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A stochastic model of maize gene dispersal

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A stochastic model of maize gene dispersal

by

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A thesis submitted to the graduate faculty
in partial fulfillment of the requirements for the degree of

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Program of Study Committee:
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This is to certify that the master's thesis of

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has met the thesis requirements of Iowa State University

Signatures have been redacted for privacy

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ABSTRACT

The spatial distribution of maize genes is predicted using a two dimensional Lagrangian stochastic (LS) model that meets sub-inertial and well-mixed conditions. Using wind data from central Iowa, a Weibull distribution is fit. From this distribution, a series of Monte Carlo simulations are run using the LS model in three forms. The final two forms of the model are based on USDA regulations for the production of pharmaceutical producing corn, demonstrating the model's suitability for analyzing the risk of transgene dispersal in maize. Special considerations are given to the large size of maize pollen, the likelihood of pollen seepage given the use of biological inhibitors of gene release, and the possibility of contemporaneous fertility of neighboring corn fields.

CHAPTER 1. INTRODUCTION

Segregation of genetic material has long been an objective of those involved in production agriculture. Plant breeders desire first to isolate specific traits and then buildup seed supplies while retaining marketable traits for distribution. Producers of niche products often follow predetermined processes that may include minimizing or eliminating the amount of exposure of their produce to transgenes. Commodity crops grown for use as either food or feed are jeopardized by the possible introduction of certain transgenes by small-scale operations.

The introduction of Starlink corn, a variety limited to use as animal feed, entered the food supply in 2000. Widespread concern about the impacts of the presence of Starlink resulted in the withdrawal of food products from grocery shelves and the loss of export markets most notably the European Union, Japan, and South Korea. In December of 2002, more than three years after the onset of the Starlink situation and the last time the variety was planted, grain in transit to Japan tested positive for Starlink demonstrating the long-term effects of a single event. A recent settlement between Aventis, the makers of Starlink, and affected farmers reached \$110 million (Arasu 2003).

In October of 2002, transgenic corn, specifically plant-made-pharmaceutical (PMP) producing corn, in Iowa and Nebraska nearly entered the food supply as poor management and violation of federal guidelines

resulted in uncontrolled volunteer corn in soybean plots (Callahan 2003).

In response to general concerns reinforced by the impact of Starlink and the PMP corn situation, the North American Millers' Association stipulated a zero tolerance policy for the presence of restricted transgenes in food crops. At the same time BIO, a biotechnology industry group, advocated the movement of all PMP corn outside of the corn-belt (Statement 2002). The growing concerns of industry are echoed by consumers and social groups (Fabi 2003).

APHIS, the Animal and Plant Health Inspection Service, is responsible for regulating the import, export, and field testing of genetically engineered crops. The process to acquire permission to field test such crops is lengthy and thorough. Minimum management practices are outlined by the agency, with more rigorous demands placed on certain organisms as warranted.

Isolation distances for a number of crops grown under different circumstances follow a minimum allowable practice rule (APHIS 2002). The determination of isolation distances has relied on research using either the collection of pollen or counting of unique kernels at various distances from a source. This arrangement works relatively well for reducing the likelihood of genetic interaction to a predetermined level. However, it is unlikely to eliminate the risks of genetic contamination, a goal of many consumers and those in the food industry.

The following model provides significant advantages over previous studies of pollen dispersal which used sample data to estimate the true underlying spatial distribution. The construction of a physical model allows one to use quantifiable physical relationships to build the true spatial distribution of maize pollen. The model also allows for scenario testing based on different case parameters, something that would be virtually impossible in the field. Before constructing a physical model, relevant research in the fields of maize pollen dispersal, non-spatial methods of genetic control, and the modeling of the dispersal of small biological particles will be reviewed.

CHAPTER 2. REVIEW OF LITERATURE

To achieve greater understanding of the diverse areas of study that are necessary to construct an appropriate mathematical model, literature in the fields of maize pollen dispersal, mathematical models of pollen and spore dispersal, and biological and mechanical methods of gene control are reviewed. Two benefits result from the review of research on the dispersal of maize pollen: being able to see the shortcomings of the experiments and to provide an understanding for the relationship between distance and gene flow in these settings. The review looks at three families of mathematical models and their recent applications to biological dispersal in order to provide insight into relative benefits of the models and techniques used to fit them to specific situations. Developments in the field of biotechnology that may someday make spatial isolation of crops irrelevant are also considered.

Pollen and Gene Dispersal in Maize

Recent interest in the establishment of acceptable isolation distances of genetically modified (gm) varieties has rekindled interest in decades old research of pollen dispersal in field crops. Studies by Emberlin (1999), Treu and Emberlin (2000), Ingram (2000), and Fiel and Schmidt (2002) have surveyed the literature. The latter two had the explicit purpose of using the previous research to determine isolation distances in crops.

Feil and Schmid (2002) provide summaries of the methods used by pollen dispersal studies as well as the factors that support or hinder the movement of pollen by wind. The authors also recognize the difference between the dispersal of pollen versus the dispersal of genes. This is the distinction between a spatial distribution of pollen and the combined spatial and temporal distribution of viable pollen. Techniques to determine the travel of pollen fall into two groups: using sticky objects at varying distances and counting the amount of pollen present and allowing actual pollination of other plants. The presence of source pollen in the second category can be displayed by visually observable dominant traits, in the laboratory, or by the use of herbicide to check for genetic resistance present in offspring.

In their outline of determinants of pollen flow, Feil and Schmid denote a number of variables impacting pollen and gene dispersal. To increase the distance traveled, the receptor crop is located downwind from the source, convection is present, and the source is relatively elevated. With regards to pollination, pollen should be released at the same time the receptor crop flowers. Male or self sterility of the receptor crop, reducing the competition for pollination also increases the likelihood of gene dispersal.

Studies Utilizing Microscopic Slides

Jones and Newell (1946) looked at both the pollination cycles and dispersal of pollen in nearly thirty grass species. In 1944 and 1945, they collected pollen on greased microscopic slides placed at distances five to

sixty rods downwind from the pollen source at heights of two-and-one-half, five, and ten feet. Initially, slides were placed every forty five degrees from one another, but it was soon found that the predominant southerly winds resulted in countable pollen only on the slides in the north and west quadrants.

The following table presents the average daily pollen count for given distances for both years of the study. The first row shows the distances at which the pollen was counted, starting at the center of the corn field, the pollen source. There is a strong and immediate drop in the amount of pollen collected as one moves from the center of the field. It is of note that countable pollen did reach distances of 40 and 60 rods, the furthest distances at which pollen was collected.

Table 1. Average Daily Maize Pollen Count per Slide
(Jones and Newell, 1946)

Distance In Rods	Center	5	15	25	40	60
Count	203	18	5	4	1	1

As part of his three article study on the contamination of seed crops, A.J. Bateman looked at the role of wind pollination in a number of crops, including maize (1947). Bateman collected pollen on greased slides to the north, south, and east of a maize plot. Over the course of five exposures, he

laid out his slides for periods ranging from 24 to 48 hours under differing weather conditions.

The first two sites were located within the maize plot: Site 0 at the plot center with only one slide and Site 1 at the plots' edge with two slides one facing the center of the plot the other facing away. All other sites, positioned away from the plot center, were distanced twenty feet apart, with the same slide setup as Site 1. Table 2 shows the total number of maize pollen counted at each site, with Sites 0 and 1 within the plot itself. As in previous studies there is a quick decrease in corn pollen, yet at all distances some pollen was found.

Table 2. Total Pollen Count by Site (Bateman 1947)

Site	0	1	2	3	4	5	6	7	8	9
Count	7485	7330	1073	341	153	121	56	30	6	1

A study of pollen dispersal in maize while simultaneously researching the movement of ragweed and timothy pollen was conducted by Raynor, Ogden, and Hayes (1972). They planted corn in eighteen meter diameter plots, and then collected pollen using microscopic slide during periods of calm weather. This data was used to construct measures of plume width measured by crosswind distribution, denoted σ_y , and plume height, σ_z .

These measures are the basis for the Gaussian plume method of physical modeling described later.

Studies Utilizing Xenia

A study conducted at Iowa State University used statistical techniques to study the variation of maize kernel types in adjacent fields (Gacitua 1946). Here, thirteen fields of waxy corn were planted adjacent to starchy corn. The harvested kernels from each crop were then exposed to iodine which causes waxy corn to turn reddish-brown while starchy corn stains blue-black. Analysis of variance methods were used to infer the significance of distance on out-crossing. Gacitua's findings were similar to those of Jones and Newell described above. They included the strong decrease in contamination resulting from an increase in distance from the source and that the direction of the receptor plot from the source played a large impact on the amount of contamination.

Haskell and Dow conducted a similar experiment with rows of corn projecting out from the central plot, this time using visibly discernable kernels (1951). They went on to use analysis of variance techniques to determine the significance of both distance and direction, relative to the mean direction, on seed-setting. Haskell and Dow found that distance was significant at an alpha less-than .001 level, while the value for direction was greater than .2. A simple linear regression with distance as the independent variable versus the natural logarithm of the mean percent of seed-set

provided a good fit visually; however, the paper includes no quantitative measure of this quality.

Paterniani and Stort planted a single corn plant that was homozygous for a dominant yellow kernel trait in four fields with corn that was homozygous recessive for white kernels (1974). Plants were categorized by distance in meters from the center plot and the percent of kernels expressing the dominant yellow trait were reported. In only one of the four fields did the corn plants immediately adjacent to the homozygous dominant plant exhibit the highest effective rate of pollination. The other three plots had the highest effective pollination at distances of two, three, and five meters.

Das (1983) planted a male corn plant surrounded by eight rows of females planted away from the male. He found pollen concentrations of 274 grains per square inch at 50 meters, 103 at 100 meters, and 2.25 at 200 meters. The average seed set percentage for those distances was 51, 11, and 0.016.

A study using genetic markers was conducted in Mexico from 1997 to 1999 (Luna et al. 2001). There a 4000 square meter plot was encompassed by four row wide, four meter long rows planted to a white kernel hybrid. In the first experiment, the central plot was planted with dominant genetic markers for purple leaves that would be exhibited in the F1 generation; the second had a plot planted to corn with markers for

purple kernels. In both cases there was very little out-crossing which the authors attributed to the arid location and absence of strong winds during pollination.

Maize Pollen Dispersal and Buffers

The impact of buffers between crops was studied by Jones and Brooks (1950). They planted two varieties, one a yellow dent field corn, the other a white sweet corn, which can be separated by appearance. For the first part of their study, yellow dent was planted at 25 rods wide by fifty yards long, while sweet corn was planted in 100 square foot plots at distances of 0, 5, 15, 25, 40, 60, 80, and 100 rods. To ensure overlapping periods of fertility, greased slides were placed in the area to identify both varieties pollen presence during the experiment.

Some of their results are presented in Table 3. The level of contamination is similar to other studies, including the one published two years earlier by Jones, but in this case pollination is allowed to occur. It is once again of note that all distances experienced some degree of contamination.

Table 3. Average Percentage of Outcrossed Seed (Jones and Brooks 1948)

Distance									
In Rods	0	5	15	25	40	60	80	100	
Percent	28.62	14.21	5.79	2.33	1.19	0.48	0.23	0.2	

The second part of the study looked at the impact of buffer rows on seed contamination. Jones and Brooks did this by comparing the number of yellow dent and white sweet corn in each row within a number of 100 square foot plots. They found that the first rows in each sweet corn plot contained relatively higher levels of out-crossing than other rows in the plot in most cases. In a 1949 plot, at a distance of fifteen rods, over twenty-eight percent of the first row kernels were yellow dent, while the levels for the eleventh and twenty first rows, were 4.55 and 1.85 percent respectively. There were some significant deviations from this overall trend including one plot 80 rods from the source had 1.42 percent out-crossing in the eleventh more than twice that in the first row of the plot.

Jones and Brooks continued their research on pollen dispersal in corn publishing a study on the effect of tree buffers in 1952. The methods mirrored much of their 1950 study except that they added an additional trial where source and receptor plots were divided by a stand of elm trees with a nearby field without a tree barrier serving as a control. At five rods, the kernel count was about half the level in the first row for that including the tree buffer versus the control group without it. At fifteen rods that ratio fell to about one-eighth in the first row of the two groups. However, these ratios did rise slightly as one moved further into each plot.

Studies on Pollen Viability

It is important to distinguish between pollen and gene dispersal (Feil and Schmidt 2002). Pollen dispersal is the physical distribution of pollen. Gene dispersal; however, occurs only when the gene is carried on to successive generations and is highly affected by weather conditions (Treu and Emberlin 2000). While a pollen grain may become located some distance from its source, unless it retains some level of vigor and comes into contact with a corn's silk, the genes contained in the pollen have not been transferred spatially.

Concurrent with their study of pollen distribution, Jones and Newell studied the viability of pollen (1948). They began by placing pollen, still in the tassel, in a refrigerator at varying temperatures and humidity levels and then exposing the pollen to the silks of a corn plant. Over the course of eight days they found diminishing rates of pollination with treatments at lower temperatures and higher levels of humidity showing higher amounts of pollination especially past the first few days.

In a simultaneous experiment, Jones and Newell collected pollen which was then placed in pollen bags. The first group, placed under the sun at a temperature of 36 degrees Celsius, retained its vigor for only three hours. The second group, placed in the shade at a temperature of 28 degrees Celsius, remained viable for thirty hours.

Herrero and Johnson focused their research on the impact of temperature stress on pollen viability (1980). They began by removing tassels from fifteen different varieties of field-grown corn which were then stored in growing chambers at 27, 32, and 38 degrees Celsius. After 24 and 48 hours, the pollen was used to perform in vitro germination. The following table summarizes information from the second year of their study, denoted Experiment II.

Table 4. In vitro Pollen Germination by Percent for Experiment II
(Herrero and Johnson 1980)

Genotypes	27 C		32 C		38 C	
	24 Hours	48 Hours	24 Hours	48 Hours	24 Hours	48 Hours
B73	26	40	43	37	8	4
FR19	76	45	56	24	17	0
H100	34	29	18	33	7	0
Mo17	60	43	74	34	28	1
N6	48	39	53	16	8	0
Oh43	21	31	49	25	13	0
Mo17 x B73	57	53	59	59	9	4
Mean Across 15 genotypes	45	37	44	30	12	1

The data shows a strong pattern of increasing time and increasing temperature resulting in lower rates of germination measured as a percent. This is most evident in the extreme case where at 38 C and time equal to 48 hours only one percent of the samples resulted in germination. Also of note

is the increase in rates of germination for varieties B73 and Oh43 at 27 C. when time increased from 24 to 48 hours.

A more recent study looked at the relationship between atmospheric water status, ψ_{atm} , and its impact on pollination (Luna et al. 2001). Here pollen from a dominant white kernelled variety was collected and allowed to dry for an amount of time varying by treatment. This pollen was then exposed to silks of a variety with recessive yellow kernels. After harvest the kernels on each corn ear were counted.

The findings were as expected with a decrease in pollination over time and as ψ_{atm} decreased. During the first experiment, pollen viability fell by 58 percent after one hour. Data from the second trial which was conducted during a period of higher temperatures and lower humidity found a decline in pollination from 100 percent immediately after anthesis to zero percent, one hour later.

Modeling Spore and Pollen Movement

A number of fields have looked at the movement of particles by wind using mathematical models. McCartney and Fitt classify those used by modelers of spore dispersion into three groups: gradient transfer models, Gaussian plume models, and random walk models (1985). Though different in construction, the general models of each type can be shown to mathematically equivalent to the others.

Gradient Transfer Theory Models

Gradient transfer theory, also referred to as K-theory, models are based on the assumption that relatively small particles follow the movement of the gas in which they are located and thus obey Fick's Law. They are modeled using partial differential equations with particles moving at a rate proportional to the gradient. Through the establishment of initial and boundary conditions combined with flexible wind and diffusivity techniques, the models can address problems with relatively specific requirements, though numerical solutions can be hard to solve (McCartney and Fitt 1985).

This is the type of model used by Legg and Powell (1979) to predict the movement of *Erysiphe graminis* and *Lycopodium* spores in barley fields. Legg and Powell constructed a two-dimensional model with the aim of comparing their numerical results to those collected experimentally by Bainbridge and Stedman (1979). As research on spore dispersal focuses on the attachment of spores to vegetation, the model relies on area densities of plant parts such as leaves, heads and awns. The model also accommodates the fact that spores that travel through vegetation at wind speeds slower than those above the crop canopy. Their prediction of *Lycopodium* movement was more accurate compared to that of *E. graminis*. The authors hypothesized that this may be the result of *E. graminis* spores clumping together and consequently falling more rapidly than if they had remained separate.

Aylor (1982) further improved the Legg and Powell model of *Erysiphe graminis* dispersal. By including a simple conditional flow statement, Aylor set a threshold speed, u_t , below which no spores would leave their source. Setting the threshold speed to 0 m s^{-1} , the model's predictions were quite similar to those found by Legg and Powell (1979). For speeds of 5 m s^{-1} and 1 m s^{-1} there was even greater deposition of pollen at distances near the source than when it was set to 0 m s^{-1} .

McCartney studied the movement of oilseed rape pollen using gradient transfer theory (1991). Using a two dimensional steady state model he calculated the dispersal of pollen which was compared with data gathered in previous field trials. Only trace amounts of pollen were collected beyond fifty meters from the source. However, the model predicted that 60 percent of the pollen released was still airborne 100 meters from the source.

Two studies looked at the movement of spores used as herbicides using gradient transfer theory (de Jong et al. 1991; de Jong 2002). Both studies were concerned with the possibility of damage to non target plants. The first paper looked at the unwanted effects of *Chondrostereum purpureum* spores on plants other than the *Prunus serotina* it was meant to target. The authors refer to their model as a second generation model, with Legg and Powell (1979) being an example of a first generation model. It uses microweather data to calculate wind speed and dispersal, the vegetative

canopy is divided into layers, and makes use of a computationally effective method of solving the partial differential equation.

In the second paper, the movement of *Sclerotinia sclerotiorum* above a grass canopy, *Lycopodium clavatum* spores in a wheat crop, and *Venturia inaequalis* spores in an apple orchard and in a grassland were predicted using the model introduced by de Jong et al. (1991). Each scenario presented unique requirements which were addressed showing the flexibility of the model used. A variant of the model was also used to determine the escape of *Sclerotinia sclerotiorum* which the authors state could be used to find minimum isolation distances for growing non-target crops using a Gaussian plume model, a class of dispersion models which is presented below.

A great deal of research has focused on the various numerical techniques that can be used to solve gradient transfer theory models. In a series of three papers, fifteen numerical methods ability used to solve a two dimensional advection equation were compared with focus on accuracy, speed, and storage requirements (Chocker 1983; Chocker and Dunker 1985; Chocker 1991). More recently, Yang (1998) compared results from three numerical methods to an equation, which had an easily calculated analytical solution, used to predict insect and spore dispersal. With knowledge of the true solution, inferences about proper time and grid steps

as well as the shortcomings of each of the three numerical methods were made.

Gaussian Plume Models

Gaussian plume models assume that concentrations of particles downwind from a source follow a Gaussian distribution most often conditioned on distance from the source (Pasquill 1983). This class of models is commonly used to study the movement of air pollution (Dobbins 1979). However the models lack the flexibility to address complex situations, including movement over nonhomogeneous crops (McCartney and Fitt 1985).

Giddings (2000) uses the Gaussian plume model to predict the dispersal of *Lolium perenne* pollen and the risks associated with the release of transgenic pollen into the wind. Small, 2 meters in diameter, regions are located on a circle one kilometer from transgenic source. Using parameters estimated in a previous study, Giddings estimates the amount of transgenic pollen as a percentage of all pollen for each 2 meter region. He found that approximately four percent of the pollen located one kilometer from the source was transgenic when the source was also 2 meters in diameter. When the source diameter was increased to 320 meters between 29 and 99 percent of the pollen was from the transgenic plants.

A general Gaussian plume model was also used to model the movement of spores above a potato canopy and adapt the general model to

improve its ability to model spore dispersal (Spijkerboer 2002). Using a general model that used broad classifications of climatic phenomena the correlation between actual and predicted dispersal of *Lycopodium clavatum* was only $R=0.4$. The authors improved model focused on calculating the risk of spore dispersal on specific areas based on weather data collected around the times of actual spore escape. With this data the correlation between actual and predicted dispersal rose to $R=0.8$.

Stochastic Models

Stochastic models attempt to describe the Brownian motion of small particles using a stochastic process. Early models of particle dispersion made use of Markov chain and elementary random walk models to explain this movement (Csanady 1973; Hall 1975). More recently, models have used Langevin equations that meet subinertial and well-mixed conditions (Thomson 1987). These criteria require that the time step be short enough to capture the effects of Brownian motion and that particles follow the distribution of the larger mass of which they are part as they move away from the source.

Aylor and Flesch (2002) used such a two dimensional stochastic model to explain the dispersion of *Lycopodium* and *V. inaequalis* spores above and within different crops. Aylor used the approach of Legg and Powell (1979) regarding plant area densities, to model deposition of spores onto vegetation. For deposition onto the ground, a probability function,

based on the settling velocity, v_s , and the vertical velocity, w , determined by the stochastic process is used. As the sizes of the spores modeled were relatively large, the study adjusted the time step used as described by Sawford and Guest (1991).

The authors found the model to be in excellent agreement with previous collections of spores in the modeled situations. However, they expressed concern for the accuracy of dispersal within plant canopies. They also discussed the importance of gusts and lulls on vertical movement of the crop with the absence of gusts resulting in quick deposition of the spores near their source.

A paper by Klein et al. (2003), used a simple random walk model to predict the movement of corn pollen which was then compared to data collected during previous field studies. While the model did not meet subinertial time requirements or the well-mixed conditions, it did find a good fit between model predictions and data collected during field experiments. The process of pollination is modeled by having fertilization occur when a pollen grain first crosses a height, h . The authors also raised concern for impact of relative heights of male and female plants, as well as the minor role vegetation had on the dispersal of pollen.

Preventing Gene Movement

Spatial segregation of crop varieties has traditionally been used to minimize the amount of unwanted genetic movement. Other procedures,

such as detasseling and bagging of ear shoots and tassels have been used in combination with physical isolation in corn seed development for years. However, recent advances in biotechnology may prove to be the next preferred method of controlling the presence and expression of restricted genes.

To the present time most attempts at limiting the movement of genes at the molecular level have focused on maternal inheritance, male sterility, and seed sterility. Other techniques including asexual reproduction, self-fertilization in the form of cleistogamy, genetic incompatibility, chemical manipulation of transgene flow, and transgenic mitigation have shown promise either theoretically or in limited trials (Daniell 2002). A brief summary of these techniques based on Daniell (2002) follows.

Certain genetic material is contained only within cytoplasmic material, chloroplast, and is not transferred by male nuclear genes during fertilization resulting in maternal inheritance of such genes. At least 30 transgenes including those showing promise with use as biopharmaceuticals have been integrated in chloroplast genomes (Daniell et al. 2001; Daniell and Dhingra 2002). One particular benefit of this technique is that transgene would be that tissue specific, at the location of the functioning plastid (Daniell 2002).

Altering the reproductive system of a transgenic crop is also a possibility, with most research focusing on damaging the function of the

anther. Daniell (2002) points out that there exists a risk that a male sterile plant could be fertilized by another plant resulting in the next generation containing a respective transgene. Another alternative is the use of the Technology Protection System (TPS) more commonly referred to as 'terminator' technology. In the case of transgenes, the application of tetracycline or other antibiotic on harvested seed results in the destruction of specific plant tissue (Russell 1992).

Methods taking advantage of cleistogamy, self-fertilization before the flower opens, and apomixes, the use of vegetative seed, have little application in corn. The remaining approaches, genetic incompatibility, chemical manipulation of transgene flow, and transgenic mitigation, show promise to minimize the risk of transgene introduction into weed populations (Daniell 2002). Unlike other field crops, corn lacks close wild relatives and these concerns are of little relevance.

CHAPTER 3. THEORETICAL CONSIDERATIONS

In this chapter, the theoretical foundation of a maize gene dispersal model is presented. First a simple physical model and then a more appropriate advection-diffusion representation that allows for both the movement and expansion of a pollen cloud are addressed. Unique requirements are met by referring to the literature of mathematical modeling of pollen and spores. Also presented is a maximum likelihood method for determining the parameters that define the Weibull distribution which is used to describe the behavior of wind speed.

A Simple Physical Model

A simple physical model of pollen dispersal by wind (1) is presented by Johnson (1981). Here the distance (D) traveled by a particle is dependent on the initial height (H) of the pollen cloud, the wind speed (u) in a horizontal (x) dimension, and the terminal velocity of the particle (V_s), an object's rate of fall through the air. One can imagine this as a vector with horizontal (x -dimension) and vertical (z -dimension) components with D being the distance from the origin of the source where the vector crosses the $z=0$ line.

$$D = \frac{u \times H}{V_s} \quad (1)$$

Since the height of the crop and the terminal velocity are readily available for most pollen, the equation becomes dependent on the wind

speed (2). Thus, for any given value of u , equation (1) is simple arithmetic. The estimation of a wind speed distribution for a location allows one to calculate the distribution of the distance pollen would likely travel.

$$D = d(u; H = H', V_s')$$
 (2)

Constructing a Weibull Distribution

The Weibull distribution is used to represent the wind speed distribution (Seguro and Lambert 2000). The following equation (3) shows the construction of the model with k , the Weibull shape parameter; c , the Weibull scale parameter; and u , wind speed. It is important to note that only nonzero wind speeds should be used in the calculation.

$$P(u < u_i < u + du) = P(u > 0) \left(\frac{k}{c} \right) \left(\frac{u_i}{c} \right)^{k-1} \exp \left[- \left(\frac{u_i}{c} \right)^k \right] du$$
 (3)

The estimation of the shape and scale parameters is estimated using the maximum likelihood method described by Stevens and Smuther (1976). Equations (4) and (5) present the equations used to estimate the k and c parameters respectively. As the shape parameter, k , is located on both sides of equation (4), the solution can be found iteratively.

$$k = \left(\frac{\sum_{i=1}^n u_i^k \ln(u_i)}{\sum_{i=1}^n u_i^k} - \frac{\sum_{i=1}^n \ln(u_i)}{n} \right)^{-1}$$
 (4)

$$c = \left(\frac{1}{n} \sum_{i=1}^n u_i^k \right)^{1/k}$$
 (5)

The data used for the calculation for the Weibull distribution are those from the Boone Municipal Airport in Central Iowa. Wind speeds in knots collected at twenty minute intervals in mid-morning from mid to late July from 1995 to 2002 are used. This is the approximate time of pollination during recent years in Iowa (Miller 1985; NASS 2002).

There are numerous shortcomings of the simple model. Foremost is the exclusion of seemingly obvious and necessary variables in the movement of particles in wind: the role of turbulence, the differing wind speed just above and through a field, the contact of pollen and silk downwind. In addition, the simple model recognizes pollen as a single unit rather than a concentration of particles across the temporal and spatial dimensions. Using the simple model, intuition would be led one to believe that no pollination would occur more than a few meters from the source as it would be too low vertically to come into contact with the silks of neighboring corn.

An Advection-Diffusion Model

The deficiencies of the simple model are addressed by using an advection-diffusion representation. This model is used by plant pathologists to model spore dispersal, atmospheric scientists interested in the movement of pollution, and economists studying the pricing of options, among others. We use the gradient transfer theory model presented by McCartney and Fitt as a guide (1985). The classical diffusion model, described in Cartesian coordinates and time, is presented in (6).

$$\begin{aligned} \frac{dC}{dt} = S(x, y, z) + \frac{\partial}{\partial x} \left(K_x \frac{\partial C}{\partial x} \right) + \frac{\partial}{\partial y} \left(K_y \frac{\partial C}{\partial y} \right) + \frac{\partial}{\partial z} \left(K_z \frac{\partial C}{\partial z} \right) \\ + v_s \frac{\partial C}{\partial z} - u \frac{\partial C}{\partial x} - v \frac{\partial C}{\partial y} - w \frac{\partial C}{\partial z} \end{aligned} \quad (6)$$

Here the change in concentration for a given location, C , given a change in time, t , is dependent on the source concentration, S , and is affected by movement caused by wind speed, u , v , w , and diffusion K_x , K_y , K_z , in the x , y , and z directions. V_s , as in the simple model, is the terminal velocity of the pollen in the vertical direction. This equation is difficult to solve, but can be simplified based on a few reasonable assumptions.

Cross-wind, v , and diffusivity in the y -plane, K_y , are ignored as the focus is on movement in the x -direction, to which they are assumed unrelated. Vertical wind speed, w , is also assumed to be zero, the equivalent of a non-intense thermal convection situation. Diffusion in the mean wind direction, K_x , is also set equal to zero, as it is understood to be overpowered by the wind moving in that direction. As wind speeds, u , are positive, the refined model is of a semi-infinite line source type (7).

$$\frac{dC}{dt} = S(x, z) + \frac{\partial}{\partial z} \left(K_z \frac{\partial C}{\partial z} \right) + v_s \frac{\partial C}{\partial z} - u \frac{\partial C}{\partial x} \quad (7)$$

Boundary Conditions

Boundary conditions for the advection-diffusion model are defined as follows. The concentration approaches zero as one moves vertically and horizontally away from the source, $C \rightarrow 0$ as $X \rightarrow \infty$, and $C \rightarrow 0$ as $Z \rightarrow \infty$.

The ground is assumed to be a less than perfect sink as described in equation (8) with v_d being the velocity of deposition (Chamberlain 1975).

$$K_z \frac{\partial C}{\partial z} + v_s C = v_d C \text{ at } z = 0 \quad (8)$$

Vertical Diffusion

The wind speed profile represented by the Weibull distribution provides the basis for the derivation of other necessary atmospheric values. The first of these is the frictionless wind speed, u^* , that will be used to calculate vertical diffusion. Equation (9) provides the basis for calculating the frictionless wind speed given the distance from ground level, z , the zero displacement level, d , and the roughness length, z_0 (Monteith 1973). The zero displacement level for maize has been estimated at 1.7 and the roughness length to be .3 (Hosker and Lingberg 1982). The value, k , von Karman's constant is approximately .4.

$$u(z) = (u^*/k) \ln[(z-d)/z_0] \quad (9)$$

Using the value of frictionless wind speed determined above we can determine the vertical diffusion under normal atmospheric conditions (10) (Monteith 1973). Von Karman's constant, distance from the ground, and the zero displacement level are again used.

$$K_z = ku^*(z-d) \quad (10)$$

Stochastic Modeling of Dispersion

The model of advection-diffusion can be solved numerically by conducting a Monte Carlo simulation using a Lagrangian stochastic (LS) model. This is done by simulating the path of individual particles repeatedly rather than by determining the concentration of a mass of particles. This technique is more readily adaptable to meet the requirements of Scenarios 3 and 4. It also drastically reduces the computation time relative to finding the solution to the partial differential equation (7) directly, such as by the finite difference method.

A generalized first-order LS model takes the form of (8). Here the change in velocity at time i , du_i ; is determined by the drift coefficient, a_i ; multiplied by the time step, dt ; and b_{ij} , a diffusion coefficient, times $d\xi_j$, increments from a Wiener process. The model and its variants are often used in finance to describe the asset price behavior over time.

The values of $d\xi_j$ follow a Gaussian distribution with mean zero and variance dt , with values of $d\xi_j$ independent of $d\xi_k$, for $j \neq k$. So for each time period, dt , the movement of a particle, dx , is a function of the stochastic process (8a). The stochastic process in (8a) is akin to diffusion in (7), with both measuring the impact of Brownian motion on the concentration of particles.

$$du_i = a_i dt + b_{ij} d\xi_j \quad (8a)$$

$$dx = udt \quad (8b)$$

The following system of equations (13) defines our LS model. The model differs from the generalized model in that restrictions are placed on a_i and b_{ij} . The coefficient b_{ij} must uphold Kolmogorov's similarity theory assumption of choosing time increments within the sub-inertial time range. The term a_i is uniquely determined in order to meet the well-mixed condition outlined by Thomson (1987).

The well-mixed condition is the most stringent criterion for such models and states that a particle that is well-mixed within a mass of particles will remain well mixed. Meeting this condition is the equivalent of the LS model being compatible with the Eulerian equations, the form of (7). This accounts for the inclusion of the second group of terms within the brackets of (9a) as well as the scalar before the mean dimensional movement, W .

$$dW = \left[-\frac{b_w^2}{2\sigma_w^2}W + \frac{1}{2} \frac{\partial \sigma_w^2}{\partial z} \left(\frac{W^2}{\sigma_w^2} + 1 \right) \right] dt + b_w d\xi_w \quad (9a)$$

$$b_w = \left(\frac{2\sigma_w^2}{\tau} \right)^{1/2} \quad (9b)$$

$$dX = Udt \quad (9c)$$

$$dZ = (W - v_s)dt \quad (9d)$$

Theoretical Relationships among Parameters

The wind speed profile represented by the Weibull distribution provides the basis for the derivation of other necessary atmospheric values. The first of these is the frictionless wind speed, u^* , needed to find σ_u , σ_w , and T_l . Rearranging equation (10) one can calculate the frictionless wind speed given the distance from ground level, z , the zero displacement level, d , and the roughness length, z_0 (Monteith 1973). The zero displacement level for maize has been estimated at 1.7 and the roughness length to be .3 (Hosker and Lingberg 1982). The value, k , von Karman's constant is approximately .4.

$$u(z) = (u^*/k) \ln[(z-d)/z_0] \quad (10)$$

Using the value of frictionless wind velocity, one is able to determine the values of σ_w , and T_l , the deviations of horizontal and vertical wind velocity and the Lagrangian time scale, using (11).

$$\sigma_w = 1.3u^* \quad (11b)$$

$$T_l = \frac{ku^*z}{\sigma_w^2} \quad (11c)$$

The model is modified to address the relatively large size of the corn pollen. Walklate (1987) recommended the adjustment of the variance of the fluid downward to reflect the drag associated with larger particle sizes. Wilson (2000); however, found that the difference between those with altered

and unaltered variance is negligible and this adjustment is ignored by the model. Nor do we simultaneously solve the equations for the particle and gas which Wilson (2000) also found unnecessary.

We do include a scaled time step using the equation derived by Sawford and Guest (1991) and used by Aylor (2002) in his study of spore dispersal. Equations (12a) and (12b) present the relationships between the the Lagrangian time scale, T_l , and τ , the velocity time scale. In (12b), β is a constant relating the Eulerian timescale to the Lagrangian timescale and is placed equal to 1.5 in this study as by Sawford and Guest (1991) and Aylor (2002).

$$\tau = fT_l \quad (12a)$$

$$f = \frac{1}{\sqrt{1 + (\beta v_s / \sigma_w)^2}} \quad (12b)$$

Pollen Deposition

The final segment of the model concerning the movement of pollen concerns its deposition from the air onto either open ground or other crops. The process of deposition will be described by two mechanisms. The first represents the depositing of pollen onto open ground and non-fertile corn, the remainder onto fertile corn.

The deposition of pollen onto non-fertile corn areas is done by deposition occurring when the pollen crosses a certain height, z_s . This height is assumed to be the same as the zero plane displacement level, d .

From the literature these values are taken to be 0 for open ground, .47 for soybeans, and 1.7 for corn (McIntosh and Thom 1972, Perrier et al. 1972, Hosker and Lindberg 1982).

For the deposit onto fertile corn plants, the biological processes involved are discounted. The probability of pollination of a plant by source pollen is defined as a ratio (13) (Emberlin 1999). However, not all pollen will still viable by the time it comes in contact with the fertile corn. This is accommodated by classifying all pollen that has been airborne for more than two hours as an unviable transmitter of transgenes. Given Iowa's warm humid climate during the time of pollination this is a strict condition placed on the model.

The temporal condition is described by (13a) with Q_T being the amount of viable transgenic pollen from the source. $Q_R(0, H, 0)$ is the quantity of pollen released at time zero at the source where x is equal to zero, z is the initial height, H . Pollen is considered viable if it reaches the roughness length, z_0 , with t less than or equal to 7200 seconds. Given that condition, equation (13b) describes the pollination process with Q_A being the total amount of viable pollen in the vicinity of the plant.

$$Q_T = \{Q_R(0, H, 0) : Q_R(x, z_0, t), t \leq 7200\} \quad (13a)$$

$$P = Q_T / Q_A \quad (13b)$$

CHAPTER 4. EMPIRICAL RESULTS

Using the models described in Chapter 3, the dispersal of maize pollen is predicted under four scenarios. The first of these utilizes the simple physical model while the remaining scenarios use the advection-diffusion representation. The baseline model of advection-diffusion is for the most part defined within Chapter 3. Scenarios three and four make subtle adjustments to the baseline model based on the minimum production practices for pharmaceutical corn outlined by APHIS (2002).

Scenario 1: The Simple Physical Model

The first scenario uses the model of Johnson (1981) to describe the dispersal of corn pollen. Each of the ten thousand pollen clouds to be modeled begins with the independent random drawing of a wind speed from the Weibull distribution calculated using data from Boone, Iowa. Due to the construction of Johnson's formula the distribution of gene dispersal found in this scenario is essentially a scaled version of the Weibull distribution.

Figure 1 illustrates the scenario. A plume of corn pollen is released 1.7 meters above the ground at the source, where x is equal to 0. The pollen is then carried by the wind until its deposition on the ground. With the absence of diffusion of the pollen cloud, the model predicts a linear trajectory for each pollen plume and consequently a finite point of deposition for each wind speed drawn from the Weibull distribution.

The viability of pollen is not a limiting condition in this scenario. Another byproduct of the construction of the simple model, all pollen falls after 12.5 seconds of being airborne regardless of climatic conditions. This, as described by equation (14), arises since all pollen falls at a constant rate, v_s , equal to .2 from an initial height of 1.7 meters.

$$t = \frac{H}{v_s} = \frac{1.7}{.2} = 12.5 \quad (14)$$

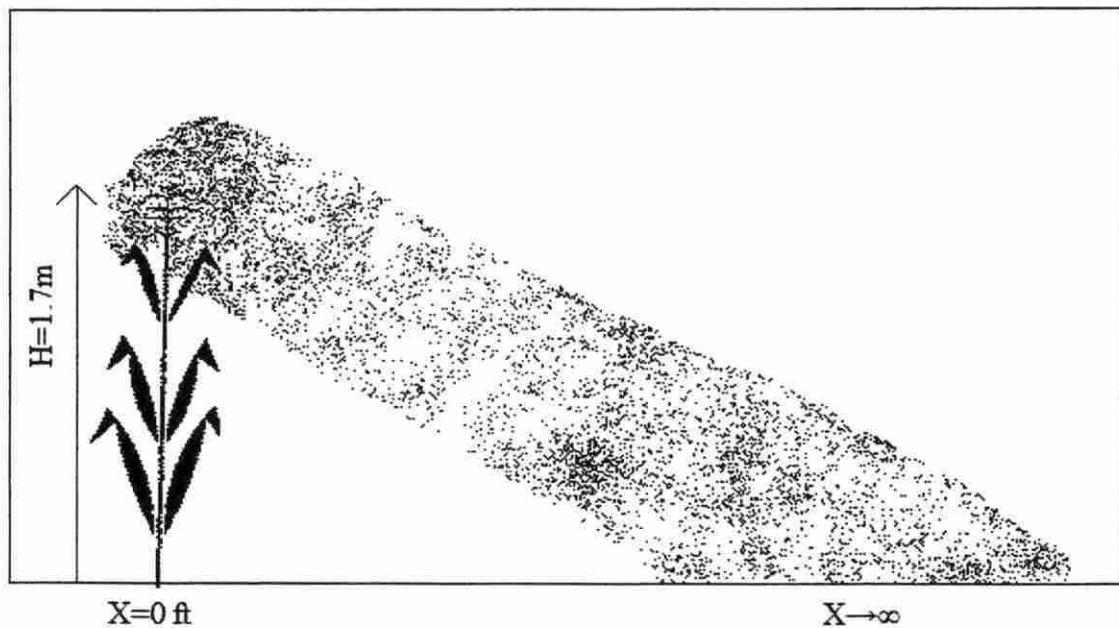


Figure 1. Illustration of Gene Dispersal Using the Simple Physical Model

The following figure displays the results of the simulation using the simple physical model. With no diffusion, all pollen was predicted to fall within 80 meters of the source, approximately half deposited within 20 meters. The absence of diffusion results in underestimation of genes near

the source with the model placing just under 5 percent of the pollen within 5 meters of the source.

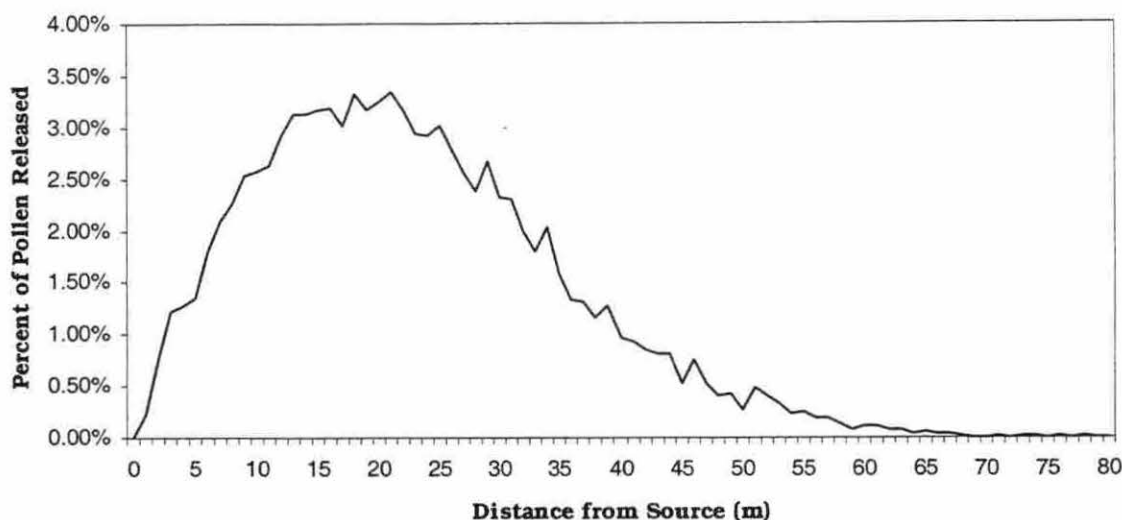


Figure 2. Maize Gene Dispersal Using the Simple Physical Model

Scenario 2: The Baseline Advection-Diffusion Model

The baseline scenario of the advection-diffusion model is based on the theoretical representation outlined in Chapter Three. As in the first scenario, the plume of pollen is initially located 1.7 meters above the ground, where x is equal to zero. With no plants located downwind from the source, the zero displacement plane, d , has a value of zero at all locations. For the same reason the roughness length, z_0 , also remains a constant .01. Figure 3 illustrates the problem similar to the simple physical model except for diffusion shown by the expanding plume.

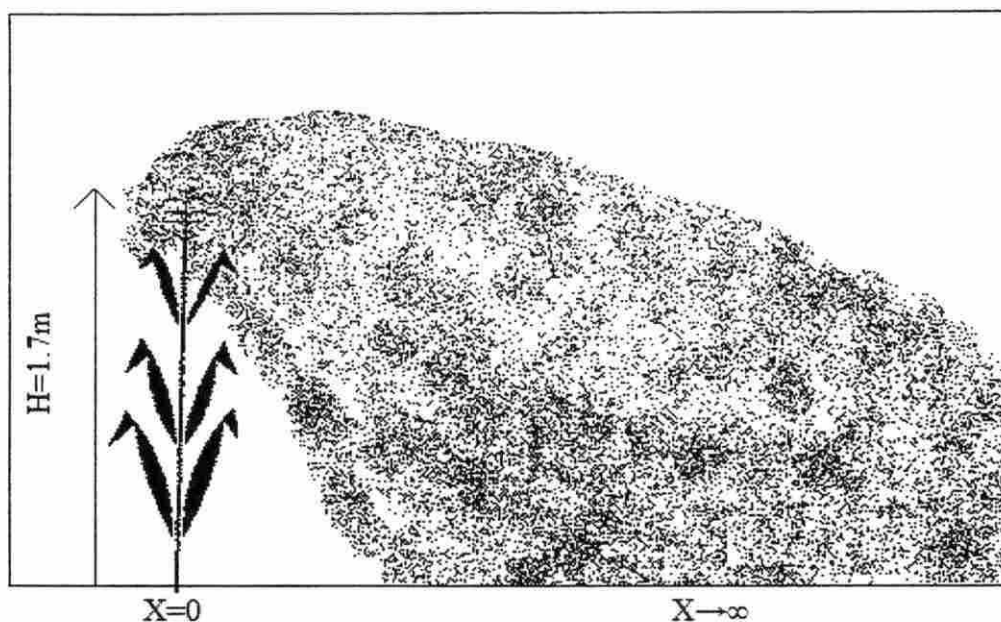


Figure 3. Illustration of Maize Gene Dispersal Using the Baseline Advection-Diffusion Model

Calibration of the Baseline Advection-Diffusion Problem

The baseline advection-diffusion problem is calibrated using data from Bateman (1947). Of the three papers that collected pollen using microscopic slides, this is the only study that included both a description of wind behavior and precise results. In this study, maize pollen was collected over the course of five days at sites spaced twenty feet apart up to 120 feet away from the source plot in three cardinal directions.

The calibration is done by comparing the ratio of pollen located downwind from plot relative to the amount of pollen found at the first site outside the maize plot, twenty feet from the source. These ratios are located in parenthesis in the following tables, the first showing the experimental

data from Bateman (1947). It should be noted that the first three experiments were conducted under relatively mild winds, the later two under stronger ones. This is evident in the much greater amount of normalized pollen that is present at distances of 80 and 100 feet for the last two trials.

Table 5. Number of Pollen Grains Collected (Bateman 1947)

Experiment	Distance from Plot in Feet					
	20	40	60	80	100	120
I	125 (100)	43 (34)	17 (13)	10 (8)	2 (2)	3 (2)
II	271 (100)	95 (35)	37 (14)	16 (6)	12 (4)	3 (1)
III	91 (100)	31 (34)	8 (9)	7 (8)	2 (2)	1 (1)
IV	174 (100)	76 (43)	29 (17)	39 (22)	17 (10)	9 (5)
V	21 (100)	7 (33)	6 (29)	4 (19)	4 (19)	1 (5)

Simulations using the advection-diffusion model were conducted by creating sinks elevated three feet from the ground and spaced twenty feet apart away from the plot. For each of four wind speeds, 100,000 grains of pollen were released at a height of 1.7 meters above the ground. The amount of pollen captured by each sink is presented in Table 6.

As the amount of pollen released from the experimental plants is not known, ratios of simulated pollen at each distance, relative to the amount at the first site, are included in Table 6. This provides the ability to compare the experimental and simulated findings. Bateman did not provide numerical descriptions of weather behavior, but rather verbal accounts for the day of pollen collection. As a result, simulations run for four wind speeds are assumed to include those that occurred during the field experiments.

Table 6. Simulated Number of Pollen Grains Collected

Wind Speed (m/s)	Distance from Plot in Feet					
	20	40	60	80	100	120
0.5	69 (100)	6 (8)	3 (4)	2 (2)	1 (1)	1 (1)
1	610 (100)	85 (14)	31 (5)	19 (3)	16 (3)	6 (2)
2	1942 (100)	228 (12)	90 (5)	45 (2)	25 (1)	15 (1)
2.5	2545 (100)	349 (14)	94 (4)	46 (2)	30 (1)	18 (1)

For each simulation, the model underestimated the relative amount of pollen compared to that collected by Bateman. For the most part pollen presence was under predicted by a factor of 2. This may have arisen due to four conditions that hinder Bateman's study. First, pollen was collecting in

only three directions, none of which aligned with the mean wind direction for any of the experiments. Second, wind speed and direction were only vaguely described. Also, for each trial the slides were exposed for most of the day and were likely subject to varying climatic. Finally, the experiment was bounded on two sides by high walls and on the other two sides by trees. This likely resulted in turbulence as the wind's behavior was altered as it entered the trial area.

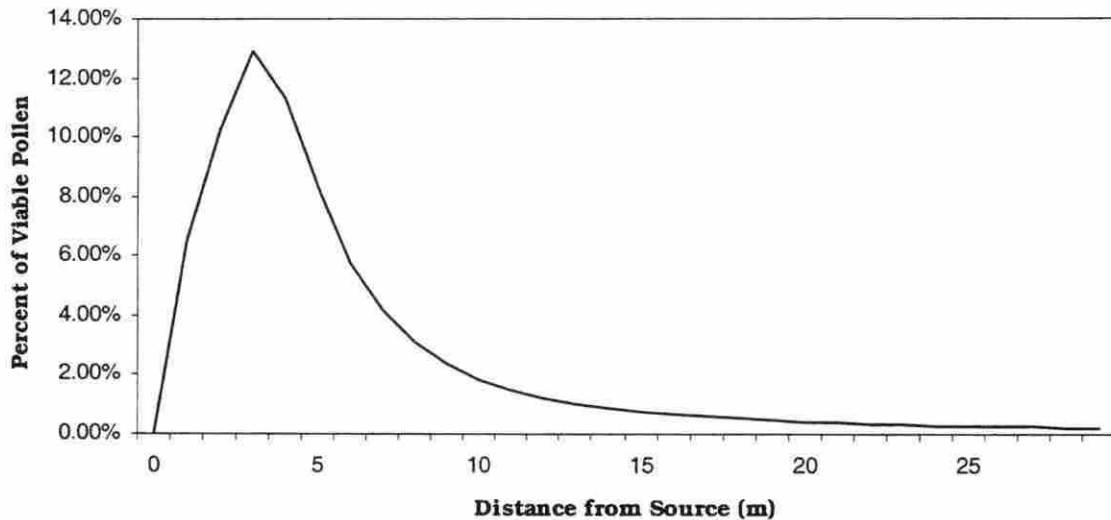
Despite the fact that the model underestimated the amount of pollen concentration, it does not discount its usefulness. Instead, using these findings it can be assumed that model will under predict pollen dispersal, possibly by amounts even greater than that found in this section. The analysis of pollen and gene dispersal away from a transgenic source can make use of the model, keeping in mind its findings are likely on the low side.

Gene Dispersal Using the Baseline Advection-Diffusion Model

A Monte Carlo simulation is run using the baseline advection-diffusion model. It begins with the draw of 1,000 wind speeds from the Weibull distribution describing the behavior of wind in Central Iowa. For each of these wind speeds, the dispersal of 1,000 grains of pollen is simulated and the location of their point of deposition recorded.

Figure 4. presents the distribution of viable pollen predicted by the baseline model within 30 meters of the source. In this case, approximately

five percent of the pollen lost its vigor before being deposition on the ground. Of the pollen that retained its viability, 77 percent of the pollen is deposited within this range, the maximum amount in the vicinity of five meters from the source. While the data on this table is of value for comparison to other experimental studies, this study's emphasis is on long range dispersal.



**Figure 4. Maize Gene Dispersal Near the Source
Using the Baseline Advection-Diffusion Model**

Dispersal at more distant location is presented in Figure 5 which displays the deposition of viable pollen at distances from 1000 to 5000 meters. Unlike in the chart presenting nearby deposition of pollen, the stochastic nature of the model is evident in this range due to the small number of pollen grains found for each unit of space. For the baseline

model, the true dispersal for a set of parameters should behave as a continuously decreasing exponential function.

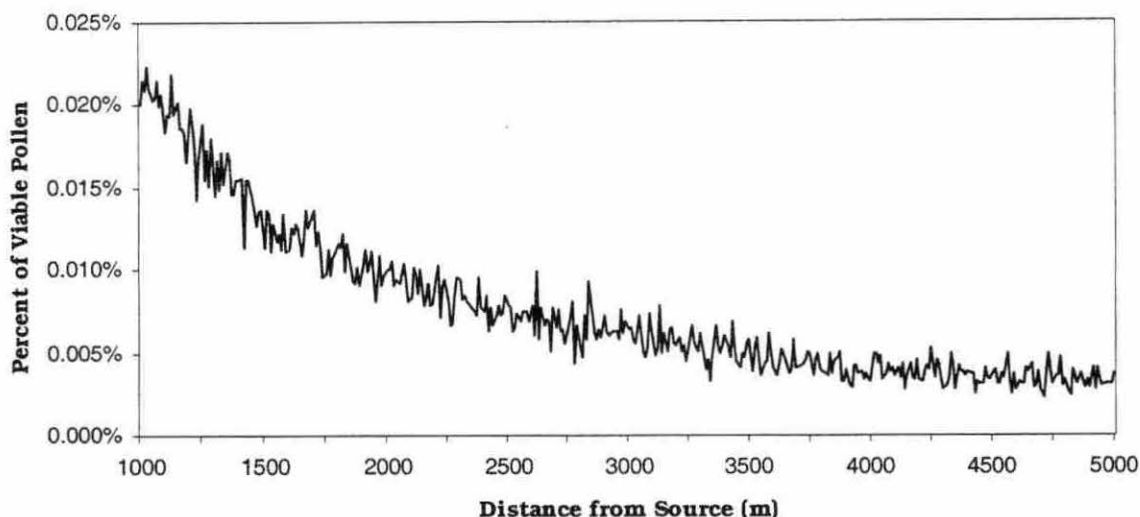


Figure 5. Long Distance Maize Gene Dispersal Using the Baseline Advection-Diffusion Model

Scenarios 3 and 4: The Advection-Diffusion Model Pharmaceutical Maize Gene Dispersal

These scenarios adjust the baseline model to meet the APHIS requirements for the production of pharmaceutical corn. Scenario 3 does not involve the presence of male fertile non-transgenic corn, while Scenario 4 does. The planting of such rows result in a different set of restrictions regarding the types and location of plant production within a specified distance of the pharmaceutical corn plots..

For the third scenario, these regulations require that the twenty-five feet immediately adjacent to each plot of pharmaceutical corn be left fallow

and no corn planted within one-half mile. Corn may be planted between one-half mile and one mile if it is seeded 21 days before or after the planting of the transgenic corn (APHIS 2002). Figure 6 illustrates the scenario including the assumption that soybeans are planted between the fallow ground and the infertile corn, from distances 25 feet to one-half mile.

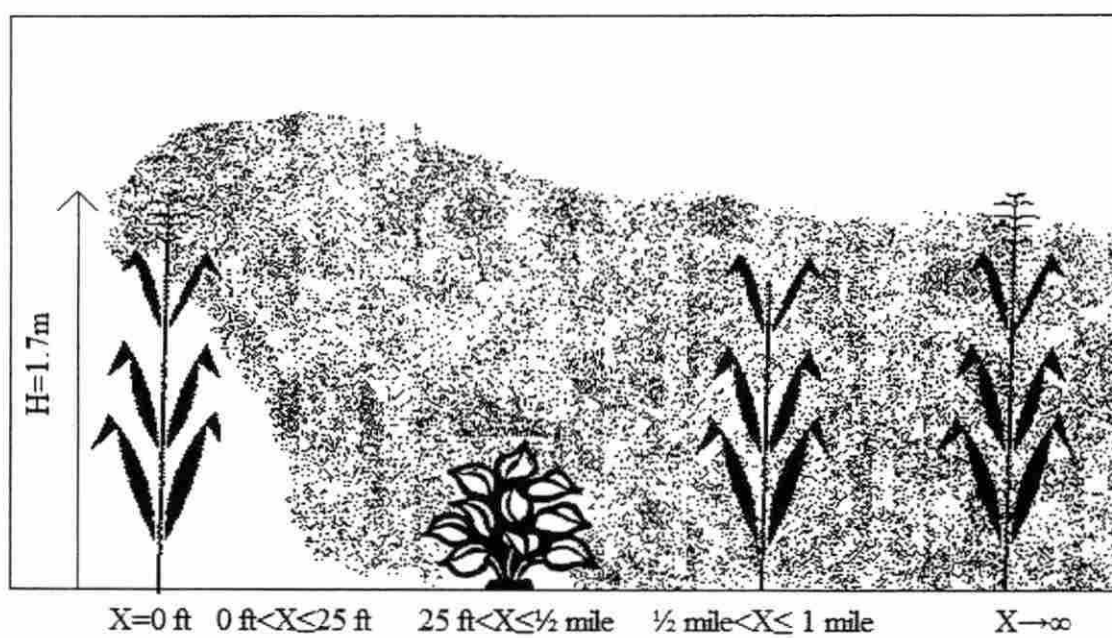


Figure 6. Illustration of Pharmaceutical Maize Gene Dispersal with No Buffer

In the case where buffer rows are present, the distance that remains fallow is the same, but corn can now be planted as close as one-fourth mile from the pharmaceutical crop if it is sown fourteen days before or after the pharmaceutical crop. At distances beyond one-half mile there are no further restrictions. As in Scenario 3, it is assumed that the area between the fallow portion of the field and the start of corn production, which is

grown from one-fourth mile distance to infinity, is planted to soybeans. The illustration below presents this visually.

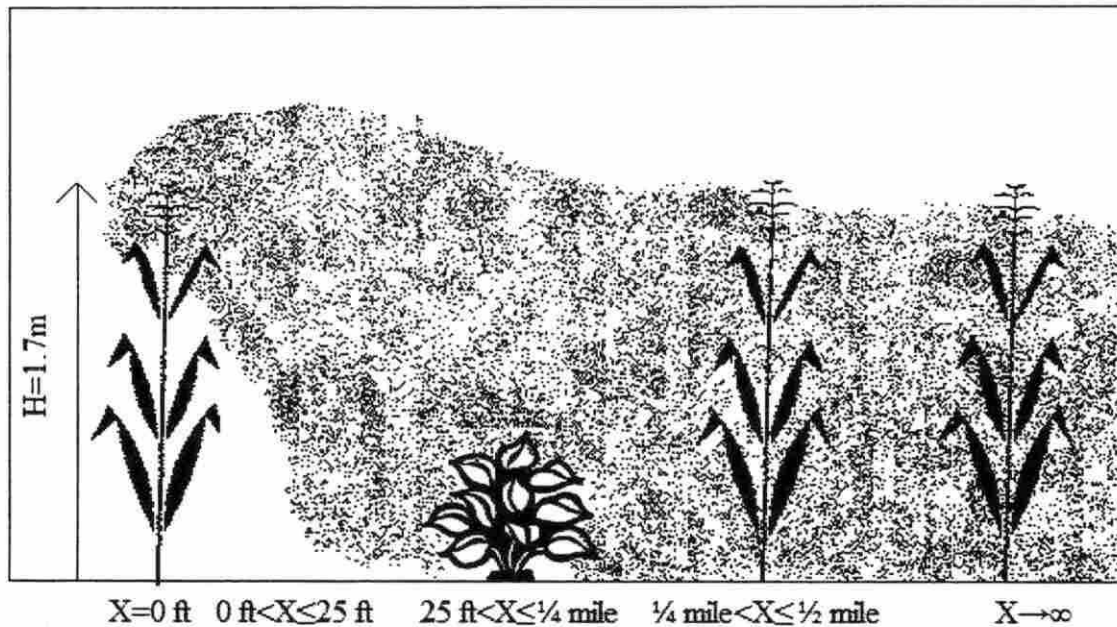


Figure 7. Illustration of Pharmaceutical Maize Gene Dispersal with Buffer Rows

For these scenarios the advection-diffusion model is modified to account for the change in wind speed above the varying vegetation. Specifically, the roughness length, z_0 , and zero displacement plane, d , above the soybean and corn plants values are adjusted. In the case of soybeans, z_0 and d take values of .13 and .47 meters. For corn, the roughness length is .3 meters and the zero displacement plane is 1.7 meters above the ground.

Genetic Seepage

It is assumed that the pharmaceutical corn seed used will make use of a biological mechanism, such as those described by Daniell (2002), to

reduce the likelihood of gene dispersal. The mechanism used is assumed to be imperfect, failing one percent of the time. This failure results in the release of pollen containing restricted transgenes that can be transferred to other corn plants. This can be considered in one of two ways: either the mechanism may attempt to prohibit the presence of restricted genes in pollen or it may try to eliminate the production of pollen altogether. While the manual detasseling of corn would theoretically eliminate the dispersal of pollen, this procedure is not common practice as producers of pharmaceutical corn avoid physical entry into such fields as it introduces the possibility of further contamination of other corn production.

Transgenic Planting and Neighborhood Crop Fertility

The federal regulations that require a temporal separation of planting times of corn plants likely have farther reaching effects. This results from the assumption that producers of pharmaceutical corn do not dictate the time of corn planting outside their field, but rather delay their planting relative to surrounding corn production by the time denoted by federal regulations. While federal timing conditions must be met within designated distances, it is possible that corn production beyond this distance, due to being planted relatively late or developing more slowly, shares a period of fertility with the pharmaceutical producing crop.

To accommodate for the contemporaneous fertility of the pharmaceutical and neighboring corn crops, a distribution of periods of

fertility is constructed. This distribution is derived using USDA Crop Progress Reports' data on silk emergence, a proxy for corn fertility, in the state of Iowa. The time of silk emergence is assumed to follow a normal distribution.

Figure 7 presents two overlapping distributions of silk emergence. The first describes the behavior of non-pharmaceutical crops, the second, pharmaceutical crops which in this case are fertile on average three weeks after that of neighboring corn. The shaded area is that of the overlap of the two distributions that occurs three weeks after the first crops peak period of fertility. About two percent of non-pharmaceutical pollen is fertile three weeks after the statewide peak, five percent two weeks afterwards.

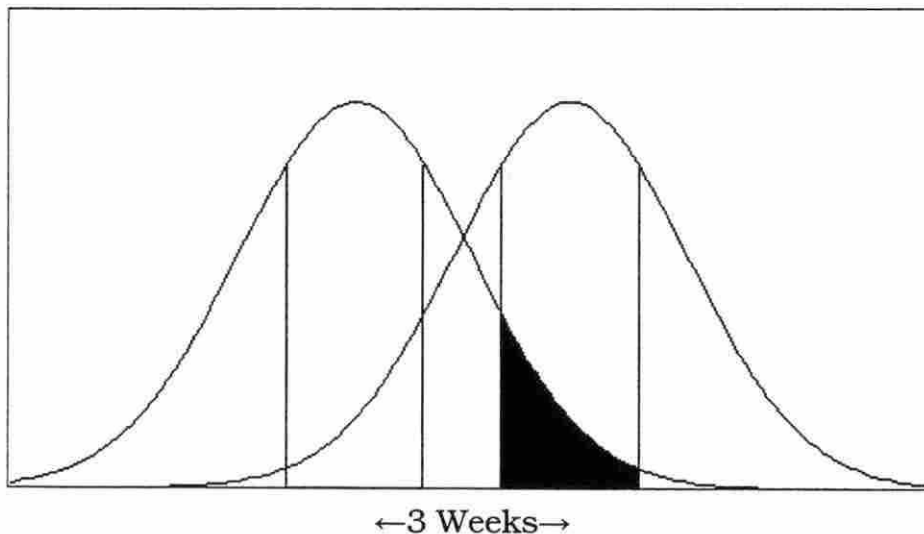


Figure 8. Temporal Distribution of Corn Fertility

Gene Dispersal Under Scenarios 3 and 4

The impact of plant presence on wind speed and height of deposition has already been described. The inclusion of soybean and corn plants also

decrease the percent of pollen that loses its viability before deposition, as pollen has a shorter distance to fall before settling. In the baseline case, 5.3 percent of the pollen loses its vigor compared to 4.5 and 4.4 percent in Scenario 3 and Scenario 4.

The next chart presents the percent of gene deposited at a given distance from the source under the baseline and Scenario 3 conditions. For the first part of the chart they mimic each other closely, but a spike approximately 7 meters from the source occurs as a result of the soybean plants found just beyond the fallow area in Scenario 3. As a result of those plants, over eighty four percent of viable pollen is deposited in this range compared to seventy-seven percent in the baseline. The deposition of pollen in Scenario 4 closely follows that of Scenario 3 over this range of distances.

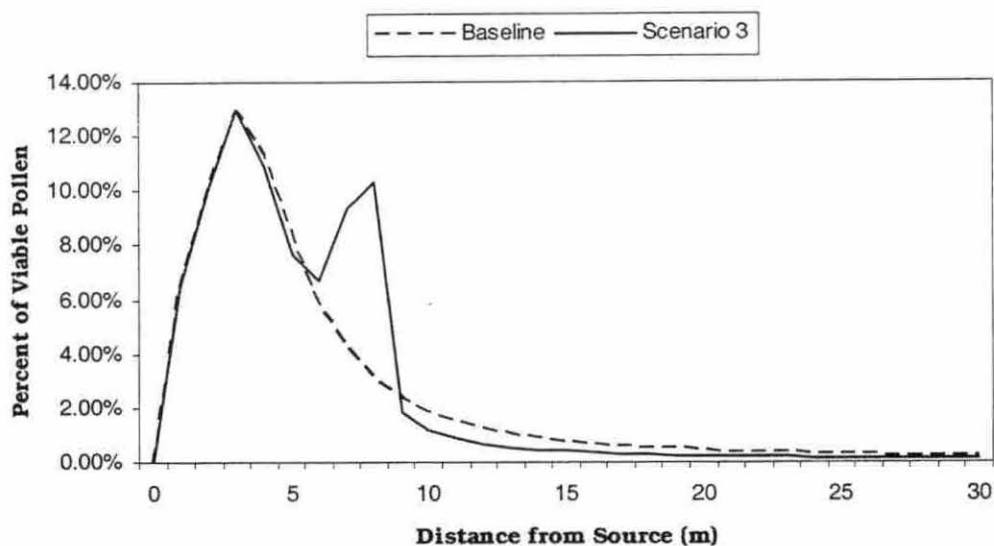


Figure 9. Maize Gene Dispersal Near the Source

Further downwind, spikes again occur as there is a transition from soybean to corn production. This occurs at one-half mile in the case of Scenario 3 and one-quarter mile under Scenario 4. This can be seen in Figures 9 and 10 with small, but noticeable increases in the percentage of pollen deposited in the vicinity of production changes. Only at locations of vegetative transition does the deposition of pollen in Scenarios 3 and 4 exceed that of the Baseline. These high points are offset by lower level of dispersal through the remaining locations.

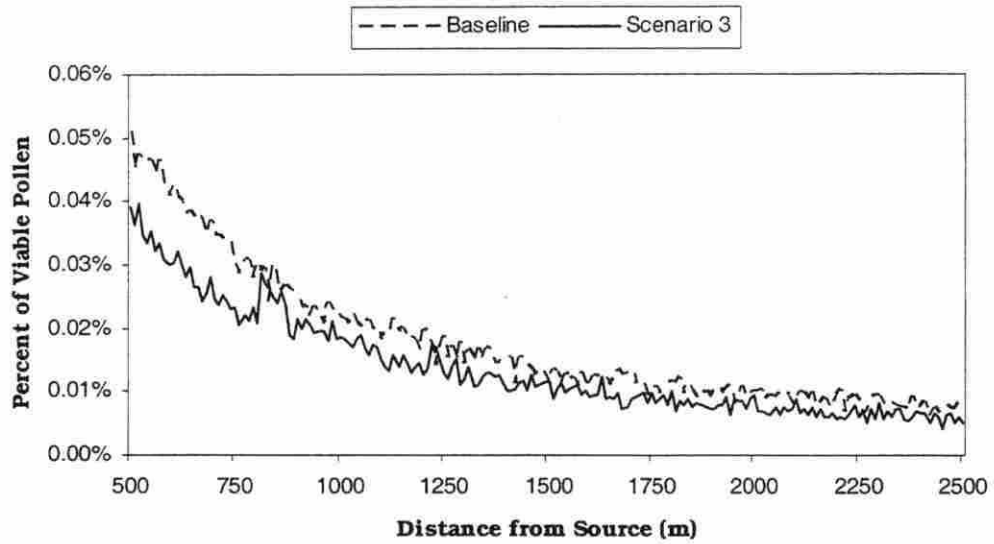


Figure 10. Long Distance Maize Gene Dispersal I

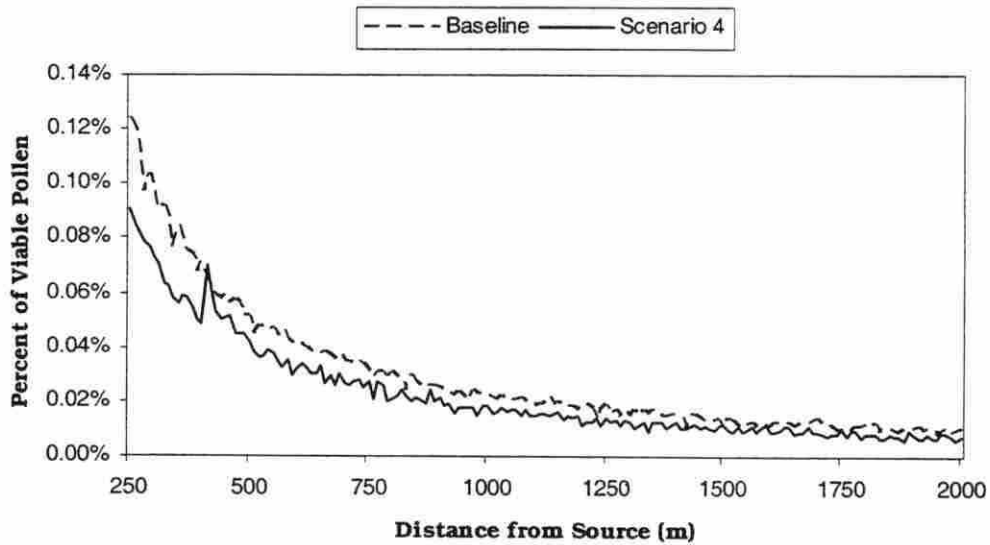


Figure 11. Long Distance Maize Gene Dispersal II

Long Distance Gene Dispersal

The final portion of the analysis is to compare the amount of viable pharmaceutical pollen to the amount of non-pharmaceutical pollen located in the vicinity of receptive non-pharmaceutical corn silks. Adjustments to equation (13b) must be made to account for the dispersal of pharmaceutical producing corn grown in accordance with federal regulations (14). The first modification is made to address the one-percent likelihood that genetic seepage occurs, P_s . The second adjustment is made to account for the possibility of contemporaneous fertility, represented by the probability P_f , with values of .02 and .05 for the third and fourth scenarios, respectively.

$$P = P_f \left(\frac{P_s Q_T}{Q_A} \right) \quad (14)$$

Assuming that twelve rows of pharmaceutical corn are planted .8 meters apart and that neighboring corn releases the same amount of pollen as the pharmaceutical corn, equation 14 is easily approximated. The first step in solving (14) is to model the movement of non-pharmaceutical pollen. Integrating the amount of all viable pollen deposited over the non-pharmaceutical crop yields Q_A .

Figure 11 presents values of P, the probability of pollination of non-pharmaceutical corn plants, located beyond the distance controlled by federal regulators referred to as the receptor crop, by pollen containing pharmaceutical producing genes, for Scenario 3. These data are dependant

on the size of the receptor crop, which impacts the relative amounts of pollen deposited. An increase in the size of the receptor crop coincides with larger amounts of non-pharmaceutical-pollen being released and later deposited, as well as slighter higher amounts of pharmaceutical pollen settling over the larger area. The probability of pollination by pharmaceutical pollen ranges from 1 in 50 million at the edge of the receptor crop to 1 in 3 billion 800 meters into the crop for Scenario 3.

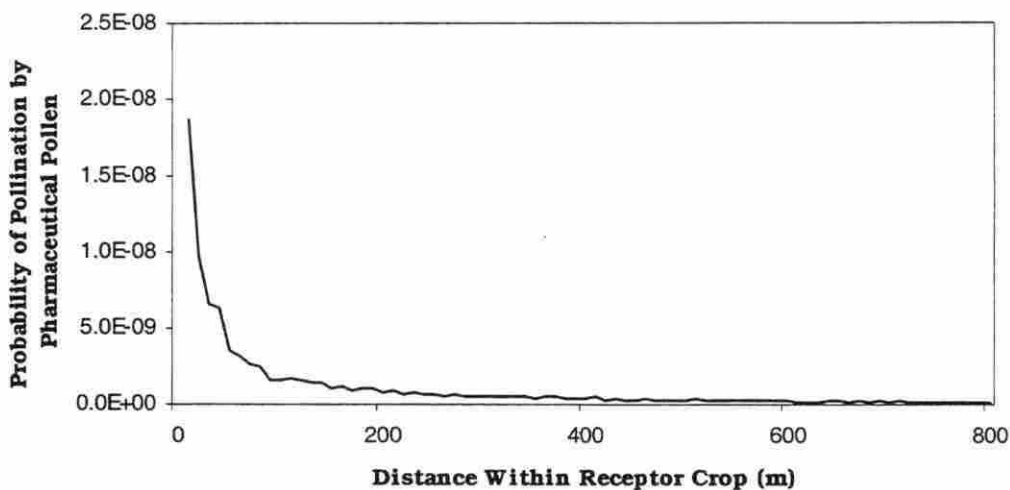


Figure 12. Percent of Viable Pharmaceutical Pollen Above Receptor Crop with No Buffer Rows

This same information found using parameter values under Scenario 4 is presented in Figure 12. Interestingly, in this case where buffers are present there is much higher likelihood of pollination by pharmaceutical pollen than for Scenario 3. At the edge of the field 1 in 10 million pollen

grains are from a pharmaceutical corn plant. This probability falls to 1 in 700 million, 800 meters into the field.

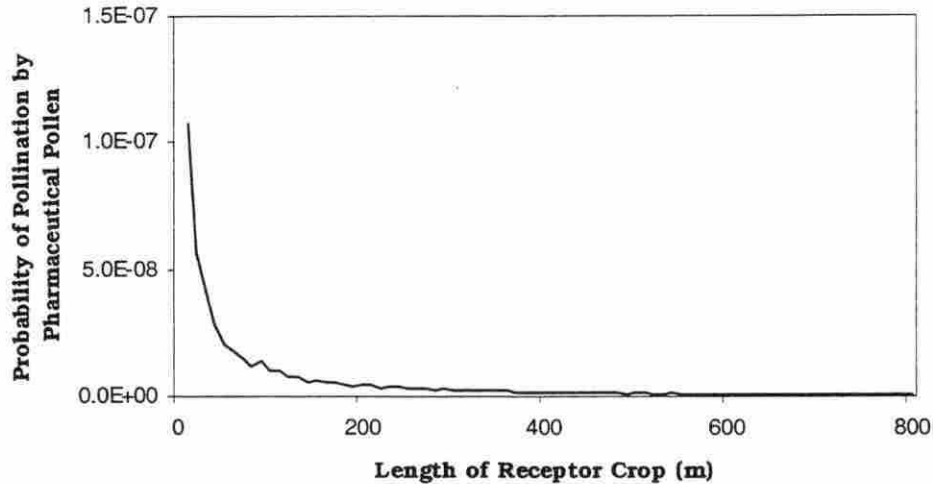


Figure 13. Percent of Viable Pharmaceutical Pollen Above Receptor Crop With Buffer Rows

Probability of Contamination

Industry groups such as the North American Millers Association have set zero tolerance levels for contamination. This probability of any contamination can be calculated using P , the probability of pollination described previously. It is determined by equation 15 where P_c is the probability of contamination and K is the number of kernels harvest from the receptor crop.

$$P_c = (1 - (1 - P)^K) \quad (15)$$

The probability of any contamination is calculated for forty acre plots just outside federally designated isolation distances for pharmaceutical corn production. The source crop has the same length as the receptor crop, 1320 feet, and has a width of 12 rows spaced 30 inches apart. The number of kernels harvested from the receptor crop is assumed to be 800 million, with each corn plant producing the same number of kernels.

For pharmaceutical corn grown without buffer rows, the probability of contamination occurring is .7957. When buffer rows are present, the probability rises to .9998. This level of gene dispersal may be acceptable for transgenes with minimal adverse affects. In the case of the introduction of pharmaceutical producing genes into the feed and food supply where there is an official zero tolerance standard, these results demonstrate a shortcoming of federal policy concerning the production of such crops. However, APHIS regulations specifically state that minimum guidelines can be emboldened or permits for production denied on the nature of the risk involved.

CHAPTER 5. SUMMARY

A Lagrangian Stochastic (LS) model was constructed to estimate the dispersal of maize genes. Unlike previous studies which relied on the gathering of field data, the theoretical model provides the ability to easily replicate situations while changing one or more parameters, without having to exert the effort required to conduct a field experiment. This flexibility allows for examining the behavior of corn pollen grown in accordance with APHIS regulations for pharmaceutical corn over the course of 1,000 years of wind speeds drawn from a fitted Weibull distribution.

Before these scenarios were dealt with, a baseline model was constructed and calibrated using experimental data where pollen was collected on greased slides. The baseline model underestimated the relative amount of pollen at all distances. This led to the realization that model predictions are likely on the low side.

For all but the mildest wind speeds, pollen reached distances of greater than one mile in situations with and without vegetative buffers. The probability of pollination by pharmaceutical crops outside federally designated distances is also calculated which while small exceed the zero tolerance policy espoused by those in the food processing industry. With this in mind, great caution should be placed on the production of transgenic corn plants that transmit restricted genetic material via pollen near other corn production.

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