

Farm scale evaluations of GM crops: monitoring gene flow from GM crops to non-GM equivalent crops in the vicinity (contract reference EPG 1/5/138). Part I: Forage Maize.

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EXECUTIVE SUMMARY

1. This study was commissioned to monitor gene flow from genetically modified (GM) crops to adjacent non-GM equivalent crops. It was undertaken on behalf of Defra to validate the assumptions made in the original risk assessments concerning gene flow from GM plants.
2. Gene flow was monitored at the farm-scale evaluation (FSE) sites of winter and spring oilseed rape and fodder maize, genetically modified to be herbicide tolerant (HT) and released under the authority of the Genetically Modified Organisms (Deliberate Release) Regulations. This report describes the sampling and analysis of the FSE fodder maize crops.
3. Maize is wind pollinated and demonstrates an outcrossing rate of around 95%. Factors such as wind speed and direction, surface turbulence as well as synchronisation of flowering and the density of the pollen clouds can all affect the distance that pollen travels and the extent of gene flow (cross-pollination). The current recommended separation distance for conventional crops from GM forage maize is 80m (SCIMAC).
4. Both field and laboratory based methodologies were developed to sample and analyse seed for the detection of gene transfer between the two crop types. The analysis of seeds collected at the FSE sites, using real-time PCR, has demonstrated the occurrence of gene flow events between GM and conventional crops. In addition, quantitative data on the extent of the gene flow has been obtained.
5. Overall results showed that there was a rapid decrease in the rate of cross-pollination within the first 20m from the donor crop and beyond this distance the rate of decrease was much slower. There was significant variation in levels of GM: non-GM cross-pollination between sites in each year ($p < 0.01$), although the variation between years across all sites was not significant ($p > 0.05$).
6. Results from individual fields could be correlated to both wind direction during the flowering period, synchrony of flowering between the two (GM and conventional) crops and to separation distance between the crops.
7. Evidence of gene flow was detected beyond both the 80m and 200m separation distances recommended for forage maize and sweetcorn respectively. The significance of this in relation to current EU regulations on GM adventitious presence is discussed.

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INTRODUCTION

The last two decades have witnessed a revolution in the techniques of genetic modification, with associated optimism about the benefits to be gained from the construction of genetically modified (GM) plants. Improved weed management and a reduction in costs and amount of herbicide applied are some of the proposed benefits from these novel crop varieties. However, the release (and management) of such organisms into the environment may have far reaching side effects on farmland biodiversity.

There is currently a moratorium on the commercial planting of GM crops in the UK. The government will make a decision on whether or not GM crops should be cultivated, following publication of the results of the farm-scale evaluations, reviews of the costs and benefits of GM crops (Strategy Unit of the Cabinet Office, 2003) and a review of the science relevant to GM crops and food based on interests and concerns of the public (King *et al.*, 2003).

In 1999, the Department for Environment, Transport and the Regions (DETR; now Defra) established farm-scale evaluations (FSE) to assess the effects of the agricultural management of field-scale releases of GM herbicide tolerant (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 the GM to conventional crops was also commissioned, using the FSE sites of winter and spring oilseed rape and fodder maize, genetically modified to be tolerant to the herbicide glufosinate ammonium and released under the authority of the Genetically Modified Organisms (Deliberate Release) Regulations. This report presents quantitative results from years 2000 to 2002 for the extent of transfer of the GM herbicide tolerance gene to conventional fodder maize at different distances from the GM crop.

A review was published by the National Institute of Agricultural Botany (NIAB), which addressed the issue of separation distances between GM and other crops (Ingram, 2000). Currently the minimum separation distance in the European Union is 200m for all categories of seed production, which is believed to be sufficient to maintain inbred lines at 99.9% purity (Ingram, 2000). The recommended separation distances for non-GM crops from the Supply Chain Initiative on Modified Agricultural Crops (SCIMAC) guidelines for growing GM HT crops are 200m for sweetcorn and 80m for forage maize (this distance was increased from 50m in 2001). In the UK most of the maize is for fodder, although some sweetcorn is also grown in some areas. No maize is currently grown for seed production in the UK.

There are a number of factors that affect pollination rates in maize. Most of the pollen is shed from the plants before the silks are receptive, but there is some overlap, resulting in up to 5% self pollination (at least 95% of ovules are fertilised by pollen from other plants). Pollination rates can also be affected by competition from pollen from other sources. Pollen viability can vary between 2h and 8 days, depending on environmental conditions. The impaction rate (settling velocity) of maize pollen is 30-40 cm s⁻¹ so the pollen normally only travels short distances. Finally, wind speed and direction and surface turbulence can also affect pollination rates and these factors make it difficult to predict the effect of one maize field on another. A higher wind speed may cause the pollen to travel further downwind but the impaction rate of the pollen will also

increase. Factors affecting the rate of cross-pollination between fields include synchronisation of flowering, the relative concentration of the pollen in the donor and receptor plot (the protective strength of the field pollen cloud), the levels of selfing and the density of the stands.

There is limited literature available on gene flow from pollen, and hybrid corn production practices have remained basically unchanged for the last thirty years (Burris, 2001). Reports of outcrossing rates range from 40% at 2.5m (Bateman, 1947), 4.5% at 3m (Jugenheimer, 1976), 1.11% at 200m (Burris, 2001) and 2.47% at 200m (Jones & Brookes; 1950). Under very arid, calm conditions, outcrossing was not detected beyond 200m (Baltazar and Schoper, 2002). Previous studies on gene flow from maize have not been carried out on a commercial scale (with the exception of Burris). The FSE trials offered the opportunity to sample a large number of fields in a wide range of locations and environments in England.

Gene flow can be defined as the movement of genetic information among individuals, populations or taxa. Gene flow is thought to take place between plants through two routes, the movement of pollen and consequent fertilisation of sexually compatible individuals and the dispersal and establishment of seeds. A third possible route for gene flow might be via bacteria in the soil rhizosphere (horizontal gene transfer). Currently there is a shortage of firm data on which to evaluate the levels of risk of gene flow via bacteria in the soil rhizosphere (Gebhard & Smalla, 1999).

All GM crops grown in the FSE are grown in accordance with SCIMAC guidelines. These set out the principles of good practice in relation to specific husbandry and management of GMHT crops and clearly specify the need to control unintentional dispersal of seeds on farm machinery through spillage. With the possible exception of the occurrence of volunteers in the crops that follow, gene flow from GMHT crops is most likely to occur *via* pollen. The SCIMAC guidelines specify that HT crops should observe prescribed separation distances in order to reduce the levels of cross-pollination.

The objective of this exercise was to test assumptions made in risk assessments concerning gene flow by pollen from the farm-scale evaluations and to ensure that the guidelines issued by SCIMAC stipulate an effective separation distance for each of the crop types.

METHODS

Overall Approach of the Farm-scale Evaluations

For the biodiversity studies, a split-field design was used and differences in biodiversity measured between an area planted with a GM crop and one with a comparable non-GM crop, placed close together on the same site (see Figure 1). Sites were chosen to represent fully the range of variation in soil, climate, species occurrence and farm management that is likely to be found in the commercial growing of GM crops. The pairing of the GM and non-GM areas within each site ensures this variation is accounted for within the analyses, and does not affect the comparison between the crops.

The FSE offered the opportunity to sample a large number of fields in a wide range of locations (Figure 2) and environments in England (and Scotland for oilseed rape). The maize trials were located at a range of sites across England, covering fifteen counties from Dorset to North Yorkshire and from Shropshire to Lincolnshire.

The sites were well distributed throughout the country and a number of them were used for more than one year. Several were clustered in some locations. Some of the sites were still sampled despite being vandalised earlier in the season.

Sites consisted of a split field design, half planted with Liberty Link™, line T25 (containing the *pat* gene), which is tolerant to Liberty™, a broad spectrum, non-residual, glufosinate ammonium herbicide and the other half with an equivalent conventional maize variety.

Covariate data

Additional information was collected to aid in describing patterns of gene flow. Field information describing the crop flowering times, orientation and field boundary features (such as hedges, woods and the shape of fields) were obtained from the FSE biodiversity dataset and meteorological data recording wind direction were obtained from the nearest meteorological station to each field.

The meteorological data were used in conjunction with FSE data describing the flowering stages of the GM and conventional crops, to compile diagrams (Figures 6 and 7) showing the wind direction during the overlapping flowering period when both the GM and conventional maize were in flower.

From a previous study (Daniels & Boffey, 2001) capturing maize pollen over a two-week period using a Burkard trap, the results suggested that the majority of the pollen is shed between the hours of approximately 7am and 11pm (Figure 3). This agrees with data published by Miller (1985), which states that dehiscence occurs between 6:30 am and 11:00 am. The wind rose diagrams were correspondingly refined to include only wind direction during these hours. Flowering stage was recorded for the FSE sites at two-weekly intervals, giving approximate dates for onset and finish of pollen production. More frequent recordings may have been preferable if the methodology had been set up primarily for studying gene flow. However, data from a previous study (Daniels & Boffey, 2001) show a peak in flowering in the middle of the two weeks and tailing off at the start and end of the fortnight, so the FSE flowering data were

considered reliable enough to give an indication of wind direction over the main flowering period.

Sampling Strategy

A total of 55 maize FSE sites were used in this study, from which cobs at 1152 sample points within the conventional crop were collected and tested during the course of three years. Each sample consisted of 3-5 cobs (>1000 seeds), each from a separate plant in the sampling location. Samples were collected from three transects in the conventional crop at distances of approximately a quarter, half and three-quarters (Figure 1) of the way across the field (6 transects were sampled in year 2000; see Figure 1). Along each transect, cob samples were collected at the following distances: 2m, 5, 10, 20 or 25m, 50 and 150m away from the junction with the GM crop. Where the field length was too short to have a 150m-sample point, the cobs were collected at 5m within the field margin (the furthest edge from the GM crop) and the distance noted. Control samples were collected from 2m distances into the GM crop. At each sample point one maize cob was collected from each of three neighbouring maize plants along a single row, or from the next suitable plant if these cobs were unripe.

DNA Extraction and Real-Time PCR

The maize grains were removed from the cobs and ground up. Genomic DNA was extracted from the maize using the Promega Wizard® Magnetic DNA purification system and the Labsystems KingFisher ml Magnetic Particle Processor.

The maize samples were tested in duplicate using real-time (TaqMan) polymerase chain reaction (PCR). Briefly, a reporter dye and a quencher dye are attached to the 5' and 3' ends of a TaqMan probe. When both dyes are attached to the probe, reporter dye emission is quenched. During each PCR extension cycle, the *Taq* DNA polymerase cleaves the probe when bound to the template ahead of the *Taq*, which separates the dyes. Once separated from the quencher, the reporter dye emits its characteristic fluorescence. The fluorescence is detected using an ABI Prism 7700 sequence detection system. Results are recorded at the point where an increase in reporter fluorescence can be first detected, this is known as the Ct (cycle threshold) value.

Two sets of primers and probe were used. One set was specific for the *pat* gene (target gene; to detect the T25 transformation event) and the other set was specific to the *Zea mays cdc2* gene (the endogenous control). The endogenous control primers served two purposes. Firstly, to indicate whether the DNA extraction and PCR were successful for each sample, and secondly as a normaliser against which the amount of GM in each sample could be quantified. In other words, the endogenous control provided a quantity for the total amount of DNA in each sample analysis.

Samples were quantified using standard curves, which were prepared from known amounts of DNA from GM positive control material. Aventis (now Bayer CropScience) provided the positive control material (T25 maize seed). The T25 maize was heterozygous for the *pat* gene and this was taken as being 100% (i.e. 1:1 ratio) reference material.

Standard curves were created by plotting the Ct values of the known standards against the log of the concentration of DNA. Data for the unknown samples was

then calculated from the standard curve. A normalized amount of target DNA was obtained by dividing the amount of GM DNA by the amount of the endogenous control. The normalized TaqMan data was expressed as a GM: non-GM ratio.

Statistical Analysis

To stabilise variances all results of the proportion of GM DNA (*pat* gene) detected in the field, samples were subjected to a probit transformation (Armitage, 1983). To determine the effect of year and site on proportion of GM cross-pollination, field sites that had been sampled in more than one year were chosen. The transformed results were analysed using General Linear Model (GLM) Analysis of Variance (ANOVA) (McCullagh & Nelder, 1989). To determine the spatial spread of the *pat* gene, results collected at different distances along transects established in the fields were used. The results were subjected to non-linear regression analysis to estimate the extent of gene flow with distance from source.

Following the analysis described above, data from the field sites in 2000 (where there were 36 sampling points) were analysed further. The distribution of GM: non-GM hybridisation was examined for each group using SADIE (Spatial Analysis by Distance IndiciEs). This utilises an innovative class of techniques to detect and measure the degree of spatial pattern in spatially referenced data.

RESULTS

Overall the results showed a decrease in the rate of cross-pollination with increasing distance from the GM crop crop (Figure 4). There was a rapid decrease in the rate of cross-pollination within the first 20m from the donor crop and beyond this distance the rate of decrease was much slower. The molecular data was analysed to look for variation between years and fields, also significant relationships with distance (between pollen source and sink) and wind direction were investigated. Correlations between the varieties of conventional crop and cross-pollination levels were not possible due to the very wide range of maize varieties (37) planted at the sites: the GM maize variety was the same in all cases.

Results of the GLM ANOVA indicated that the GM contamination was significantly different between sampling locations on the field transects with distance ($t = -5.67$; d.f = 65; $p < 0.001$) and between fields ($t = -3.32$; d.f. = 65; $p = 0.001$) but not between years ($t = -1.18$; d.f. = 65; $p = 0.241$). A comparison of different non-linear equations indicated that the inverse power regression explained most of the variation in the experimental results and thus, was chosen for subsequent analysis. Results of the non-linear regression analysis further indicated that contamination was highly dependent upon distance from the source of GM DNA ($F = 30.4$; d.f = 2,8; $p < 0.001$; Figure 4).

The proportion of the GM *pat* gene detected in the samples, expressed as percentage DNA, was calculated at the furthest distances from the GM source. In 2000, evidence of cross-pollination was found up to 200m from the GM crop in two of the three sites where samples at this distance were tested, and in one of these sites values on two of the transects were particularly high (0.42% and 0.14%). In a separate study, samples were taken from the nearest facing edge of adjacent fields at two of the sites and analysis of these samples provided one positive result (0.14%) at a distance of 650m from the GM source field.

The regression equation was validated against field results not used in its derivation. The model predicted that at 650m from a source of GM maize, contamination would be 0.04% whereas a mean value of 0.02% was recorded. Further examination of the predicted equation indicated that at a distance of 80m contamination levels would be less than 0.3% (0.298%), and that to ensure contamination levels of less than 0.9% and 0.1% crops would need to be located at distances greater than 24.4m and 257.7m respectively.

The analysis of the results (from the year 2000) using SADIE provided another means by which to visualize the levels of gene flow across the fields (Figure 5). Once again this analysis showed that at the majority of the field sites levels of cross-hybridisation were highly aggregated and were spatially clustered towards the GM source. This method of analysis was not used on the samples taken in 2001 and 2002 because the sampling strategy was altered from 36 samples per site to 18 per site.

Looking at the results from the fields in all three years, at 50m into the conventional crop, evidence of cross-pollination was found in 43 out of the 55 fields tested and, of these, 34 had GM DNA detected at quantities greater than 0.1% and 23 quantities greater than 0.3% (Table 1). Samples taken from 150m into the conventional maize showed evidence of cross-pollination in 19 out of 44

fields and of these, 12 had $\geq 0.1\%$ GM DNA and 7 had $\geq 0.3\%$ (Table 2). Consistency across the fields was observed as in all cases where GM DNA was recorded at 150m it was also present at 50m.

In order to look at how separation distances would affect the whole crop (not just individual sampling points), the average GM content for the whole field was calculated both with and without the first 80m of the crop. Out of all the sites 26 of them had GM quantities $\geq 1\%$ across the whole field. After removing the data from the first 80m of each field only 2 of the sites had quantities $\geq 1\%$.

Several fields were identified as having particularly high cross-pollination rates at the further distances from the GM source and the data for these individual fields have been examined more fully.

Correlation was observed between extensive amounts of cross-pollination, up to and beyond 150m, and wind direction during the flowering period for three of the fields. Two of these were the same field, sown with GM maize in the FSE in years 2000 and 2002. They differed slightly in the area of GM crop being narrower in one year, however in both years the orientation of the GM and non-GM crops was approximately the same and the meteorological data for both years showed a high percentage of winds flowing from the GM to the conventional side of the crop during the flowering period (Figure 6).

Most of the other fields for which meteorological data were collected showed either very little or no major wind flow in the GM to conventional direction. One exception was a field (Figure 7) where cross pollination rates were low but the wind direction was consistently in the direction of GM to conventional, however there were only two days of overlap for the flowering times and thus limited opportunity for cross pollination.

It would appear however that wind direction might not explain the extent of gene flow in all situations, as the other fields, which showed high cross-pollination at greater distances, did not show corresponding predominance of winds from the GM to non-GM sides of the fields. In one field however, although the majority of the wind during the overlapping flowering period (Figure 7) was flowing in the conventional to GM direction, for 10% of the time the wind direction was from the GM direction. The shape of the field may be a factor in this situation as this was a long rectangular field, providing a wide front of pollen. Two other sites that were notable in having no evidence of cross-pollination beyond 10m into the conventional crop were 'L-shaped' fields where the width of conventional crop was far less than the adjoining GM side so potentially producing a reduced GM pollen cloud directly adjacent to the conventional crop.

Wooded areas or hedges around fields may influence the patterns and extent of gene flow by creating turbulence, or by reducing wind speed as it reaches the wooded area so potentially depositing any pollen suspended in the air. One pattern emerging from this study was greater cross-pollination at distance in the fields that tended to have wooded areas or hedges at the edge of the conventional sides, in contrast to the fields showing less gene flow where there was a general absence of wooded boundaries. Convection currents may also play a part in affecting wind flow as the warmer crop heats up the air above it and affects wind flow patterns.

The very low hybridization levels detected at one field could be explained by the 142m of set-aside crop between the GM and the conventional. These data show some gene flow at 2m into the conventional crop (0.1% and 0.026%) but none thereafter except at one sample point at 50m (0.06%). It has been suggested in the literature (Ingram, 2000; Burris, 2002) that when crops are isolated by open ground or low growing crops, the first few rows intercept a high proportion of the incoming pollen and cross-pollination decreases exponentially with distance.

In addition to patterns in gene flow related to wind direction and isolation distance we also noted several fields where the levels of gene flow showed a marked increase at distances of 100-150m from the GM source. One example of uneven gene flow is shown in Figure 8. Assuming that the conventional crop was completely free of GM adventitious presence, the effects of the landscape and of air movement over the crop (as mentioned above) could explain these 'hot spots'. For example pockets of airborne pollen may have been blown up into the air and then deposited at a greater distance away from the GM source.

CONCLUSIONS

This study is unique both in the number and range of the trial sites and in the molecular approach to quantification of gene flow. The FSE were set up to investigate different effects on biodiversity between GMHT and conventional farming practices and **not explicitly** to determine the extent of gene flow. However, these trials represent the potential for gene flow under realistic farming practices rather than in either small-scale trial plots or from monitoring a number of GM plants in the middle of a conventional field.

The conclusion of the work by Burris (2002) was that distance to the contaminant source is important but its contribution to reducing adventitious pollen intrusion is often overshadowed by other factors such as wind intensity, direction and the protective strength of the field pollen cloud. Theoretical scenarios for pollen dispersal e.g. Emberlin, 1999, and models such as that presented by Klein *et al.* (2003), have been produced. However, the effects of the agricultural landscape on daily variations in wind speed and turbulence are difficult to predict.

Looking at the results from individual fields in this study it is evident that the extent of hybridisation is very variable between fields and that isolation distance alone cannot account for gene flow levels between the two crops. As mentioned in the introduction, there are a number of factors that affect the rate and extent of cross-pollination. The analysis completed so far has highlighted not only the effect of isolation distance on gene flow but also the effect of wind direction.

If the aim is to maintain a 99.9% purity level then an 80m-separation distance will not be enough. The current proposed threshold for the adventitious presence of GM seeds in certified seed lots is now 0.3% for authorized events and 0.1%-nil for unauthorized events (under part C of Directive 2001/18/EC). A recent report published by the European Commission (Block *et al.*, 2002) suggested that for maize, a threshold of 0.1% would be extremely difficult to achieve for any farming scenarios (conventional and organic farms).

Maize seed is not produced in this country, therefore it is more important to consider the threshold for food and feed, which is currently set at 0.9 %. Based on the results presented here it would be possible to meet this threshold but an increase to the current isolation distances would increase the certainty with which this could be achieved. In addition the results have demonstrated that even with a large isolation distance (e.g. 142m) there is evidence of GM: non-GM hybridization in the GM-facing stands. In addition, sampling from an adjacent field at one of the locations revealed evidence of hybridization 650m away from the GM crop. This 'edge effect' should be taken into consideration when making recommendations for co-existence and crop management, for example the removal of the first few GM-facing rows of the crop prior to harvest might be worthwhile.

The original aim of this project was to validate assumptions made in risk assessments for gene flow by pollen from the farm-scale evaluations and to ensure that the guidelines issued by SCIMAC stipulate an effective separation distance for the crop. It is evident from the results that cross-pollination events occurred not only beyond the 80m isolation distance recommended for forage/fodder crops, but also beyond the 200m distance recommended for sweet

corn and organic crops. Although these trials did not use sweet corn, it is reasonable to assume that pollen flow between the two crops would be the same assuming flowering times coincide. It is important to emphasise that the whole of the plant is harvested in forage crops and thus any cross-pollination events will be 'diluted' out. Sweet corn presents more of a problem in that individual cobs will be consumed. So even if a field was well below the threshold, individual cobs may not be.

Summary of Conclusions

- A quantitative molecular assay (TaqMan PCR) was used to detect GM presence in conventional maize seed collected from 55 FSE field sites.
- Evidence of GM: non-GM hybridisation was detected at all of the field sites.
- 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.
- High levels of gene flow were linked to the prevailing wind direction (GM to conventional) during the overlapping flowering period. Low levels of gene flow were linked to a large isolation distance and also to a lack of synchrony in the flowering times of the two crops.
- Overall the data suggests that an isolation distance of 24.4m would be required to meet the 0.9% threshold recommended by the EU for food and feed.
- The 80m isolation distance recommended by SCIMAC would, in most cases, be sufficient to ensure that levels were below a threshold of 0.3%.
- The results also indicate that the 200m-separation distance (recommended by SCIMAC for sweetcorn and organic crops) would be sufficient although 'edge effect' and removal of the first few GM facing stands prior to harvest should be considered.

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Table 1. Maize samples tested at 50m into the conventional crop.

	Total no. fields sampled	<i>pat</i> gene absent	<i>pat</i> gene present	≥ 0.1% DNA	≥0.3% DNA
YEAR					
2000	9	1	8	6	4
2001	20	6	14	12	9
2002	26	5	21	16	10
All years	55				

Table 2. Maize samples tested at 150m into conventional crop.

	Total no. fields sampled	<i>pat</i> gene absent	<i>pat</i> gene present	≥ 0.1% DNA	≥0.3% DNA
YEAR					
2000	3	0	3	2	1
2001	20	14	6	4	3
2002	21	11	10	7	2
All years	44				

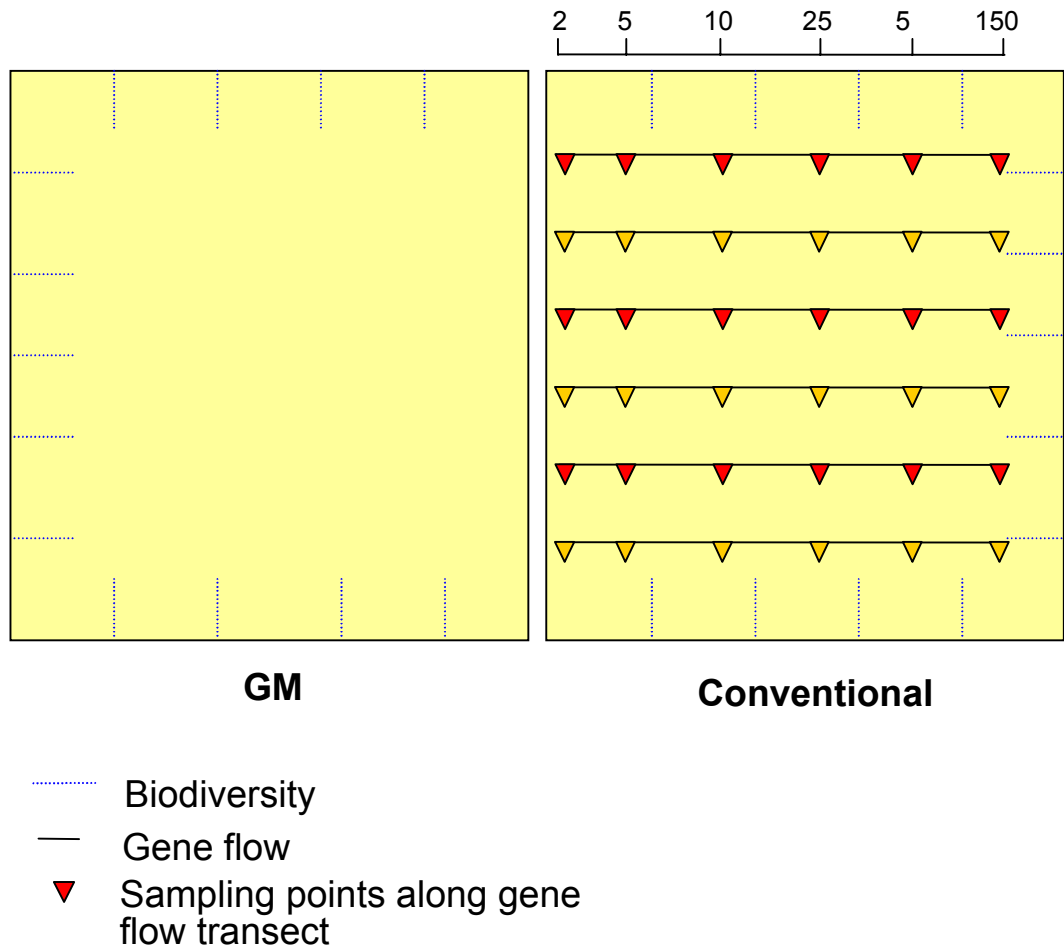


Figure 1. A diagrammatic representation of the sampling points for the gene flow study (this diagram is not to scale). A total of 6 transects were used in year 1 and in the following 2 years 3 transects were used. The position of the biodiversity transects are also indicated (dotted lines).

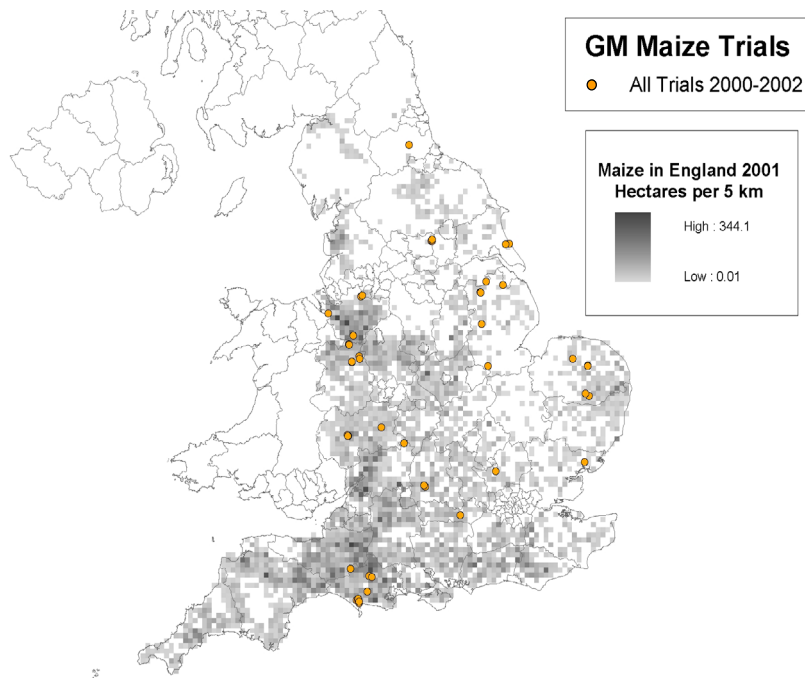


Figure 2. Map of England indicating the position of the maize FSE sites relative to the main maize growing areas across the country.

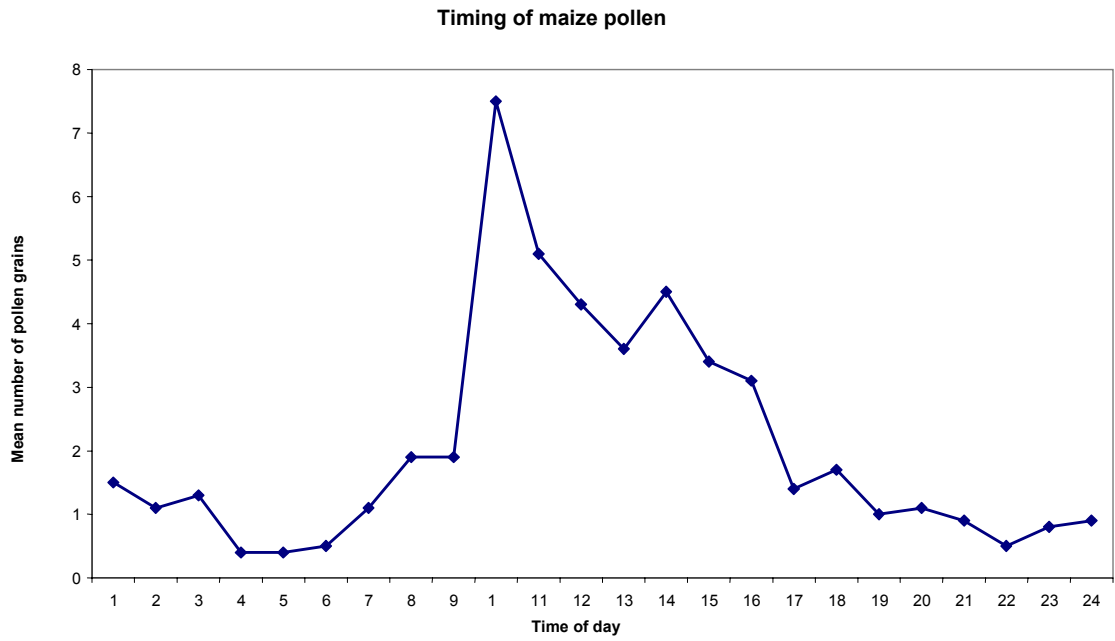


Figure 3. Mean hourly pollen capture over 24 hour period, using data collected over two weeks.

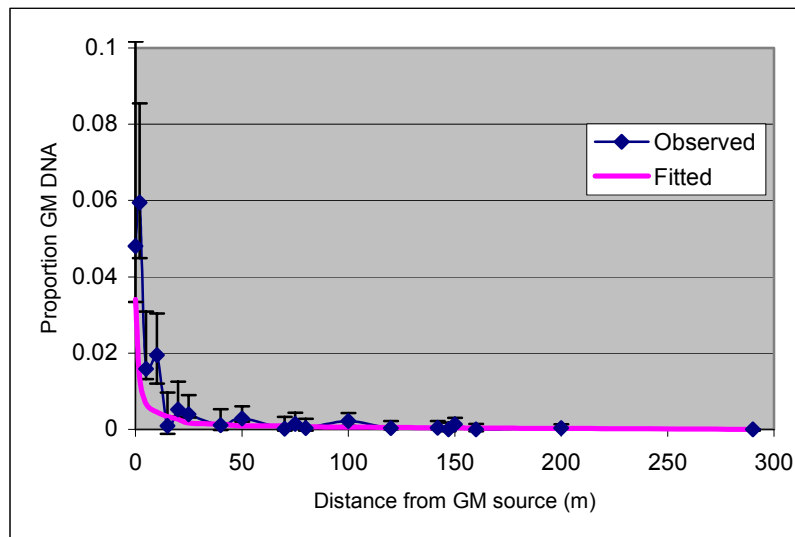


Figure 4. Comparison of fitted and observed GM: non-GM cross-pollination (gene flow) against distance from GM source in metres. The level of cross-pollination is indicated as the proportion of GM DNA detected in each sample.

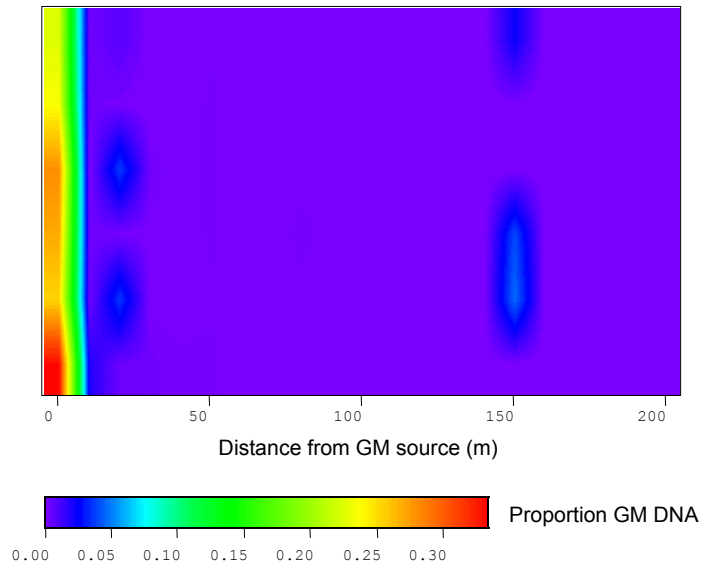


Figure 5. Frequency of GM/non-GM cross-pollination in maize fields analysed using SADIE software. The colour scale represents the proportion of GM DNA.

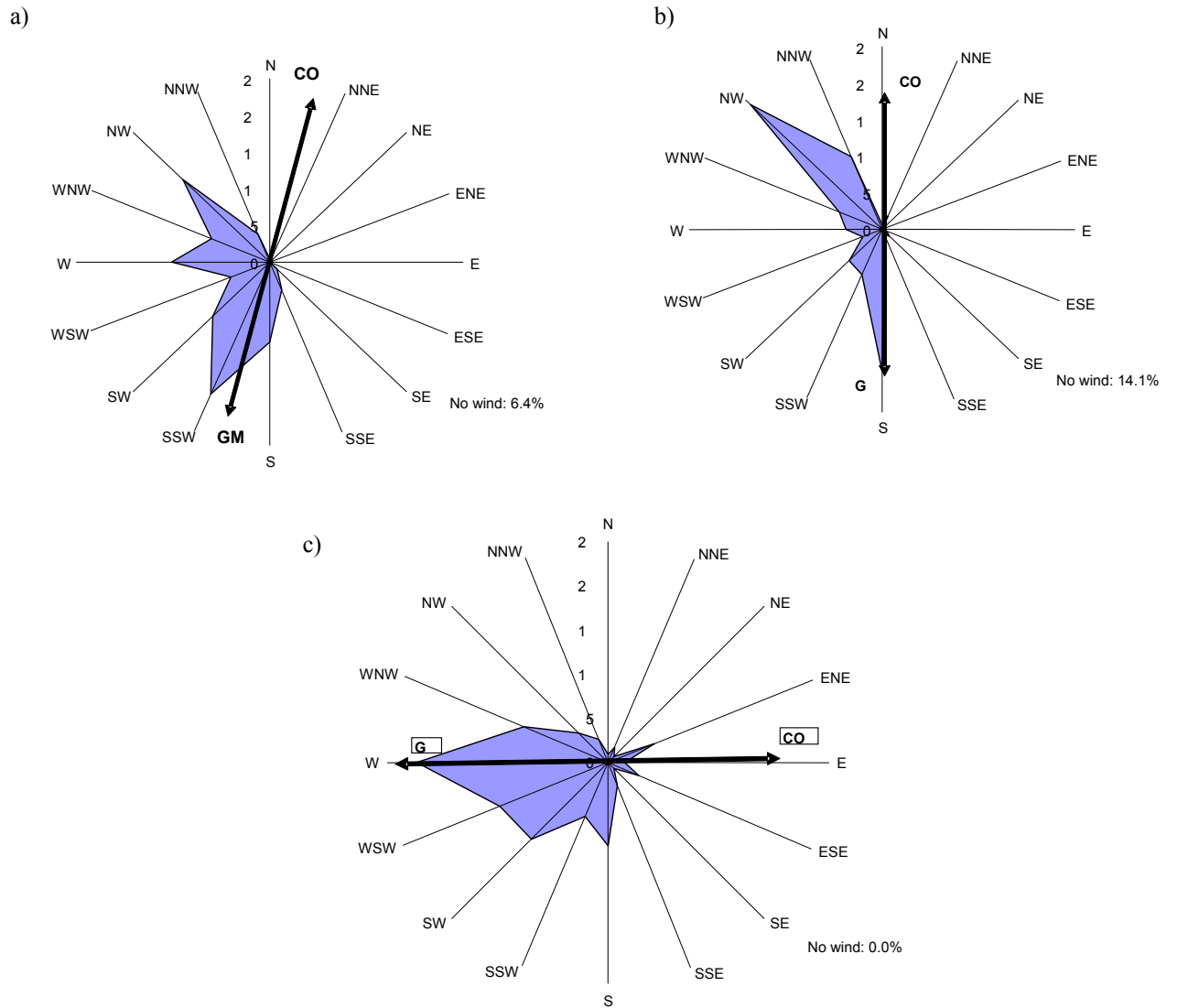


Figure 6. Wind roses representing the percentage wind direction during the overlapping flowering period at sites a) , b) and c). The shaded area represents the direction that the wind was blowing from. The orientation of the GM and conventional (CON) crops are denoted by an arrow.

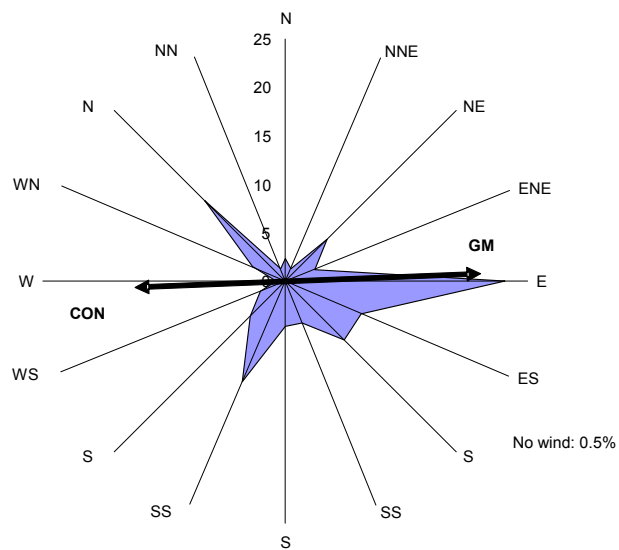
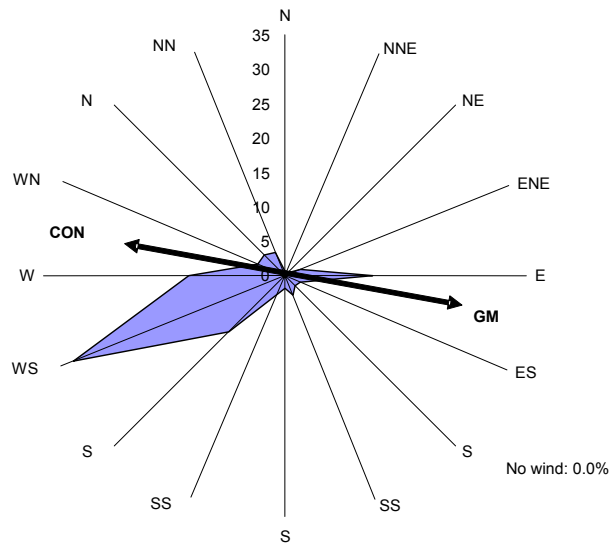


Figure 7. Wind rose representing the percentage wind direction during the overlapping flowering period at the sites d) and e). The shaded area represents the direction that the wind was blowing from. The orientation of the GM and conventional (CON) crops are denoted by an arrow.

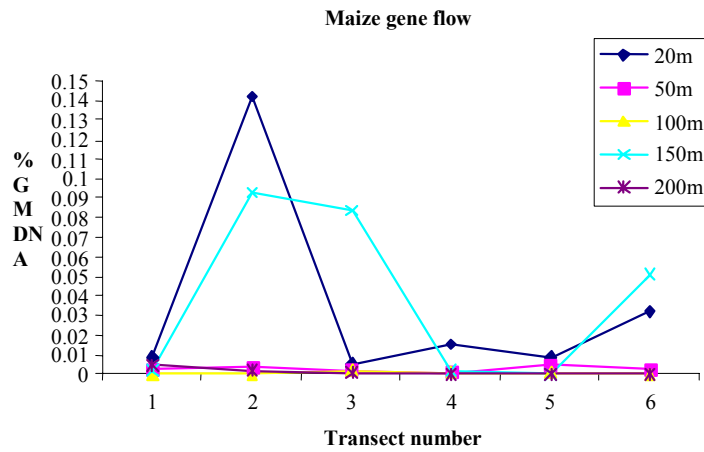


Figure 8. Percentage GM DNA at different distances into the conventional crop, along 6 transects, for one of the fields.