Management of Hybrid Maize Seed Production

David L. Beck
CIMMYT, August, 2002

I. MAIZE HYBRID TYPES
II. SELECTION, MAINTENANCE, AND PRODUCTION OF PARENT SEED STOCKS
   Inbred line development and characterization
   Female inbred lines
   Male inbred lines
   Inbred line maintenance
   Practical suggestions on maintaining early generation lines
   Practical suggestions on maintaining highly inbred lines
   Production of parent seed stocks
   Foundation seed stock increase

III. AGRO-CLIMATE

IV. SITE SELECTION CRITERIA AND CONTRACT FARMERS

V. AGRONOMIC MANAGEMENT OF SEED PRODUCTION FIELDS
   Field preparation and planting dates
   Planting density
   Machine planting
   Fertilizer
   Irrigation
   Weed control
   Insect control
   Disease control

VI. MAIZE POLLEN AND ISOLATION
   Pollen production and movement
   Isolation
   Isolation distances
   Time isolation

VII. PLANTING HYBRID SEED MAIZE
   Avoiding mixtures of male and female lines
   Female:male ratios
   Compact or inter-planting
   Male row removal
   Promoting pollen movement
   Achieving synchronization in time between male and female parents

VIII. VARIETAL CHARACTERIZATION
   How to
   Biotechnology

IX. ROGUING
   Contaminants removed by roguing
   Selection of land to minimize roguing
   Procedures for roguing
   When to rogue
X. POLLEN CONTROL
   Detasseling
   Duration of the detasseling period
   Potential complications
   Hand or manual detasseling
   Mechanical detasseling
   Pollen control with cytoplasmic male sterility
   Pollen control with chemical agents
   Pollen control with biotechnology assisted methods

XI. HARVESTING
   Appropriate harvest time
   Hand harvesting
   Machine harvesting

XII. RECEPTION, HUSKING, AND SORTING

XIII. DRYING
   Sun dryers
   Drying temperatures
   Artificial drying systems

XIV. SHELLING, CLEANING, AND SIZING
   Shelling
   Cleaning
   Sizing
   Cleaning sized seed

XV. TREATING SEED
   Fungicides
   Insecticides
   Seed coatings

XVI. BAGGING

XVII. STORAGE
   Bulk and bagged storage
   Storage temperature and relative humidity
   Storage cleanliness and organization

XVIII. QUALITY ASSURANCE
   Procedures and standards
   Quality control laboratories
   Field inspections
   Post-harvest inspections
   Seed quality tests
   Moisture content determination
   Genetic purity analysis
   Germination test
   Vigor tests
   Seed health tests
   Seed tests for genetically modified organisms
   Quality assurance and genetically modified organisms

XIX. SUMMARY

XX. ACKNOWLEDGEMENTS

XXI. REFERENCES
I. Maize Hybrid Types

Numerous types of both conventional (based only on inbred lines) and non-conventional (where at least one parent is not an inbred line) maize hybrids may be produced (Vasal, 1988). Conventional hybrid types include single crosses, three way crosses and double crosses (Figure 1.). Single cross hybrids are based on two parental inbred lines whereas three way crosses first require the production of a single cross hybrid followed by its use as a female parent crossed to a male inbred line. Double cross hybrids are the product of crossing two distinct single crosses. Single crosses are popular in the developed world because of their high yield performance and uniformity. However, they are expensive and difficult to produce as the female parent on which the hybrid seed is produced is typically a relatively low yielding inbred line. Three way and double cross hybrids overcome this difficulty as the female parent in these conventional hybrid types is a single-cross hybrid. Disadvantages of double cross hybrids is that they need seven separate production fields including four blocks to maintain and produce the inbred lines, two fields to produce the two single-cross hybrids, and finally a production field to form the double cross hybrid (Figure 1.). At present, three way cross hybrids are the most common maize hybrids types grown in much of the developing world.

Figure. 1. Production of different kinds of maize hybrids

From: Maize seed industries in developing countries, 1998. Morris, M.L.(ed.).

Non-conventional hybrids can be classified into four major categories including varietal hybrids, family hybrids, topcross hybrids, and double topcross hybrids (Vasal and Gonzalez, 1999a). Most non-conventional hybrids are based on two components. This is true with varietal hybrids
which are based on a cross between two varieties, synthetics, and/or populations. Family hybrids consist of two either full-sib or half-sib families coming from the same or different populations. Topcross hybrids may include crosses between an inbred line and a variety, synthetic, population, or family. Double topcross hybrids involve a single cross in combination with a variety, synthetic, population or family. The fact that none or only one component in non-conventional hybrids is an inbred line helps overcome some of the difficulties associated with producing conventional hybrids. However, non-conventional hybrids typically are less uniform and lower yielding than their conventional hybrid counterparts.

II. Selection, Maintenance and Production of Parent Seed Stocks

Selection, maintenance and production of OPV, synthetic, population, or family parental seed will be addressed in a separate paper. Management of inbred parental seed stocks will be discussed here.

Inbred parental seed is the foundation for the production of all conventional and some non-conventional maize hybrid types. Developing good inbred lines is an important but difficult and costly process. This is illustrated by a statement by distinguished maize breeder Dr. A.R. Hallauer when he said “only 1 in 10,000 S2 or S3 lines tested eventually were used to any extent in commercial hybrids.” (Hallauer and Miranda, 1997). It has been estimated that in the U.S. it costs about one half million dollars to develop a commercial inbred line when all costs are included (T. Little, personal communication, 1992 cited in Lopez-Pereira and Garcia, 1997). The difficulty and cost arises from various factors including: a) the decrease in vigor along with the exposure of deleterious characters which occurs during the inbreeding process rendering most maize lines unusable, b) the extensive and costly evaluations of inbreds in hybrid combination, and c) the fact that in addition to yield and combining ability, inbreds must possess various production characteristics to facilitate their use particularly in single cross hybrid seed production.

Compared to maize varieties or hybrids, inbred lines typically have shorter height, less vigor, thinner stalks, smaller tassels, smaller ears, and lower yields. Associated with this, they are often more susceptible to stresses such as drought, diseases, and insects. This reduction in vigor and yield, and generally greater susceptibility to stress often creates problems in seed production.

Inbred line development and characterization

Lack of proper characterization of inbred progenitors can be the cause of serious losses in seed production. As maize inbred lines typically have a large G x E interaction compared to other crops, it is essential to obtain sufficient performance information on inbreds in their
planned production environments. Of special importance is to obtain information on floral synchronization (nicking), seed yields in female lines, pollen production capability of male lines, and responses of inbreds to fertility, pests, diseases, and pesticides. Table 1 shows the dramatic differences in flowering dates that can occur when inbreds are grown in a range of environments. With U.S. Corn Belt adapted inbreds, as little as 3 to as many as 50 locations of flowering data may be needed to make accurate predictions of nicking between male and female parents (Lauer, M., pers. comm., 1998).

Table 1. Time and length of flowering period in days for maize lines and hybrids (mean of three sites in each area)

<table>
<thead>
<tr>
<th>Seed Parent</th>
<th>Flower</th>
<th>Glendale (1100 m)</th>
<th></th>
<th>Harare West (1500 m)</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Days to 1st Flower</td>
<td>Flowering Period</td>
<td>Days to 1st flower</td>
<td>Flowering Period</td>
</tr>
<tr>
<td>N3</td>
<td>Silks</td>
<td>64</td>
<td>17</td>
<td>73</td>
<td>19</td>
</tr>
<tr>
<td>Sc</td>
<td>Pollen</td>
<td>67</td>
<td>11</td>
<td>75</td>
<td>16</td>
</tr>
<tr>
<td>SX6</td>
<td>Silks</td>
<td>63</td>
<td>14</td>
<td>73</td>
<td>21</td>
</tr>
<tr>
<td>K64r/RL7</td>
<td>Pollen</td>
<td>64</td>
<td>16</td>
<td>74</td>
<td>22</td>
</tr>
<tr>
<td>SR52</td>
<td>Pollen</td>
<td>65</td>
<td>15</td>
<td>70</td>
<td>16</td>
</tr>
</tbody>
</table>


Female inbred lines

Developing productive female lines is one of the most important aspects of profitable maize seed production. Hoegemeyer and Gutormsen (1996) identified large genetic differences for germinability and vigor in a set of inbred lines sown under cool, wet conditions. Frey (1981) emphasized the importance of heat and drought tolerance in female lines to better achieve production goals in terms of quantity and quality. Additional desirable characteristics of female (seed) parent lines include:

a) High seed yield
b) Good seed grade-out
c) Lodging resistance
d) Good silk emergence
e) Uniformity in flowering
f) Good tassel exertion prior to pollen shed
g) Ear rot resistant

Male inbred lines

Dodd (1998) commented on “the remarkable trend toward more outcrossing” in hybrid maize seed production in the past 10 years and associated this with less pollen in male lines. He indicated that the decrease in pollen production is in some ways unavoidable as we push up the yield levels in maize hybrids thus increasing within plant
competition between grain and pollen. However, he suggested an additional reason being the over attention in production research on female lines and under attention on male pollen production. Male (pollen) parent lines with good pollen production and a long pollen shedding period allow growers to use higher female:male ratios and often result in less nicking problems. Methods to evaluate pollen producing ability include: a) taking and weighing a 15 tassel sample from a given line just prior to pollen liberation and then doing the same after pollen release. The difference in weight is an indication of pollen production (Wych, pers. comm., 1998), b) placing 10 pollinating bags per line and collecting pollen daily for 5-10 days. Pollen collected is measured in a graduate cylinder and checked under a microscope for viability, c) tagging individual plants upon initiation of pollen shedding and then simply checking each plant daily by tapping the tassel against a black sheet of paper and comparing pollen amount to a standard check (Thielen, 1986). Additional desirable characteristics of male lines include adequate plant height and good standability.

Inbred line maintenance

The goal in inbred line maintenance is to maintain the performance, appearance, and genetic integrity of the original lines while limiting roguing expenses. It is essential to maintain the highest levels of field execution including using properly isolated plots, rigorous elimination of off-types, care in pollination procedures, and using accurate pedigree records and labels (Vasal and Gonzalez, 1999b). However, despite all these precautions, changes in the breeding behavior of lines may occur usually caused by delayed segregation or relic heterozygosity, mutation, transposable elements, epigenetic modification, and out-crossing.

Limiting the number of line increases through the use of good cold storage and other means will help reduce the risk of out-crossing and genetic drift.

Maintaining inbreds may involve self-pollination, sib-pollination, or a combination of these procedures. Selfing aids in maintaining inbreds in a pure condition. Sibbing tends to prevent excessive loss of vigor. Parental inbreds may be planted ear-to-row, or ears from individual inbreds may be bulked for increase. Off-type plants from individual ears may be detected and discarded more easily using the ear-to-row method. Increase of inbred by sib-pollination may be done by hand-pollination or in an isolated field. All off-type plants should be removed during the growing season before pollen is shed.

Practical suggestions on maintaining early generation lines (S₁ – S₄)

Plant a minimum of 75-100 plants in 3-4 5m rows. Rogue out off-type
plants. Make plant to plant sib crosses among selected plants. At harvest select ears from plants with characteristics consistent with the varietal description. Bulk seed from selected ears. If large quantities of seed are needed, one alternative is to plant out the line in an isolation block. Rogue out off-type plants prior to flowering. Allow maize to random pollinate. At harvest, select ears consistent with the varietal description.

Practical suggestions on maintaining highly inbred lines ($S_5+$)

Alternative 1 – Self-pollination with progeny testing. Plants of the inbred line are self-pollinated. Plants with uniform characteristics consistent with the varietal description are harvested individually. Ears consistent with the varietal description are selected and shelled individually. In the second season, a progeny row from each selected ear is sown. Eliminate off-type rows, followed by self-pollinations of all plants in selected rows consistent with the varietal description. Harvest self-pollinated ears individually and discard off-types. Shell each ear separately. A portion of each ear will be used for future progeny testing and the remainder from all ears bulked as breeder seed.

Alternative 2 – Sib-mating. The inbred line is planted out with bulked seed. Non-uniform plants are rogued out prior to flowering. Pollen is bulked and used to pollinate an equal number of plants, or plant-to-plant crosses are made, or plants are grown in isolation and allowed to cross pollinate. Plants with uniform characteristics consistent with the varietal description are bulked. Part of the seed is used for future maintenance of the inbred and the remainder is used as breeder seed.

Other modifications may be introduced to the two alternatives described. It is critical to remember that, in order to maintain the genetic integrity and vigor of early generation lines, a sib-mating scheme rather than a self-mating procedure is recommended.

Production of parent seed stocks

Large quantities of parent seed stocks are required annually to plant the hundreds of thousands of hectares of commercial hybrid maize seed (Wych, 1988). Large seed companies commonly have parent or foundation seed departments responsible for the production and inventory of inbred and single cross parents needed for commercial seed production. Smaller companies may purchase parental or foundation seed from various suppliers.

Foundation seed stock increase
Foundation or basic seed stock increase involves the maintenance and increase of inbred lines and single cross parent seed used to produce commercial hybrids. Inbreds must be maintained and increased under rigid control to ensure good final product performance (Vasal and Gonzalez, 1999b). Three key steps in this process include: a) establishing and maintaining a supply of breeder seed, b) increasing inbred line seed, and c) producing single cross parental seed for use in three way and double cross production.

Although early generation lines may be used, it is more common to use advanced generation lines (particularly in more developed breeding programs) which have gone through six or more cycles of inbreeding. The main advantage of using highly inbred lines is their uniformity. The breeder has the responsibility to ensure that the inbreds in production are homozygous, uniform for plant type, and that they adequately represent the genetic constitution of the inbred. All inbred increases are made from a base population of breeder seed maintained and supplied by the breeder.

Foundation seed production is conducted under a set of more rigid standards than that used for commercial hybrid production. Standards are stricter for factors such as isolation distances, number of border rows, minimum number of off-types acceptable, with foundation as compared to commercial seed. Quality control evaluations are commonly more extensive with foundation seed. Many of these themes will be discussed in the subsequent sections describing commercial maize seed production.

III. Agroclimate

In maize seed production it is essential to select an agroclimate that is conducive to proper development of the lines, synthetics, hybrids, and OPVs in production. It is critical to select sites which allow adequate reproduction of all plants to reduce the risk of shifts in the genetic make-up of a given material. Suitable environmental conditions for successful maize seed production include good soil type with adequate radiation, temperature and rainfall (McDonald and Copeland, 1997). Well-drained fertile soils with good water holding capacity are preferred. Level and uniform fields with economically controllable natural weed populations are also desirable. Maize prefers environments with abundant sunshine during the growing season and sufficient but not excessive rainfall with good distribution particularly during the flowering period (Desai, 1997). Excessive rainfall can result in poor pollination and seed set and higher incidence of diseases and pests. Low moisture availability especially when accompanied by high temperatures can be detrimental to pollination mainly by reducing growth and receptivity of female silks. These conditions can also complicate the seed producers’ goal to achieve a good synchronization or nick between male and female parents.
Intermediate to large-scale production of maize seed will often require the use of several locations. Many of these sites are commonly owned and managed by commercial farmers. Therefore, an essential aspect of successful maize seed production, is the selection of growing areas and contract farmers.

IV. Site Selection Criteria and Contract Farmers

Proper isolation in time, distance, or with other means is essential to reduce the possibility of genetic contamination by outcrossing. The locations chosen must take into account the conditions in neighboring fields including first if maize will be sown, and then the planned planting date in order to determine if there will be an isolation problem. If possible, avoid planting in fields that were previously sown to maize to avoid problems with a volunteer crop. Other considerations to reduce physical contamination are to avoid areas with significant movement of people, animals, or equipment.

Choosing sites with good transport access is important to facilitate the delivery of inputs such as seeds and fertilizers, the transport of people during labor, intensive periods such as roguing and detasseling, as well as to deliver the harvested seed. Selecting sites in the proximity of processing facilities is advantageous to limit seed deterioration and reduce costs between harvest and processing.

Collaborative work between seed enterprises and farmers in the production and sale of hybrid maize seed has been critical to the success of the U.S. maize seed industry. Since maize seed activities are costly due to the size, labor-intensity, and high level of supervision required, most U.S. based seed companies conduct their commercial seed production using land and labor contracted from private farmers (Wych, 1988). Both in the U.S. and worldwide, selecting the appropriate contract farmers is essential and should include consideration of the following factors:

a) Generally prefer the most progressive, innovative, and flexible farmers.
   The sensitivity of inbreds to a range of environmental conditions require that contract growers must be willing to alter their cultural practices including the timing, rate, and kind of pesticides applied. Equipment modifications may also be necessary.

b) The inherent capability of the farm (i.e. soil type) plus the capacity of the farmer to irrigate, successfully control pests, etc.

c) Farmers position within the isolation block.

d) Farmers social relationships in the area. This is especially important regarding hiring of people for the roguing and detasseling operations.

e) Farmers previous experience in seed production.
V. Agronomic Management of Seed Production Fields

Agronomic practices in maize seed fields are in general similar to those used in producing a commercial grain crop. However, there are some additional requirements unique to seed production. First, we must remember that the value of good seed is higher than that of grain. Therefore, a seed crop warrants greater care and more inputs than a grain crop. Second, we must keep in mind our goal of obtaining the maximum number of high quality genetically pure seeds while at the same time minimizing the risk. Third, care shown in the selection and preparation of the seed fields to obtain the most uniform growing environment possible will greatly facilitate identification of off-type plants in future roguing operations. Fourth, careful consideration must be given to the area planted taking into account projected sales, labor availability during critical times such as detasseling, and production research data on the yield potential of the given hybrids or inbreds. Fifth, when conventional maize hybrids are being produced, inbred lines are involved. Inbreds characteristically have relatively poor vigor and above average susceptibility to environmental stress. Therefore, the level of agronomic management must be raised in order to support the development of these sensitive yet valuable materials.

Field preparation and planting dates

It may be advantageous to prepare seed fields several weeks prior to planting to help in the pre-germination of weed seeds and volunteer maize. Alternatively, a delay in the last tillage until volunteers and weeds have germinated may be desirable. Planting dates should be chosen to avoid risky environmental conditions such as excessively cool or hot temperatures and isolation problems. Various studies have demonstrated the advantage of early plantings to achieve better yields (Wych, 1988; Craig, 1977). Separate planting dates may be used to achieve a proper nick between the male and female parents. A winter season seed crop may be necessary to meet production goals but will likely introduce new environmental risks. Planting the production field starts with a thorough seedbed preparation to help ensure even germination and rapid seedling development. The seedbed should be plowed and harrowed properly to produce a fine tilth.

Planting density

Planting density in the seed field should be chosen to produce maximum yields of high quality seed of saleable kernel size. Optimum population is principally a function of soil moisture and fertility, and the parental material being used. For maize inbreds, these densities can vary considerably from 40,000 to 80,000 plants ha\(^{-1}\) (Wych, 1988). In general, slightly lower population densities are recommended for female rows. This will help ensure good seed set and development while avoiding problems of plant stress which can increase silk delay and result in more disease and lodging problems. However, too low female populations can result in sub-optimal
seed yields, poor grade-out including excessive production of large round kernels, and significant tiller development which can complicate detasseling operations. Male rows are frequently sown at higher densities as male gamete production is less sensitive to density and environmental stress, and hopefully greater pollen production can be achieved per unit area. Excessive population densities should be avoided to facilitate full expression of the plant type to aid in identification of true-to-type and to eliminate off-type plants.

*Machine planting*

When machine planting ensure that the planter is properly calibrated for the size and shape of the seed leading to the attainment of a proper planting density. Planting depth should coincide with common practices in the area but may need to be adjusted due to inbred sensitivity to emergence conditions and to delay flowering in one of the parental lines to help achieve a better nick. Also, it is essential to thoroughly check the planter to ensure that it is completely clean and free of other maize seeds.

*Fertilizer*

Fertilizer should be applied to achieve maximum seed production while considering economic and environmental factors. Appropriate fertilizer applications will vary depending on the soil and environmental conditions and the material being produced. In general, inbreds have poorer rooting capacity than hybrids and therefore are more vulnerable to nutrient imbalances and deficiencies. Soil tests along with production data on inbred response to fertility are recommended. Proper levels of N, P, K, are essential for adequate plant growth. Several recent studies looking at optimal N fertilizer rates in maize seed fields have shown that best seed yields and grade-outs can be achieved with lower N rates (55 – 110 kg ha\(^{-1}\)) than that recommended by many companies and commonly used by seed producers (Peterson and Corak, 1993; Wilhelm and Johnson, 1997). In addition to N-P-K, several micronutrients are of special importance for seed development processes including magnesium (essential for germination), boron (flower development and pollen germination), zinc (seed formation), and molibdenum (endosperm development).

*Irrigation*

If irrigation is available, it should be applied based on soil texture and depth, climactic conditions, and the needs of the crop. Irrigation early in the season may be beneficial to help establish a uniform stand. Water availability during flowering in maize is especially critical to help achieve good seed set. Scheduling irrigations just ahead of flowering is important to both meet the crop needs but also to avoid complications with machines and/or personal involved in the detasseling operation (Taylor, 1979).
general, inbred lines require less water at a given time but more frequently than full-vigor hybrids or varieties.

**Weed control**

Proper control of weeds, insects, and diseases are an integral and necessary part of maize seed production. Proper weed control is particularly important, as it will greatly facilitate roguing, detasseling, field inspection, and harvest operations. Weed control with inbreds is especially critical as they often have low vigor thus are not well suited to compete with aggressive broad-leaf weeds and grasses. Harvesting an excessive amount of weeds in a seed crop can complicate the cleaning and conditioning process. Additionally, weeds can be hosts of undesirable diseases and insects which can reduce seed quality. Numerous pre- and post-emergence herbicides are available for weed control in maize seed production fields but caution should be applied in their selection as inbred toxicity to certain compounds has been observed (Wych and Schoper, 1987). Additional precautions for herbicide and other chemical applications include ensuring that spray tanks are clean before use and properly calibrated to apply proper rates while limiting human contact.

**Insect control**

Control of both above and below ground insects is essential in maize seed production. Most below ground insects are controlled with either seed treatments or granular insecticides applied at planting. Genetically engineered maize producing the \( Bt \) or other toxins are available to control above ground insects and soon will be available to control rootworms and other below ground pests. Chemical applications may be necessary to control insects particularly which feed on the silks, ears and tassels. Heavy populations of borers, rootworm beetles, or aphids can result in poor seed set and development and thus a reduction in both the quality and the quantity of maize seed (Culy et al., 1992). An integrated pest management program (IPM) is recommended including proper scouting to determine if and when insecticides are needed. Selection of the proper insecticide will depend on the specific insect to be controlled, the level of infestation, the development stage of the seed crop, safety considerations, and the reentry period.

**Disease control**

Proper disease control is important to obtain good seed quality and avoid spreading seed born pathogens. Genetic resistance in parental lines is preferred but sometimes otherwise outstanding lines are susceptible to particular pathogens present in production fields. As with insect control, an
IPM program is desirable to monitor disease development and to determine if and when chemical application is necessary.

VI. Maize pollen and isolation

The cross-pollinated nature of maize, along with its abundant production of wind carried pollen, often make it very difficult for seed producers to avoid contamination. Proper isolation is essential to limit contamination from foreign pollen, thus promoting the production of genetically pure seed.

Pollen production and movement

Understanding maize pollen production and movement will help us better plan to achieve the appropriate isolation. A maize tassel can produce between 2 to 25 million pollen grains over a period typically of from 5 to 12 days (Hall et al., 1982; Poehlman, 1979). At peak production, the pollen density in a given field can exceed 500 grains cm\(^{-2}\) day\(^{-1}\). Determination of pollen viability is problematic with some considerable range in accepted values depending on the genetics and the methods used to determine viability. Burris (2001) stated “it is safe to assume that pollen can maintain viability from a few hours to several days.” Maize pollen while within the tassel appears to be fairly tolerant to stress conditions, with drought stress shown to have little effect on pollen viability, although germinability of pollen often declines significantly when temperatures exceed 38°C (Westgate and Boyer, 1986 a,b; Herrero and Johnson, 1981). However, once maize pollen is shed from the tassel, viability declines rapidly under typical ambient temperatures. Luna et al. (2001) found that pollen exposed to ambient field conditions in Nayarit, Mexico, declined in viability by an average of 69% after one hour over two years, and was 100% non-viable in two hours. Jones and Newell (1948) reported pollen survival for three hours in a pollination bag under mid-summer field conditions in Nebraska. Although maize pollen is abundantly produced, its movement in the field is often limited. This is because maize pollen is relatively large and heavy compared to pollen of other crops. Di-Giovanni et al. (1995) measured maize pollen settling velocity at 31 cm s\(^{-1}\) or an order of magnitude greater than that reported for other wind pollinated pollen species. Table 2 and Figure 2 show that most maize pollen will commonly fall within 60 m of the source. However, if windy conditions exist these distances could expand considerably.
Figure 2. Typical concentration and deposition patterns from a corn pollen area source represented by the shaded figures. Concentrations are shown in the horizontal at a height of 1.5 m and in the vertical along the plume centerline.
Table 2. Variation with distance of mean, standard deviation, and coefficient of variation of centerline concentrations at a height of 1.5 m expressed as a percentage of the 1-m concentration.

<table>
<thead>
<tr>
<th>Source</th>
<th>Sampling Distance (m)</th>
<th>Mean</th>
<th>Stand. Dev. (SD)</th>
<th>SD/Mean</th>
<th>n</th>
</tr>
</thead>
<tbody>
<tr>
<td>1963 maize</td>
<td>1.0</td>
<td>100</td>
<td>-</td>
<td>-</td>
<td>15</td>
</tr>
<tr>
<td></td>
<td>9.2</td>
<td>29.2</td>
<td>10.7</td>
<td>0.37</td>
<td>15</td>
</tr>
<tr>
<td></td>
<td>27.5</td>
<td>8.0</td>
<td>6.4</td>
<td>0.80</td>
<td>15</td>
</tr>
<tr>
<td></td>
<td>54.9</td>
<td>1.3</td>
<td>3.3</td>
<td>2.54</td>
<td>15</td>
</tr>
<tr>
<td>1964 maize</td>
<td>1.0</td>
<td>100</td>
<td>-</td>
<td>-</td>
<td>13</td>
</tr>
<tr>
<td></td>
<td>7.7</td>
<td>34.0</td>
<td>17.5</td>
<td>0.51</td>
<td>13</td>
</tr>
<tr>
<td></td>
<td>15.3</td>
<td>16.3</td>
<td>10.4</td>
<td>0.64</td>
<td>13</td>
</tr>
<tr>
<td></td>
<td>32.0</td>
<td>4.6</td>
<td>4.3</td>
<td>0.94</td>
<td>13</td>
</tr>
<tr>
<td></td>
<td>59.5</td>
<td>1.1</td>
<td>2.1</td>
<td>1.91</td>
<td>12</td>
</tr>
</tbody>
</table>


Isolation

Proper isolation is basically accomplished in three ways including: a) distance b) time, and c) good synchronization. Seed producers often say that the best isolation is a perfect nick, that is, when the pollen parent starts shedding just before silk emergence in the female parent. An additional option may be used when it is difficult to get suitable distance or time isolation. This includes planting a barrier of foundation or certified seed of the same variety on all sides about 50 m from the production block (Cordova et al., 1999).

Isolation distances

Factors likely to affect isolation distances include the following (Wych, 1988):

a) The greatest contamination occurs within 50 to 75 m of the contaminating maize.

b) Pollen from border rows dilutes contamination.

c) Natural barriers (particularly trees) may reduce contamination.

d) An abundant supply of pollen from the male parent at the right time reduces contamination.

e) The risk of contamination is generally less the larger the production field.

f) Direction from contaminating pollen and prevailing winds influence the amount of contamination.

g) Differential flowering times are effective in isolation if silks of female parents are not receptive when contaminating pollen is present.

h) The genetic purity desired.

i) Environmental conditions during pollination. For example, if drought is present during flowering, chances of contamination would increase...
because pollen from contaminating maize would likely be more competitive than pollen from an inbred line.

Standards for minimum isolation distances have been developed based on research investigating flower morphology and pollen weight and movement. The major factors affecting the standards presented here for maize include:

a) Whether the crop is a hybrid or open-pollinated variety (Table 3).
b) Whether breeder, foundation, or certified seed is being produced.
c) The use of border rows (Tables 4 & 5).
d) The grain color/texture of the contaminating maize.
e) The size of the field (Table 5).

Minimum isolation distances normally range from 100 – 400m (Table 3). The big unpredictable environmental factor which often is the cause of contamination from undesirable pollen is the wind which can carry maize pollen for several kilometers. Certainly, the greater the distance and number of male border rows, the less chance of contaminant pollen entering your seed field.

<table>
<thead>
<tr>
<th>Category</th>
<th>Hybrids</th>
<th>OPV</th>
</tr>
</thead>
<tbody>
<tr>
<td>Breeders</td>
<td>Absolute</td>
<td>300 m</td>
</tr>
<tr>
<td>Foundation</td>
<td>400 m</td>
<td>300 m</td>
</tr>
<tr>
<td>Certified</td>
<td>200 m</td>
<td>200 m</td>
</tr>
</tbody>
</table>

Table 4. Minimum standards for foundation single cross and inbred line maize seed production fields.

<table>
<thead>
<tr>
<th>Minimum Distance from other Maize (m)*</th>
<th>Border Rows (min.)</th>
</tr>
</thead>
<tbody>
<tr>
<td>&gt; 300</td>
<td>2</td>
</tr>
<tr>
<td>275 – 299</td>
<td>4</td>
</tr>
<tr>
<td>250 – 274</td>
<td>6</td>
</tr>
<tr>
<td>225 – 249</td>
<td>8</td>
</tr>
<tr>
<td>200 – 224</td>
<td>10</td>
</tr>
</tbody>
</table>

* With the same grain color and texture

Source: Ohio State University, 1980.

Table 5. Minimum standards for certified single cross maize seed production fields.

<table>
<thead>
<tr>
<th>Minimum distance from other maize (m)*</th>
<th>Border rows (min.)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1 – 20 Acres</td>
</tr>
<tr>
<td></td>
<td>21 Acres or more</td>
</tr>
<tr>
<td>125.0</td>
<td>0</td>
</tr>
<tr>
<td>112.5</td>
<td>2</td>
</tr>
<tr>
<td>100.0</td>
<td>4</td>
</tr>
<tr>
<td>87.5</td>
<td>6</td>
</tr>
<tr>
<td>75.0</td>
<td>8</td>
</tr>
<tr>
<td>62.5</td>
<td>10</td>
</tr>
<tr>
<td>50.0</td>
<td>12</td>
</tr>
<tr>
<td>37.5</td>
<td>14</td>
</tr>
<tr>
<td>25.0</td>
<td>16</td>
</tr>
<tr>
<td>0.0</td>
<td>Not permissible</td>
</tr>
</tbody>
</table>

* With the same grain color and texture.

Source: Ohio State University, 1980.

Time isolation

When using time isolation, it is best to have a minimum of three to four weeks difference between the planting of your maize seed crop and potential contaminant sources. At the Rattray Arnold Research Station in Zimbabwe, Africa, a study was conducted to evaluate the effect of time isolation on the degree of contamination in the N3 seed parent (Havazvidi, 1990). N3 is a white grained female line used in the popular single-cross hybrid SR52. This line was planted at seven day intervals up to 35 days after the control. A yellow-grained three-way cross hybrid, ZS206, was planted on the same day as the control treatment surrounding the N3 seed parent planted at different dates. Results
comparing time to flower and the amount of yellow contamination in N3 by mass is shown in Table 6. The level of contamination was significant when the time isolation between 95% silking of N3 and 5% pollen shed of ZS206 was less than 14 days. This occurred when the N3 seed parent was planted less than 3 weeks before ZS206.

**Table 6. The effect of time isolation on the degree of contamination of the N3 seed parent.**

<table>
<thead>
<tr>
<th>Delay in planting N3 Seed Parent</th>
<th>DS 5 = Days from planting ZS206 to 5% silking in N3</th>
<th>DS 5 in N3 minus DP 95 In ZS206</th>
<th>% Yellow contamination in N3 by mass</th>
</tr>
</thead>
<tbody>
<tr>
<td>0 days</td>
<td>84</td>
<td>0</td>
<td>86.21</td>
</tr>
<tr>
<td>7 days</td>
<td>88</td>
<td>+ 4</td>
<td>61.36</td>
</tr>
<tr>
<td>14 days</td>
<td>92</td>
<td>+ 8</td>
<td>3.80</td>
</tr>
<tr>
<td>21 days</td>
<td>96</td>
<td>+ 12</td>
<td>0.03</td>
</tr>
<tr>
<td>28 days</td>
<td>103</td>
<td>+19</td>
<td>0.03</td>
</tr>
</tbody>
</table>


**VII. Planting Hybrid Seed Maize**

Planting a maize seed crop differs from the planting of a commercial crop in that border rows are often required, a lower plant density is often recommended, and when producing a hybrid—two components rather than one are sown in the same field. This latter aspect can significantly complicate the planting. Following find guidelines on the planting operation for hybrid maize.

**Avoiding mixtures of males and females**

To help avoid mixtures one may color code the male and female parents. For example, female parents may be seed treated with a red dye, and male parents with a green dye. Similarly, female parents may be placed in red striped bags, and male parents in green striped bags. A practice of some seed companies is to not permit seed of male and female parents to be transported in the same vehicle or trailer or to be placed in the same area of the field. Staking the field with different color codes for the male and female rows may help reduce errors.

**Female:male ratios**

It is important to recognize that hybrid seed is only harvested off of female rows. Therefore, we want a minimum number of male rows, but sufficient pollen production to ensure a good seed set in the entire field. The female:male ratios typically range from 2:1 to 6:1 depending principally on the pollen producing ability of the male parent (Wych, 1988). The most
popular ratio used today in the U.S. Corn Belt is 4:1 (Burris, personal communication, August, 2001). Some producers choose to use altered row ratios such as 3:1/4:1. A denser planting of the male rows is common since pollen production is less sensitive to density stress than seed production. The type of planting, detasseling, and harvest equipment available may also affect a grower’s decision on female:male ratios. Production research data in various locations is critical to make appropriate male:female row recommendations for the hybrids in production. Tables 7 and 8 show results from trials in the U.S. and Central America comparing different planting types and patterns. Production research should include a careful evaluation of the production fields for degree of pollination, seed set, and grade-out by row position (Culy et al., 1991).

Table 7. Maize seed yield with different planting types and patterns.

<table>
<thead>
<tr>
<th>Pattern</th>
<th>Area (% Female)</th>
<th>Yield (kg/ha)</th>
</tr>
</thead>
<tbody>
<tr>
<td>4:1 Standard</td>
<td>80</td>
<td>3692</td>
</tr>
<tr>
<td>6:2 Standard</td>
<td>75</td>
<td>3476</td>
</tr>
<tr>
<td>6:2 Squeeze</td>
<td>80</td>
<td>3775</td>
</tr>
<tr>
<td>4:1 Standard</td>
<td>80</td>
<td>3898</td>
</tr>
<tr>
<td>4:1 Interplant</td>
<td>100</td>
<td>4173</td>
</tr>
</tbody>
</table>

Source: Newsource Pioneer

Table 8. Maize seed yield (Mg ha⁻¹) using different female-male patterns in three Central American countries, 1986.

<table>
<thead>
<tr>
<th>Pattern</th>
<th>El Salvador H-5</th>
<th>Honduras H-27</th>
<th>Guatemala H-B 83</th>
</tr>
</thead>
<tbody>
<tr>
<td>4:1</td>
<td>4500</td>
<td>3523</td>
<td>4000</td>
</tr>
<tr>
<td>5:1</td>
<td>5265</td>
<td>4028</td>
<td>4520</td>
</tr>
<tr>
<td>6:1</td>
<td>4320</td>
<td>4150</td>
<td>4140</td>
</tr>
</tbody>
</table>


Compact or inter-planting

This method involves planting the female parent throughout the seed field followed by squeezing in the male rows between the females every two, three, or four rows. The advantage is a fuller utilization of the land area for the female parent (where the hybrid seed is produced) and the placement of the male rows physically closer to the female rows.

Compact planting is usually limited to crosses including a female parent with intermediate vigor such that the male parent is not overshadowed thereby affecting pollen shed. For example, it would not be used in three-way cross hybrid production. Additionally, it is often limited to crosses with
short male delays. Use of this technique may include planting female rows in slightly wider row spacing. Other requirements include good root and stalk quality in both parents, and good pollen production in the male. With the inter-planting method, it is essential to plant straight rows and to have good weed control.

**Male row removal**

Male rows should be eliminated after flowering which is a common practice of many hybrid maize seed producers. This is often done by cutting, chopping, or running down the male rows after pollination is complete. Advantages of male row removal include less competition for the developing female plants resulting in better seed yields and quality, less chance of seed mixing at harvest, and less risk of theft. Dodd (1998) suggested that removing males after pollination becomes more important when the female line is carrying a specialty trait gene characteristic of the new GMO hybrids. Root lodging of the male is the most common cause of mixing the male with hybrid seed at harvest.

**Promoting pollen movement**

Normally, sufficient pollen is transported by the wind from male tassels into the female canopy. However, under certain circumstances such as drought or high temperatures, it may be necessary to intervene to help achieve sufficient pollination and seed set. Various techniques from the simple to the expensive are used to promote male pollen movement into the female canopy. These include manually shaking the male rows, carrying pollen in pollination bags, passing through male rows with back-pack sprayers or fans, and the use of airplanes and helicopters. These methods have various advantages and disadvantages including their cost and practicality. One or more of these methods may be necessary particularly when a good nick is not achieved between male and female parents.

**Achieving synchronization in time between male and female flowering**

One critical challenge facing breeders and seed producers of hybrid maize is the use of parents that differ in their time of flowering. Ideally, one would select parents of similar maturity for hybrid production. However, in the real world, many of the best hybrid combinations involve parents of different maturity. In the case of three-way cross production, the inbred male parent is usually later maturing than the female single cross. When producing hybrids involving parents of differing maturities, it is common to plant the parents using different dates to help synchronize flowering. However, various complications arise in field management. Not only must one enter the field at least twice for planting but multiple applications of fertilizer, irrigation, and pesticides may be necessary. Inclement weather may create further difficulties.
For maize seed producers, there is a big need for methodologies to regulate plant growth such that an entire field could be planted at one time. However, to date, the most popular and practical method is to use different planting dates.

Other methods used to achieve a good synchronization or nick in male and female flowering include:

a) Double planting of male rows where a second planting of the male rows is delayed and sown several inches to the side of the first male planting resulting in a greater time spread in pollen production.

b) Variable depth of planting. Deeper planting is only recommended for parents with good seedling vigor. As a guide, consider that for each 2.5 cm increased depth of planting, expect a two day delay.

c) Variable use of starter fertilizers. This may be used to advance or delay development for one to two days. Results have been less consistent in highly fertile soils, while better in low fertile soils. High phosphate ratio fertilizers tend to accelerate plant development, while high potassium ratios tend to delay plant development. Caution in application is warranted as too much fertilizer placed close to the seed can result in burning and reduced germination.

d) Foliar fertilizer applications which normally include P to accelerate and N to delay flowering.

e) Seed coat treatments. The major problems encountered in the use of chemical seed coat treatments is the cost and difficulty associated with achieving a uniform application of chemical around the seed (Burris, 1992). Certain chemicals have been used to inhibit imbibition of moisture resulting in delayed germination. For example, polyurethane coatings have been successfully applied to achieve delays of up to seven days depending on the thickness of the coating. However, these materials are dangerous to use when planting in soils subject to compaction or crusting. Recently, several products of this type have been commercialized. One called Pollinator Plus manufactured by Landec Ag. Inc. contains a heat-unit activated polymer to predictably delay seed germination. The seed treatment shields the seed from moisture delaying germination until the polymer absorbs a preset amount of heat from the soil. Male seed treated with Pollinator Plus can achieve a wide pollination window using just one planting date (Hicks et al., 1996). Other chemicals have been used to accelerate germination and growth rate. Examples here include the use of plant hormones such as gibberellins and cytokinins.

f) Clipping or mowing has been effectively used to save a maize crop when inclement weather occurred after the first parent was planted but before the second parent was sown (Cloninger et al., 1974). It generally is not used to delay the female parent because significant seed yield reductions often result. It is best to mow when plants are about 25 to 30 cm tall, cutting away half of the plant material. An example of the delay that one
might expect from cutting based on different treatment times is shown in Table 9.

g) Flaming involves using gas burners to physically burn back the earlier maturing parent. It is recommended for use on the male rather than the female parent. It can effectively be used to delay pollen production 3 to 7 days. As a guide, it is best to flame plants when they are about 15 cm tall as the growing point remains below the soil surface. Flamer units should have two burners, one for each side of the row. The flame should be directed at the base of the plant. Gas pressure, ground speed, and height of the flamer should be adjusted to burn most of the exposed plant material. Burning back of vegetation may also be achieved using liquid nitrogen fertilizer or various herbicides. Caution need be applied when burning weak males that are poor pollen shedders (Thielen, 1986; Pinter, 1989).

h) Plastic sheets or bitumen used as a warming mulch. This could only be effectively used when planting in cool soils.

i) Increasing plant population density. Most practical in gaining a short delay in male flowering.

j) Water soaking seeds. May advance flowering 1-2 days.

k) Mechanically cutting back female shoots. Female flowering may be advanced 1-2 days by cutting 2.5-4.0 cm off the silk channel.

Table 9. The effect of cutting on flowering delay when applied at different growth stages.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Delay in Flowering</th>
</tr>
</thead>
<tbody>
<tr>
<td>20 cm cutback to 10 cm</td>
<td>4 days</td>
</tr>
<tr>
<td>60 cm cutback to 30 cm</td>
<td>7 days</td>
</tr>
<tr>
<td>90 cm cutback to 45 cm</td>
<td>7 days</td>
</tr>
</tbody>
</table>

Source: Cox, 1980.

Some of these treatments are impractical for use on a large scale and others generate problems because they supply a significant stress on the maize plants, particularly inbred lines. Shoultz (1985) surveyed U.S. seed producers regarding the parent delay techniques used in hybrid maize seed production. Table 10 summarizes the results of 70 U.S. seed producers when they were asked both what parental delay techniques they have used successfully and unsuccessfully. It is interesting to observe that the techniques reported as used most frequently were the same techniques other producers reported as being the most unsuccessful. Other notable results from the survey include the large number of hybrids in production that require some type of parental delay treatment and the common practice of delaying at least one half the male even if it appears that both parents would synchronize if planted at the same time.
Table 10. Survey response to the question “What techniques do you use in addition to delayed planting?"

<table>
<thead>
<tr>
<th>Techniques</th>
<th>% Companies Using</th>
<th>% Not Using Because Unsuccessful</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mowing</td>
<td>19</td>
<td>17</td>
</tr>
<tr>
<td>Adjusting planting depth</td>
<td>19</td>
<td>9</td>
</tr>
<tr>
<td>Adjust starter fertilizer</td>
<td>17</td>
<td>9</td>
</tr>
<tr>
<td>Flaming</td>
<td>11</td>
<td>4</td>
</tr>
<tr>
<td>Adjusting planting</td>
<td></td>
<td></td>
</tr>
<tr>
<td>population</td>
<td>2</td>
<td>1</td>
</tr>
<tr>
<td>Nitrogen burndown</td>
<td>2</td>
<td>1</td>
</tr>
<tr>
<td>Seed coating</td>
<td>1</td>
<td>3</td>
</tr>
<tr>
<td>Water soaking</td>
<td>1</td>
<td>0</td>
</tr>
</tbody>
</table>


At CIMMYT, we recently conducted a study examining flower synchronization techniques in highland hybrid maize production. Genetic material for this investigation included three highland inbred and three single-crosses. Techniques used to either delay or accelerate flowering included: a) foliar fertilizer applications (N, P, gibberellic acid, micro-nutrients), b) depth of planting, c) density of planting, d) cutting, and e) flaming. Table 4 shows the results of these treatments across the six genotypes tested. None of the foliar fertilizer treatments accelerated flowering as hoped (Table 11). Deeper planting depths resulted in 2-4 day flowering delays with minimal effect on grain yield. No significant differences were observed in flowering at the two plant densities (33,333 and 66,666 plants/ha) tested. The cutting treatments significantly delayed flowering from 3.5 to 14 days however plant development was stunted and yield reduced. Interestingly, the flaming treatment delayed flowering on average by 8 days with minimal negative effects on plant development and yield (Table 11). Greater changes in flowering were observed with inbreds as compared to hybrids with the flaming treatment (Table 12); inbreds were also more sensitive than hybrids to many of the other treatments (data not shown).
Table 11. The effect of different synchronization techniques on flowering and grain yield in six highland maize genotypes (Batan 1999).

<table>
<thead>
<tr>
<th>Pedigree</th>
<th>Grain Yield (t/ha)</th>
<th>Male Flower (days)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Foliar application of N (1 or 2 dosis)</td>
<td>0.24</td>
<td>0.0</td>
</tr>
<tr>
<td>Foliar application of P (1 or 2 dosis)</td>
<td>-0.34</td>
<td>0.0</td>
</tr>
<tr>
<td>Foliar application of gibberelic acid</td>
<td>-0.51</td>
<td>0.2</td>
</tr>
<tr>
<td>Foliar application of micro-nutrients</td>
<td>-2.13</td>
<td>1.5</td>
</tr>
<tr>
<td>Planting depth (5-7 cm)</td>
<td>-0.06</td>
<td>1.8</td>
</tr>
<tr>
<td>Planting depth (5-10 cm)</td>
<td>-0.33</td>
<td>3.6</td>
</tr>
<tr>
<td>Flaming</td>
<td>-0.46</td>
<td>8.0</td>
</tr>
<tr>
<td>Cutting (4 leaf cut at ground level)</td>
<td>-3.21</td>
<td>13.7</td>
</tr>
<tr>
<td>Cutting (4 leaf removing half plant)</td>
<td>-0.53</td>
<td>3.5</td>
</tr>
<tr>
<td>Cutting (8 leaf removing half plant)</td>
<td>-2.63</td>
<td>3.8</td>
</tr>
<tr>
<td>Density (33,333 - 66,666)</td>
<td>0.40</td>
<td>2.8</td>
</tr>
</tbody>
</table>

Source: Torres et al., 2002.

Table 12. The effect of flaming on flowering and grain yield of highland maize inbred lines and hybrids.

<table>
<thead>
<tr>
<th>Pedigree</th>
<th>Male Flowering (days)</th>
<th>Yield (t ha⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control</td>
<td>Flaming</td>
</tr>
<tr>
<td>CML 244</td>
<td>83</td>
<td>94</td>
</tr>
<tr>
<td>CML246</td>
<td>94</td>
<td>106</td>
</tr>
<tr>
<td>CML349</td>
<td>85</td>
<td>95</td>
</tr>
<tr>
<td>CML246 x CML242</td>
<td>82</td>
<td>90</td>
</tr>
<tr>
<td>CML244 x CML349</td>
<td>79</td>
<td>80</td>
</tr>
<tr>
<td>CML240 x IML8</td>
<td>86</td>
<td>89</td>
</tr>
<tr>
<td>Mean</td>
<td>85</td>
<td>92</td>
</tr>
</tbody>
</table>

Source: Torres et al., 2002.
VIII. Varietal Characterization

Accurate descriptions of morphological traits are important as a guide for maintenance and multiplication of inbreds, hybrids, and open-pollinated varieties (CIMMYT, 1999). They are essential for roguing and field inspections operations. Failure to develop appropriate varietal descriptions is often a source of conflict between breeders and seed producers. Breeders must recognize their responsibility to describe their released material in an accurate and timely fashion. It is important to avoid extremes where in some cases seed producers and inspectors do not have varietal descriptions (or ones that are so poor that they are of no value) and the other extreme where breeders are forced to provide highly detailed descriptions that require extensive amounts of time and resources to develop. Medium to large-scale companies typically have foundation seed departments which are responsible for inbred line descriptions.

How to

No easy standard formula exists on how to develop an appropriate varietal description. Standards chosen should not be too stringent, but rather realistic and appropriate for the conditions in a given country. Characteristics that may be included in a variety description are plant height, ear height, plant architecture, and resistance to insect and disease pests. Other possible descriptors are listed in Table 13.

The number of descriptors chosen and the number of observations taken per descriptor will depend on various factors including the uniformity of the germplasm, the uniqueness of the germplasm, the observation environment, and the resources available. One suggested example would be to describe 12-15 plant and ear traits evaluated in 2 environments, with 3 replications per environment, and 20 samples per replication.

Descriptors of quantitative characters should include expected standard deviations from the mean in order to indicate acceptable variation. Expected variation for qualitative characters should be given in percentages. Quantitative descriptors are generally preferred in the maintenance of the genotype and production of breeder seed, whereas qualitative descriptors are generally preferred for subsequent seed multiplication and for certification standards. Overall, qualitative descriptors are preferred because they tend to show less interaction with the environment.
Table 13. Characteristics that may be considered in a maize varietal description.

<table>
<thead>
<tr>
<th>Plant Parts</th>
<th>Qualitative</th>
<th>Quantitative</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Characteristics</td>
<td></td>
</tr>
<tr>
<td>Stem</td>
<td>Color</td>
<td>Height</td>
</tr>
<tr>
<td></td>
<td></td>
<td>No. of nodes</td>
</tr>
<tr>
<td></td>
<td></td>
<td>No. of tillers</td>
</tr>
<tr>
<td></td>
<td>Color of leaves</td>
<td>Total no. of leaves</td>
</tr>
<tr>
<td></td>
<td>Color of central vein</td>
<td>No. leaves above ear</td>
</tr>
<tr>
<td>Leaves</td>
<td>Color of leaf sheath</td>
<td>Leaf angle</td>
</tr>
<tr>
<td></td>
<td>Pubescence of sheath</td>
<td>Width ear leaf</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Length ear leaf</td>
</tr>
<tr>
<td></td>
<td>Color of glumes</td>
<td>Length of pedicle</td>
</tr>
<tr>
<td>Tassel</td>
<td>Color of anthers</td>
<td>Length of central axis</td>
</tr>
<tr>
<td></td>
<td>Compact or open</td>
<td>No. of branches</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Days 50% plants with pollen</td>
</tr>
<tr>
<td></td>
<td>Color of stigmas</td>
<td>No. per plant</td>
</tr>
<tr>
<td></td>
<td>Color of dry husks</td>
<td>Insertion angle</td>
</tr>
<tr>
<td></td>
<td>Husk pubescence</td>
<td>Length of ear peduncle</td>
</tr>
<tr>
<td>Ear</td>
<td>Husk texture</td>
<td>No. of kernel rows</td>
</tr>
<tr>
<td></td>
<td>Ear shape</td>
<td>Length</td>
</tr>
<tr>
<td></td>
<td>Kernel row arrangement</td>
<td>Diameter</td>
</tr>
<tr>
<td></td>
<td>Cob color</td>
<td>Weight</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Shelling percentage</td>
</tr>
<tr>
<td></td>
<td>Color of pericarp</td>
<td>Cob diameter</td>
</tr>
<tr>
<td></td>
<td>Color of aleurone</td>
<td>Length</td>
</tr>
<tr>
<td>Seed</td>
<td>Color of endosperm</td>
<td>Width</td>
</tr>
<tr>
<td></td>
<td>Texture (dent, flint)</td>
<td>Weight 1000 seeds</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Thickness of seed</td>
</tr>
</tbody>
</table>


**Biotechnology**

Biotechnology tools such as electrophoresis, restriction fragment length polymorphisms (RFLPs) and the polymerase chain reaction (PCR) can be used to characterize maize inbreds. Their principal advantage is that one can characterize material at the genome level. An additional advantage may be to avoid the G x E interaction common with use of morphological descriptions. These tools may also provide additional information such as genetic relationships and genetic diversity (Melchinger, 1999). The disadvantage of such tools lies in their cost and the associated complexity in conducting the laboratory analysis and interpreting the results (Smith and Register, 1998).
IX. Roguing

Roguing may be defined as careful and systematic evaluation of a seed production field and the removal of all undesirable plants. A plant is classified as a “rogue” if it is atypical—in other words any plant which does not conform to the varietal description. The goal of this operation is to assure the desired varietal, genetic and physical purity in the seed production field. It is the most effective and important method of contaminant removal because it both prevents contamination and eliminates existing contaminants. At the same time, it is a means of reducing the incidence of seed transmitted diseases and also eliminating weeds and other crop plants not controlled by cultivation or herbicides.

Although removal of 100% of offtypes is desirable, this goal is difficult to achieve. Planting parental seed with high levels of genetic purity will significantly reduce labor requirements for roguing.

Contaminates removed by roguing

Two major types of contamination that can occur in maize seed fields are genetic and mechanical contamination. Genetic contamination is caused by cross-pollination with compatible plants growing in the field or in the isolation area or by accidental self-pollination of female parents in hybrid blocks due to improper detasseling. Mechanical contamination is caused by mixing seed of other varieties.

The source of contaminants may include:

a) Genetic contamination of the crop in the previous seed production field
b) Mechanical mixture of undesirable seed in the prior production fields or in the seed lots.
c) Volunteer plants resulting from seed left by the prior crop.
d) Seed brought to the field by water, wind, birds, animals, people or agricultural equipment.

Selection of land to minimize roguing

Careful selection of land can prevent many problems and minimize the work of roguing. Consider the following when selecting land:

a) If possible, avoid planting into a field previously sown to maize.
b) Avoid planting into a field previously sown with a crop that easily could leave seeds or plant parts, capable of forming volunteer plants.
c) On sloped land the seed production field should not be located on a lower level than a field to be planted with a crop that could infest it.
or a field that is already infested with undesirable seeds.
d) Avoid land crossed by roads or animal trails.
e) Avoid land close to seed warehouses or other installations.

Procedures for roguing

To eliminate off-type plants a visual observation is made systematically and uniformly over all the plants in the field by the roguing team. When undesirable or doubtful plants are observed they are manually removed from the field.

For efficient roguing, the following recommendations should be observed:

a) Limit the roguing team to 10 to 12 people including the supervisor because larger groups tend to talk too much and get easily distracted. If more people are needed to rogue the fields, divide into several groups.
b) Select responsible people for the roguing team and provide proper training on the identification and removal of contaminant plants.
c) The team should start in a corner of the field and work through it slowly, walking parallel and in the same direction down the rows.
d) Each member of the roguing team should have a narrow zone to observe. For maize, consider a maximum of 2 rows.
e) Clearly mark areas of the field that have been rogued with large stakes or by other means.
f) Position of the sun and wind movement can affect the teams ability to identify undesirable plants.
g) After several hours of roguing, a person tires and becomes less efficient. Thus it is advisable to rogue relatively few hours per day. Since most off-type plants are easier to identify when the suns’ rays are at a low angle, roguing teams work most efficiently during the early morning or late afternoon hours.
h) The supervisor should concentrate on overseeing and inspecting the roguing teams activities rather than on actually doing the roguing.

When to rogue

Roguing should be conducted before genetic or physical contamination occurs and during times favorable for visual identification.

Some considerations during the plants life cycle include:

a) Post emergence - rogue volunteer plants that are often easily identified due to their difference in size and their position out of the rows.
b) Vegetative development - rogue off-type plants which deviate from
the given genotype in respect to root and stalk development, plant
type, pigmentation, leaf and stem pubescence, etc. Effective
roguing during this period will help reduce the work load during the
critical flowering period.
c) Flowering - in this phase, important agronomic and morphological
characteristics can be easily identified. This is the critical phase to
prevent genetic contamination of the crop.
d) Post flowering – roguing during the grain filling period makes
possible the removal of off-type plants distinguished by maturity,
color, drydown characteristics, disease reaction, etc.
e) Pre-Harvest -a final rogue prior to harvest should be completed to
eliminate diseased plants and those showing atypical vegetative and
reproductive characteristics.

X. Pollen Control

Detasseling

In an article entitled “Profitability through agronomic practices”, Ron
Thielen makes the following statements about maize detasseling:

- “Detasseling is the time of year that we all somewhat dread.”
- “During detasseling we all suffer from stress.”
- “I have always said that if we had to detassel 12 months of the year,
  I don’t think that you’d have anybody working in seed maize
  production.” (Thielen, 1986).

The detasseling period is usually the most critical and difficult to
manage period in hybrid maize seed production (Wych, 1988). To
achieve the necessary genetic purity standards all tassels from the
female parent rows must be removed prior to shedding and/or before
silk emergence. This will force cross-pollination between the male and
female rows in hybrid production. The detasseling operation involves
a physical removal of tassels either manually or in combination with
mechanical devices. Other options to eliminate male pollen from the
female rows include genetic and cytoplasmic male sterile systems,
chemical hybridizing agents, and some new techniques based on
biotechnology.

Duration of the detasseling period

The detasseling period usually lasts about two weeks but may range
from one to five or more weeks. The detasseling period may be
prolonged in fields which have delayed and non-uniform germination,
variation in soil fertility, waterlogging in early stages, significant pre-
flowering water stress, heavy insect infestation resulting in plant
stunting, and high incidence of foliar diseases.
**Potential complications**

The detasseling process may be complicated by various factors which require close attention (Wych, 1988). These factors include:

a) Tassels must be removed from all female plants before shedding and silk emergence.

b) Under favorable weather conditions for plant growth, fields must be worked daily, meaning 7-day workweeks, rain or shine.

c) Some female parent plant types are more easily detasseled than others. For example, the tassels of some female lines are physically hard to pull or have tassels which break easily. Other female parents whose tassels begin shedding pollen before fully emerging from the upper leaves, or which extrude silks at about the same time as pollen shed occurs, create difficult detasseling supervision and management problems.

d) Weather conditions can significantly aid or complicate the detasseling season. A windstorm or heavy rain can lodge and tangle the female parent just as tassels emerge, and make driving or walking through the field difficult. High temperatures can affect both the emergence of silks and tassels as well as the performance of the detasseling crew.

**Hand or manual detasseling**

At the onset of flowering (tassel and silk emergence) workers must begin a thorough and consistent inspection of the maize seed fields to remove tassels from the female rows. Detasseling teams should be limited to a maximum of 12-20 workers including a supervisor, because larger teams are more difficult to manage and are more easily distracted (Airy, 1955). The supervisor is responsible for recruiting, transporting, training, and managing the detasselers in his team. If more detasselers are needed, form several teams each headed by a supervisor. The detasseling team should work systematically through the field with each individual removing tassels preferably from only one female row at a time. Tassels should be pulled when they are well out of the "boot". This often occurs 1 or 2 days after the tassels are first visible. If the pulling is done prematurely, 1 or 2 leaves may be removed with the tassel, or the tassel may break off and not be completely removed. This is undesirable because loss of leaves will reduce seed yields and incomplete removal of tassels necessitates additional labor.

Hunter et al. (1973) in a study with 10 inbred lines found average yield reductions of 1.5, 4.9, and 13.5% when one, two, and three leaves respectively were removed with the tassel (Table 14). In a more recent study, Wilhelm et al. (1995) looked at the effect of leaf removal during
detasseling on yield, yield components, and seed quality in the inbred line N192. They found yield reductions of 3, 9, 14, and 24% when one, two, three, and four leaves were removed with the tassel (Table 15). Most of the yield decline was associated with a decrease in kernel number although seed size and grade out were affected. A decline in seed quality as measured by warm and cold germination tests was not observed.

Table 14. Mean yield changes of 10 inbreds following tassel and leaf removal.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Yield Change (vs. tassel)</th>
<th>Yield Change (vs. control)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tassel only</td>
<td>------</td>
<td>+ 6.9%</td>
</tr>
<tr>
<td>Tassel + 1 leaf</td>
<td>- 1.5 %</td>
<td>+ 5.8 %</td>
</tr>
<tr>
<td>Tassel + 2 leaves</td>
<td>- 4.9 %</td>
<td>+ 2.1 %</td>
</tr>
<tr>
<td>Tassel + 3 leaves</td>
<td>- 13.5 %</td>
<td>- 6.8 %</td>
</tr>
</tbody>
</table>


Table 15. The effect of leaf removal during detasseling on yield and yield components in the inbred line N192.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Grain Yield (Mg ha^{-1})</th>
<th>% Reduction</th>
<th>Kernel No.</th>
<th>Salable Seed ≠ Diameter between 6.35 and 10.32 mm</th>
<th>Germination</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>4.93</td>
<td>310</td>
<td>4.42</td>
<td>243</td>
<td>93.4 93.5</td>
</tr>
<tr>
<td>Tassel only</td>
<td>4.81</td>
<td>302</td>
<td>4.38</td>
<td>233</td>
<td>95.8 94.7</td>
</tr>
<tr>
<td>Tassel + 1 leaf</td>
<td>4.80</td>
<td>299</td>
<td>4.33</td>
<td>234</td>
<td>95.1 93.7</td>
</tr>
<tr>
<td>Tassel + 2 leaves</td>
<td>4.47</td>
<td>298</td>
<td>4.01</td>
<td>221</td>
<td>95.4 92.0</td>
</tr>
<tr>
<td>Tassel + 3 leaves</td>
<td>4.23</td>
<td>297</td>
<td>3.70</td>
<td>214</td>
<td>95.4 92.5</td>
</tr>
<tr>
<td>Tassel + 4 leaves</td>
<td>3.68</td>
<td>284</td>
<td>3.13</td>
<td>192</td>
<td>95.0 93.8</td>
</tr>
</tbody>
</table>

Standard Error 0.1

* 80,000 seeds
≠ Diameter between 6.35 and 10.32 mm


Workers should make sure that tassels are not left hanging on the maize plant because they may still be able to shed pollen resulting in contamination. When 90 to 95% of the tassels have been pulled, it is probably more economical to remove all of the tassels from the remaining plants at one time even though several leaves may be removed. It is important not to neglect to pull tassels on tillers or suckers.
Mechanical detasseling

The susceptibility of T-cms cytoplasm to race T of *Helminthosporium maydis* and the increasing labor costs and shortages in various countries has encouraged the development and use of mechanical detasslers. The major advantage of mechanical detasslers is their speed of operation (Table 16).

**Table 16. Detasseling time: mechanical vs. manual.**

<table>
<thead>
<tr>
<th>Method</th>
<th>Required Time</th>
<th>Distance Covered</th>
<th>Tassels Pulled</th>
</tr>
</thead>
<tbody>
<tr>
<td>Machine</td>
<td>6 min.</td>
<td>1/4 mile</td>
<td>88%</td>
</tr>
<tr>
<td>4 Detasslers</td>
<td>90 min.</td>
<td>1/4 mile</td>
<td>80%</td>
</tr>
</tbody>
</table>

Source: Cox, 1980.

Mechanical detasslers mounted on high clearance machines consist of two general types:

a) "Cutters" - a rotating blade or knife operates at various planes from horizontal to vertical, adjustable in height, to cut or shred the top of the maize plant including the tassel.

b) "Pullers" - usually 2 small wheels or rollers, adjustable in height, that rotate in opposite directions and grasp the tassel and upper leaves, pulling them upward in a manner comparable to a hand detasseling operation.

The efficiency of mechanical detasseling is affected by female parent morphology (leaf and tassel orientation), uniformity of female plant height and development, and the skill of the operator (Wych, 1988). Best results are achieved in uniform fields in which the tassel is well exerted ahead of pollen shedding. Under less favorable conditions, the percent of tassels removed per pass will decrease and leaf damage will increase. Normally, mechanical detasseling is delayed as long as possible prior to silk emergence to permit maximum exertion of tassels so that removal can be accomplished with minimum leaf damage. A skilled operator will remove 70% or more of the tassels in one pass with minimal leaf damage. Pullers are generally preferred to cutters because they typically inflict less damage to upper leaves resulting in higher seed yields. However, use of pullers may result in more genetic contamination as tassels removed with these devices sometimes end up in the leaf canopy where they can shed pollen. Some producers prefer to do a light cut first to help the tassels to exert above the upper leaves, followed by a pull. In all cases, hand detasseling is necessary to remove tassels remaining on missed, late, or short plants, or
suckers in the field.

In a comparison of hand vs. mechanical detasseling with common Corn Belt inbred lines, Cox showed that both yield and grade-out was superior in the hand detasseled treatments (Table 17).

**Table 17. Detasseling: Hand vs. machine**

<table>
<thead>
<tr>
<th>Method</th>
<th>Inbred</th>
<th>Yield (bu A⁻¹)</th>
<th>Harvest moisture</th>
<th>Test wt (lb)</th>
<th>Seeds/lb</th>
<th>MVK/acre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hand</td>
<td>Mo17Ht</td>
<td>50.5</td>
<td>24.1</td>
<td>59.0</td>
<td>1420</td>
<td>4040</td>
</tr>
<tr>
<td>Machine</td>
<td>Mo17Ht</td>
<td>47.2</td>
<td>23.5</td>
<td>58.0</td>
<td>1480</td>
<td>3920</td>
</tr>
<tr>
<td>Hand</td>
<td>B73</td>
<td>70.2</td>
<td>26.1</td>
<td>59.0</td>
<td>1860</td>
<td>7300</td>
</tr>
<tr>
<td>Machine</td>
<td>B73</td>
<td>67.0</td>
<td>25.1</td>
<td>58.0</td>
<td>1980</td>
<td>7440</td>
</tr>
<tr>
<td>Hand</td>
<td>A632</td>
<td>55.8</td>
<td>18.5</td>
<td>61.5</td>
<td>1760</td>
<td>5520</td>
</tr>
<tr>
<td>Machine</td>
<td>A632</td>
<td>44.0</td>
<td>18.2</td>
<td>61.0</td>
<td>1860</td>
<td>4580</td>
</tr>
</tbody>
</table>

Source: Cox, 1980.

*Pollen control with cytoplasmic male sterility*

In the U.S. in the 1950’s a cytoplasmic male sterility (*cms*) genetic system began to replace hand detasseling in hybrid maize seed production. This occurred for various reasons including the discovery of a stable type of *cms* with reliable fertility restoring genes, the perfection of techniques for utilizing these fertility regulating mechanisms, and the scarcity of labor and rising costs associated with detasseling (Poehlman, 1979). The most reliable form of *cms* was obtained from the maize variety, Mexican June. It was later identified as Texas-type cytoplasm or *cms*-T. Fertility was restored to inbreds with *cms*-T by two dominant fertility restoring genes, \( R_{f1} \) and \( R_{f2} \). Most U.S. inbreds were found to have the \( R_{f2} \) gene; thus only the \( R_{f1} \) gene needed to be crossed into such inbreds to convert them into fertility restoring lines. The \( R_{f1} \) and \( R_{f2} \) genes normally gave complete fertility restoration to *cms*-T, except under very adverse weather conditions such as very high temperatures and low humidity. However, in 1970 it was found that hybrids based on the *cms*-T system were highly susceptible to a strain of Southern Corn Leaf Blight (*Helminthosporium maydis*). Damage in the U.S. was extensive as approximately 90% of the hybrid maize at that time contained *cms*-T. After 1970, use of *cms*-T was discontinued and maize seed producers returned to their original practice of hand detasseling. However, researchers continued to develop other types of *cms*. Due to the cost-effectiveness of *cms*, increased use of other types (principally C and S types) commenced in the late 1970’s to the early 1980’s. Currently, the C and S types are being used on about 20 to 30% of the
maize seed acreage in the U.S. (Burris, personal communication, August, 2001).

The procedures for the use of \textit{cms} and fertility restoring systems in hybrid maize seed production will vary with the type of cross being made. For simplicity, in the following examples it is assumed that the inbreds entering into the hybrid will have either sterile (\textit{cms}) or normal (n) cytoplasm, with fertility restored by a dominant gene \textit{Rf}. In reality, maize plants with \textit{cms}-T require two fertility restoring genes, and the situation with \textit{cms}-S is more complex.

\textit{Cms} has been used to facilitate the crossing of two inbreds in the following ways:

a) \textit{Cms} conversion resulting in completely male sterile seed parents eliminating the need for detasseling.

b) Combinations of C- or S- cytoplasms in certain genetic backgrounds result in only partial male sterility. Anther exertion is delayed 1 to 10 days usually beginning after the tassel is fully extended above the leaves. Hand or mechanical detasseling can then be easily accomplished with minimal leaf removal.

Maintenance of inbred A-\textit{cms}:

\[
\begin{array}{c}
\text{A-}\text{-}\text{cms} \\
\text{\textit{rfrf}} \\
\text{male-sterile}
\end{array}
\times
\begin{array}{c}
\text{A-n} \\
\text{\textit{rfrf}} \\
\text{male-fertile}
\end{array}
\rightarrow
\begin{array}{c}
\text{A-}\text{-}\text{cms} \\
\text{\textit{rfrf}} \\
\text{male-sterile}
\end{array}
\]

The male-sterile line, A-\textit{cms}, is maintained by pollination from inbred A in normal cytoplasm. Neither inbred will have dominant restorer genes. The progeny will be male sterile since the cytoplasm is transmitted mainly through the female parent.

Production of the single cross, A x B, involves the following:

\[
\begin{array}{c}
\text{A-}\text{-}\text{cms} \\
\text{\textit{rfrf}} \\
\text{male-sterile}
\end{array}
\times
\begin{array}{c}
\text{B-n (or \textit{cms})} \\
\text{\textit{RfRf}} \\
\text{male-fertile}
\end{array}
\rightarrow
\begin{array}{c}
\text{AB-}\text{-}\text{cms} \\
\text{\textit{Rfrf}} \\
\text{male-fertile}
\end{array}
\]

Put male sterile cytoplasm in inbred A (the female) by backcrossing.
Inbred A should be of the non-restorer genotype (rf/rf). Inbred B (the male) should carry dominant restorer genes (Rf/Rf) in a normal or *cms* background. Plant blocks of female A-*cms* alternating with blocks of male inbred B and produce completely cross pollinated seed on inbred A-*cms* without detasseling.

It is a common practice to produce an identical hybrid by detasseling (without *cms*), and to mix or blend 25 to 50% of the fertile seed with 50 to 75% of the seed produced with *cms*. This reduces the risk if the restorer parent does not provide adequate pollen, and also to a potential disease epidemic such as occurred in 1970.

**Pollen control with chemical agents**

The use of chemically induced male sterility in the commercial production of hybrid maize seed is an alternative to the *cms*, hand and mechanical detasseling methods. Terminology used for such chemicals includes gametocide, male sterilant, pollen suppressant, and chemical hybridizing agent (CHA). The latter term is most commonly used in the current literature. The general procedure would be to apply a foliar spray to female rows prior to flowering to inhibit production of viable pollen. Another alternative is to develop chemicals which can be applied more easily as a seed treatment. Numerous chemicals have been studied including auxins, antiauxins, halogenated aliphatic acids, gibberellins, ethepon, arsenicals, etc. (McRae, 1985). Even coating tassels with liquid resins has been tried (Newlin, 1971). However, despite extensive research conducted to identify effective CHA successful commercial utilization in maize remains limited. One major problem in their use has been the failure to obtain complete pollen sterility, due principally to variations in response with different genotypes and in different environments. Other problems encountered include only temporary pollen control and associated damage to the female flower resulting in reduced seed production.

One product for maize called Detasselor™ is commercially available from ICI Seeds (Newhouse et al., 1994). Results from 34 production trials in the U.S. and France in 1991 showed that hybrid purity for two hybrids produced was similar between hand detasseled and chemical Detasselor™ treatments (Table 18). However, in production blocks yields of one hybrid were about 15% lower than the same hybrid produced using hand detasseling. Follow-up studies with additional lines showed that in some cases when the chemical is applied to some female lines there are minor yield reductions. However, with other lines yield reductions exceeding 20% were observed Table 19. Other challenges with this product include the narrow window of application time (3-5 days), its rainfastness, and its stability across environments. More recently, the manufacturers’ of Detasselor™ have provided
additional technical recommendations to overcome some of the difficulties with this product (Bayliss, 1996).

**Table 18. Results comparing hand detasseling (HDT) and Detasselor™ in maize hybrid seed production trials.**

<table>
<thead>
<tr>
<th>Hybrid Produced</th>
<th>Treatment</th>
<th># of Trials</th>
<th>Production Yields (bu/A)</th>
<th>Yield % of HDT</th>
<th>Purity 100%-selfs</th>
</tr>
</thead>
<tbody>
<tr>
<td>ICI 8344</td>
<td>HDT</td>
<td>3</td>
<td>67.7</td>
<td>--</td>
<td>100.0%</td>
</tr>
<tr>
<td></td>
<td>Detasselor</td>
<td>3</td>
<td>66.0</td>
<td>97.5</td>
<td>99.8%</td>
</tr>
<tr>
<td>ICI 8532</td>
<td>HDT</td>
<td>3</td>
<td>63.7</td>
<td>--</td>
<td>98.7%</td>
</tr>
<tr>
<td></td>
<td>Detasselor</td>
<td>3</td>
<td>54.3</td>
<td>85.2</td>
<td>98.8%</td>
</tr>
<tr>
<td>Mean across all trials</td>
<td>HDT</td>
<td>12</td>
<td>63.4</td>
<td>--</td>
<td>99.2%</td>
</tr>
<tr>
<td></td>
<td>Detasselor</td>
<td>12</td>
<td>60.5</td>
<td>95.4</td>
<td>99.5%</td>
</tr>
</tbody>
</table>

Source: Newhouse, et al. 1994

**Table 19. Mean yields from 1994 “Detasselor™” production trials.**

<table>
<thead>
<tr>
<th>No. Trials</th>
<th>HDT* Yield (bu A⁻¹)</th>
<th>Detasselor Yield (bu A⁻¹)</th>
<th>Detasselor Yield % of HDT</th>
</tr>
</thead>
<tbody>
<tr>
<td>Across females with acceptable yields (3 inbreds)</td>
<td>13</td>
<td>87.8</td>
<td>85.3</td>
</tr>
<tr>
<td>Across females with unacceptable yields (4 inbreds)</td>
<td>17</td>
<td>78.4</td>
<td>60.1</td>
</tr>
</tbody>
</table>

* Hand detasseling treatment
Source: Newhouse, et al. 1994

*Pollen control with biotechnology assisted methods*

The first commercially available maize male sterile system based on biotechnology assisted approach was the SeedLink™ system developed at Plant Genetic Systems in Belgium. The researchers used transformation technology to develop a MS3 event which consists of an
RNase nuclear gene called *barnase* and a glufosinate-tolerance gene called *bar* (Newhouse et al., 1996). In this event the RNase enzyme is expressed only in the anther cells where it destroys tapetal cells during anther development thereby preventing pollen formation. The nuclear *bar* gene confers tolerance to the herbicide glufosinate-ammonium or Liberty™. Treatment of maize plants at an early stage with the herbicide can be used to eliminate the 50% of the population which is male-fertile and herbicide sensitive. Plants that survive the Liberty™ application then comprise a uniform population of plants that are male-sterile (hemizygous) and glufosinate tolerant. Hybrid seed produced on such plants used as female seed parents, when pollinated by a fertile male, will produce 50% male-sterile/glufosinate-tolerant plants and 50% male-fertile/glufosinate-susceptible plants in the next generation.

This system requires over-planting the female rows as about 50% will be removed following the herbicide application. Timing and rate of application of the herbicide is critical with this technology to eliminate fertile females, avoid escapes and prevent drift onto the male rows. Sterility in numerous inbred backgrounds has been shown to be complete across multi-locations. Toxic effects reducing grain yield in resulting hybrids have not been found. Irregular fields due to the uneven distribution of herbicide sensitive female plants frequently occur with this system sometimes resulting in more large round and less flat seeds (Newhouse et al., 1996).

Another biotechnology based system in the developmental stage has been named Pioneer Constitutive Sterility (PCS) and involves a cloned male-sterile gene and an inducible chemical promoter. Meticulous work involving the Ac transposable element family and inverse-PCR was used to isolate a male sterile gene designated as *Ms*45 and its natural promoter (Albertson et al., 1993). These researchers than replaced the native promoter in the male fertility gene with a chemically-induced gene to accomplish two things. First, to “turn off” the male fertility gene since its natural promoter is not present. Second, to enable the gene to “turn on” when stimulated by the chemical application. The gene construct with inducible promoter is then transformed into an elite line. Through gene targeting manipulations, the native gene is replaced with the new construct rendering the female lines male sterile. The inducing chemical agent can be applied to stimulate pollen production for inbred increase although the timing of application in different inbred lines has proven to be tricky (Albertson et al., 2000). Hybrid seed produced using this system is fully fertile in the heterozygous condition. Two different chemically-inducible genetic systems are being developed; one originates from maize and uses an endogenous dicamba chemical inducible system. The second originates from the European Corn Borer (*Ostrinia nubilalis*) and involves a exogenously inducible
chemical system based on ecdysteroid chemistry (Albertson et al., 2000)

A second system termed Reversible Dominant Male Sterility (RDMS) is also under development. This system differs from PCS in that male sterility is conditioned by dominant gene action vs. recessive gene action in the PCS system. With RDMS, fertility is restored from the temporary negation of the effect of the expressed dominant gene (Albertson et al., 2000). Biotin or Vitamin H was selected as the sterility reversing chemical for this system.

Albertson et al. (1993) described various advantages of these systems including: a) Most if not all females should work unlike the conventional cms system where natural restorer genes limits the use of certain females, b) Constitutive male sterility is achieved since the female lines do not have their native promoter, c) Risk of system failure is in inbred increase fields, not in production fields, d) System failure is defined as a lack of fertility, not a lack of sterility, e) No restoring system is needed. Every male is a restoring genotype so no genetic modification of the male parent is necessary.

Assuming that one or more of the biotechnology assisted approached prove to be practical and economical, these systems can be expected to revolutionize current methods of pollen control.

XI. Harvesting

Appropriate harvest time

Harvesting may begin as early as when developing kernels approach physiological maturity which is often in the 30 to 38% moisture range with maize (Knittle and Burris, 1976). At physiological maturity seeds develop to complete functional maturity and express maximum quality potential. If possible, a prompt harvest of the maize seed crop after it reaches physiological maturity is recommended, as delays will unduly expose the seed to possible undesirable elements in the field including temperature extremes, rainfall, diseases and insects, bird damage, and theft (Figure 3 – see Annex). However, this is not practical under many situations due to the high handling and drying costs. Maize seed is commonly harvested with 15 to 25% moisture content.

Harvesting fully mature seed will result in maximum yield, improved appearance of seed, better seedling vigor in the subsequent crop, greater resistance to mechanical injury while handling in the conditioning plant, and reduced susceptibility to injury from high drying temperatures.
Hand harvesting

When hand harvesting consider the following:

a) Labor force available and cost.
b) How large is the area to be harvested along with estimates of the time required to complete the harvest.
c) That less injury to the seed will result than with mechanical harvesting.
d) That a selective harvest can be conducted.

Machine harvesting:

Important considerations when machine harvesting include:

a) Number of standing plants in the field.
b) Extent of uniform maturity in the field.
c) Overall field conditions.
In the U.S., it is now popular to use sweet-corn huskers to harvest maize seed. Traditional sweet corn harvesting units are designed to handle very high moisture ear corn in a gentle fashion so as to minimize mechanical damage (Burris, 1992). These objectives are closely aligned with that sought by the seed corn industry. The overall advantages of this system include more rapid harvests and less mechanical damage to the seed. Mechanical damage is decreased for several reasons including: a) the ears receive less damage to the basal region because the stalk is cut instead of removing the ear through friction, b) ears are harvested with complete husks, c) additional harvester parts are designed to gently handle seed. Other advantages of this equipment include speed and flexibility as the sweet corn harvesters are more aggressive in cutting the stalk below the ear and thus are able to function efficiently under a broader range of moisture conditions including mud and rain soaked fields. Disadvantages of the system include the initial start-up costs which can include modifications to processing plants and the additional transportation and handling costs (Larson, 1992).

Regardless of the method of harvest, prompt delivery of the maize seed in a carefully planned and coordinated fashion to the conditioning facility is highly recommended (Wych, 1988).

**XII. Reception, Husking, and Sorting**

Conditioning maize seed following harvest may take place in a mechanized processing facility, in a farmer’s backyard, or somewhere in between. Regardless of the scale and sophistication of the processing facility, the objective of conditioning is basically identical: to ensure the timely production of uniform, clean seed lots of healthy, viable, and genetically pure seed (Agrawal et al., 1998).

Maize seed delivered to a processing facility is first weighed typically followed by a sampling of the production as the ears are being deposited onto a conveyor mechanism to move them into the conditioning facility. It is critical to use gentle conveyor systems when moving seed both into and throughout the processing plant to limit damage.

Ears harvested either using the sweet corn equipment or with the older “two-step” husking system will arrive to processing facility with husks. It is important that the husking equipment is able to remove as much husks as possible without damaging the seed. Most husking units need to be adjusted for different seed parents and for hourly changes in moisture content of the seed (Jugenheimer, 1985). Removing a maximum amount of husks as possible will improve
drying capacity and efficiency, and facilitate the sorting, drying and shelling operations.

Following husking, the corn ears are sorted or rogued as they pass over a variable speed wide belt conveyor. In this operation, ears are visually examined to identify any off-type, diseased, or damaged ears. Practical suggestions for sorting include placing the sorting belt in a comfortable position, operating it at a reasonable speed, and allowing staff the flexibility to stop the flow if necessary. The sorting operation is an essential step in quality control and it is important that staff selected are well-trained and dedicated to this operation.

XIII. Drying

Maize seed must be dried in order to prepare it for storage and distribution including protecting seed from disease organisms. Unfortunately, the drying process has been a frequent cause of seed injury and significant economic loss (Burris, 1995).

The basic methods used for drying maize seed include: a) natural drying of seed on the cob prior to harvest while plants are still standing in the field, b) sun drying of seed on the cob following harvest, and c) artificial drying (Agrawal et al., 1998). Natural drying in the field is commonly used in regions characterized by subtropical and tropical climates and is particularly popular among small-scale seed producers who lack the resources to establish an artificial drying facility. The main advantage of natural field drying is the low cost. However, the method is risky as the seed may be exposed to numerous undesirable elements including temperature extremes, rainfall, pests, etc. Sun drying involves spreading cobs on a drying floor and exposing them to the sun for certain periods (Agrawal et al., 1998). Although more labor-intensive than natural field drying, this method represents an improvement over natural drying in that it allows the seed to be covered in the event of unexpected rainfall. However, sun drying is still subject to the vagaries of the weather and extended periods of cool, cloudy, or humid weather can significantly impede the drying process. Artificial drying of seed is the most reliable and precise method but involves significant costs in both the development and operation of the drying facility.

Seed drying involves first the transfer of moisture from the seed surface to the air around the seed and then second the movement of moisture from inside the seed to the seed surface. This is accomplished by raising the temperature and lowering the relative humidity of air and passing it through the seed mass. The heat from the air is transferred to the seed increasing the vapor pressure in the seed. Due to vapor pressure differences between the seed and air, moisture is forced out of the seed and carried away by the air. If the temperature of the air is very high, the drying process may be too
violent resulting in damage to the seed. Too low temperatures will delay the drying process often resulting in damage caused by diseases, insects, and excessive respiration. Sufficient airflow through the seed mass is essential to complete the drying operation.

Sun dryers

Sun dryers may be a good alternative to dry maize seed. In a study conducted in Guatemala with the maize variety ICTA B-5, researchers compared drying rates over five days between a sun dryer and seed dried either on concrete patios or on black plastic. Results showed that the sun dryer reduced seed moisture content by 7.7%, whereas on a concrete patio and black plastic it was reduced by 11% (Cordova et al., 1999). Percent germination remained at 90-96% with all three treatments.

Drying temperatures

Ideal corn seed drying temperatures will vary depending on the genotype, harvest moisture and environmental conditions but typically range between 35° and 45° C. The higher the initial moisture content, the more susceptible the seed is to drying damage. Therefore it is recommended that corn seed with 35 to 40% moisture be initially dried at temperatures less than 40° C. When moisture content is lowered to 20% or less, drying temperatures may be increased up to 45° C to complete drying to the desired seed moisture content of 12 to 13%.

Artificial drying systems

Three types of artificial drying systems are in current use with maize seed. These include: a) single-pass, b) single-pass reversing, and c) double-pass (Misra, 1995). With the single-pass system, heated air is passed through the wet seed in one direction and then is exhausted (Figure 4.). This is the simplest to manage but the least efficient of the three systems. The single-pass method requires use of the maximum heat and airflow permissible in order to dry all the seed in the bin. Higher airflow rates help increase the drying zone width ideally extending it completely through the seed bed. However, what frequently happens is that the bottom seed layers close to the heat source are over-dried and the upper layers are under dried.
Figure 4. Schematic of a single pass drying system.

The deficiencies of this system have been overcome with the single-pass reverse drying method. Here the moisture gradient problem is alleviated by changing the direction of the air every 12 to 24 hours facilitating more uniform drying. Another advantage of the single-pass reverse drying system is that it allows the flexibility of using different drying temperatures for each drying bin. This is useful for reducing drying time and maintaining better seed quality particularly for more heat sensitive hybrids.

The double-pass drying system is more complicated and includes both a hot-air and transfer tunnel (Figure 5.). Air from the hot air tunnel (40° to 45° degrees C) is forced through bins containing relatively dry ear corn to finish the drying process. The exhaust air which has likely cooled to about 35° to 40° C, is then passed through bins with high moisture seeds. Because relatively cooler air is less damaging to wet seeds, the physiological quality of the seed is better protected. The energy efficiency of this system is often superior to both of the single-pass methods. However, management of the double-pass drying system can be rather complex (Primus, 1995).
Regardless of the drying system used it is critical to regularly monitor drying temperatures, relative humidity, air flow rates, and moisture content of seeds in different parts of the bins (O’Leary, 1995). Additional considerations include taking precaution to properly fill the bins while limiting impact damage of the seed upon entrance into the bin. Finally, remember that corn inbreds have a range of sensitivities to drying temperature. Burris and Navratil (1980) found significant differences in the effect of drying temperatures on the warm and cold germination of a heat sensitive (Mo17) and more tolerant (A632) inbred line (Tables 20 & 21).
Table 20. The effect of drying temperature at different moisture contents on cold test emergence percentage of seed grown from different inbred parents.

<table>
<thead>
<tr>
<th>Quality Measurement</th>
<th>Inbred Seed Parents*</th>
<th>A632</th>
<th></th>
<th></th>
<th></th>
<th>Harvest Moist %</th>
<th>Drying Temp. (° C)</th>
<th>Harvest Moist %</th>
<th>Drying Temp. (° C)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>50</td>
<td>45</td>
<td>40</td>
<td>35</td>
<td>50</td>
<td>45</td>
<td>40</td>
<td>35</td>
</tr>
<tr>
<td>Cold Test Emergence (%)</td>
<td></td>
<td>40</td>
<td>26</td>
<td>18</td>
<td></td>
<td>36</td>
<td>17</td>
<td>17</td>
<td>17</td>
</tr>
<tr>
<td></td>
<td></td>
<td>42</td>
<td>63</td>
<td>99</td>
<td></td>
<td>34</td>
<td>61</td>
<td>61</td>
<td>61</td>
</tr>
<tr>
<td></td>
<td></td>
<td>87</td>
<td>95</td>
<td>99</td>
<td></td>
<td>29</td>
<td>84</td>
<td>84</td>
<td>84</td>
</tr>
<tr>
<td></td>
<td></td>
<td>96</td>
<td>99</td>
<td>99</td>
<td></td>
<td>25</td>
<td>89</td>
<td>89</td>
<td>89</td>
</tr>
<tr>
<td></td>
<td></td>
<td>97</td>
<td>99</td>
<td>99</td>
<td></td>
<td>71</td>
<td>98</td>
<td>98</td>
<td>98</td>
</tr>
<tr>
<td></td>
<td></td>
<td>98</td>
<td>98</td>
<td>98</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*L.S.D. 0.05 = 9 and 13 for A632 and Mo17, respectively.
Source: Burris and Navratil, 1980.

Table 21. The effect of drying temperature at different moisture contents on germination percentage of seed grown from different inbred parents.

<table>
<thead>
<tr>
<th>Quality Measurement</th>
<th>Inbred Seed Parents*</th>
<th>A632</th>
<th></th>
<th></th>
<th></th>
<th>Harvest Moist %</th>
<th>Drying Temp. (° C)</th>
<th>Harvest Moist %</th>
<th>Drying Temp. (° C)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>50</td>
<td>45</td>
<td>40</td>
<td>35</td>
<td>50</td>
<td>45</td>
<td>40</td>
<td>35</td>
</tr>
<tr>
<td>Germination (%)</td>
<td></td>
<td>52</td>
<td>40</td>
<td>32</td>
<td></td>
<td>49</td>
<td>49</td>
<td>49</td>
<td>49</td>
</tr>
<tr>
<td></td>
<td></td>
<td>80</td>
<td>96</td>
<td>72</td>
<td></td>
<td>49</td>
<td>95</td>
<td>95</td>
<td>95</td>
</tr>
<tr>
<td></td>
<td></td>
<td>99</td>
<td>98</td>
<td>99</td>
<td></td>
<td>49</td>
<td>95</td>
<td>95</td>
<td>95</td>
</tr>
<tr>
<td></td>
<td></td>
<td>97</td>
<td>99</td>
<td>99</td>
<td></td>
<td>49</td>
<td>95</td>
<td>95</td>
<td>95</td>
</tr>
<tr>
<td></td>
<td></td>
<td>98</td>
<td>99</td>
<td>99</td>
<td></td>
<td>49</td>
<td>95</td>
<td>95</td>
<td>95</td>
</tr>
</tbody>
</table>

*L.S.D. 0.05 = 9 for A632 and Mo17.
Source: Burris and Navratil, 1980.

XIV. Shelling, Cleaning and Sizing

Shelling

Following the drying of maize ears, seeds must be removed from the cob by a process called shelling. Although this may be done by hand, mechanical means are often used to speed up the process. However, mechanical shelling presents a great risk particularly for physical damage to the seed.
The two most widely used types of maize shellers are the cylinder and the cone. Both types rely on compression, rubbing, and twisting to remove seed from the cob (Johnston, 1996). The specific design of the sheller including the base material and presence of smoothed parts and rubber coatings can have a big influence on the success of the operation (Jugenheimer, 1985; Stanfield, 1996). Additionally, slowing sheller shaft speeds to 250 to 300 rpm and keeping the sheller full including maintaining a consistent flow of ears into the device and seed out, should help reduce mechanical damage. Airy (1955) reported that by slowing a cylinder sheller down from 720 to 240 rpm, cold test germination results in a seed lot increased from 50 to 74% with untreated seed, and from 64 to 79% in treated seed. Slowing the sheller down may have the added benefit of placing maize ears more in contact with each other rather than the typically harder sheller parts resulting in a more gently rubbing action and less mechanical damage. Ideal moisture content of ear maize for efficient shelling is between 14 and 15%, although for practical reasons (largely to dry seed to a safe storage moisture content) seed is often dried and shelled at about 12% moisture content. Inbred line differences for shelling efficiency and damage have been observed. Ideally, one would hope to limit damage to 1-2% or less of the seed.

Cleaning

Seed maize leaving the sheller often contains varying amounts of foreign material including bits of cob, husk, broken kernels, stones, dirt, weed seeds, insect larvae, etc. This debris must be removed to improve seed appearance, and to promote good storability and plantability. This may be done manually through a laborious separation operation or more efficiently though the use of hand-held screens. In larger seed operations, mechanical air screen cleaners or scalperators are commonly used. Air screen machines convey shelled maize to a series of sloping shaker screens. Large debris including oversized kernels, broken pieces of cob, stones, etc. are removed as they pass over the first screen while the good seed passes through. Subsequent screens can be used to separate out different sized kernels, while the final screen is used to remove very small seed including weed seed. Alternatively, scalperators may be used to clean maize seed. These devices consist of rotating wire mesh drums into which unclean seed is fed. Desirable kernels pass through the mesh leaving behind unwanted large debris. The final cleaning step with both the scalperator and air screen machine is to pass through an air blast that lifts away fines, small cob pieces, dirt and dust which may have escaped the earlier screening action.
**Sizing**

One of the unique aspects of seeds on a maize ear is their dramatic difference in size and shape due largely to their position on the ear. Large round seeds are often found at the base of the ear and small rounds at the tip. About 75% of the seeds in between these round types are flattened as a result of their tightly packed position. These flats typically range in size from small to large. Historically, these seed size differences were recognized by the seed maize industry, which marketed according to uniform sizes so that they would fit specific plates found in maize planters. Today, “plateless” precision air planters, which do not require uniform size seed, are in common use. However, the sizing of maize seed is still a common practice.

Wych (1988) described in detail the process and equipment used in sizing maize seed. Briefly, maize seed is sized based on width, thickness, and sometimes length through the use of a series of flat or cylindrical metal screens which possess either round or oblong holes. Thickness separation is achieved using slotted openings, while width separations are achieved using round perforations. Seed maize may be separated into a large number of size/shape categories depending on the processing plants capability and demands of the farmer. However, all size/shape categories will fall into one of six including large rounds, large flats, medium rounds, medium flats, small rounds, and small flats.

**Cleaning sized seed**

A final mechanical cleaning operation may be conducted through the use of a specific gravity separator. This device functions on the principle that heavier seed is of higher quality than light seed which is often damaged or diseased. The gravity table consists of a rough perforated deck through which air is introduced on the lower side and seed placed on the upper side. Maize seed on the deck are typically from three to seven layers deep. Air flow is adjusted to stratify the seeds on the table surface with the light seeds floating on top and the heavy seeds on the bottom in contact with the deck. The deck is inclined and made to vibrate by an eccentric gear. The heavy seeds in contact with the table surface are moved upward with the motion of the deck while the floating light seeds are essentially unaffected. However, the light seeds are affected by the downward incline of the deck and commonly migrate off the low side. Numerous adjustments are necessary with a gravity table including airflow, speed, feed, slopes and dividers to separate the seed categories (Misra, 1994). These are precision machines which can do a fine separation operation but require skilled operators.
Shelling, cleaning, and sizing maize seed are necessary to prepare a uniform, high-quality product, with satisfactory appearance, and one that is easily planted by the farmer using his existing equipment. Airy (1955) reported that typically only about 65 – 75% of the maize seed crop delivered from the field to the processing plant is of salable seed size and quality. He estimated that about 15% of the original crop is lost in sorting, handling, and drying, with an additional 15% lost in the cleaning and sizing operations.

XV. Treating Seed

Much of the hybrid maize seed sold worldwide is treated with a fungicide or a combination fungicide/insecticide. The purpose of this treatment is to protect the seed and developing seedling from diseases and to give short-term protection against storage insects. Fungicides are particularly helpful when sowing in conditions where soils are heavy, crusted, and/or cold and wet. Some of these conditions are commonly associated with the increased use of conservation tillage practices. Seed treatment chemicals may also help offset vulnerability to disease of mechanically chipped or cracked seeds occurring during the harvest and/or conditioning operations.

The fungicides and insecticides are commonly mixed into a liquid slurry and metered onto the seed using a seed treator device. Dyes are often added to the slurry to impart a distinctive color indicating that the treatment has been added. A uniform coverage of the seed along with safety precautions are critical in the treatment operation.

Fungicides

The most common fungicide used today in treating hybrid maize seed is Maxim XL™. This product contains Maxim (active ingredient – fludioxonil) and Apron XL™ (active ingredient – metalaxyl) to give broad spectrum fungicidal activity against numerous seed-borne and soil-borne pathogens. Additional advantages of these fungicides is that they are applied at very low rates thus reducing the dust-off problem. Other fungicides still in use for maize seed treatment include Captan and Thiram.

Insecticides

Insecticides frequently applied to commercial maize seed include several new compounds labeled as Gaucho™ or Prescribe™, Cruiser™, and ForceSt™. Gaucho™ (a.i. imidacloprid) is a relatively new chemical that has shown to be quite toxic to important maize insect pests but relatively benign to other organisms and the environment (Rushing, 1993; Knake, 2001). Cruiser™ (a.i. thiamethoxam) is a systemic insecticide which controls a broad range
of seed, soil, and foliar chewing and sucking insects. ForceST™ has been shown to give effective control of important below ground maize insect pests (Wall, 2001).

Seed coatings

Although more commonly used with horticultural crops, increasing interest has been shown in the use of seed enhancement agents with hybrid maize seed (Burris, 1992, 1994). Seeds may be coated for various reasons including: a) to promote more uniform and vigorous germination particularly under stressful planting conditions, b) to obtain better accuracy and efficacy of pesticides especially with many of the new compounds which are applied in very low dosages, c) to achieve better flowability and plantability by adjusting seed size and shape, d) for safety reasons such as to reduce dust-off, and e) for environmental reasons (Tyron, 1993; Tyron, 1994a,b). Additional benefits mentioned in a previous section are to allow simultaneous planting of both parental lines in maize hybrid production when they differ in flowering maturity by applying a plastic polymer to the earlier parent. Seed enhancement agents have been used with sweet corn but their commercial use with field maize production remains limited.

XVI. Bagging

The final step in the conditioning process is the packaging of seed. A seed package accomplishes several essential functions including; a) serving as a convenient unit for handling, transport, and storage, b) protecting seed against contamination and mechanical damage, c) providing a suitable environment for storage, d) providing a barrier against seed loss and escape of pesticides, and e) serving as a sales promoter (Thomson, 1979).

Seed packaging material may consist of cloth, jute, plastic, paper, metal or various combinations (Warham, 1986). Each material has characteristics that make it suitable for a particular type of package and use. Plastic, paper, or plastic/paper combinations are the materials of choice for packaging maize seed. In the U.S. maize hybrid seed is commonly packaged in multi-ply paper bags with a polyethylene plastic lining to protect against external moisture. Plastic bags are preferred in some areas because farmers can see the seed. However, use of plastic bags may be risky if seed is exposed to the sun and warm temperatures resulting in accelerated respiration and likely seed deterioration (Cordova et al., 1999). Seed may be packed by hand or through the use of semi-automatic or automatic equipment. This process starts with a specific amount of seed being weighed out. Then the bag is hung and filled, the bag is sewn or heat sealed, and a tag is attached. The tag commonly includes information about the variety, seed lot number, the physical and genetic purity, germination test
results, and the seed treatment chemicals used. Maize seed package size varies depending on the farmers requirements normally ranging from as little as one to as large as 25 kilos.

**XVII. Storage**

As agricultural production is seasonal in nature, storage of seed is inevitable. Maize seed harvested at the end of one cropping cycle will be stored at least until the beginning of the next planting season, a period that can range from a few days up to several months. In some instances, longer term seed storage may be necessary. For example, in cases of overproduction or to guard against possible shortages or with parental seed of inbreds and single crosses to most efficiently utilize supply produced and better maintain genetic purity (Agrawal et al., 1998).

*Bulk and bagged storage*

Maize seed is typically stored either in bulk or bagged. Seed conditioning facilities commonly possess some bulk storage capacity, but this often proves inadequate when large quantities of seed must be stored for extended periods. Thus, seed is commonly bagged which has the added benefit of ease of movement.

The principal objective in seed storage is to maintain seed quality which unfortunately is quite susceptible to deterioration. Ideal conditions to accomplish this are dry, cool locations which are relatively free of diseases, storage insects, and rodents.

*Storage temperature and relative humidity*

Seed moisture content and temperature are the two most important factors influencing seed storability (Delouche, 1973). Seed biological activity increase as seed moisture content and temperature increase. Similarly, the growth and reproduction of undesirable seed molds and storage insects are increased under higher moisture and temperature conditions. Both temperature and moisture strongly interact to affect seed quality, although moisture is the most critical factor.

As seeds have the ability to take up or give off moisture, it is essential to store them in environments with low relative humidity. Many years ago, Harrington (1959), proposed a “rule of thumb” which stated that for proper seed storage the relative humidity percentage and the storage temperature in degrees F should add up to no more than 100. Examples would include storage conditions of 50% RH and 50º F or 60% RH and 40º F, etc. Such conditions would be quite suitable to maintain maize seed quality in storage for a period of one year or more.
In tropical environments it is quite difficult to comply with Harrington’s rule of thumb and the achievement of a 60% RH with 60° F is more practical and should be sufficient to store maize seed for 4-6 months.

Numerous studies have been conducted confirming the important influence of storage temperature and moisture on maize seed germination (Abba and Lovato, 1999; Airy, 1955; Sayre, 1948). Interesting differences in the storage life of maize parental lines have also been observed dramatically illustrated in the comparison between WF9 and H55 (Figure 6.) (Delouche, 1973).

![Figure 6. Differences in longevity of seed of two inbred lines of corn and the single cross hybrid under conditions of 86° F and 75% relative humidity. The seed were produced at the same time and the same place.](image-url)
Storage cleanliness and organization

Finally, cleanliness and good organization are essential for proper maize seed storage. Unnecessary debris can often harbor disease and insect pests and should be removed. Use of wooden pallets can have various benefits including facilitating the movement of bagged seed with use of fork-lift trucks, raising seed off of concrete floors through which moisture can be absorbed, and lowering stacking height thus reducing compression pressure on seed. Properly labeled bags and seed lots, good inventory control, plus organizing the seed lots based on popularity in demand and observing the first in, first out rule will help in achieving an efficient seed storage operation (Boyd, 1976).

XVIII. Quality Assurance

As this article demonstrates, maize seed production involves many different operations which require intensive management, meticulous planning, impeccable timing, a high level of technical expertise, and considerable attention to detail. Most seed enterprises have a system of internal quality control to monitor and help coordinate all aspects of maize seed production. In small companies, the seed production manager may double as his own quality control expert, whereas large companies have a separate department whose sole function is quality control.

Procedures and standards

The quality assurance (QA) program should have well-defined procedures and standards that are understood and applied by all levels of management. These procedures and standards are often similar or more strict than those outlined in Rules and Testing of Seeds (AOSA, 1993), the AOSCA Certification Handbook (AOSCA, 1992), and the International Rules of Seed Testing (ISTA, 1995).

Quality control laboratories

The quality control laboratory is the hub of an effective quality assurance program (Grabe, 1980). However, it should be emphasized that the best quality control program is an effective production program. The QA department should link closely with essentially all major divisions of the company including research, production, processing, marketing, agronomic services, etc. (Figure 7.).
Field Inspections

High quality maize seed begins in the field with the selection and training of contract growers. QA inspectors should work with contract growers and field production staff during numerous aspects of the cropping season. These inspectors should also conduct detailed evaluations based on a series of standards previously mentioned. Inspection counts should follow a random but pre-planned pattern that is both representative and well distributed throughout the field. The goal is to get maximum coverage of a field while walking a minimum distance (Figure 8.). The number of samples taken will depend on the size of the field and by the maximum tolerance levels accepted. The larger the field and the stricter the tolerance levels accepted, the more samples need be taken.
Figure 8. Possible walking and sampling patterns of maize seed fields.

The initial inspection should verify that the seed field:

a) Was sown from an approved seed source
b) Meets the prescribed land requirements as to the previous crop.
c) Is in compliance with prescribed isolation standards and number of border rows.
d) Is planted with the prescribed ratios of female and male parents.

Additionally, the inspector should look for potential problem areas such as low spots, weedy areas, and off-type plants.

In the subsequent inspection prior to flowering the inspector should verify that the seed field is properly rogued and that the plant material is true to the varietal characterization. Observations on isolation or border problems, growth problems, and any unusual circumstances should be made. At this time, the inspector should carefully examine
crop growth to plan his/her next visit during the critical flowering period.

The flowering inspection should begin when there are 1-2% receptive silks in the female parent. During flowering, the hybrid seed fields should be inspected periodically as many times as deemed necessary and as resources permit. With hybrid seed fields, observations should be taken on male and female rows separately. It is recommended that several samples of from 10-50 plants be taken in different parts of the field. For the male and female rows, counts should be taken for plants shedding pollen, off-types, and off-types shedding pollen. Additionally for the females, counts will include the number of plants with receptive silks. At this time it is essential to aid the seed producer in properly detasseling the female rows, and eliminating off-type plants particularly from the male rows. Any isolation problems should also be noted and dealt with.

A final inspection prior to or during harvest can be made to identify off-types, diseased plants, and to assure that the crop is harvested at the proper time while avoiding mixtures between the male and female parents.

It is essential that during all field visits, the inspector accurately record his observations on standard forms. These forms may include the following information:

- Field location, number, etc.
- Grower
- Area of field inspected
- Planting dates
- Report number (no. of inspection)
- Type of seed (basic, certified, etc.)
- Seed source
- Planting rate
- Previous crop
- Isolation distance
- Border row status
- Field counts
- Comments regarding reported observations compared with prescribed standards for acceptance or rejection of the field.

*Post-harvest inspections*

Inspectors typically are involved in the harvest operation to help ensure timely harvest with no mechanical mixtures. They also should monitor various post-harvest processes including the green sort and drying operations. At different stages of the seed conditioning operation,
representative samples based on standard seed testing rules should be taken for submission to the quality control laboratory for analysis.

Seed quality tests

A range of quality tests may be conducted with these samples to evaluate the physical quality, genetic purity, viability, and vigor.

Moisture content determination

Since moisture content has such a large influence on the life and quality of the seed, the initial quality control test is often simply an evaluation of moisture content. This is typically followed by a physical purity analysis which examines the amount and type of damaged or off-type kernels along with the quantity of inert matter and weed seeds present in the sample.

Genetic purity analysis

A genetic purity analysis is particularly important with breeder and foundation seed. The conventional approach to monitoring genetic purity has been through “grow-out” tests. These tests involve planting out a sample of the seed lot in the greenhouse, winter-nursery, and/or summer season field to evaluate plant development in comparison to the varietal description. Disadvantages of this system include time, and the fact that the genetic material may be evaluated in an environment where it is not well adapted resulting in greater difficulties in identifying off-type vs. true-to-type plants. Today, sophisticated laboratory techniques such as starch gel electrophoresis (Smith and Weissinger, 1984) are available to conduct purity analysis. These techniques have the advantages of increased precision and speed, but the disadvantages of cost and complexity. Biochemical isozyme analysis remains as the most useful and cost effective method for genetic purity analysis despite the availability of newer DNA-based technology (Smith and Register, 1998). Several companies such as BioDiagnostics (www.biodiagnostics.net), Biogenetic Services, Inc. (www.biogeneticservices.com), and the Indiana Crop Improvement Association (www.indianacrop.org) offer electrophoresis isozyme purity tests in the U.S.

Germination test

The most common seed quality test is the germination test which measures seed viability under ideal conditions. For a maize seed lot, 4 reps of 100 seeds each are sown either in sand or a paper substrate and placed under adequate moisture conditions and either at 25° C (with 12 h light/day) or 20° and 30° C alternating. The number of normal and abnormal seedlings, and ungerminated seeds are determined at 4 and 7 days following the initiation of the test.
Vigor tests

Vigor tests have been developed to evaluate the ability of a seed lot to emerge in a timely, uniform, and adequate fashion under a range of field conditions. The most common vigor evaluation conducted by the U.S. maize seed industry is the cold test (Ferguson, 1990). The cold test is a method of simulating adverse field conditions such as when temperatures are low and soils are wet. However, one major problem with the cold test is that a standardized method has yet to be developed (TeKrony and Woltz, 1997). The AOSA (1983) and ISTA (1995) suggested two methods to conduct the cold test which include the rolled towel and tray cold test. Briefly, both methods involve placing maize seed in a moist sand:soil mixture (1:1) and exposing them to a cool 10°C temperature for 7 days. The samples are then placed in a 25°C chamber and evaluated after 4 and 7 days in a similar fashion to the germination test. More recent modifications of this technique include placing the maize seeds in a water saturated environment for the duration of the 7 day 10°C treatment (Martin et al., 1988). Various studies have reported variability in methodology and results among seed testing laboratories conducting the cold test (Burris and Navratil, 1980; Nijenstein, 1995; TeKrony, 1987). Recently, TeKrony and Woltz (1997) concluded that the major variables influencing cold test germination results such as soil moisture, temperature, seed treatment, and oxygen supply can be controlled in the laboratory which should aid in uniformity and standardization of this important test. However, standardizing the type and amount of pathogens in the soils used for this test is more difficult.

In warmer, more tropical climates the accelerated aging test has been used as an indication of seed vigor. The conditions for this test are to expose maize seed to 42°C (108°F) and 100% RH for 96 hours, followed by a 7 day treatment at 25°C and evaluation similar to the germination test. As with the cold test, the accelerated aging test has suffered from problems of standardization and variability in results among seed labs (Santipracha et al., 1997).

Numerous indirect tests of seed vigor have been developed; the most widely used with maize is the tetrazolium test. The tetrazolium test is a biochemical test which differentiates the living and dead tissues of a seed by the presence or absence of a red stain known as formazan (Pili-Sevilla, 1987). Enzymatic activity in living seed tissues turns the colorless tetrazolium red while loss of enzymatic activity in dead tissue leaves the tetrazolium colorless. The main advantage of this test is speed as the evaluation is typically completed in about 24 hours. Additional advantages include its use to identify maize seed with internal damage caused by insects, frost, or other factors (Iowa State Seed Science Center, 1997; Bennett and Loomis, 1949; Moore, 1958). The limitations of this test is that it is labor intensive and requires
staff with specialized training and experience (Pili-Sevilla, 1987).

Issues of health, safety, and justice are much in the news today in discussions about globalization, economic opportunity, and new science such as biotechnology. As biotechnology and world trade are having a major impact on the maize seed industry, it has become increasingly necessary to closely monitor the quality of these new genetic products to help ensure they meet the customers needs while satisfying the public at large. Quality control tests to evaluate seed health have been available for decades while tests for the presence of new genes in genetically modified organisms (GMO) have been developed more recently.

Seed health tests

To reduce the possibility of spreading harmful pathogens via seeds to other countries and areas, a series of seed health tests have been developed and implemented. These tests are commonly of four kinds including: a) a physical examination of the internal and external seed parts including use of a microscope to identify the presence of pathogens, b) plating seeds on agar followed by identification of the emergent organisms, c) germinating seeds and growing the seedlings in conditions known to encourage the development of diagnostic symptoms (blotter or incubation test), and d) washing seed samples with distilled water followed by centrifugation and observation under a microscope (wash test) (Warham, 1985). A comprehensive guide to maize diseases including information on the distribution and economic importance of the pathogens, pathogen transmission, control practices, effect on seed quality, seed treatments, and seed health tests has been published by McGee (1988).

Seed tests for genetically modified organisms (GMO)

Goggi (2000) described three categories of seed tests for GMO including bioassay, immunoassay and molecular genetic screening. The bioassay tests are commonly used in herbicide tolerance screening and involve exposing plant material to these chemicals and evaluating the degree of damage inflicted. Specific techniques include: a) soaking seeds in a dilute herbicide/water solution prior to planting, b) growing seedlings in media moistened with a low-concentration herbicide, and c) growing seedlings to different stages of development and then spraying them with herbicide.

The immunoassay tests determine the presence or absence of a gene product (typically a protein) using serological techniques such as the enzyme-linked immunosorbent assay or ELISA. With this technique, an antibody and substrate are brought in contact with ground plant tissue in a liquid buffer solution placed in plastic micro-well plates or
other containers. If the new gene is present a chemical reaction occurs resulting in the production of a distinct color or bands (Goggi, 2000). Commercial ELISA kits are now available for identification of the Bt and Round-Up Ready genes. These tests are easy to perform and do not require much training or special expensive equipment (Bhushan, 2001). However, spurious false positive or false negative results are sometimes obtained with particular genotypes when grown in certain environmental conditions (www.biogeneticservices.com).

Molecular genetic screening includes numerous laboratory techniques to identify the presence of a gene in a plant's genome. Currently, the simplest and most economical technique for seed testing is the polymerase chain reaction (PCR) (Goggi, 2000). Primers of known amino acid sequence are used to amplify (reproduce numerous copies of) a specified region in the DNA. Primers are chosen to flank the DNA region to be amplified; the amplified target fragment can then be evaluated on special staining gels. The PCR technique is highly accurate, sensitive, specific, and robust but requires a well-equipped laboratory with skilled technicians to be used effectively in a QA program (Bhushan, 2001).

Martin (1998) described the quality control techniques used by one large seed company to detect the presence of herbicide tolerant (Liberty, Roundup, Sethoxidim, Imidazolinone) and insecticidal (two Bt cry) genes.

**Quality Assurance and GMOs**

Quality control standards, although quite high in the U.S. maize seed industry, may need to be raised further in the production and sale of GMOs. This is true for various reasons including: a) most of the GMO maize products available today such as Bt and herbicide resistance are single gene traits (Dodd, 1998), b) to help assuage public safety concerns, c) to help reduce the chance of liability claims against seed companies, and d) because GMO hybrids are priced at a premium and customers will likely have higher quality expectations for these products (Christensen, 1998). One example is a conventional hybrid with 5% off-types which may be acceptable to a customer but a herbicide tolerant hybrid with the same level of dead plants after application of the herbicide will likely be unacceptable.

**XIX. Summary**

Production of hybrid maize seed is a unique and dynamic industry worldwide. The foundation of the industry is productive inbred lines which are developed by research programs which must invest significant quantities of time and resources. These lines are then used in field production to
produce hybrid seed. This is followed by proper conditioning and marketing of the seed. Many steps are involved in the production, processing, and marketing chain.

Good field management of hybrid maize seed production requires giving attention to adequate site selection including isolation, use of the best agronomic management practices, using appropriate female:male ratios, achieving a good nick between parental lines, properly controlling pollen production in female rows through detasseling or other means, effectively removing off-type plants, and harvesting the crop in a timely fashion. This work is often done under contract with private farmers, thus the selection, training, and collaboration with the best farmers is essential. Once the seed crop has been harvested it must be transported to the processing facility where it is de-husked, dried, sorted, cleaned, sized, treated, and packaged. Storage and then marketing and sale of the product follows. This long chain of activities requires intensive management, meticulous planning, impeccable timing, a high level of technical expertise, and the cooperation of weather and other factors. An effective quality control program should monitor and help coordinate all aspects of the maize seed program.

XX. ACKNOWLEDGEMENTS

I would like to thank the numerous authors whose research, experience, and ideas form the backbone of this chapter. Of special note are the large contributions in their papers and personal communication made by Dr. Bob Wych and Dr. Joseph Burris.
XXI. REFERENCES


Iowa State Seed Science Center, 1997. Tetrazolium test for maize seed. Seed Testing Laboratory, Seed Science Center, Iowa State University, SL-103, pp. 1-2.


Pinter, L. 1989. Improvement for maize (Zea mays L.) seed production. Georgicon Agric. 2:151-158


Thorp, F. 2000. Some things independent seed companies are doing to adapt to the GMO technology era, in S. Goggi (ed.) *Proceedings of the 22nd Ann. Seed Technology Conference*. Seed Science Center, Ames, Iowa, pp. 28-34.


