Application of an atmospheric gene flow model for assessing environmental risks from transgenic corn crops

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Abstract: Gene flow data from experiments under limited environmental conditions (e.g. wind speed and direction, atmospheric stability) have only provided limited information for gene flow risk management. It is necessary to apply models to predict the gene flow under a complete set of possible environmental conditions to inform farmers, seed companies, government agencies, and researchers about the risks and potential prevention and precaution methods. In this paper, the previous validated gene flow model developed by the authors was used to predict gene flow from genetically modified (GM) corn crops. The model was used to simulate potential gene flow from GM corn sources of different sizes from one plant area of 0.1 m^2 to an area 3.1×10^6 m^2 under normal weather conditions. In addition, the model was also used to predict the potential gene flow for different source strengths, atmospheric conditions, buffer heights, buffer field sizes, and pollen settling speeds from 10,000 m^2 sources. The model simulations have provided gene flow information for risk management for the above conditions and have shown that the source sizes, source strengths, buffer heights, buffer sizes, atmospheric conditions, and pollen settling speeds had important effects on gene flow. While the atmospheric conditions and pollen settling speeds cannot be controlled, choosing appropriate buffer heights and sizes will effectively prevent gene flow. The lost seed control is crucial to limit gene flow because even a GM corn plant can result in a grand total deposition flux of 646,272 grains/m^2, an outcrossing ratio of 0.016, and outcrossed seed of 110 kernels/m^2 at 0.8 m from the plant in the non-target field under normal atmospheric conditions.

Keywords: model, risk management, crops, corn, pollen, gene flow, random walk, outcrossing
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1 Introduction

Since the first introduction of a genetically altered microscopic bacterium for devouring oil spills in 1971[1], DNA technology (popularly referred to as genetic engineering or genetic modification) development and application have rapidly accelerated, especially in agricultural and pharmaceutical processes and products. In agriculture, scientists use recombinant DNA technology to introduce genes for a desired trait from either the same or different species to produce novel (transgenic) plants with special characteristics to resist particular diseases, chemicals, or environmental stress for higher yields and/or better quality (U.S. Congress Office of Technology Assessment, 1992). It is estimated that in 2006, approximately 53.4 million hectares of land were planted with transgenic plants in the United States[2].

Transgenic corn was one of the first four pest-resistant crops to flow from the industrial R&D pipeline to commercial production. It is estimated that one-third of all cornfields in the United States are planted

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to Bt corn[1]. This genetically modified crop (GMC) contains the *Bacillus thuringiensis* crystal toxin (Bt cry) transgenes to overcome constraints and stresses of insect pests. According to Sweet et al.[3], transformations that significantly overcome constraints and stresses of pests, diseases, or adverse environmental factors (such as drought) in general have the highest potential to affect the environment. Although corn pollen is relatively heavy, the large-scale of production of the plants makes transgenic corn a crop with a high level of environmental impact from pollen dispersion.

Effective management of environmental hazards is heavily dependent on comprehensive knowledge of risk assessment. Since 1992, the United States Department of Agriculture (USDA), Environmental Protection Agency (EPA), and other government agencies have initialized research programs to develop and assemble such badly needed knowledge. Many research projects have been or are being conducted, which have generated sizable data, valuable experiences, reliable protocols, and useful information in this regard. For example, numerous experiments have been conducted to estimate gene flow from maize crops, starting as early as the first half of the past century[4-8]. However, most of the experiments were with small source plant fields under specific environmental conditions (e.g., atmospheric stability, wind speed and direction). Because of differences between experiment designs and environmental conditions, these experiments had significantly different experimental results one from another. Comprehensive field studies on gene flow under different conditions of GMC field size, source production, wind speed, and atmospheric stability are costly and not practical. This demands development and application of models in gene flow studies.

Mechanistic models have been developed for simulating gene flow[4, 9-11]. However, these models have rarely been applied to predict the gene flow risks under different environmental conditions to provide farmers, seed companies, researchers, and government agencies with information for management purposes and to provide potential prevention methods.

Wang and Yang[12] developed and validated a gene flow model for GM corn crops. The inputs of the model include friction velocity (wind speed parameter), wind direction, atmospheric stability, precipitation, and field sizes and species of source, buffer and receptor. The simulation domain was defined as circular areas; the source was in the inner circle area, the receptor was in the outer circle area, and the buffer was in between (Figure 1). The model can predict the dynamic pollen release, 3-D dispersion, 2-D deposition at silk height, and final 2-D outcrossing in the receptor field. The dynamic pollen release, dispersion, and deposition are based on the weather condition (precipitation), canopy structure, and wind field in the simulation domain. The wind field is calculated based on the atmospheric and canopy structure inputs. Finally, the model accumulates the pollen deposition at silk height in the receptor field over the whole pollination season, and the outcrossing ratio is calculated as a function of the grand total pollen deposition.

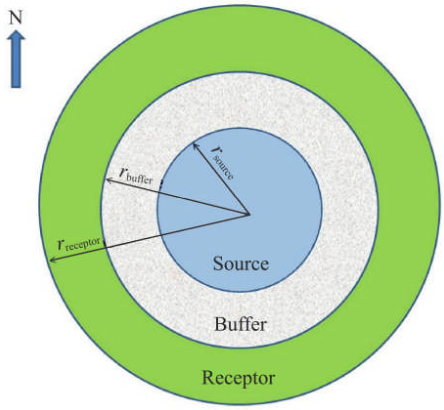

Figure 1 Simulation domain of the gene flow model in Wang and Yang[12] buffer

The objective of this paper was to use the validated gene flow model of Wang and Yang[12] to predict the risks of gene flow from GM corn crops to determine possible prevention methods to aid in gene flow and seed contamination management.

2 Materials and methods

The validated gene flow model[12] was used to predict the potential gene flow from GM corn crops with source size ranging from 0.2 m to 1,000 m in radius (from a single plant to a 3.0×10^6 m^2 area) under normal

atmospheric conditions (see Table 1 for the default atmospheric and plant conditions). The model was also used to simulate potential gene flow from a 10,000 m^2 GMC field with different source strengths (1 to 500 grains/m^2/s), buffer heights (0 to 5 m), buffer sizes (0 to 200 m), wind strengths (u_* from 0.05 to 1.5 m/s), atmospheric stabilities (L from -1,000 to 1,000 m), and pollen settling speeds (0.05 to 0.4 m/s).

Table 1 Default conditions used for simulations of different source field sizes, source strengths, buffer heights, buffer field sizes, atmospheric conditions, and pollen sizes

Input	u_* /m · s^{-1}	L /m	Source strength /grains · m^{-2} · s^{-1}	Source radius/m	Buffer radius /m	Source plant	Receptor plant	Buffer plant	Pollen release height/m	Deposition predicted height/m	Pollen settling Speed/m · s^{-1}
Value	0.2	-15	136	56 (1 ha)	56 (no buffer)	8,464 Wx Grain corn	8,419 W Grain corn	none	2.9	1.8	0.31

The average daytime source strength of 136 grains/m^2/s during pollination season, measured in Wang and Yang[12], was used as a default. Canopy characteristics of the source and receptor plants were represented by those of the 8,464 Wx and 8,419 W plants, respectively (Garst Seeds Company, Slater, IA)[12]. In all the simulations, receptor plants were assumed to surround the source with a buffer. The buffer size ranged from 0 to 200 m radially.

In simulations with different buffer heights, the buffer size was set to 40 m radially. The buffer was set to different heights of 0, 0.75, 1.5, 2.9, and 5 m. The height of 0 m represented the bare ground buffer. The canopy characteristics for buffer with a height of 0.75 m used wheat data in Aylor[13]. The plant characteristics for buffer with height of 1.5 m and above used 8,419 W corn data, proportionally allocated into eight layers in the vertical direction.

The domain of the simulations was set by the radius in the receptor field where the deposition flux density was reduced to 0.1% of that at the edge of the source area. The total simulation period was from 15 min to 2 h corresponding to the size of the source and receptor fields.

The output of the model includes 3-D concentration (grains/m^3), 2-D deposition flux density (grains/m^2/s) at 1.8 m silk height, and 2-D outcrossing ratio from source plant. The deposition output was accumulated over time for the whole pollination season. The 1% and 0.1% distances (at which the predicted pollen deposition flux density was 1% and 0.1% of the source strength, respectively) were determined along the prevailing wind direction.

For all simulations except for different source sizes, the total deposition flux (TDF, grains/m^2) during pollen viable period (2 h[14]) was estimated at each prediction location by multiplying the deposition flux density by 2 h. For simulations for different source sizes, the grand total pollen deposition flux for the whole pollination season was estimated by multiplying the deposition flux density by daily shedding time of 12.5 h (0700-1930) and the pollination season of nine days observed in Wang and Yang[12]; the outcrossing ratio (OutR, outcrossed seed number divided by the total seed number on a receptor ear) was estimated by Equation 10 in Wang and Yang[12]. By assuming receptor field with density of 71,000 plants/ha with two ears per plant and 500 kernels per ear observed in Wang and Yang[12], the actual outcrossed seed number (OSN) was also estimated (kernels/m^2).

$$OSN = 71000/10000 \times 2 \times 500 \times OutR \quad (1)$$

The 1% and 0.1% distances for grand total deposition in the prevailing wind direction were determined as the distance where the deposition was 1% and 0.1% of the deposition at the source field edge. Similarly, the 1% and 0.1% distances in the prevailing wind direction for outcrossing ratio are the distances where the outcrossing ratio was 1% and 0.1%.

3 Results and discussion

As expected, source size had important effects on pollen dispersion. The bigger the size, the longer the 1% and 0.1% distances. With the increase of source area from 0.1 to 3.14×10^6 m^2, the 1% and 0.1% distances increased from 0.8 m to 98 m and 1.5 m to

1,435 m, respectively (Table 2). Larger-scale source plants produce more potential of gene flow as stated in Squire et al.[15] "The flow of genes from some crops to some wild relatives is possible and will happen if GM crops are deployed on a large scale." When there was one GM corn plant in a non-target field (source area of 0.1 m^2), the 0.1% distance was at 1.5 m away from the plant, with potential grand total deposition flux of 64,627 grains/m^2, outcrossing ratio 0.002, and outcrossed seed of 10 kernel/m^2; the 1% distance was at 0.8 m, with grand total deposition flux of 646,272 grains/m^2, outcrossing ratio of 0.016, and outcrossed seed of 110 kernels/m^2. Gene flow from lost seeds was reported in the paper by Palaudelmàs et al.[16]. The paper stated that lost rice seeds could potentially contribute to adventitious GM levels, especially at high initial densities (i.e. above 1,000 volunteers/ha). Lost oil-rape seeds can persist up to four years in fields, in normal cropping conditions and in the absence of cultivation one experiment has confirmed persistence for over eleven years[17] .

Table 2 1% and 0.1% distances (where the grand total deposition fluxes were 646,272 and 64,627 grains/m^2, outcrossing ratios were 0.016 and 0.002, and outcrossed seed numbers were 110 and 10 kernels/m^2, respectively) simulated from different size source fields (see Table 1 for the default weather and plant conditions)

Source radius /m	Source area /m^2	1% Distance/m	0.1% Distance/m
0.2	0.1 (one plant)	0.8	1.5
1	3	1.1	3.5
5	79	6	37
10	314	8	50
20	1,256	23	68
56	10,000	43	148
100	31,400	44	227
200	125,600	51	525
400	502,400	90	628
1,000	3,140,000	98	1,435

Source strength did not have effects on the 1% and 0.1% distances. However, the total deposition flux at the distances was linearly related to the source strength (Table 3) because the source strength is a linear scaling factor in deposition calculation equation (see Equation 6 in Wang and Yang[12]).

Table 3 Predicted total deposition flux (TDF, grains/m^2) during 2-h period at 1% and 0.1% distances from 10,000 m^2 GM corn plants for different source strengths under normal atmospheric conditions (see Table 1 for the default weather and plant conditions)

Source strength /grains · m^{-2} · s^{-1}	TDF at 1% distance of 43 m/grains · m^{-2}	TDF at 0.1% distance of 148 m /grains · m^{-2}
1	72	7
50	3,600	360
100	7,200	720
200	14,400	1,440
500	36,000	3,600

Wind speed had strong effects on corn pollen transport (Table 4). At weak wind speed (u_* = 0.05 m/s, i.e., mean wind speed was 0.24 m/s at one meter above canopy), the 0.1% distance was shorter than 2 m from 10,000 m^2 source. In the range of normal wind speed of from u_* = 0.1 to 0.3 m/s (wind speed 0.48 to 1.45 m/s), the distances increased quickly with u_*. The 0.1% distance increased from 7 m to 245 m. With strong wind of u_* = 1.5 m/s (wind speed = 7.27 m/s), the 0.1% distance can reach to 370 m with 98 pollen grains/m^2 in 2 h.

Table 4 Predicted 1% and 0.1% distances during two-hour simulation period from 10,000 m^2 GM corn plants for different u_* (see Table 1 for the default weather and plant conditions)

u_* /m · s^{-1}	Wind speed at one meter above canopy/m · s^{-1}	1% distance (TDF = 9,800)/m	0.1% distance (TDF = 980)/m
0.05	0.24	1.2	1.5
0.1	0.48	1.6	7
0.2	0.97	43	148
0.3	1.45	68	245
0.8	3.88	69	308
1.5	7.27	70	370

Atmospheric stability conditions had weaker effects on corn pollen transport than wind speed. Table 5 shows that, under normal wind speed (u_* = 0.2 m/s, i.e., wind speed = 1 m/s), atmospheric stability varied from very stable to very unstable. Under the very unstable condition (L = -5 m), the distances were the maximum; the 1% and 0.1% distances were 43 and 163 m, respectively, with total deposition flux of 9,800 and 980 pollen grains/m^2, respectively, during two-hour period. Under neutral to stable condition, the distances were the minimum.

Table 5 Predicted 1% and 0.1% distances with total deposition flux (TDF, grains/m^2) during 2-hr period from 10,000 m^2 GM corn source plants for different atmospheric stabilities (see Table 1 for the default weather and plant conditions)

L/m	Atmospheric state	1% distance (TDF = 9,800)/m	0.1% distance (TDF = 980)/m
-5	Very unstable	43	163
-15	Moderate unstable	43	148
-50	Unstable	31	96
-1,000	Neutral	29	82
1,000	Slightly stable	27	82
50	Stable	26	83

Buffer heights had important effects on pollen transport (Table 6). With the increase of buffer height from 0 to 5 m, the 1% and 0.1% distances from the leading edge of the receptor decreased from 48 to 0 m and from 206 to 75 m, respectively. Buffer size also had important effects on pollen transport (Table 7). With the buffer size increased from 0 to 200 m, the 1% and 0.1% distances decreased from 43 to 0 m and from 148 to 0 m, respectively. Literature supported these opinions. For example, Song et al.[18] suggested that tall sugarcane buffer zone can be planted between transgenic rice and non-target fields to prevent gene flow. Damgaard and Kjellsson.[19] stated that the use of a width of 5 m buffer zone (the buffer zone used receptor plants) surrounding the non-oil-rape-GM-field is expected to reduce GM-pollination by about a third.

Table 6 Predicted 1% and 0.1% distances from receptor leading edge with total deposition flux (TDF, grains/m^2) during two-hour simulation period from 10,000 m^2 GM corn source plants for different buffer heights under normal atmospheric condition (buffer size was 40 m radially; see Table 1 for the default weather and plant conditions)

Buffer height /m	Buffer plant	1% distance (TDF = 9,800)/m	0.1% distance (TDF = 980)/m
0	Bare ground	48	206
0.75	Wheat	18	135
1.5	Short corn	5	109
2.9	Tall corn	3	108
5	5 m tall plant[(a)]	0	75

Note: [a] The 1.5, 2.9, and 5 m tall plant buffers used 8419W plant characteristics in each corresponding layer of 8 total layers.

Table 7 Predicted 1% and 0.1% distances from receptor leading edge with total deposition flux (TDF, grains/m^2) during two-hour simulation period from 10,000 m^2 GM corn source plants for different buffer sizes (buffer plant characteristics used 8419W data; see Table 1 for the default weather and plant conditions)

Buffer size/m	1% distance (TDF = 9,800)/m	0.1% distance (TDF = 980)/m
0	43	148
50	0	98
100	0	0
200	0	0

Pollen settling speeds had important effects on transport (Table 8). The smaller the speed, the longer the 1% and 0.1% distances. With the decrease of the pollen settling speed from 0.4 to 0.05 m/s, the 1% and 0.1% distances increased from 22 to 99 m and from 72 to 394 m, respectively.

Table 8 Predicted 1% and 0.1% distances during two-hour period from 10,000 m^2 GM plants for different pollen settling speeds under normal atmospheric conditions (see Table 1 for the default weather and plant conditions; TDF = total deposition flux, grains/m^2)

Settling speed /m·s^{-1}	1% distance (TDF = 9,800)/m	0.1% distance (TDF = 980)/m
0.4	22	72
0.3	43	148
0.2	75	276
0.1	76	280
0.05	99	394

4 Conclusions

The 1% and 0.1% distances of pollen deposition were obtained, and outcrossing ratios and outcrossed seed numbers at the distances were estimated by model simulations for different size GM corn sources from one plant area of 0.1 m^2 to an area 3.1×10^6 m^2 under normal weather conditions. The simulations provided the 1% and 0.1% distances of deposition for different source strengths, buffer heights, buffer field sizes, wind strengths, and atmospheric stabilities from 10,000 m^2 GM corn source field; the simulation also provided the distances for different pollen settling speeds for other 10,000 m^2 GM species. The application of the model has shown that the source sizes, source strengths, buffer heights,

buffer sizes, atmospheric conditions, and pollen settling speeds had important effects on gene flow. While the atmospheric conditions and pollen settling speeds cannot be controlled, choosing appropriate buffer heights and sizes will effectively prevent gene flow.

Gene flow experimental results from small-size sources are not appropriate to give guidelines for gene flow control for large-scale GM sources because gene flow rate is sensitive to source size. More attention should be paid to large-scale GM field conditions. For a 3.14×10^6 m^2 GM cornfield during the whole pollination season under normal atmospheric conditions, at 1,435 m distance, the non-target plants may obtain pollen of 64,627 grains/ m^2, outcrossing ratio of 0.002, outcrossed seeds of 10 kernels/m^2.

Gene flow experiments under different atmospheric conditions and canopy structures or for different species may not be comparable. The lost seed control during harvest or processing is crucial to limit gene flow because even a GM corn plant can result in grand total deposition flux of 646,272 grains/m^2, outcrossing ratio of 0.016, and outcrossed seed of 110 kernels/m^2 at 0.8 m from the plant in the non-target field under normal atmospheric conditions.

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