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# Mathematical Models for Gene Flow from GM Crops in the Environment

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**Abstract:** Risk assessment of gene flow from GM crops into the environment requires both the development of physical transport models and biological models for the assessment of outcrossing probabilities. Our starting point is a Lagrangian approach for pollen dispersal, which describes the concentration statistics in terms of the stochastic properties of the paths of ensembles of particles. Transport of a particle from a location  $(x',y',z')$  to a location  $(x,y,z)$  is mediated by a probability density or transfer function  $Q(x,y,z/x',y',z')$ . The transfer function depends on the statistics of the wind field during pollination. The total amount of pollen, which reaches a single plant, is then derived by the integral over all donators. In the context of gene flow, particle transport is but one aspect. The target variable is not primarily pollen density but the amount of outcrossing. The transfer function  $Q$  thus has to take into account both transport and biological processes and is devised to combine a transport submodel capable of integrating the statistics of wind velocities, a pollen viability submodel, a phenological submodel, a submodel for pollen redistribution by insects and a pollen competition submodel. Model parameters are estimated from data of outcrossing studies of maize and oil seed rape. The model is then applied to study the effect of field geometries on outcrossing rates.

**Keywords:** gene flow modelling, spatial spread of genetic information

## 1. INTRODUCTION

Since October 2002, the European Environmental directive 2001/18/EG is brought into force. It regulates the permit for commercial and experimental release of genetically modified (GM) plants. An important feature of the directive is the strict risk assessment with regard to direct and indirect environmental effects. Furthermore, threshold values of tolerated contaminations by GM material in food and food ingredients are established (Commission Regulations No 49/2000). The unwanted dispersal of genetic information from GM crops to neighbouring conventional crops or related weeds is possible and negative consequences are not excluded by now. Fundamental aim of all efforts is to minimize the risk and to enable coexistence between GMO and non-GMO agriculture.

Prerequisite of risk assessments and management programs is the estimation of pollen transport. With the model presented here, simulations of pollen transport and outcrossing rates are possible.

The model takes into account physical transport processes as well as biological influences. Biological influences are for instance the degree of cross-pollination, the influence of insects or the overlap of fertile periods of donor and target population. After some simulation studies to demonstrate the model characteristics, the simulation results of outcrossing experiments with maize and oil seed rape are shown below.

## 2. THE MODEL

### 2.1 List of Symbols

$x, y, z$	coordinates of the observation point
$x', y', z'$	coordinates of the source point
$\phi$	wind direction
$a_x, a_y, a_z$	dispersion coefficient in x-, y- and z-direction
$q$	pollen release rate [kg m <sup>-3</sup> s <sup>-1</sup> ]

$\alpha, \beta$	parameter of the wind distribution [°]
$s$	standard deviation of the wind distribution [°]
$w$	mean wind direction [°]
$u$	wind speed [m/s]
$Q(x, y, z x', y', z')$	probability density function for transport from location $(x', y', z')$ to $(x, y, z)$
$G$	Source region
$S(x, y, z)$	source density [kg m <sup>-3</sup> s <sup>-1</sup> ]
$f(u, \phi)$	Probability density function for wind speed and direction
$C_{wind}(x, y)$	particle concentration after averaging over the wind distribution [kg m <sup>-3</sup> ]
$GM, REC$	indices for transgenic and not-transgenic donor populations
$INSECT$	index for influence of insects
$b(x, y)$	insect activity
$P(x, y)$	outcrossing probability [%]
$c_b(x, y)$	pollen density of background sources [kg m <sup>-3</sup> ]
$K$	efficiency factor
$\eta$	portion of fertile target plants
$\gamma$	efficiency factor for fertilization by background sources
$\mu$	efficiency factor for outcrossing

## 2.2 General concept

The Lagrangian theory is used to describe the dispersal of particles in the atmosphere (generally in fluids). It is based on the stochastic description of distances of a group of particles by way of using a probability density function  $Q(x, y, z|x', y', z')$  for the transport from location  $(x', y', z')$  to  $(x, y, z)$ . The concentration at location  $(x, y, z)$  is given by the integral over the product of the source  $S(x', y', z')$  and the density function.

$$c(x, y, z) = \iiint_G Q(x, y, z|x', y', z') S(x', y', z') dx' dy' dz' \quad (1)$$

The intention, however, is not to calculate pollen density, but to calculate the outcrossing rate which not only depends on a single transport event, but integrates the entire previous history, i.e. weather and competition during the entire fertile period. In addition, pollen is distributed by insects. An outcrossing event depends on

- The pollen concentration at the location  $(x, y, z)$  of donor and target population.
- The degree of self-fertilization of the target plants
- The overlap of the fertile period of the donor population and the sensitive phase of the target population.

The goal is to find a formulation within the scope of the Lagrangian approach which provides the probability of gene transfer at location  $(x, y, z)$ ,  $p_g(x, y, z|M, B, G)$ , in dependence on meteorological variables  $M$ , biological variables  $B$  and geometrical parameters  $G$ .

## 2.3 The Transfer function

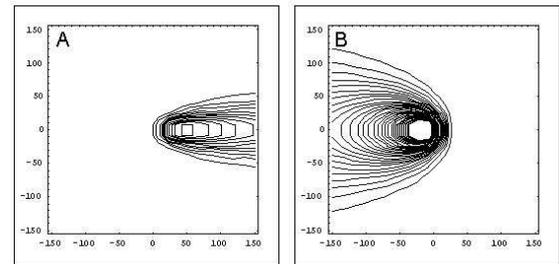
For the probability density function in the Lagrangian approach, a Gauss dispersal model is used. It provides a stationary particle transport from a point source at location  $(x', y', z')$  in a stationary wind field. The resulting concentration field is given by

$$C(x, y, z|x', y', z', u(\phi)) = \frac{q}{2\pi\sqrt{a_y \cdot a_z}} \exp\left[\frac{u(\phi)}{a_x} r - (x-x')\cos\phi - (y-y')\sin\phi\right]$$

with (2)

$$r^2 = ((x-x')\cos\phi + (y-y')\sin\phi)^2 + \left(\frac{a_x}{a_y}\right) \left(- (x-x')\sin\phi + (y-y')\cos\phi\right)^2 + \left(\frac{a_x}{a_z}\right) z^2$$

Figures 1A and 1B show projections of the plume for two different wind directions and wind velocities  $u$ .



**Figure 1.** Projections of plume concentrations (arbitrary units). Spatial units are m. A: In main wind direction with  $u = 4$  m/s. B: In opposite wind direction with  $u = 0.5$  m/s.

## 2.4 Influence of wind velocity and wind direction

Equation (2) is valid for a point source in a wind field with velocity  $u$  and direction  $\phi$ . However, the entire previous history of weather during the fertile period has to be taken into account. This is summarized in a bivariate probability density

function  $f(u, \phi) = f_\phi(\phi) f_u(u, \phi)$ , which is derived from the histograms of wind direction and wind velocity. The single plume equation (2) is weighted according to wind direction and strength via the integral

$$c_{wind}(x, y|x', y') = \int_0^\infty \int_0^{2\pi} c(x, y|x', y', \phi) \cdot f(u, \phi) du d\phi \quad (3)$$

For the evaluation of the density function  $f(u, \phi)$  only those readings are taken into account, which fall into periods when pollen emission is possible (daytime, favourable moisture conditions). Technical remark: the density functions  $f_u(u, \phi)$  were modelled by truncated normal distributions and the density function  $f_\phi(\phi)$  was approximated by a periodic interpolation polynomial. Figure 3 shows an example.

## 2.5 Influence of field geometry

The outcrossing probability is determined by the geometry of the fields of the donor and target populations. The GMO-pollen concentration at location  $(x, y)$  is obtained by integrating over the GMO plot with the source density  $S(x', y')$ .

$$c_{GM}(x, y) = \iint_G c_{wind}(x, y|x', y') S_{GM}(x', y') dx' dy' \quad (4)$$

In the case of a constant source density

$$S(x', y') = \begin{cases} 1 & (x', y') \in G \\ 0 & \text{else} \end{cases}$$

## 2.6 Competition between donor and target pollen

The outcrossing probability is determined by the competition between the pollen of donor and target populations

$$P(x, y) = \kappa \frac{c_{GM}(x, y)}{c_{GM}(x, y) + \eta c_{REC}(x, y) + \gamma c_b(x, y)} \quad (5)$$

while  $c_{REC}(x, y)$  is to be computed by an integral analogously to equation (4). The efficiency factor  $\kappa$  reflects possible advantages of pollen of the recipient population over the donor population.

If  $c_{GM} \ll c_{REC}$ ,  $c_{REC}$  is constant and pollen from background sources  $c_b$  can be neglected, then the outcrossing probability is approximately proportional to the pollen concentration of the donor population with proportionality factor  $\mu$

$$P(x, y) = \mu c_{GM}(x, y). \quad (6)$$

For the analysis of the experimental data (cf. section 4) this form was used.

## 2.7 Influence of cross-pollination by insects

The pollination by insects can easily be integrated into the Lagrange approach [Walklate et al., 2004]. The activity of insects leads to a redistribution of the pollen originally distributed by the wind. This process is described in the frame of Lagrangian theory by

$$C_{INSECT}(x, y) = \iint_G Q_{INSECT}(x, y|x', y') C_{GM}(x', y') dx' dy' \quad (7)$$

where  $Q_{INSECT}(x, y|x', y')$  is a suitable transfer function for redistribution of pollen by insects.

The transfer function takes into account that only part of the pollen is redistributed and is therefore formulated as the sum of a redistribution function  $g(x-x', y-y')$  and a dirac delta distribution.

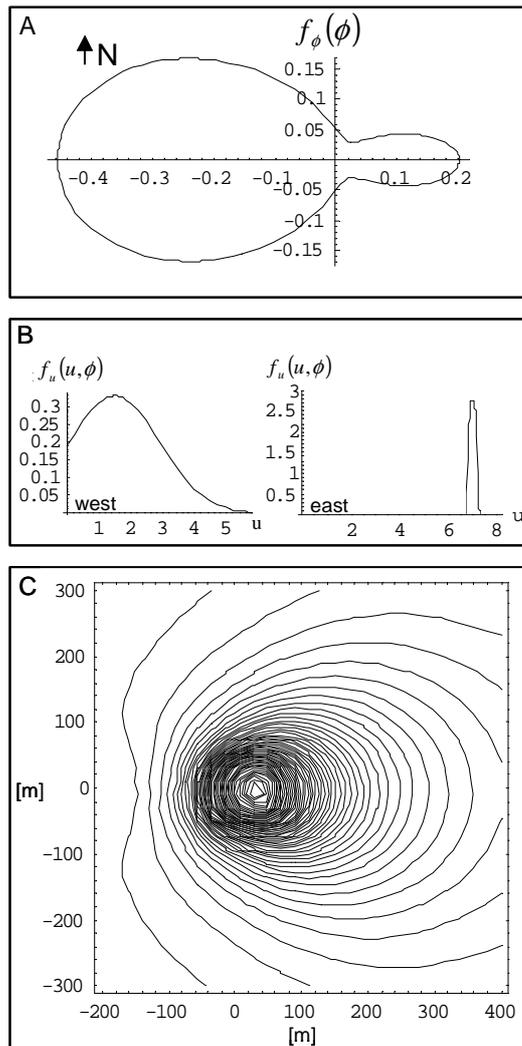
$$Q_{INSECT}(x, y|x', y') = b g(x-x', y-y') + (1-b) \delta(x-x', y-y') \quad (8)$$

In our simulation we followed the approach of Walklate [2003], who used a rectangle transfer function with border lengths  $\Delta x$  and  $\Delta y$ . This approach was further modified by the superposition of a large scale insect activity distribution function  $b(x, y)$ .

## 3. SIMULATION STUDIES

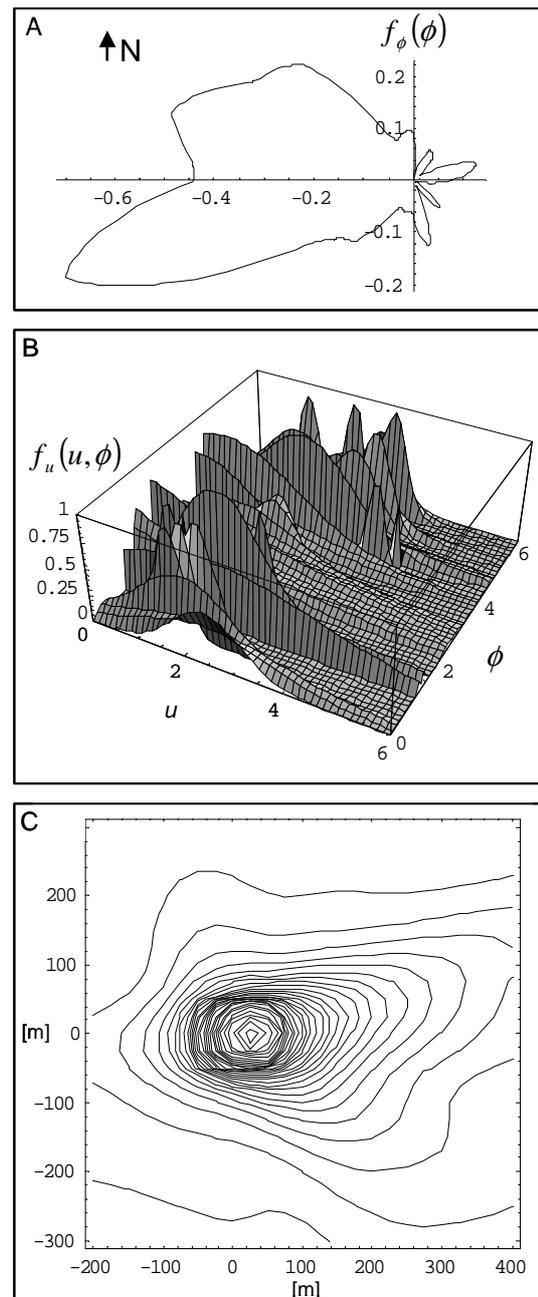
### 3.1 Influence of wind velocity and wind direction

A wind field with main wind direction from west and a short but strong wind event from east direction is constructed. Figures 2A and 2B show the corresponding wind rose and the two opposite distributions of wind velocities. The resulting pattern of pollen distribution (Figure 2C) clearly reflects the form of the wind distribution.



**Figure 2.** Wind field and corresponding pollen dispersal. A: Wind rose with local maxima in west and east direction (polar plot). B: Distributions of the wind velocities; gentle shape in the west and strong wind forces in the easterly wind direction. C: Resulting dispersal pattern of pollen concentration, projection.

Figure 3 shows the same kind of plots based on real wind data. The wind distribution function (Figure 3B) is based on 24 empirical velocity distributions (cf. 2.4). The corresponding dispersal pattern (Figure 3C) is considerably distorted but is still showing the predominance of south westerly winds.

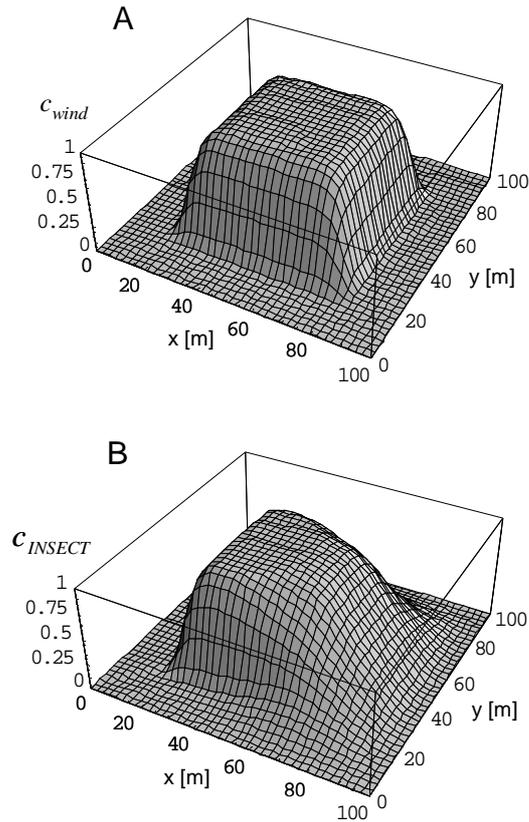


**Figure 3.** Wind field and pollen dispersal based on a real data set (data courtesy of P. Zwerger and A. Dietz-Pfeilstetter, Biological Research Centre for Agriculture and Forestry, Braunschweig, Germany).

A: Wind rose (polar plot). B: Distributions of wind velocities. C: Resulting dispersal pattern of pollen concentration.

### 3.2 Pollination by insects

A fictive pollen distribution is shown in Figure 4A. The redistribution by insects (Figure 4B) is carried out by an increasing insect activity from  $f(x,y)=0$  to  $f(x,y)=1$  in east direction. The original strong decline of concentrations is smoothed with increasing insect activity



**Figure 4.** Redistribution of pollen by insects. A: Original pollen distribution by wind transport. B: Redistribution with increasing insect activity in easterly direction.

#### 4. COMPARISON WITH EXPERIMENTAL DATA

In the following examples, the parameter identification proved to be cumbersome because of large computer time. The reason is that the calculating the objective function

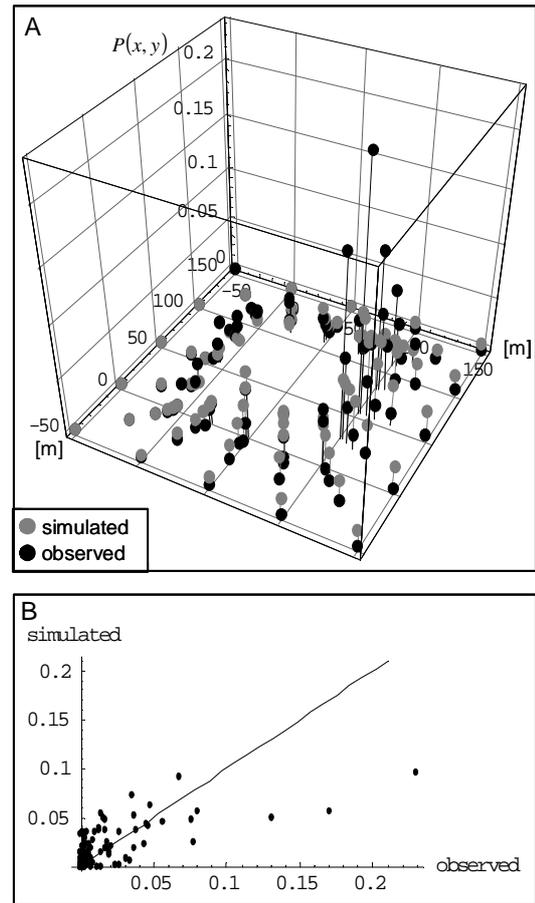
$$SQ(a_x, a_y, q) = \sum_{i=1}^n [c(x_i, y_i | a_x, a_y, q) - P(x_i, y_i)]^2 \quad (9)$$

necessitates the evaluation of multiple integrals for each measurement point. For the parameter identification the FindMinimum procedure in Mathematica was employed, which is based on the Levenberg-Marquardt algorithm. The multiple integrals were evaluated by the adaptive Genz-Malik algorithm, which is also implemented in Mathematica.

##### 4.1 Maize

The model was applied to cross-pollination data from a monitoring farm scale experiment with maize carried out at the federal Biological Research Centre for Agriculture and Forestry (BBA) in Braunschweig, Germany. Objective of

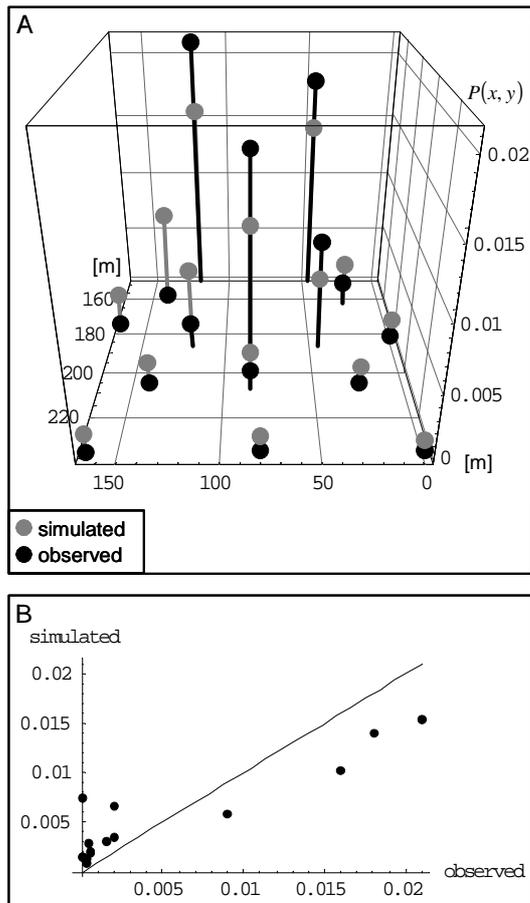
the experiment was to establish methods for quantifying transgenic contamination in the crop by outcrossing [Schiemann et al., 2002]. In the year 2000, the field trial was carried out with an herbicide-resistant line of maize (*Zea mays*). The transgenic square with a size of 100- by 100 m was surrounded by crops of ordinary maize of the same variety. Altogether 96 sampling points were monitored. The results of the simulations are shown in Figure 5.



**Figure 5.** Gene flow from transgenic maize. A: Direct comparison between simulation and data (percent outcrossing). B: Correlation between data and model predictions.

##### 4.2 Oil seed rape

Furthermore the model was applied to the data of an experiment with oil seed rape (*Brassica napus*) in the year 1999/2000, carried out by the BBA Braunschweig as well. Two different herbicide-resistant lines were used (Glufosinat- and Glyphosat-resistance) and the outcrossing in the neighbouring transgenic field and in the surrounding non-transgenic seed was examined [Dietz-Pfeilstetter et al. (2004)]. The data shown here refer to the transference of Glufosinat-Resistance (Liberty Link, LL).



**Figure 6:** Gene flow from transgenic oil seed rape. A: Direct comparison between simulation and data (percent outcrossing). B: Correlation between data and model predictions.

## 5. DISCUSSION

The modelling approach presented here allows to explicitly incorporate the statistics of wind velocity and wind speed into Lagrangian based transfer models. Although the general dispersal patterns are fitted quite well, systematic deviations between observed and simulated values occur. At large distances from the source the model tends to overestimate outcrossing rates, whereas outcrossing rates are underestimated at very short distances. This discrepancy can be removed, as was shown by Loos et al. (2003) for a simple Lagrangian approach by a superposition of two Gaussian plumes distinguishing between far and near transport processes. Further model developments concern the incorporation of landscape structures and an improvement of the fitting algorithms.

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