ [number of plants per m^2]
	Autumn	
End of XI.	8.8	38.0
	Spring	
Beginning of IV.	0.1	2.9
Beginning of V.		
Winter wheat	6.2	40.0
Spring barley	2.0	14.0
Maize	8.4	85.0
Beginning of VII.		
Maize	0.6	3.0

After oilseed rape harvest, different timing of stubble tillage and than different depth of soil tillage was used and numbers of rape seeds in different depths were counted (Table 2).

Table 2. The influence of different post-harvest management onto number of rape seeds in different soil depths.

Variant	Depth		Total
	0-10 cm	10-20 cm	
1	50	70	120
2	75	110	185
3	55	120	175
4	50	5	55
5	50	15	65
6	35	0	35

Variants:
1- No stubble tillage, ploughing.
2- Stubble tillage 1 week after harvest, ploughing.
3- Stubble tillage 3 weeks after harvest, ploughing.
4- No stubble tillage, shallow tillage before sowing.
5- Stubble tillage 1 week after harvest, shallow tillage before sowing.

This study was supported by the National Agency for Agricultural Research grant No. QD1360.

Measured airborne concentration and deposition rate of pollen downwind of maize crops

N. Jarosz[1], B. Loubet[1], B. Durand[1] & X. Foueillassar[2]
[1]INRA-UMR EGC, 78850 Thiverval-Grignon
France
E-mail: nathalie.jarosz@grignon.inra.fr
[2]Arvalis, Institut du végétal
Chemin de Pau, 64121 Montardon
France
E-mail: x.foueillassar@arvalisinstitutduvegetal.fr

In recent years there has been increasing interest in pollen dispersal, particularly in relation to gene flow from genetically modified crops and the maintenance of seed quality. Maize (*Zea mays* L.) is primarily wind pollinated and is one of the most important cereal crops grown in many parts of the world. However, there have been surprisingly few studies reporting pollen dispersal from maize crops. Many of these studies were performed between the 1940s and the 1970s over short downwind distances and in a limited range of weather conditions. Three experiments are presented here. Vertical and horizontal profiles of airborne maize pollen concentrations and deposition rates were measured within and downwind from a 20 × 20 m, a 24 × 48 m and a 500 × 1000 m maize crop coupled with micrometeorological measurements over the whole pollination period. Concentration and deposition rates decrease rapidly downwind from the source. On average the deposition rates at 30 m were less than 5% of the rate at 1 m, and less than 1% at distances greater than 60 m. However, pollen grains are still observed at 1000 m. Moreover, the pollen emission in the morning appears to coincide with the drying of the crop. The results of this study concur broadly with the few published studies for maize pollen dispersal. Coupled with full micrometeorological measurements, these results will be used for testing and validating pollen dispersal models.

Modelling long-distance wind dispersal of GM pollen from oilseed rape

P. Løfstrøm[1*], C. Geels[1], J. Brandt[1], L.M. Frohn[1], G. Kjellsson[2], C. Damgaard[2] & V. Simonsen[2]

[1]National Environmental Research Institute, Department of Atmospheric Environment, Frederiksborgvej, 399, P. O. Box 358, DK-4000 Roskilde, Denmark

[*]E-mail: pl@dmu.dk

[2]National Environmental Research Institute, Department of Terrestrial Ecology, Vejlsøvej 25, DK-8600 Silkeborg, Denmark

The focus on the application of genetically modified organisms (GMO) and the consequences for conventional and organic farming has increased in the recent years. One of the earliest GM types of crops is oilseed rape that is known to be partly insect-pollinated and partly wind pollinated. This has lead to a discussion on the risk for genetic pollution of organic fields through the dispersal of pollen. In the Danish project "Tool for protection against contamination by GMO" (TOPRO) one objective is to develop a computer model for simulations of wind dispersal of pollen. In this project a modified version of the Gaussian OML model (Berkowicz *et al*., 1986), developed at the Danish National Environmental Research Institute, is applied. The OML model is a local scale operational air pollution model used by the Danish authorities for regulation of industry by calculating dispersion of passive gasses or particles from point and area sources.

The model has been further developed for simulations of pollen transport by including a parameterization of the deposition velocity and using the principle of surface depletion (Horst, 1977). Here the focus is on oilseed rape pollen and the seasonal and diurnal variation of the pollen emission/production is based on the observed pollen values from McCartney and Lacey (1991). The temporal and spatial variability of the hourly pollen concentration and deposition is calculated for 5 years at three sites in Denmark. Results are presented as the field average ratio of GM to non-GM pollen at canopy height inside non-GM fields. Scenarios for the size of and distance between GM and non-GM fields varies have been performed. The average seasonal ratio for e.g. two 300 m squares of GM and non-GM filed is modelled to be 1% and 0.1% at a distance of 100 m and 750 m, respectively.

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Evaluation of gene flow in a commercial field of maize

J. Messeguer[1], J. Ballester[2], G. Peñas[1], J. Olivar[3], E. Alcalde[4] & E. Melé[1]
[1]Departament de Genètica Vegetal. Centre de Cabrils. IRTA. Carretera de Cabrils s/n. Cabrils 08348, Barcelona, Spain
E-mail: joaquima.messeguer@irta.es
[2]IrtaGen. Centre de Cabrils. IRTA. Carretera de Cabrils s/n. Cabrils 08348, Barcelona, Spain
[3]Servei de Producció Agrícola. Direcció General de Producció Agraria i Innovació Rural. Generalitat de Catalunya DARP. Gran Via de les Corts Catalanes 612-614 Barcelona 08007 Spain
[4]Syngenta Seeds. C/ Balmes, 117, 08008 Barcelona, Spain

Collaboration between IRTA Institute, Syngenta Seeds and the Catalan Government was established with the aim to evaluate the pollen mediated gene flow in a commercial field of maize, to provide data to assess on the agronomic practices facilitating the co-existence between GMs and non-GMs maize crops.

A nucleus of transgenic maize (Compa cultivar) of 50 x 50 m was planted in the middle of a field and surrounded by a non-transgenic cultivar Brasco. The total surface of the experimental field was about 7.5 Ha. Panicle samples placed at 1, 2, 5 and 10 m distance from transgenic nucleus were collected by hand taking into account its geographic orientation. The rest of the field was divided in squares of 30 x 30 m by using UMT coordinates with the GPS System, and panicle samples were collected in all the compass card directions. Each sample (3 panicles) was grounded and analysed with the RT-PCR technique to detect Zein and Bt-176 genes. Dominant wind direction during flowering time was recorded in a Meteorological station placed near the field. This field trial is still in progress but it is expected that experimental data obtained will contribute to the establishment of practical regulations to assure the proper co-existence of maize crops.

GMO residues in organic farming products

B. Oehen & K. Nowack-Heimgartner
Research Institut of Organic Agriculture, Postfach, Ackerstrasse, CH-5070 Frick
Schweiz
E-mail: bernadette.oehen@fibl.org; karin.nowack@fibl.org

Organic farmers have decided not to use neither genetically modified organisms (GMOs) nor their derivatives. As a consequence of the worldwide spread and use of genetically modified organisms in conventional agriculture and food production, there is an increasing risk of unwanted contamination of organic products by GMOs. We have identified the various pathways by which GMOs or their products can enter the organic farming system: Organic fields may become contaminated through pollen drift (wind or insects), the spread of transgenic seeds and plants. Planting or harvesting machines and transporters are additional sources of contaminations. If genetically modified and organic products are processed in the same installation, there is a risk of impurity, since complete cleaning is not possible for dusty goods. To show the risk of mixing with GMOs during processing, we carried out trials in a corn mill. After the milling of GMO corn and the usual cleaning, GMO contamination was found in all milling products of conventional non-GMO corn.

Based on this analysis, a further study identified measures to guarantee GMO-free organic production that can be applied by organic farmers and food processors. These measures include separation distances between fields of GM and organic crops, spatial separation of organic products, complete documentation of produce flow and exclusion of critical substances.

Moreover, we analysed the quality management system of the food, feed and seed industries in Switzerland. To check the effect of the measures, GMO contamination in conventional and organic corn and soybeans (food and feed) were investigated. Traces of GMOs were detected in 25% of organic corn and soybeans and in 33% of conventional imported from over sea's countries to Switzerland for food use. In 90% of all organic samples, the contaminations were less than 0.1% GMO-DNA. In 10% of the samples, we found GMO-DNA between 0.1% and 1%. In conventional corn and soybeans for food and feed use, the contaminations were higher and more frequent.

Looking for a seed bank in feral populations of oilseed rape

S. Pivard[1], J. Lecomte[1], C. Lavigne[1], E.K. Klein[1,2] & P.H. Gouyon[1]
[1] Laboratoire ESE, bâtiment 360, Université Paris-Sud, CNRS UMR 8079, 91405 Orsay Cedex, France
E-mail: sandrine.pivard@ese.u-psud.fr
[2] UMR 578, INRA/INAPG, 16 rue Claude Bernard, 75005 Paris, France

Feral populations of oilseed rape are frequently met on roadsides, slopes or fallows. Their presence increases the risks of unintentional spread of transgenes from GM into non-GM crop varieties thus making the co-existence of GM and non-GM crops difficult. They could behave as reservoirs and/or relays of pollution via pollen and seeds, thus increasing long-distance dispersal of transgenes. This is particularly plausible if these populations can persist for many years.

Our team has been studying the dynamics of feral populations of oilseed rape in the region of Selommes (Loir-et-Cher, France) for five years. Some results from this field work plead in favour of populations persistence during several years. Computer simulations furthermore indicate that seed bank is an important factor in this persistence. However, the models use parameter estimates from measurements of seed banks from cultivated fields (Pekrun *et al*., 1999). To our knowledge, oilseed rape seed bank has never been studied in feral populations, and parameter estimates might prove very different since the soil is generally packed and not turned-over.

We thus conducted a study to check the existence and quantify the seed bank of 41 feral populations in our study region. These populations differed by their proximity with roads, oilseed rape fields and feral populations for the last three years. Sampling was performed one month before the new seeds were mature. Our results confirm the existence of a seed bank but in a low number. They thus indicate that the majority of seeds tend to germinate or die in the first year. Although the seed bank can behave as a reservoir of (trans)genes, our results strengthen the importance of external seed flow (crops, seed transport) and population self-recruitment in the maintenance of feral populations. These results will be use in our population dynamic models to determine suitable management of feral populations.

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Is a significant outcrossing-rate of weedy rice an obstacle to co-existence with herbicide resistant cultivars in northern Latin America?

E. Skov[1], H. Wellendorf[3], B.E. Valverde[2] & S.B. Andersen[1]
Royal Veterinary- and Agricultural University
[1] Department of Agricultural Sciences, Plant breeding
E-mail: els@kvl.dk
[2] Department of Agricultural Sciences, Weed Science
E-mail: bev@kvl.dk
[3]Department of Forest and Landscape, Arboretum
Denmark

Weedy rice in northern Latin America is predominantly *Oryza sativa* and thereby belonging to the same species as the widely cultivated varieties of this Asian species. Other *Oryza* species, however, also occur as weeds in rice fields, among those the assumed Asian ancestor to *O. sativa, O. rufipogon* as well as the native American species *O. latifolia*. Cultivated *O. sativa* is able to hybridize with weedy *O. sativa* and *O. rufipogon* and also with the native *O. glumaepatula*.

The study was conducted to characterize the relationship and diversity among a sample of 73 weedy and cultivated *O. sativa* populations from northern Latin America and to estimate outcrossing rates of weedy rice. The analytical tools were 15 published microsatellite markers combined with cluster analysis and basic population genetic theory.

73 bulk-analyzed accessions could be grouped into 3 clusters: cluster 1: 39 accessions of weedy rice from all parts of the region, large within-accession diversity; cluster 2: 21 accessions of weedy as well as cultivars from all parts of the region, restricted within-accession diversity, and cluster 3: 7 accessions of cultivars from Venezuela, restricted within-accession diversity.

A sub-sample of 23 weedy rice accessions represented in cluster 1 and 2 showed an average out-crossing rate of 46%.

The obvious lack of a geographic pattern of relationship between weedy and cultivar accessions over most of the region indicate a widespread moving of more or less domesticated cultivars in the past. The documented significant level of out-crossing rates of weedy rice suggests, that genes behind specific herbicide resistance in cultivars sooner or later will migrate to weedy relatives in the neighbourhood. So, co-existence of herbicide resistant cultivars and weedy rice lacking herbicide resistance may not sustain without repeated effort to renew the genetic background for resistance of the cultivars.

Importance of popularizing the knowledge about GMOs, and the museum's role in this process

D. Šubová
Slovak Museum of Nature Protection and Speleology
Školská Street 4, 03101 Liptovský Mikuláš
Slovakia
E-mail: subova@smopaj.sk

At the turn of the 20th and the 21st centuries the modern biology represents the biology changing from a theoretical stage to the discipline of technical engineering thanks to new technologies of the genetic engineering (methods of DNA isolation, C-DNA anti-sense m-RNA construction technologies, vector constructions). Now these technologies more and more enable everybody to get in contact with new organisms many of which cannot originate naturally in the nature and even by traditional breeding methods (hybridization and selection). In a broader sense these organisms are generally named genetically modified organisms. But their utilization brings many questions as well.

Consequently the Slovak Museum of Nature Protection and Speleology which is in establishing competence of the Ministry of Environment of the Slovak Republic cannot have activities in classic fields of nature protection only, but must quickly respond on actual questions of environment such as the questions related to GMO's. This is realized through exhibitions, talks, and inquiries. At the end of 2002 our museum opened the exhibition GMO – hope or threat of the 21st century? This exhibition presents history of genetics, basic knowledge about molecular genetics (models of DNA structure, a principle of the genetic code, a principle of transcription and translation of genetic information) all on a popular form. The exhibition is devoted to the main roles of genetic engineering technologies in plant, animal, and human fields also introducing the perspectives and problems like resistance to herbicides and pests, production of new substances, blocking of specific substances, technology of extended in vitro cultivation of embryos, cloning technologies, etc. The exhibition on 25 exhibition panels has been open to the public at our Ministry of Environment, museums, health centres and schools all over Slovakia up to now. It completes many talks organized for the broad public and at schools, and also inquiries, which already call attention to necessity of popularization of these problems in primary schools.

Our activities also resulted into publishing the handbook of methods for teachers, which were distributed to Slovakian schools.

(A part of my presentation is demonstration of exhibition panels in a poster form or their presentation in PC programme "power point".)

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Transgene escape through natural cross pollination in tomato

L. Tomassoli, V. Lumia, A. Zaccaria & M. Barba
Plant Pathology Research Institute – Via C.G. Bertero, 22 - 00156 Rome
Italy
E-mail: l.tomassoli@ispave.it

Pollen-mediated gene flow is an important part of risk assessment procedure to determinate the potential environmental and agronomic impact of genetically modified crops. Tomato (*Lycopersicon esculentum* L.) is predominately a self-pollinated crop. Natural cross-pollination may occur with percentages ranging from 0.07% to 12% depending on environmental condition and variety characteristics. In our laboratory, different tomato varieties were genetically modified for cucumber mosaic virus (CMV) resistance using the coat-protein (*CP*) strategy and neomycin phosphotransferase II (*NPTII*) gene as selectable marker. In our first study, no cross-pollination between transgenic and non-transgenic tomato of UC82B variety was ascertained. Recently, frequency of transgene transfer by pollen was estimated for a fresh-market tomato type, named INB, and the typical Italian San Marzano. The transgenic tomato lines were released in an experimental field for evaluating their resistance to CMV and seeds were collected from untransformed plants to screen them *in vitro* for the kanamycin resistance on selective medium. More than 3.500 and 1300 seeds, respectively, of INB and San Marzano were screened and after two months of culturing, two INB plantlets and 46 San Marzano ones resulted kanamycin resistant (KR). The presence of *CP* gene and *NPTII* gene in KR seedlings was detected by PCR and the expression of the transgenic coat protein by serological methods. Analysis of progenies attested the hereditary of transgenes in a segregation ratio of 3:1, as Mendel's First Law claims, indicating that KR seedlings were "transgenic hybrids" obtained by natural cross-pollination in open field. Our assessed rates of hybridization (INB: 0.056%; San Marzano: 3.3%) fit the very low natural cross-pollination estimated in tomato in most past and recent experiences. Our results demonstrated the significantly influence of style length on cross-pollination. Thereby, transgenic tomato varieties having protruding style (e.g. San Marzano) need case-by-case evaluation to adopt effective safeguards for experimental or commercial cultivations.

List of authors arranged alphabetically