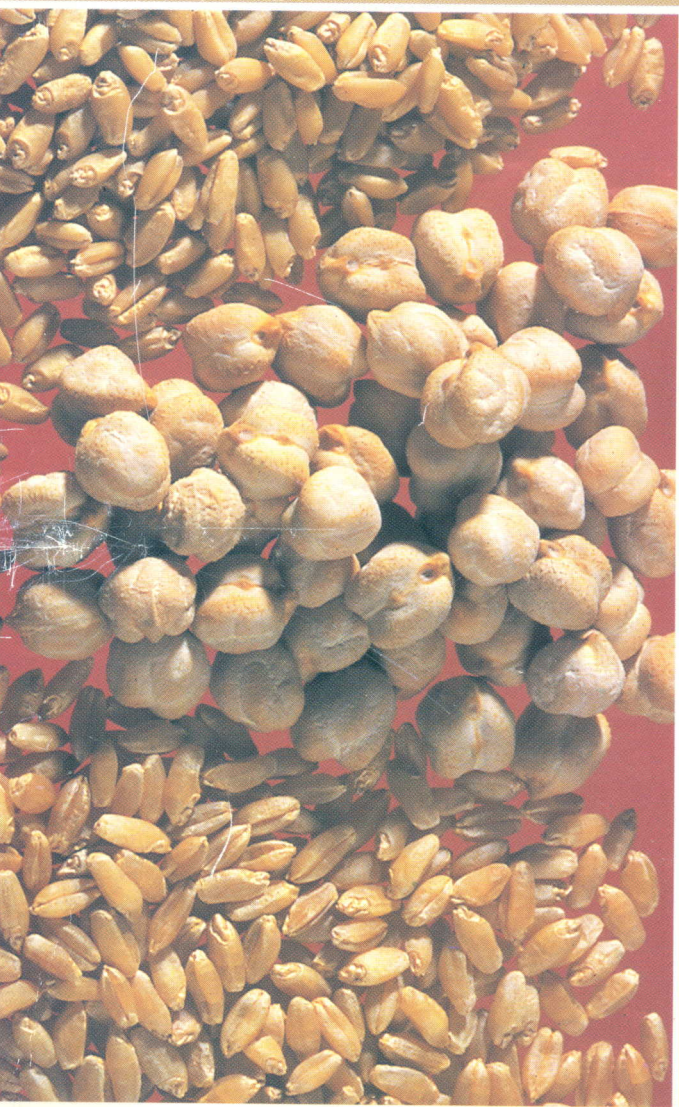


Seed Science and Technology



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Agricultural Research
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Seed Science and Technology

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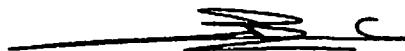
Foreword

Quality seed of improved varieties is an important input for any crop production program—and, as it is often the least expensive input, it represents an affordable way to reach cost-conscious farmers. A well-organized national seed program must exist to move seed of new varieties to the farmers' fields. The ultimate payoff is more and better food for the world's increasing population.

Seed Science and Technology is intended as a reference for those concerned with seed production, processing, marketing, and distribution, whether in a policy, organizational, or technical context. It also aims to fill an important gap for university instructors who wish to include seed technology in their curricula.

The authors have attempted to produce a practical handbook, in which the "ground rules" for seed production are laid out, while providing enough theoretical background to keep abreast of technological changes.

Above all else, the authors hope that the present work will contribute to a better understanding of the complexity of seed programs and the key role they play in the food production chain.



Prof. Dr Adel El-Beltagy
Director General

Editors' Note

Seed technology is a relatively new discipline, in terms of academic study, having only in recent years emerged as an integrated subject. Although highly specialized, it is a broad subject, which requires understanding of many disciplines. In the developing world there are very few seed technologists, and most general agriculturists lack the basic knowledge of seed technology. Unfortunately, there are only a very few universities in the world that offer B.Sc., M.Sc., and Ph.D. degrees in Seed Science and Technology. Moreover, these universities are all located in the developed world, where studies are expensive and restricted to a limited number of students. Although seed science and technology is a crucial element of agricultural production, it is seldom adequately covered in plant breeding and agronomy curricula.

Aware of this situation, and as a first step towards improving it, a workshop was held at the University of Jordan, Faculty of Agriculture, Amman, Jordan, from April 22 to May 10, 1993. The workshop was sponsored by MEDCAMPUS, and EU program designed to stimulate cooperation between EU universities and Mediterranean Non-Community (MNC) countries.

Nineteen participants from eight countries (Morocco, Algeria, Tunisia, Egypt, Jordan, Lebanon, Syria and Turkey) attended the workshop. The majority were university teachers in their respective countries. Resource personnel came from EU countries (Greece, Italy, Germany, the Netherlands, France, and Spain) as well as from ICARDA and the University of Jordan.

The aim of the workshop was to strengthen seed science and technology knowledge of university teachers throughout the WANA region. During the workshop, discussions were held on the need for and content of an M.Sc. course on Seed Science and Technology in the WANA region. As a result, the University of Jordan, Amman, has established a Seed Science and Technology major within its crop science master's degree course.

In addition to the papers that make up the present work, which represent the basic knowledge necessary for the study of seed science and technology, detailed country reports were also presented at the workshop. As several of these country reports are no longer up-to-date, they have been omitted.

The Editors

Keynote Address

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Introduction

Seed is the starting point of agriculture, its source of continuity, change, and restoration, as well as its most important product. Seed (here defined as all living materials used to plant a crop, whether dry seed, cuttings or vegetative parts) is too often taken for granted, or worse, neglected. Yet a sound understanding of the primary and catalytic roles of seed is important for the formulation of effective strategies for agricultural development.

Seed is the most important agricultural input. It is the basic unit for distribution and maintenance of plant populations. It carries the genetic potential of the crop plant, determining the upper limit on yield. It thus dictates the ultimate productivity of other inputs such as fertilizer, pesticide, irrigation water, etc., which build the environment that enables the plant to perform. Similarly, improved farming techniques and machinery are only as effective as the germplasm they support.

When considered as a production input, improved seed has several advantages over other inputs. It is required in relatively small quantities, it is increased rather than consumed in the production process, and its use does not require substantial changes in farming practices. On the other hand, seed has two major disadvantages. It must be kept alive to fulfill its propagative function, and its production must be planned well in advance of the time it will be needed.

Seed is an agent of change. Plant crops change as seed changes. For millennia, farmers have improved crops by saving the seed of plants that exhibit traits perceived as most useful, resulting in a slow but reliable improvement. This process has been exponentially accelerated by an understanding of genetics, especially during the second half of the present century. Seed can also be used to rapidly rehabilitate agriculture in the wake of natural disasters such as flood, drought or blight due to insects or plant disease. Seed, which can be efficiently maintained, and multiplied, is the primary means of delivering crop improvements to farmers' fields.

What Improved Seed Can Do

The potential benefits from the spread and use of improved seed are enormous. At the farm level, this means enhanced productivity, reduced risk and increased net income through higher yield, more efficient use of available nutrients, faster maturation, better resistance to pests, and higher nutrient content in the harvested crop. At the regional and national levels, more flexible and diversified agricultural production systems are made possible by multiple cropping and the spread of crops to a wider set of agro-ecological zones. This greater flexibility, together with enhanced yield and nutritional value, can contribute to increased food security. Varieties requiring fewer agro-chemicals to combat pests and disease also promote more sustainable agricultural production patterns.

Other potential benefits relate to the multiplier effect of enhanced productivity. When the diffusion of improved seed contributes to increased yield and multiple cropping, greater opportunities for farm and post-harvest employment are generated. Where improved seed contributes to higher yield and higher quality crops, both processors and consumers may benefit through more plentiful, low cost, and high quality supplies of raw materials and food. The spread of improved seed may also speed up both the adoption of agricultural production technologies and the economic return on existing or planned investments in the rural and agricultural infrastructure.

Thus, in the development and spread of improved seed, a multiple set of agricultural and rural development objectives can be pursued: economic growth, rendering the existing agricultural system more sustainable, and promoting socioeconomic welfare. For varietal development and seed production and distribution, there is a set of narrower objectives, depending upon targeted crops, location, and type of producer. Such work will likely have a substantial impact on patterns of agricultural and agro-industrial production, food consumption, and distribution of income. While there is a tendency to focus (at least initially) on major crops and more favored areas, there are also opportunities to enhance productivity in secondary crops and in remote or disadvantaged areas.

Supplying Seed

Since the first cultivator, every serious farmer has taken steps to ensure an adequate seed supply. Thus seed programs were established. If seed was not saved, there was no agriculture next season.

Traditionally, farmers grew and maintained their own seed, choosing from local landraces and exploiting natural mutations, outcrosses, and selection. This selection process resulted in locally adapted varieties which provided reliable performance patterns. Benefits were frequently spread through local communities via farmer-to-farmer exchanges of seed.

With the advent of plant breeding research, substantial improvements in varietal development have occurred, offering the potential for major advances in productivity and product quality. Advances in the seed industry (including production, processing, storage, and marketing) also provide opportunities to improve traditional seed production and distribution systems.

There are two ways to improve seed. The first is to improve the information contained within the seed itself through breeding, which results in improved varieties. These improvements in the seed's genetic makeup provide the potential for higher yield, greater pest resistance, and improved quality of harvested crops. The second source of improvement relates to the physical properties of the seed, such as size, purity, and germination capacity. These improvements stem from effective processing, quality control, chemical dressing, handling, and storage. The resulting higher quality seed provides value through enhanced performance and compatibility with other production inputs.

Cultivar Development and Diffusion

Cultivar improvement is the incorporation of genetic characteristics that enable the plant to cope better with its environment, to respond better to other agricultural inputs and practices that yield higher economic return, or to more effectively meet ecological or social demands.

Varietal Improvement

Crosses or mutations established the foundation for much of the improvement work that has followed. This work has four traditional methods:

Selection

This, in its simplest form, consists of gleaning the most promising plants from heterogeneous populations. Practiced by farmers for centuries, the technique has been adopted by plant breeders for selecting the offspring of crosses.

Varietal introduction

Varieties that have proved themselves elsewhere under similar agro-climatic conditions are imported and introduced.

Hybridization

This involves planned crosses and subsequent selection of desired plants from the segregating populations to combine the most desirable characteristics of two or more varieties.

Hybrids

The first generation of the cross between inbred lines traditionally displays hybrid vigor and uniformity, producing large quantities of seed for commercial use.

Genetic engineering can also be used to select crops for the incorporation of new traits.

International Variety Transfer

Geographical dimensions

By definition, an international transfer of technology involves two or more countries, but the transfer often involves a complex chain which may include both developed and developing countries.

Semi-dwarf wheat is a good example. It originated in East Asia, spread first to the United States, next to Mexico, and then throughout the world—including back to East Asia and the United States.

An increasingly evident aspect of the process is its two-way nature; seldom, if ever, is one country strictly a donor and another simply a recipient. Donors become recipients, and recipients may eventually become donors. The tide may run one way for a while, but ultimately it will turn. The United States, for instance, after providing Norman Borlaug, at CIMMYT in Mexico, with Noring 10 x Brevor, in time received several varieties back from CIMMYT, which were used directly or as parents in US breeding programs. Some of these varieties have undoubtedly been used for similar purposes in other countries.

Cultivar transfer through institutions

Varieties in the form of seed move internationally in many ways, ranging from the accidental and informal to the organized transfer process. Over time, this process—partly because of the large numbers involved—has become more and more formal.

The two main institutional components of the formal process are the public and private sectors. The public sector plays several roles at the international level. These include: national agricultural research systems in both developed and developing countries, international agricultural research centers, germplasm collection and exchange, international nurseries, and foreign assistance programs. The private sector includes national firms in both developed and developing nations, multinational or international firms, and trade.

The process of distributing material from international centers to national programs has changed over the years. Initially, germplasm was exchanged directly. Later, international testing programs were established, which included varieties from national and other programs.

Despite the many institutional differences, the final result has been a set of lines/varieties developed in one nation and tested and grown in many other nations. New varieties do not take hold everywhere they are tried; usually they must be adapted to local conditions through further breeding work, or they may serve as parents for other varieties. The transfer, however, is the vital key to the process. The large-scale introduction of adapted varieties of wheat initiated the green revolution from Mexico to India, Pakistan, and Turkey.

Varietal Replacement at the Farm Level

Once improved varieties have found their way to a developing nation, they become part of a replacement process at the farm level. This process has three dimensions:

- First, improved varieties may replace traditional varieties or landraces, but as this process continues, they may themselves be replaced. For wheat, the earlier improved varieties of conventional height have been replaced by higher yielding varieties that are shorter or semi-dwarf.
- Second, the varieties within each category are subject to replacement over time as they become susceptible to insects, disease, environmental factors, and other forces. The newer seed is sometimes higher yielding. In some countries this process is relatively rapid, in others it is slow. Factors influencing the rate of wheat seed replacement were recently studied in Pakistan where the rate of replacement is slow. This is a key area requiring study for other crops in other countries.
- Third, there is seed replacement of individual varieties. This is important for self-pollinating crops such as wheat, for which farmers may grow and use their own seed from year to year. The problem with this process is

that the seed may deteriorate over time because of seed or weed intermixture, loss of vigor, loss of germination potential because of poor storage, seedling infection due to untreated seed, genetic deterioration resulting from outcrossing, and other factors. At some point therefore, it benefits farmers to buy new replacement seed for their existing varieties; this also gives them the opportunity to buy new varieties.

Relative Roles of Improved and Unimproved Varieties

Crop cultivation in developing countries includes both improved and unimproved varieties, the relative roles of which vary considerably by country and region. Generally, the adoption of improved varieties is high in irrigated and better-endowed agro-climatic regions, whereas unimproved, locally adapted cultivars are common in low-input, stressed environments. The adoption rate is also influenced by the degree of risk, superiority of varieties, farmers' educational levels, efficiency of seed production and supply systems, and availability of credit and other inputs.

A related issue concerns the seed source. Although one might suppose that unimproved seed is obtained from the informal seed sector (farmer production) and improved seed from the formal sector (private and public firms), this is only partially true. The informal seed sector is an important source of improved seed, especially for self-pollinating crops. Although the informal sector is important for seed replacement, it does not usually generate new varieties; these normally originate in the formal sector.

The relative importance of sources is illustrated by improved wheat seed in Pakistan. A survey in 1985/86 showed that of the total wheat seed used, about 58 percent came from own or retained production, with about 42 percent from other sources. These other sources were: seed for currently used varieties (20 percent), seed of new varieties (22 percent), other farmers (nearly 21 percent), seed depots (over 17 percent) and miscellaneous (research/extension and grain merchants, nearly 4 percent). During this period, seed production and marketing was managed primarily by the government. Similar information for other crops and countries would be useful, especially where the private sector is more heavily involved.

Factors Influencing Transfer of Varieties

Many factors influence the rate and course of diffusion of new plant varieties, including: access to economically, socially, and ecologically acceptable varieties, conducive government policies, availability of essential agricultural inputs, adequate infrastructure, credit, and receptive farmers. Most of

these factors are well known and have been widely discussed over the past two decades. One thing that has become clear over this period is that formal research systems play a relevant role in formal seed production and dissemination. The main topics of these systems are considered below.

Availability of New Varieties

There are several sources of new varieties in developing countries.

In many countries both private and government breeders are active, though in other countries breeding and varietal release are carried out exclusively or predominantly by one sector. Much depends on government policy and regulations. For example, the attitude of private companies to the breeding of self-pollinating crops often depends on whether their product (varieties) is protected by some form of intellectual property rights. Decisions about the export of their best varieties to other countries display a similar dependence. Conversely, a company is automatically protected in the case of hybrid varieties; hence, it may be interested in establishing a market even in countries without plant variety protection. Where private activity is limited, the farmer is dependent on local public breeding stations for new and better varieties.

Development of Varieties

Continuous development of new superior varieties adopted at the farm level is the cornerstone of improved and sustainable agricultural development and growth. Formal development of varieties occurs in either the public or the private sector.

Public sector

The research activities of this sector are generally well known and significant for developing countries. There are three major components:

- Domestic research by national agricultural research systems.
- International research by international research centers.
- Research in foreign countries that is transferred to developing countries.

In the last case, relatively little technology is transferred directly from a developed to a developing country, except where both have similar ecological zones. Prospects for transfer among developing countries with similar ecologies may be considerable, but are less likely to be exploited.

The public sector has thus far accounted for virtually all of the research on major field crops.

Private sector

In most developing countries the private sector is not yet an important source of new varieties. The principal exception is hybrid maize.

In the last few years, several developing countries have begun to encourage multinational corporations to participate in research and development.

Interaction between sectors

Interaction between the public and private sectors is generally positive, though there can be some negative aspects. The major benefits arise from the complementarity, with the public sector doing most of the basic research and the private sector handling the applied research.

Another complementary relationship involves the target area and crop type. The private sector develops and serves the most ecologically favored production regions; public sector research can concentrate on less favored zones. The private sector favors research on hybrids and low-volume, high-value crops such as vegetables and hybrid seed, whereas the public sector works on low-value, high-volume crops such as wheat and rice, and on either open-pollinating crops or non-conventional hybrids.

Sometimes there is interaction between the public and private sectors involving facility use and the conducting of on-farm trials. Beyond this, the flow is generally in one direction—movement of scientists and germplasm from the public to the private sector, with little movement in the opposite direction. In addition, the existence of one sector counterbalances possible monopolization by the other. Problems can arise when one sector has an unfair or extreme advantage over the other. This often arises when public sector seed is sold through a state enterprise at an artificially low price, thereby discouraging research in the private sector. Less frequently, one sector produces such superior lines that the other sector is outclassed.

Role of imports

Seed imports and local private research are closely related. After a minimum amount of testing, companies start selling seed developed elsewhere, and then invest in research to develop new varieties for local conditions as the market expands. Initially they import seed because it is cheaper than establishing their own production and processing operation. As the market grows, companies establish local operations which produce most of the seed, but also continue to import.

Seed Supplies

Good plant breeding research and availability of improved varieties are essential, but unless good-quality seed of these varieties is produced commercially and supplied to farmers, a country will not benefit from the research. An efficient production and distribution system for seed is necessary. Some countries have not been able to exploit domestic and international plant breeding efforts because a functional and efficient seed production and distribution system does not exist.

Seed supply normally involves three main operations: production, conditioning, and distribution. The public and private sectors play somewhat different roles in these operations.

Public sector

The public seed production and supply sector is largely composed of public or parastatal firms, which conduct little, if any, research. There is a clear demarcation between these organizations and the public national agricultural research systems. There is, however, one important task that falls between the two—the production of breeder seed, which is usually carried out by a public national research organization.

Although public seed firms generally play a critical role in the production and distribution of self-pollinating crops, they often have a bad image. Many countries want low seed prices to partially offset low product prices. Seed is a prime candidate because it can be produced domestically. Parastatal organizations are set up for this task, but often evolve into sizable bureaucracies and monopolies, often subsidized. Under these conditions, they frequently become high cost and inefficient producers.

Other common problems are the lack of adequate amounts of high-quality seed, neglect of realistic demand forecasting, and the lack of an adequate distribution network and trained production and marketing personnel.

However, because self-pollinating crops—which are not very attractive to the private sector—usually fall to the public sector, the potential for profit is limited; therefore, there are inherent limitations on how well the public sector can perform.

Private sector

There are three components to the private sector seed industry: other farmers, farmer cooperatives, and commercial firms. Other farmers are a significant source of seed for self-pollinating crops, but there are no formal statistics on the overall importance of this source.

Farmer cooperatives are important in some areas, but there is no information available on their overall significance. Prevailing opinion says that generally the private seed sector is weak in most developing countries, with little effective competition among producers. However, the situation probably differs from country to country and is changing over time as more nations give increased attention to the private sector. For self-pollinating crops, some small "seedsmen" farmers are emerging as private entrepreneurs to supply seed of new varieties in limited areas.

Interactions between sectors

Interaction between the public and private seed sectors, both positive and negative, shares some of the same characteristics noted earlier. Briefly, positive interactions involve cooperation in field trials, complementarity in crop type, zone coverage, and the counterbalance function. Negative interactions are principally in pricing.

For seed supply, a balance is clearly required between the public and private sectors, which are both needed in most developing countries. Although the appropriate mix will vary over time, generally speaking the role of the private sector should expand and that of government agencies decrease. The private sector, however, cannot be expected to do the entire job itself.

Role of imports

The importance of seed imports varies between temperate and tropical developing countries. Tropical nations are not likely to benefit significantly from seed imported from temperate regions, where most of the seed industries are.

Developing nations in or with temperate zones are more likely to benefit from seed imports from developed countries.

Restrictions on seed imports and on the role of foreign seed companies are common throughout the developing world. Local firms may encourage such barriers. However, it is in the best interest of developing nations to reduce or eliminate barriers to seed trade. They can benefit in the short run from imports, and possibly in the long run from seed exports. Improvements in plant quarantine facilities may be needed, but this is a small price to pay considering the possible benefits.

Other Institutional and Policy Factors

There is a host of other institutional and policy components that influence the transfer and adoption of plant varieties.

Other production inputs

Seed is, of course, just one component of the package of improved practices that is needed to increase yield. Fertilizer is a vital component of this package, as are water control and improved cultural practices. Packages may be applied in whole or in part, at or below recommended levels. Productivity limits may be set by the levels at which other components of the package are applied.

Government price and credit programs

This is a vast subject. Narrowly defined, it can be viewed in terms of subsidies to the seed industry and the provision of other inputs and credit. Such subsidies, although well intentioned, too often distort the rural economy and discourage private sector development.

Extension

Some assume that good technology will sell itself in the countryside, via farmer-to-farmer information exchange, but this is not true everywhere. Much useful technology may not be dramatic enough to spread far or rapidly on its own.

Transportation and distribution

Supplies must be distributed at the right time and to the right place. Without improvements in transportation and distribution, vast quantities of seed may end up in warehouses, as end-of-year stocks increase.

In addition to these general factors, there are several other institutional factors that affect the seed industry. Seed legislation—especially laws related to seed quality—is usually needed. One view is that such legislation should initially be kept to a minimum to promote entry into the system, and that as the industry develops, legislative controls can be upgraded as necessary.

Adaptability, Ecology, and Spillover

The transfer of technology embodied in plant varieties is greatly facilitated when the varieties are widely adaptable and developed in similar ecological areas. The high-yielding semi-dwarf wheat and rice varieties have proved to be widely adaptable under optimal crop management. In developing countries, much of the improved wheat and rice is grown under irrigated conditions, thereby diminishing the degree to which environmental variability is a key factor. Other crops are grown under more variable dry land conditions. In each case, diffusion is more varied in stressed environments than in favorable environments.

Obviously, the return to a given investment in varietal research varies, and is thus more or less attractive.

Emerging Issues in Technology Transfer

There are several emerging issues that modify the technology transfer process for crops.

Biotechnology

Through the development of new plant varieties with novel characteristics, biotechnology is expected to have a significant impact on agriculture sometime after the year 2000. Progress in developing countries, however, will depend on the strength of plant breeding programs. New technologies with potential for early application in developing countries include new detection methods for pests and disease, genetic mapping of major crops, plant virus resistance, and biocontrol agents. It is currently unclear when these technologies will become a major factor.

Intellectual Property Rights

Intellectual property rights (IPR) are generally nonexistent in developing nations. Where patent laws exist, agricultural products are generally excluded. One benefit is that IPR facilitates joint ventures between government research institutes and private cooperatives and firms.

Views on the importance of IPR vary. On the one hand, the lack of patent protection is a major disincentive for private sector investment. Also, such laws can stifle growth rather than accelerate development. Moreover, when trained personnel and specialized implementing institutions are absent, the enactment of laws has little value and leads to further bureaucracy.

Indigenous Seed Supply Systems

Papers and public debate have often focused on the formal seed system, although the informal system—particularly other farmers—can be an important source of seed.

Farmers can efficiently produce and store seed for most varieties of self-fertilizing crops. They can also produce and store seed of some self-pollinating crop varieties, and vegetatively propagated crop plant material.

Some activities, such as plant breeding research, or the production of high quality hybrid seed, cannot be done efficiently by farmers.

Considerable capacity exists at the farm level for maintaining genetic diversity, performing informal experiments, and selection. The challenge is to strengthen local technology with elements of modern science.

Another rather different idea is oriented to "farmers' rights." Under this system, there would be a tax on seed sales to compensate farmers for their prior selection and improvement efforts. Although this proposal is problematic, both conceptually and practically, the funds thus gathered could be used to finance indigenous seed systems.

Environmental and Sustainability Considerations

The varietal transfer process can assume a different dimension when environmental and sustainability issues are considered. The transfer process can go too far. Planting of large areas to one variety can be significantly hazardous due to genetic vulnerability to pests or disease. Diverse germplasm is needed.

Research is important to improve the adaptability of varieties, but if it permits them to be grown in environmentally vulnerable areas, adverse societal externalities may result. An interesting environmental case occurred in Thailand, where high profits from growing maize persuaded farmers to expand cultivation by clearing forests, a process that was aided by the eradication of malaria in the country. Expansion of the road system and cheap fuel also contributed greatly. Consequently, there was a tremendous expansion in the maize area between 1947 and 1971. Production in high slope areas, however, has led to severe erosion problems.

Another environmental dimension is that new varieties may require relatively heavy applications of nitrogen and pesticides, and as just noted, can contribute to erosion. Depending on the extent to which chemicals are applied in developing nations, these considerations may limit the extent to which transfer of cultivars is desirable.

Implications

Although much has been written about the development and spread of high-yielding varieties, relatively little attention has been given to simultaneously examining the closely related question of the role of the seed industry.

This paper has viewed crop technology primarily in terms of research and production and distribution. Both are carried out in the public and private sectors, both have international dimensions, and both are interrelated. A

strong public research sector is essential if seed firms are to have improved varieties to sell. Improved germplasm from other countries or from international research centers can be tremendously important in generating improved varieties for distribution in individual countries.

With several dimensions involved, a balanced approach is needed. Governments and agencies should not emphasize only one aspect of the package and ignore others. Parity is needed in the attention given to the public and private sectors, and to national and international programs. Each, however, should be approached somewhat differently and with discernment.

Varietal improvement and distribution are dynamic and changing fields. Emerging issues will continue to influence technology transfer and should therefore be examined before specific actions are undertaken.

Section I

General Aspects

World Seed Market

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World Seed Market Estimates

Because of the lack of information for many countries, especially in eastern Europe, Asia and Africa, data for the world seed market can only be roughly estimated. It is practically impossible to estimate the amount of seed used worldwide. A tentative evaluation for the main commercial regions can be made using selected sources of information.

In 1987, the French organization Sanofi estimated the international agricultural supply market at 113 billion dollars, 30 percent of which was in seed. One third of the seed market was represented by commercial seed, conservatively estimated at US\$ 11 billion. In 1985, almost US\$ 5 billion was ascribed to the United States alone.

More recent figures (Table 1), furnished by Precepta, a market research company, suggest that US\$ 15 billion is a more realistic estimate for the commercial seed market, with North America accounting for at least 40 percent and EU countries for about one third. Japan and Latin America are the other two major markets.

Table 1. Estimates of the world seed market.

	1990 (by Precepta, revised)	
	Bil \$	%
Commercial seed, of which:	15	100
North America	6 (a)	40
EU	5 (b)	33
Others	4	27
Breakdown within EU		100
France	1.5	30
Germany, Italy, UK	2	40
Others (c)	1.5	30

(a) Underestimated according to note (a).

(b) Revised according to our estimates, which may approach 5.5 billion dollars.

(c) Underestimated. A more realistic estimate might be about 2 billion dollars, half from Benelux.

As far as the EU is concerned, France is the leading European market, with a turnover estimated by Precepta at US\$ 1.5 billion (30 percent of the EU market). Germany, Italy and Great Britain account for about 40 percent. Of the other EU countries, Holland is the leader in potato, flower, and some vegetable seed.

With the growth of agriculture and the expansion of agri-business companies since the beginning of the “green revolution,” and the innovations in genetics and biotechnology, the seed market has acquired strategic importance. (Antonietti, 1991; Scarascia Mugnozza, 1991).

Table 2. Seed diversification and investment regions of the main European groups.

a) Seed Diversification

Groups	Country	Grain cereals	Corn and sorghum	Forage crops	Oil seed crops	Protein crops	Flower and vegetable	Sugar beet
Ciba Geigy	Switzerland	*	*		*			
ICI	Great Britain	*	*		*			*
KWS	Germany	*	*		*	*	*	*
Limagrain	France		*	*	*	*	*	*
Orsan	France	*	*	*	*	*	*	*
Rhone poulenc	France		*		*		*	*
Sandoz	Switzerland		*	*	*		*	*
Sanoi	France	*	*		*	*	*	
Suiker Unie	Netherlands	*	*	*	*		*	*

b) Investment Regions

Groups	Country	West Europe	East Europe	Former USSR	No/Central America	South America	Africa	Asia	Oceania
Ciba Geigy	Switzerland	*	*		*	*	*	*	
ICI	Great Britain	*			*	*		*	*
KWS	Germany	*	*	*	*	*			
Limagrain	France	*	*		*	*	*	*	*
Orsan	France	*			*	*	*		
Rhone poulenc	France	*			*	*			
Sandoz	Switzerland	*	*		*	*	*	*	
Sanofi	France	*	*		*	*			
Suiker Unie	Netherlands	*	*		*	*			

Sources:

Semences et biotechnologies: les grands groupes étrangers, DPA, Paris, 1982.

Agritech decision, OCDE, April 1990.

Biotechnologies and the Seed Industry, by Asgrow, 1990.

Bulletin Semencete, winter 1991.

International Competitiveness in the Propagating Material Sector, Rabobank, Eindhoven, Sept. 1992.

Terra e Vita, (111 mondo delle sementi), by Edagricole, Bologna, 1988–1992.

Seed World, November 1992.

Only one third of the main multinational groups specialize in seed and agricultural trading: (i) Limagrain, Ragt, Sigma, and Verneuil (France), KWS (Germany), Barenbrug and Royal Sluis (Holland); (ii) Cargill, Dekalb, Geo. J. Ball, Pennington and Pioneer (the United States); and (iii) Sakata and Takii (Japan).

Groups involved in agro-chemical activities have made substantial investments in the seed business in various regions of the world, including eastern Europe, the former USSR (CIS), Asia, Africa and Oceania, with a wide diversification in many crop sectors (Tables 2 and 3).

Table 3. Seed diversification and investment regions of the main US groups.

a) Seed Diversification

Groups	Grain cereals	Corn and Sorghum	Forage	Oil seed	Rice	Flower and vegetable	Cotton
Cargill	*	*		*	*		*
Dekalb corp.		*	*	*			
Pioneer Hybrid		*	*	*			
Upjohn		*	*	*		*	

b) Investment Regions

Groups	North/Central America	South America	Western Europe	Eastern Europe	Africa	Asia	Oceania
Cargill	*	*	*		*	*	*
Dekalb corp.	*	*	*			*	
Pioneer Hybrid	*		*	*	*	*	
Upjohn	*	*	*		*		

Sources: see Table 2.

Investment by Multinational Groups

Most investment has taken place since the Seventies, with an upward trend throughout the Eighties. This is certainly due to the acknowledged synergy between seed and agro-chemical products. The many seed company acquisitions by pharmaceutical and petrochemical firms have led to extensive research efforts. For example, Ciba-Geigy, Sandoz and Sanofi from Europe, and Lubrizol, Upjohn, etc. from the United States have acquired large seed companies.

Recently, some of the larger multinational groups involved in the European food sector (Danisco, Cebeco, Suiker Unie, Unilever and Provendora-Volvo) have made substantial investment in the seed business. By 1990, 23 groups controlled approximately 40 percent of world seed sales, while 10 groups,

specializing in seed and agri-trading, controlled more than the 20 percent of the market. The five groups mentioned above accounted for about 2,730 million dollars of commercial seed value (18 percent of the world total). Table 4 summarizes the investment in human resources and in research and development programs as a percentage of seed sales.

Table 4. Economic data of the main seed group(*).

Groups	Country	turnover mil/\$	Main activity	Seed Activity			
				Sales 1990		Staff number	R&D %
				mil/\$	ranking		
Ciba Geigy	Switz.	3,029	Agro-chemical	154	17	850	12
ICI	G.Britain	2,337	Agro-chemical	250	7	2,000	8
Bayer	Germany	2,030	Agro-chemical	NA	NP	NA	4 (a)
Rhone poulenc	France	1,917	Agro-chemical	200	12	NA	NA
Monsanto	USA	1,508	Agro-chemical	NA	NP	NA	12
Dow elanco	USA	1,500	Agro-chemical	NA	NP	NA	NA
Hoechst	Germany	1,243	Agro-chemical	NA	NP	NA	NA
Pioneer hybrid	USA	1,101	Seed	1,101 (b)	1	5,000 (c)	5
Sandoz	Switz.	858	Agro chemical	640	2	5,300	5-7
Limagrain(coop)	France	NA	Agro-seed	435	3	3,050	6-8
Upjohn	USA	662	Pharma	280 (b)	4	NA	8-9
Sanofi (ELF)	France	NA	Oil-Pharma	272	5	400	NA
Cebeco (coop)	Neth.	NA	Agro-seed	267	6	NA	6 (a)
Cargill	USA	NA	Food industry	250	7	NA	NA
Pennington Inc.	USA	229	Seed	229	9	672	7
Dekalb Genetics Co.	USA	220	Seed	212	10	2,400	10
Takii	Japan	201	Seed	201	11	750	NA
Suiker Unie	Neth.	NA	Food industry	200 (b)	12	850	12 (a)
Sigma (coop)	France	1880	Trading	200	12	240	NA
Ragt (coop)	France	200	Seed	200	12	685	NA
Orsan	France	NA	Concrete	180	16	1,600	NA
Sakata	Japan	NA	Seed	137	18	600	NA
KWS	Germany	218	Seed	130	19	700	11-14
Lubrizol	USA	NA	Oil	117	20	750	5 (a)
Provendur/Volvo	Sweden	NA	Food industry	110	21	800	NA
Barenburg Holding	Neth.	110	Seed	110 (b)	21	350	NA
Royal Sluis	Neth.	104	Seed	104	23	800	6

(*) Selected from a seed turnover of +100 mil/\$ and/or a permanent seed staff of 170 people.

NA: not available; NP: not present in the first twenty groups

(a) 1982.

(b) 1991.

(c) 2800 of which in North America.

Sources: see Table 2.

R&D increased from 4-12 percent (1982) to 5-14 percent (1990). The interest shown by two US firms (Biotechnica and Calgene), which were once primarily involved in biotechnology research, joint ventures and the acqui-

sition of seed firms within the United States, should be noted. Biotech companies operating in western Europe are:

- AGC (GB), whose main activities involve plant breeding and agricultural research.
- Algene (F), which specializes in plant biotechnology research for arable crops and vegetable/flower species.
- Mogen (NL), which develops international collaboration in the field of plant biotech research.
- PGS (B) a plant genetic systems company operating in cereal, oil seed rape and vegetable crops.

Concentration and Diversification Goals

The environment in which seed research and development takes place underwent a significant change in the Seventies. The approval of the Plant Variety Protection Act (1970) in the United States, followed by similar regulations in western Europe, made it possible for seed companies to receive patents and royalties on new plant varieties. This sparked the interest of many large multinational groups with the capacity to undertake extensive research.

Several of these groups have acquired seed companies, creating concerns that a seed business oligopoly is developing. Especially disturbing to some critics is that a growing number of seed companies are now owned by pesticide manufacturers, thus creating the possibility that seed breeders will have an increasing bias towards marketing seed that requires greater chemical inputs (US Presidential Commission on World Hunger, 1980). There is no evidence, however, that this is taking place. It is unlikely that pesticide firms will ever dominate the seed market for a particular crop.

The trend towards concentration and diversification of the seed mix seems to be aimed at acquiring a larger market share, and at supporting R&D programs through economies of scale in production and marketing. The acceptance of a given variety by farmers is positively correlated with performance, even if it leads to a jump in seed prices.

Research efforts aimed at developing pest-resistant varieties have created a demand for cost-effective strategies. Companies will continue to concentrate on the production of pest-resistant varieties (Leibenluft, 1981).

The food industry is increasing its interest in the seed business, as borne out by the capital investment made in the sector as well as by various joint ventures. This can be explained as an attempt to gain control of the food chain

by directly developing quality seed. Global quality for many food products requires direct management of all stages of the food processing chain, starting with seed. For example, Del Monte Co., a well-known corporation operating in the food industry, recently invested in seed activities to obtain quality vegetable crops for its processed products.

The Seed Industry

The seed industry has changed over the last two decades, and is still changing, although the trend towards concentration has leveled off in recent years (Cailliez, 1991). As Leibenluft's states, "Plant patenting legislation has brought large multinational corporations, capable of mounting substantial research and development efforts, into the seed business." These large companies have also allowed for the development of marketing links between the USA and western Europe, the USA and Latin America, Japan and Europe, etc. The direct effect has been a reduction in the range of seed varieties available to farmers for some crops, as these companies tend to develop only a limited number of high-yield and hybrid varieties capable of finding broad market acceptance.

This trend, which was first observed for corn and sorghum hybrids, is now occurring for several vegetable crops. Significantly, many seed firms for these crops have a parent company in California, France, Holland or Japan, and have staked their existence on future development. The economic performance of Pioneer Hi Bred, a leading company in the corn seed market,

Table 5. Estimated cost structure of a typical hybrid (percent).

Net price after discounts and commissions	100
Less production costs:	
Variable:	
Detasseling	5-7
Payments to farmers for seed	30
Fixed:	
Plant overhead	15
Total	50-52
Gross margin	48-50
Less expenses:	
Research & Development	3
Selling	10
General and administrative	5
Pretax margin	30-32

Source: Goldman Sachs Research, "The Hybrid Seed Corn Industry: Implication of a Changing Environment", 1978.

illustrates this. A case study from the second half of the Seventies, but still viable, estimated the gross margin at approximately 50 percent and the pre-tax margin at 30-32 percent (Table 5). This is considered very profitable, even though a slight reduction may have occurred in the more recent years.

The margin is much lower for traditional small grain cereal and forage crop seed. But there is an emerging market for vegetable seed that is very profitable because of many specialized niches, mostly managed by plant producers, in the hobby market (Grillenzoni *et al.*, 1992). These

niches represent exciting marketing opportunities for several medium-sized firms operating in the Mediterranean.

While seed company acquisitions over the last two decades suggest some industry consolidation, the integrating firm is often a new entrant into the seed industry (ICI, Sandoz, Unilever). Unlike in other industries, where the process has been more marked, the concentration of new seed varieties among patent holders—a trend aimed at diversifying the market—has not yet led to an oligopoly.

The possible reduction in genetic diversity which may accompany seed industry evolution warrants careful scrutiny by agro-ecologists and biologists (Porceddu, 1991). One major fear is the increasing trend towards genetic uniformity. As the larger seed companies develop and market new plant varieties, farmers will abandon the traditional multitude of seed types and concentrate on a few high-yield varieties. This lack of genetic diversity may increase vulnerability of major crops to widespread disease, such as struck the corn crop in the United States in 1970.

Another danger is that as traditional varieties are dropped from the market, their germplasm may be lost.

Supporters of plant patenting agree that genetic uniformity and loss of germplasm resources are legitimate concerns. Their position, however, is that if plant patenting has any effect, it is to promote genetic diversity; they argue that germplasm can and should be preserved by seed banks, and by germplasm conservation programs.

Apart from these fears, there are other reasons supporting the opinion that anti-competitive practices are unlikely to expand.

Quality Seed vs. Farm Seed

The trend towards concentration in the seed industry is the result of legislation aimed at creating a limited number of varieties through the patenting mechanism. Such patents are a reasonable trade-off in return for greater private breeding and marketing. Sugar beet, corn and sorghum, as well as some flower and vegetable crops, are prime examples of sophisticated technologies used in seed production.

The overall increase in concentration, however, has been low, considering the many self-pollinating species and varieties, such as small grain cereals, forage crops, oil seed and protein crops as well as close substitutes for many standard vegetable seeds.

Seed growers, who are organized into successful producer associations in many countries (France and Holland in Europe, and the US), are closely linked to public institutions (universities and agricultural experimental stations in the US and in some western European countries, research organizations like INRA in France, etc.), and are therefore strongly motivated to continue research in new plant varieties. These growers have formed cooperatives or inter-professional organizations (Grillenzoni, 1988; Regazzi, 1988) to directly advertise and market seed developed by public agencies. These combined efforts are likely to provide alternative channels to the larger seed companies.

A further guarantee against oligopolistic pricing or other anti-competitive behavior with regard to the sale of seed of varieties of self-pollinating varieties is offered by farm seed through the so-called farmer's privilege. This is the option given to the farmer to set aside a part of the harvested crop for use as seed in the following year.

This topic is being debated by professional organizations involved within the EU seed industry. The average rate of renewal with regard to small grain cereal seeds, for example, is about 60 percent (Table 6), although it is far lower in Mediterranean countries, including Italy. This means that almost 40 percent (or more in some countries) of seed need is provided by farm seed rather than by commercial certified seed.

Table 6. Provisional seed figures 1989/90 CEE (small grain cereals).

Country	Certified seed production (1988) 1000 t	Certified seed use (1988/89) 1000 t	Domestic seed need (1988/89) 1000 t	Renewal rate %
France	723.2	694.2	1,217.5	57
Belgium	44.2	32.1	52.9	61
Luxembourg	4.4	4.3	4.8	89
Netherlands	47.4	26.1	31.8	82
Germany	469.8	396.8	693.0	57
Italy	322.5	327.1	658.2	50
U.K.	527.9	524.0	707.4	74
Ireland	NA	NA	61.8	NA
Denmark	262.2	263.4	279.7	94
Greece	NA	NA	236.2	NA
Portugal	NA	NA	72.5	NA
Spain	NA	NA	1114.1	NA
CEE (*)	2,401.6	2,267.9	3,645.3	62
CEE			5,129.9	

(*) Not including Ireland, Greece, Portugal, Spain.

Sources: INTERCOOP estimates on EUROSTAT statistics.

This trend is destined to continue for two reasons: (i) commercial seed prices are increasing, in some cases, to unfairly high levels; and (ii) the extensive agricultural processes underway in the EU and in some areas of the US will continue in the near future. Table 7 estimates the seed market in the United States; as can be seen, the demand for commercial seed increased from 74 (1975) to 78 percent (1985) of the estimated total. Table 8 gives a breakdown of this percentage according to crop species. Cotton, soybean, barley, wheat, oat, rye and flax seed rank below the national average of commercial seed, thus confirming the competitive structure of the overall seed market.

At the international level, however, the seed market is shifting from fragmentation to concentration. Competitors capable of handling and managing innovative biotechnology now have a chance to enter the market.

Table 7. Estimates of the US seed market (mil \$).

Seeds groups	Consumption		Commercial seed		Percent of coml. seed over consumption	
	1975 (a)	1985 (b)	1975	1985	1975	1985
Cereals	1,330.5	3,476.0	940.0	2,617.5	70.6	75.3
Forage crops	252.5	520.0	239.5	494.0	95.0	95.0
Industrial crops (soybean, cotton, peanut)	605.0	1,582.0	381.0	1,083.5	63.0	68.5
Vegetable crops (bean and pea included)	150.0	347.0	127.5	340.0	85.0	98.0
Packet, turf & others	176.0	448.0	176.0	448.0	100.0	100.0
TOTAL	2,514.0	6,373.0	1,864.0	4,983.0	74.1	78.2
of which (%):						
-cereals	52.9	54.5	50.4	52.5		
-forage crops	10.0	8.2	12.8	9.9		
-industrial crops	24.1	24.8	20.4	21.7		
-vegetable crops	6.0	5.4	6.8	6.8		
-others	7.0	7.0	9.4	9.0		

(a) Harvard Business School, Case Study for Pioneer Hi-Bred Int., 1978.

(b) L.W. Teweles & Co., The U.S. Seed Industry, 1982-85 (1985 values in 1982 dollars).

Table 8. Estimates of commercial seed as a percent of total consumption in the United States.

Selected species	1975 (a)	1985 (b)
Sugar beet, Tobacco	100	100
Packet vegetables and flowers, Turf	100	100
Corn, Sorghum	95	100
Alfalfa, Clover, Forage grasses	95	95
Bean, Pea	85	95
Peanut	70	90
Rice	70	80
Cotton	50	70
Soybean	60	63
Barley	50	50
Wheat	35	43
Oats, Rye	30	26
Flaxseed	15	15

(a) Harvard Business School, Case Study for Pioneer Hi-Bread Int., 1978.

(b) L.W. Teweles & Co., The U.S. Seed Industry, 1982-85.

Final Remarks

The major seed companies hold patents for high-tech seed. Despite market trends, which might lead one to believe that the rich get richer, and the poor get poorer, there is a “golden pond” of acceptable quality seed that can be managed by public institutions and private companies through equitable agreements and profitable marketing channels. This is confirmed by the increasing number of inter-professional agreements, which, on a country-by-country basis, aim to set up innovative technological processes consistent with the economic development of agriculture.

A graph can be drawn comparing agricultural evolution from an economic (productivity, stability, and sustainability) point of view (Crosson, 1990; Ruttan, 1991), with technological innovation (green revolution, gene and biotech innovation, agro-ecology). Diagram 1 summarizes the most significant achievements of the seed industry over the last three decades, including high-yield varieties and hybrids, seed uniformity (vs. gene diversity) and environmentally friendly seed. The blank areas of the graph are filled in by the respondents, and the positive or negative impact of the matrix combination can be evaluated, taking into account the situation in more—or in less—developed countries.

The responses in Table 9 can be verified by wider empirical study of new seed varieties developed through the application of modern biotechnology (Kalter and Tauer, 1987; Offutt and Kuchler, 1987).

Diagram 1. Steps in economic development/technological innovation.

Economic Objectives	Technology Response		
	Green Revolution	Gene and Biotech Innovation	Agro-ecology
Productivity	High- yield varieties and hybrids	Uniformity vs. Biodiversity	Environment-friendly quality seeds
Stability			
Sustainability			

Seed industry development is heavily influenced by intellectual property rights (IPR) related to biotechnological applications in agriculture (June, 1992). Although still evolving, IPR legislation has a major impact in industrialized countries. The complexity of doing business is likely to increase significantly in the future. Since a new protected variety is likely to depend on many IPRs owned by different partners, it becomes more and more difficult to share the added value between these partners (Sehgal and Van Rompaey, 1992).

The impact of these problems on developing countries is minimal, since the seed industry is in the early stages of development. Adoption of IPR laws in such countries can stifle growth, rather than accelerate development. Developing countries are more in need of know-how. Their seed industries will be able to develop if there is appropriate transfer and absorption of technology from industrialized countries (Sehgal and Van Rompaey, 1992). Technology refers not only to seed, but also to agricultural practices and extension services, as well as to goals, depending on the level of economic development (as illustrated in Diagram 1 and Table 9). One Purdue economist (Doering, 1992) states, "International cooperation in this effort appears to be drying up, and other countries are launching their own efforts. Or, they are turning their cooperative international efforts into national ones. What impact will this have on the pace of scientific discovery and technology development in the 1990s? What will this new approach do to the structure and survival prospects of technology firms, such as those in the seed business? Have we been wrong to have large public investments in basic research and technology that was then available to all? These are critical decisions that will have great impact. How we respond to the very real internationalization of agriculture depends partially upon how we receive it. We can perceive it as something we can, at least partially, shape through our own actions."

Table 9. Test response by Amman workshop participants.

Economic Objectives	Technology Response (%)					
	Green Revolution		Gene and Biotech Innovation		Agro-ecology	
	Positive	Negative	Positive	Negative	Positive	Negative
Productivity	High-yield varieties and hybrids		95	5	62	38
Stability	65	35	Uniformity vs. Biodiversity		31	69
Sustainability	42	58	65	35	Environment-friendly quality seeds	

Country Typology	Technology Response (%)					
	Green Revolution		Gene and Biotech Innovation		Agro-ecology	
	Positive	Negative	Positive	Negative	Positive	Negative
Industrialized	73	27	92	8	50	50
Developing	35	65	68	32	42	58

Agriculture and the seed industry should develop so as to ensure a positive balance of natural resources, and, as regards the former, sustainability of productivity over time. The seed industry, on the other hand, should develop its technology in a manner consistent with the different agro-economic needs of the more developed and the less developed countries operating on the international scene.

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Formal and Informal Seed Programs

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Introduction

A seed program can be defined as "...an outline of measures to be implemented and activities to be carried out to secure the timely production and supply of seed of prescribed quality in the required quantity..." (Feistritzer and Kelley, 1978). Two important approaches to seed supply exist, i.e. formal and informal seed supply. The formal seed supply is carried out by the public sector, often with private seed enterprise assistance, while with informal systems farmers produce seed for themselves or their neighbors.

Both systems are needed and the one cannot substitute for the other. The formal system produces, releases, and distributes new varieties to the farming community. The informal systems ensure that resource-poor, low-income farmers also have access to seed and benefit from public and private developments.

Formal Seed Program

The main seed supply system is the official government seed program, assisted by private seed enterprises. This program is a complex and organizational concept and has several essential components that are strongly inter-related. The most important of these are: (i) variety breeding, evaluation, and release; (ii) seed production, processing, and storage; (iii) seed quality control; and (iv) marketing and distribution (Fig. 1). Each component must be implemented at the proper time and in the correct sequence, and every component is essential. If one component is not operating, the entire seed program will not work properly.

Experience in North America, Europe and other countries has shown that a policy whereby the private seed industry is encouraged to participate in the national seed industry can be very beneficial. Private seed companies will, however, only produce and distribute seed of crops to areas in which profits can be made.

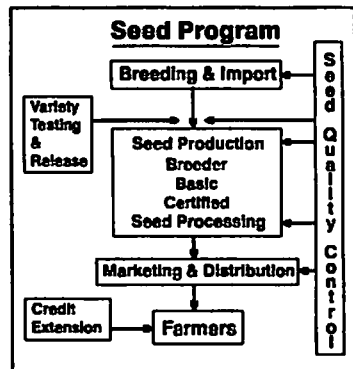


Figure 1. The most important elements of a seed program.

The most important elements have been described by Douglas (1980), Feistritzer and Kelly (1978), and Van Amstel and van Gastel (1986) and are summarized below.

Breeding

A developed seed program is supported by strong breeding activities, because production of quality seed of traditional varieties seldom generates sufficient benefits to the farmer to compensate for the increased seed cost.

The breeding program produces small quantities of what is called breeder seed. This seed is the parent material for further multiplication and the source of all certified seed.

At the initial stages of seed program development, varieties often originate from screening international collections in national trials. Imported seed (either basic or ready-to-use certified seed) is an alternative. At later stages, the national breeding programs develop new varieties for the country, at least for basic food crops.

Variety Testing

A seed program evaluates new varieties before they are released to farmers. Varieties are evaluated in field tests for about three years. In such experiments, which are conducted in the various ecological zones of the country, the agricultural value of new varieties is compared with that of existing ones. Such experiments are often carried out at different management levels under different farming systems.

When the variety number increases and the program advances beyond the initial stage, accurate variety descriptions are made. This is particularly necessary when seed certification or a system of plant breeders' rights is implemented. Varietal description is also necessary for maintenance breeding and consumer protection. In addition to agro-ecological value, varieties are tested for distinctness, uniformity, and stability (DUS tests).

Variety Release

Because governments wish to have control over the varieties multiplied (to ensure that only superior varieties are grown in the country), in well-developed seed programs, one or more national variety release committees advise on release of varieties, based on the results of performance trials and DUS tests.

Variety Maintenance

The purpose of maintenance is to produce new lots of breeder seed with the same genetic composition to start a new multiplication cycle for certified seed. It is the breeder's task to maintain the variety once it has been released. Variety maintenance is the basis for pure and disease-free seed for farmers.

A special procedure is necessary to obtain quality starting material for the multiplication cycle. For cereals, this involves collecting individual ears, planting them in single ear rows, selecting the true-to-type rows and bulking them up to constitute breeder seed (often lines are kept separate for a further generation). Through this procedure it is possible to obtain varietyally pure and disease-free breeder seed, which, if properly multiplied, will result in sufficiently pure and disease-free certified seed.

Seed Multiplication

The small amount of breeder seed is multiplied a number of times to produce the large quantity of certified seed needed to satisfy farmers' requirements. The breeder seed is first multiplied to produce basic seed; this in turn is used to produce certified seed. The certified seed (or the next generation) is sold to farmers for commercial grain production.

Throughout these multiplication cycles, a high level of genetic purity and disease-free seed is sustained to guarantee a high quality end product. Standards are slightly stricter for early than for later generations.

To prevent genetic change, the early generations are grown in areas to which the variety is adapted. No selection is attempted, other than roguing off-types. The best possible agricultural practices and inputs are applied.

Processing and Storage

Careful processing is very important in producing quality seed. This step is the most capital-intensive component of the seed program. All seed requires processing; the seed is cleaned, graded, sized, blended, treated, and packaged. In developed seed programs the entire process is a complex, largely mechanized operation using relatively sophisticated equipment.

The processed seed is stored in seed stores protected against damaging environmental conditions such as high temperature and high seed moisture content, under which viability rapidly deteriorates. In the humid tropics, where conditions are particularly unfavorable for seed storage, the investment in drying equipment and seed stores is very high.

Marketing

Marketing is vital for improved varieties to reach the farmer. The seed should be of the right quality and available at the right time, in the required quantities and at a reasonable price. Required inputs must also be available.

In countries with a developed seed program and a strong private seed sector, certified seed is marketed to the farmers through a highly organized and effective distribution network involving wholesalers and retailers. Such an organization, which takes many years to evolve, also promotes the sale of seed and helps to forecast accurate market demand, which is essential for production planning. The success of a distribution network depends largely on the selection of competent retail dealers.

Seed Quality Control

In developing systems, the seed quality control service is a central unit that carries out control at most stages. It plays an important role in ensuring that the seed is of high quality. In developed seed programs, the agency is usually not linked to the seed production organization, but functions as an independent governmental or semi-governmental organization, directly responsible to the country's Ministry of Agriculture.

Seed quality includes seed testing, seed certification and seed legislation. Aspects of quality are purity, germination, health, weed seed content, moisture content and other characteristics.

Seed testing

Seed testing is often the first step in enhancing the quality of the seed. The testing can be minimal (only for germination), or very extensive (for moisture, purity, germination, health and other characteristics). Either one national or several regional testing stations can be established. Countries should follow the procedures of the International Seed Testing Association to facilitate ISTA membership at a later stage.

Seed certification

Seed certification ensures that the seed sold to the farmers is of the indicated variety, sufficiently pure, of good germination capacity, and disease free. Certification in developed seed programs includes:

- Field inspection to verify seed source, varietal identity, previous cropping, isolation distance, impurities (off-types, weeds, other crops and/or varieties) and disease. In many less-developed certification schemes, the field inspector also assesses other aspects of seed production.

- Seed inspection at the processing plant and in the seed store. Seed samples are taken and tested at the seed testing laboratory.
- Pre- and post-control plots grown on the farm of the certification agency to allow additional verification of varietal identity, varietal purity, and seed-borne disease. Pre-control plots are grown in the same season as the main seed field, and results are used for certification. Post-control plots, grown from seed that is already certified, function as checks on the effectiveness of field inspection.

Seed legislation

Seed quality is more difficult to judge than that of any other commodity. Seed with very poor germination capacity for example, may appear good, but if planted, the farmer loses both his investment in the sowing seed and the entire value of the expected harvest. In developed seed programs, seed legislation regulates the various steps of the seed program to protect the farmer, using one of two different regulatory mechanisms:

- **Minimum standards:** Specified for all seed allowed in trade. Substandard seed is excluded from the market. This approach is often used in countries where farmers are not sufficiently educated.
- **Truth in labeling:** All seed is allowed to be marketed, but the quality is indicated on the label. The truth of the label is ensured by the quality control agency.

Other Considerations

A successful seed industry also depends upon many other outreach mechanisms such as extension, and the education of farmers in the benefit and use of improved seed. This topic is, however, completely neglected. Extension services are often the weakest programs in many developing countries; in some countries it is even considered a demotion to be transferred to the extension service. Extension services not only have the task to “bring the message” of quality seed, but must also handle many non seed-related issues. Extension services have to convince farmers to use—and educate them in the use of—improved technologies. Improvement of extension efforts is needed in many developing countries.

Often, a system of credit facilities must be established to enable farmers to purchase improved seed and complementary inputs.

It is also important that markets exist to absorb the increased yields resulting from the use of new seed.

Informal Systems

Informal systems, whereby farmers produce seed for themselves or for neighbors (Fig. 2), can play a very important role in the national seed supply system. Informal seed production systems also play a complementary role in producing seed of improved landraces, populations and mixtures. Certainly in high risk agricultural areas small farmers are not interested in uniform, homogeneous and often environment-specific varieties. Improved landraces, populations, and mixtures play an important role. Seed organizations are usually not interested in producing seed of such varieties. They have problems with uniformity, distinctness, and stability; existing standards are difficult to apply.

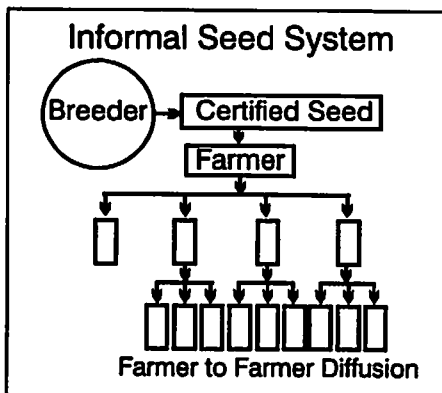


Figure 2. Alternative seed supply.

For ages farmers have been able to successfully save seed, and in North America, wheat farmers still typically meet 90 percent of their seed needs from the previous year's crop (McMullen, 1987). Stanelle *et al.* (1988) show that in Kansas, home-grown seed accounted for 68 percent of seed used for wheat plantings.

A clear description of a situation where both systems are operating is described by Tetlay *et al.* (1991) with regard to wheat seed in Pakistan. The major source of seed planted by Pakistani farmers was own-saved seed (55–62 percent), followed by seed obtained from other farmers (21–27 percent). The percentage of seed obtained from the national seed industry's seed depots was less than 10 percent. The seed industry (and other farmers) was generally the source of seed for new varieties. New seed often entered a village through the larger farmers, from whom diffusion took place to other farmers in the vicinity. Seed diffusion was highly localized.

Other examples of attempts to strengthen the informal seed supply system follow:

- Although the Indian seed industry has developed rapidly during the last 15 years, about 80 percent of the seed is still traditionally supplied. In view of the low replacement rate (8 percent) for wheat and rice, the traditional seed supply is and will remain important. In the 1960s the seed village was initiated through field demonstrations and seed multiplication in Jyonti village in Delhi State. Mobile seed cleaning plants

were made available by the local village council or by the state's department of agriculture. One of the most important constraints faced by small farmers was unavailability of seed of improved varieties at sowing time, since all seed was usually sold out soon after harvest because farmers were in need of cash. Bank credit has partly solved the problem (Agrawal, 1990).

- In Tanzania, TANSEED meets less than a third of the actual maize demand, and seed produced is all hybrid-based and too expensive. Unfortunately, the seed industry did not produce a competitive composite, suitable for the highlands, and from which seed could be retained for 4–7 years. Therefore, a village-based seed production approach was proposed, whereby small farms would be supplied annually with foundation seed from research stations. During the season extension advice would be given by a seed control unit. Furthermore, farmers would be trained. At a later stage farmers could take over maintenance and become self-sufficient (Friss-Hansen, 1989).
- The Malawi government initiated a small holder seed multiplication scheme for ground nut. Each small holder was supplied with enough foundation seed for 0.4 ha. A decentralized seed control unit inspected the crop and, after harvest, accepted or rejected it. Unfortunately, the program was only seen as a medium-term solution and it was anticipated that the normal seed program would take over in the long run (Friss-Hansen, 1989).

These systems should be given more attention because they produce a very large amount of seed. In developing countries they will probably play a larger role in the coming years, because national seed organizations will not be able to cope with the increased demand for quality seed.

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Seed Legislation

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Introduction

The development of improved agriculture is an indispensable step toward a better living standard. It depends upon a number of improved inputs, of which seed is the most important. Through plant breeding, seed can become the best technology available to the farmer. In the age of computers, plant breeders are rightly regarded as one of the most sophisticated programmers, especially when they make use of genetic engineering.

Seed is both the cause and the effect of a given level of technology. When attempting to introduce a new technology, a new machine, a new irrigation technique or a different crop management, one of the critical aspects is often the availability of seed. If the varieties do not meet the required standard, farmers' attempts at technological change may be in vain.

The replacement of landraces or ecotypes by varieties obtained through plant breeding marks the beginning of a process in which new varieties and techniques interact to increase yield and other outputs. This is a process in which plant breeding must provide the motive force. It is generally accepted, but difficult to prove, that variety improvements have accounted for at least 50 percent of the spectacular increases in production which have been obtained in many crops in recent decades.

The cost of seed in most EU countries accounts for about 3.5 percent of the cost of intermediate inputs (pesticides account for 4.5 percent, fertilizer 13.8 percent, and energy 11 percent). The relative low cost of seed and its effect on production show that it is wrong to try to save on seed costs, and at the same time, important to develop a strong and efficient seed production structure (Lorenzetti, 1988).

Seed Law Evolution

In primitive agriculture the seed that is sown is either from a previous crop or from the farmer's neighbor. In such a situation, seed legislation is not necessary.

Seed produced on the farm can be used in developed agriculture, especially for varieties whose genetic structure is not modified during subsequent generations of multiplication (pure lines and, to some extent, synthetic varieties). It is generally accepted that for autogamous species, seed of improved varieties can be multiplied and used on the farm for three to four generations. In these situations it is impractical to control and ensure that the seed sown is of high quality.

The objective of seed legislation is to regulate seed commerce. In the early stages, seed legislation tends to control agronomic characters of seed lots (mainly germination and purity). Later on, with advanced plant breeding, seed legislation also includes variety certification and variety protection.

General Principles of Seed Legislation

The purpose of a seed law is to protect the farmer against unwitting purchase of poor quality seed. Seed quality is much more difficult to judge than the quality of other merchandise. For example, it is impossible to judge, by sight, the germination capacity of seed.

Seed is also different from other forms of merchandise because poor quality is not only confined to the seed itself, but can result in the total loss of a crop, and perhaps a year's livelihood for the farmer.

Experience shows that without a legislative framework, the seed industry cannot progress satisfactorily. Progress requires safeguarding farmer interests and protecting seed producers and merchants from unfair competition.

Seed legislation aims at promoting the overall development of agriculture, but it does not guarantee that quality seed reaches the farmer. Seed laws can only achieve their aim if high-quality seed is available. Seed laws must be enforceable and must fit the social, economic, and judicial make-up of the country. A single comprehensive seed legislation model does not exist.

There are two alternative systems of seed legislation. The first is the *comprehensive regulatory system*, wherein the law prohibits the sale of seed that does not meet a minimum standard of quality. In its extreme form the system requires a list of cultivars, and only certified seed of registered cultivars can be offered for sale. Seed producers and trader companies must be registered in order to do business. The second is the *truth in labeling system*, wherein the seller must provide correct information about the seed to be sold. There are no restrictions on quality.

Either of these two philosophies can be followed by seed legislators. In practice, each country's seed legislation has its own peculiar characteristics.

Laws and Regulations

Since the development of a seed industry is an evolutionary process, seed legislation must be conceived in such a way as to regulate existing practices. In general, seed legislation consists of laws and regulations. The basic law describes the institutional framework, which is implemented by an administrative body which issues specific regulations.

This approach gives stability to the basic principles of seed production and trade, and at the same time permits the flexibility necessary for meeting the requirements of particular crops and/or new situations. The basic law establishes the object and scope of seed legislation. Laws of different countries point out specific objectives, but, in general, cover the following:

- Guarantee the identity and quality of seed sold to farmers.
- Stimulate the production of high quality seed and promote its use.
- Ensure the supply of seed in the required quantity, and improve quality.

Regulations are set up by the authority named by the law, and are intended to regulate the major aspects of the law, particularly those of certain crops. It is not advisable to incorporate too many details into the law, because they may need to be frequently adjusted or amended as new situations arise. In fact, emergency situations may require the suspension of some market regulations.

Basic Concepts and Terminology

Seed legislation should be clearly presented. Technical terms should be precisely defined to avoid controversy.

The first term to be defined in seed legislation is *seed*, which in some cases relates strictly to its botanical meaning, but more often includes vegetative means of propagation, comprising practically everything that can be planted. In some cases the term also includes seedlings.

Variety, in general, has the meaning assigned to *cultivar* by the Commission of the International Association of Biological Science on the Nomenclature of Cultivated Plants: "...an assemblage of cultivated plants which are clearly distinguished by certain morphological, physiological, cytological, chemical or other characteristic of agricultural or economic significance which may be perpetuated by reproduction."

Other terms that can be defined concern seed quality: *germinability*, *purity*, *noxious weeds*, *moisture content*, and *1,000 seed weight*.

Registration or Licensing of Persons or Firms Engaged in the Marketing of Seed

In many countries seed sellers must register with a trade registry. The acceptance of the application is subject to their technical capability and financial status. They are required to keep records of their activities. Once accepted, official registration is by a central authority or, more often, a regional authority. In Italy, the chamber of commerce of each province is in charge of registration.

Seed Testing Institutes and Laboratories

Because seed legislation aims at providing farmers with high quality seed, testing of seed available on the market must be carried out.

Factors of seed quality include: trueness to cultivar (genetic purity), mechanical damage or injury, germination, disease infection, insect damage, treatment coverage, size, moisture content, seed purity, test weight (hectoliter weight, 1,000 seed weight), as well as the presence of noxious and common weed seed, other crop seed and inert matter. Standard seed testing includes a specific set of tests which determines the level of the quality factors.

At the heart of quality control is the seed testing laboratory. In each country one or more seed testing stations may be specified as official stations. Non-official stations can be authorized to carry out tests for particular crops or under specific conditions. The stations in a country should all follow the same testing methods. Normally, the International Rules for Seed Testing defined by the International Seed Testing Association are used.

Seed Certification

Seed certification is the process which documents that the seed in a sealed container fulfills the characteristics required by seed legislation and/or as indicated on the attached label. An official certification scheme is based on:

- Approval of cultivars based on tests for distinctness, uniformity and stability. The EU scheme also requires that the cultivar have superior agronomic value to existing varieties, at least in one region of the country.
- The recommendations of an advisory board, which suggests to the Ministry of Agriculture which cultivars should be approved.
- A statutory list of approved cultivars, on which a cultivar is registered for a specific time (generally 10 years).

- Before a cultivar is listed, its name should be approved to avoid confusion with other cultivars. The International Code of Botanical Nomenclature for Cultivated Plants states recommends that:
 - Numerals and symbols should not be used.
 - Names should not exaggerate the merits of the variety.
 - Names should consist of no more than two words.
 - Names should be simple, easy to pronounce, and unlikely to be misspelled.
 - Names should not be reused for at least for ten years after a cultivar has ceased to be listed.
- The seed categories to be marketed under the certification scheme are defined as:
 - Basic seed (the progeny of breeder seed), the production of which, under the EU scheme, is the responsibility of the breeder.
 - Certified seed is produced from basic seed for one generation (certified seed of first reproduction) or two (certified seed of second reproduction). In the USA the terms foundation, registered and certified are used for the first, second and third generation following breeder seed.
 - Commercial seed is certified as being true-to-species. No cultivar purity is guaranteed
- During production and marketing, a special control operated by a state agency guarantees that seed is of the required quality. The certifying authority can only certify that, to the best of its knowledge, the various rules have been followed, and that the seed meets the prescribed standards. The producer must ensure that the rules are followed when the authority cannot be present.

Legal Protection for Crop Varieties: Plant Breeders' Rights

New varieties can be produced by public institutions or by private individuals or firms. In France and Germany, public institutions tend to concentrate on basic research, leaving the task of obtaining improved varieties to private firms, while in other countries (Britain, Sweden), public institutions also produce new cultivars for direct release to farmers.

The cost of developing new cultivars—low at the beginning for certain crops because the process is very simple—has tended to increase with time. Private breeders recover these costs through the sale of seed, and they find it increasingly difficult, as the cost of varieties increases.

Recovering the cost of new cultivars depends on the genetic structure of the variety. For F_1 hybrids, the breeder has a monopoly if he or she produces the inbred lines. For varieties based on pure lines (self-pollinating crops), the breeder loses control as soon as the variety is released. He can only recover his costs by charging for the original release, or for the basic seed required to start new cycles of multiplication.

Incentives to promote private breeders, and their drawbacks, include:

- The cultivar name is registered as a trademark. This has the effect of protecting the name rather than the cultivar.
- Subsidies are granted to breeders who produce registered cultivars. The subsidies, however, do not take into account the cultivar's true value.
- Varieties are patented and protected as industrial products. However, living organisms normally show phenotypic variability, and the act of creating the same variety is never repeated.
- The variety is protected by controlling its use. A variety resembles a musical work more than a mechanically manufactured product. A musical composition is created once, but not freely available to the public. The protection of new varieties should therefore follow existing copyright schemes (plant breeders' rights). Royalties should be paid by the users in proportion to the quantity of seed used.

The concept of plant breeders' rights was first introduced in an international convention signed in Paris in 1961. To be protected, a variety has to fulfill the following requirements: (i) distinctiveness; (ii) uniformity; (iii) stability; and (iv) no previous commercialization.

The protection lasts for a period not less than 15 years, and the royalty payments are, in general, calculated on the quantity of seed sold. An international union for the protection of new varieties of plants has been established (UPOV: Union Internationale pour la Protection des Obtentions Vegetales) in Geneva, made up of countries adhering to the convention. UPOV coordinates the administrative and technical procedures followed in member countries for the granting of rights.

The original convention, signed on 2/12/1961, has since been amended.

Imports

Control of imports is often included in seed legislation to protect the national seed industry and farmers, or for quarantine purposes. Imported seed must comply with the laws and regulations applied to home-produced seed.

Quarantine restrictions may refer to seed-borne disease and pests and to weed seed. If improperly used, they can distort the international seed trade. In the USA, for example, imported lucerne or red clover seed must be stained to indicate that it is of foreign origin; a similar rule has been applied in Italy. The aim is to protect farmers, because imported seed of these species is generally not adapted to new environments.

Infringements of Seed Legislation and Penalties

After a seed act has become law, certain authorities should be empowered by the government to implement the seed law. Generally speaking, the competent authority for seed matters is the Ministry of Agriculture. Sometimes the seed authority has only an advisory function.

Coordination between the Ministry of Agriculture and seed producers, tradesmen and users is achieved through the representation of various organizations. Law enforcement is assured by a limited number of seed inspectors. In many cases the inspectors are civil servants who are responsible for the collection of samples and laboratory analysis.

Some seed laws require special control of goods entering and leaving the premises of seed producers and merchants. In most cases the main infringements of the law are explicitly mentioned. Sometimes a general statement is made to the effect that any act of contravention is deemed to be an offense punishable with specific penalties, which usually take the form of fines and confiscation of the seed involved in the offense.

Step by Step Development of Seed Legislation

In primitive agriculture, there is almost no commerce in seed, and seed legislation is not required.

When seed is sold at the village level, laws can only be concerned with characters such as purity, mechanical integrity and germination, for which minimum standards can be set.

As improved varieties become available and agricultural development takes place, a more sophisticated seed trade develops, and a stronger seed legislation is needed. At this stage labeling of seed for quality is the first step; labels must truthfully represent the actual seed quality. Seed testing laboratories must be available, and farmers and seedsmen should be encouraged to use these services. As seed trade gains sophistication and becomes more intricate, seed legislation tends to become more complex and restrictive.

The last steps in seed legislation include seed certification and regulation of plant breeders' rights.

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Plant Breeders, Seed Production and Distribution—Who is the Boss?

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Introduction

This paper contributes to the connection between plant breeders and the seed industry in the broadest sense.

We shall emphasize the need for a professional seed industry, the demands on seed production and, finally, the necessity for a proper seed distribution system if the farmer and ultimately the consumer are to benefit from the plant breeder's work.

Plant Breeders

The plant breeder working in food crops has as his or her first priority the increase of crop yield. In food crops the relation to seed yield is often direct; a variety with a higher grain yield, for instance, is a variety with better seed yield. This is not necessarily so with other crops, such as hybrid maize, vegetable or forage crops. The issue becomes quickly more complicated when breeders' aims and objectives are concentrated on other characteristics. When high lysine maize became a topic 25 years ago, it appeared that the seed quality of this maize was poor—so poor in fact that the seed could hardly be sold under the high quality label.

Numerous examples can be quoted in which plant breeders "overshot the mark" by creating a new variety or hybrid which could not be economically reproduced on a large scale. There are also positive examples of certain characteristics contributing towards good seed quality—straw strength, for instance. A heavy but lodged crop can, under adverse weather conditions, produce a crop the grain of which is not suitable for sowing. Therefore, straw strength generally contributes towards good seed quality, all other conditions being equal.

From the point of view of the seed industry, the difference might be one between life and death or, economically speaking, between bankruptcy and prosperity. For this reason it is worth asking who is in charge, who decides on priorities, or, in brief "who is the boss." The answer to this question is not as simple as it sounds.

In a public institute, national or international, the man or woman in charge is usually clearly identified by means of an organizational chart. The goals for a plant breeder are more often than not complex, and therefore his or her role is not so clear. An increase in yield of 10 percent may be possible after the work of several seasons. But a 10 percent yield increase in combination with a number of other characteristics may be extremely difficult. Therefore, the priorities and the criteria must be relevant, particularly now that economic realities increasingly force us to take chances. When the budgets of plant breeders are under scrutiny, and the difference between publicly or privately funded programs becomes small, the plant breeder's priorities can become urgent, and the "scientific freedom" to maneuver may narrow. Under this kind of pressure one will increasingly hear the question: "Who is in charge here?"

Despite the stories about surplus food production in some parts of the world, in areas where food is really needed, the demand to obtain higher yielding varieties remains unaltered. In addition, the following battle cry is more and more frequently heard: higher levels of food production should not come at the expense of the environment. Environmental concerns are diverse, ranging from the threat of genetic erosion to the prohibition of the use of certain crop production chemicals and even some fertilizers. This adds to the complexity of the breeders' objectives, and to the urgency of the question of who decides those objectives.

In practice, the answer is probably not as complex as in theory. Plants can only be manipulated so far, and breeders' priorities will ultimately be set by what is technically possible and economically feasible. These two criteria are difficult to fulfill. Plant breeders should therefore concentrate on those aims that are practically achievable, creating varieties which are of practical use. To achieve this, they have to stay in touch with the wishes of the consumer. To do this, they have to occasionally leave their offices and breeders' plots to ensure that they are still in touch with the market.

This is not to say that the breeder should not stay within his limits; breeders should not, in my opinion, get directly involved with seed production, processing and marketing, since when they do, their breeding work will suffer. In a seed company organized as a commercial enterprise, all job descriptions are well-defined and usually well-balanced. When plant breeders work on a specialized breeding station, this balance is not so natural.

Seed Production

Seed production deserves careful and professional attention. The reason is clear: seed has to be kept healthy and alive under conditions that are often far from favorable. Threats to seed health come from many directions. After treatment by humans comes attack by animals, insects and vermin, heat, moisture, etc. Since all those threats are real, we have successfully developed all sorts of techniques—some very scientific, others just clever, some simple, others very complicated, some very cheap, others very expensive—and the application of these techniques greatly advances a successful crop. This must be foremost in our mind. Seed production is never a purpose unto itself, it is not a “final destination.” The purpose of seed production is to produce a crop for the ultimate benefit of humankind.

It is not necessary to dwell on the details of seed technology here. I would like to discuss some of the techniques which ought to be used in deciding how much seed should be produced, this being the most difficult task surrounding the issue of seed production. Many different models have been developed and used.

But most people, one must conclude, are not very good at this particular job. Many years of experience have shown that it is no good trying to plan exactly the right quantity of seed. First we have to ask ourselves, who within the organization is responsible for seed production, and should we try to produce too little seed or more than we need? One can never produce the right quantity—if one does it is luck, not judgment.

The basic question therefore—too much or too little—is relevant; both have pros and cons. Both are also heavily dependent on circumstances. For example, in northwestern Europe, where rainfall is not a limiting factor, seed production is quite reliable. One does not have to plan for a large carry-over in annual crops. The same could be said if seed production were done under irrigation. If growing conditions are manageable, seed production can be tuned accordingly. Given the risk and costs of storing seed under any circumstances, the advice is clear—do not produce too much. However, particularly with food crops, there may be compelling reasons to plan production in excess of foreseeable need. Some reasons for this may be technical, others may be political. If weather conditions, for instance, are not reliable, it may be decided to produce a slight excess, thus aiming for a surplus. This is in order as long as one counts all the potential costs of doing so.

Another reason for more production is political. In some countries the whole food production chain may steal the political limelight and, in times of serious shortage, seed supply may be pointed at as the culprit. This can

happen if only a few farmers claim that they could not get seed in time, even if those farmers have never bought seed in their lives.

This is one of the main causes of the falling supply of open-pollinated crop seed in marginal areas. In good crop years most farmers will use their own farm-produced seed again next season. In poor years the whole crop is eaten or sold, and when planting time comes again, there is a run on certified seed supplies and inevitable complaints of shortages.

This cycle is bound to repeat itself, as the author has experienced. Still, the risk of over-supply is on balance far greater than the risk of shortage. Many more seed production schemes have withered for reasons of unsold stock than for being regularly sold out.

The boss again makes the final judgment and carries the responsibility. It has to be clear, therefore, who is in charge: the technician, the entrepreneur or the politician.

Distribution and Sales

Strange as it sounds, this may be the most difficult area of the entire seed industry. This is not hard to explain. For all the earlier mentioned links in the seed chain, basic techniques have been developed and can be applied virtually everywhere, as long as the people and the capital are available. Plant breeding, seed testing, storage, etc. can be carried out with known and well-proven methods. There is, however, no single technique or basic rule of thumb when it comes to seed distribution. On one basic principle we all agree: without effective distribution, the spreading of the plant breeders' efforts becomes problematic.

Quality

Seed must be of high, reliable quality when it is used for planting, not only when it is tested in the laboratory. Therefore conditions during transport and storage at the distribution point must be of high quality so that the product does not deteriorate in the final stretch. (Remember, it may have taken many years to arrive at this point.) Rain or heat during this period may prove disastrous.

Packing

The packing of seed deserves a lot of attention. Packaging must be strong and of the proper size but, on the other hand, need not be luxurious.

Treatment

The treatment (or dressing) of seed is the subject of much controversy. In the past, methods varied widely from smoking to hot water treatments. Currently, chemicals are generally used, and it is precisely these chemicals that are so much discussed and in the end forbidden. DDT, once the savior of the human race, was declared a killer and is now almost forgotten. However, effective and low toxicity seed treatment chemicals are still available and should be applied when necessary, firstly to protect the seed when it is still in storage and secondly when it is planted. Both are equally important.

Time of Delivery

It sounds so obvious to say that seed has to be delivered on time. The success or failure of a crop may have a direct link with the time of planting and therefore it is essential that the seed is available to the farmer before the optimal planting date.

Pricing

Seed should be sold at the right price. This statement sounds familiar and will immediately be followed by the question of what is this right price. This question too is not difficult to answer. The right price to the farmer is a price he or she can afford and, more important, which will be to his or her benefit. For a long time the idea prevailed that most farmers could not afford the purchase price of good seed and therefore the price had to be kept low by some form of subsidy.

There is only one valid argument against this: subsidies cannot be guaranteed to last. Therefore, when they are withdrawn, the carefully constructed seed distribution network may collapse, at least temporarily. Having lived through this experience, attitudes have changed.

In seed distribution, the options vary widely and depend very much on the crop, the region and on general local circumstances. Whatever the variations, a few well-tryed principles apply.

Payment

Under all circumstance, the method of payment for seed must receive due attention. Local commercial practices will have to be considered when deciding on a payment policy. Whatever method is decided, administrative procedures will have to be kept at a minimum, otherwise the cost of administration may become a burden on the whole exercise.

Planning the Distribution

Seed distribution and sales should be a carefully planned and coordinated exercise, receiving as much attention as seed production. This is often not the case, and therefore distribution goes wrong for reasons explained earlier. There is no basic technique available, and therefore distribution requires invention and flexibility and a lot of management attention.

Management and Ownership

In this final section we shall pay some attention to the question of who is in charge. It is perhaps easier to answer the question who should *not* be in charge. In this respect, attitudes have changed substantially during the last decade or so. Virtually every interested party will now answer that this is a question of ownership, not government. Or, as I read the other day, the job of the state is neither to fix nor to stabilize prices, but to create conditions of free competition. As far as seed supply is concerned, one could add, provide supporting services such as independent seed testing and related technical services.

It would be simple to conclude that the seed industry will prosper as long as governments do not get involved. There is no easy solution available, only a sound understanding of the needs and opportunities. Recognizing the limits and the problems will usually lead to the solution.

This solution will often consist of a combination of publicly funded research and private initiatives of production, processing, storage and distribution supported by a commonly available infrastructure. When all those resources are put to proper use, one can speak of a successful seed enterprise, and the question of who owns all those assets becomes less relevant.

The synchronization of the work of plant breeders, seed producers and distributors needs careful planning and management, and the final answer as to who's the boss can be heard loud and clear: the market.

Private Seed Companies and Public Seed Program

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In many places where private companies are lacking or are rare, people think that such companies are best able to solve all seed business problems. If that were the case, the number of new seed companies would explode in a short time. This does not occur. In this paper, a clearer picture of the role of private seed companies in seed program development is presented.

Companies need good organization, with integrated research and development for seed production, marketing and sales. The necessary components are listed below.

R&D aims:

- Develop a breeding plan based on market requirements and maintain genetic resources.
- Improve genetic variability.
- Develop experimental varieties.
- Control stock seed.
- Control the parent lines for F₁ hybrid seed production.
- Organize internal and external screening and market-oriented trials.
- Describe and register varieties.

Production guidelines:

- Demand healthy and pure seed sources.
- Demand information about the parent lines and the hybrid.
- Know how to multiply seed, including synchronized flowering, pollination requirements, seed/plant health requirements, nutrition requirements, and harvesting technology.
- Develop a production manual including the conditions for (sub)optimal production.
- Search for appropriate production locations.
- Search for qualified production personnel.
- Create a network for new production technologies.

- Develop facilities for testing, processing and bagging.
- Develop field inspections including health assessment.
- Develop facilities for seed quality control.

Marketing needs:

- Product policy.
- Product form policy.
- Stock policy and variety protection.
- Price policy.
- Introduction and variety life cycle policy.
- Packing and distribution policy.
- Knowledge of national and international market developments.

Sales needs:

- Knowledge of products.
- Knowledge of procedures and rules (export/import).
- Knowledge of needs and desires of clients.
- Good, adequate distribution system for each country.
- Price conditions per variety per country.
- Good overview of actual production situation and of the stock situation.
- Enough clients/customers to economically and profitably carry out all these activities.

Is it possible to have all of the above in one company? Is it different for a public program? A good working situation has developed in the Netherlands (Van der Burg, 1986). However, original basic breeding activity of government institutes has ended. Private companies have taken over that area of research.

In several countries, there are private companies and public programs. Public activities oversee the seed program. Small private companies should get more support, but the public sector should always be aware of the possibility of minimizing its support. A minimum level of support by the public sector is needed to create a competitive environment for private companies.

In addition to the above, there should be clarity about who will carry out research on breeding methods, protection, post-harvest activities such as storing, treatment, and testing. Where the stimulation and control of the public sector starts is still open to debate; inspection and certification could

be a self-supporting activity of the private sector, but are usually carried out by the public sector, at least in developing countries. Research should be carried out by the private or public sector, or by both sectors in partnership.

An example of this can be seen in Egypt. In 1975, Pioneer started its activities. They developed so well that by 1985 a commercial organization, 70 percent owned by Pioneer and 30 percent owned by local Egyptian investors, had been established (Rihm, 1989).

Reasons for cooperation in technological investigation are: shared cost, shared risk, additional technological knowledge, as well as market know-how to serve an international market and the development of (i.e. European) industrial standards.

Cooperation should not be competitive, and both partners should have an equal interest. Partners should agree upon goals, and on how to reach them. Partners should also have the same level of commitment.

For successful cooperation in technological innovation, all parties should have a joint interest with no contradictions. Public organizations are also needed in an integrated network to regulating seed programs. Public breeding research must be stimulated, with priorities and research aimed at meeting the needs of private breeding firms, farmers and growers.

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Section II

Seed Physiology

Apomixis and Seed Production

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Apomixis: Definition and Main Implications

Apomixis is a mode of plant reproduction. In its broadest, most comprehensive meaning, it indicates "reproduction without sexual fusion." Therefore, it refers both to vegetative reproduction in all its forms (bulbs, corms, tubers, runners) and to *agamospermy*, which is the production of fertile seed (not vegetative organs) without fusion between an egg and a sperm: "seeds without sex." But, if no sexual fusion occurs, where does the embryo come from? It derives from mitotic division of a nucleus in a somatic cell of the ovule. If the derivation of the embryo is from a somatic cell, it follows that its genotype is the same as the sporophyte. Furthermore, all embryos derived from the same plant will share exactly the same genotype—the maternal one—and can be considered clones. This has enormous practical implications that will be discussed later on.

Although vegetative reproduction is in theory a form of apomixis, this term is in practice most often referred to as *agamospermy*. Thus, in this paper, apomixis will be used as a synonym of *agamospermy*.

Apomicts—those genotypes that reproduce apomictically—can be either obligate or facultative. In the latter case, reproduction can occur both sexually and apomictically, even in the same ovule. No such possibility exists for obligate apomicts, where no hybridization, i.e., sexual fusion, takes place. In obligate apomicts, mutation remains the only alternative to generating variation, although it will be seen that, in reality, sexual plants can be found in most apomictic species.

Darlington (1939), in a dramatization of the genetic status of obligate apomicts, wrote: "Apomixis...is an escape from sterility...into a blind alley of evolution." Apomixis is the absence of meiosis, so there is no chance of forming sterile gametes, genomically unbalanced, derived from irregular meioses. Those gametes are normally selected against (meiotic sieve). Therefore, reproduction through apomixis is a guarantee against what amounts to agametic waste, allowing more effective reproduction. However, the absence of meiosis in truly obligate apomicts would also prevent any release of new variability, thus leading to the "blind alley of evolution."

Without meiosis there is no variation and no capability to adapt. It is either extinction or blind conservation.

Apomixis and Breeding

Sexual plants can be observed even in species thought to be completely apomictic. Thus, apomixis is better considered not as a hindrance, but as a plant breeding tool in an overall system characterized by:

- Generation of variation, at least in the offspring of sexual plants, with the consequent possibility for the species to adapt by changing.
- Efficiency of reproduction, as with vegetative organs.
- Convenience of propagation, as with seed (dispersal in space, dormancy, etc.).

Distribution of Apomixis

Apomictic reproduction has been observed so far in at least 30 families of higher plants, including more than 300 species. Apomixis is more than a sporadic occurrence. For instance, it plays a crucial role in the reproduction of *Citrus*, several berries, guayule, numerous perennial forage grasses, etc. Furthermore, it has recently been reported in grain sorghum and pearl millet. Following is a more comprehensive view of genera and species showing apomixis, subdivided according to specific mechanisms of apomixis considered in this paper.

Adventitious embryony

- *Citrus* (most species and hybrids)
- *Mangifera* (mango)
- *Spathifillum*
- *Capparis* (caper)
- *Eugenia* (rose apple)
- *Nigritella*
- *Garcinia* (mangosteen)
- *Euonymous*
- *Pachira*
- *Opuntia*

Apospory

- *Poa pratensis/alpina*
- *Panicum maximum*
- *Potentilla argentea*
- *Dicanthium/bothriochloa*
- *Cortaderia* spp.
- *Paspalum* spp.
- *Hieracium sub. pilosella*
- *Pennisetum* spp.
- *Urochloa* spp.
- *Malus* spp.
- *Cotoneaster* spp.
- *Crataegus* spp.
- *Crepis* spp.

Diplospory

- *Antennaria*
- *Erigeron* spp.
- *Taraxacum*
- *Limonium*
- *Ixeris*
- *Elatostema*
- *Chondrilla*
- *Arnica* spp.
- *Balanophora*
- *Rudbeckia*
- *automictic Rubus*

Mechanisms of Apomixis

Apomixis can follow several mechanisms, as presented above. They must be understood if breeding is to be carried out on those species. The distinction between mechanisms is based on: (i) site of origin of the embryo mother cell; and (ii) the developmental pattern of that cell.

Four major mechanisms of apomixis are known:

- Apospory.
- Diplospory.
- Adventitious embryony.
- Parthenogenesis.

There are also a couple of minor, less common mechanisms.

Since, for ease of reference, apomixis is considered a deviation from typical sexual reproduction, the latter will be described first, and then, for each apomictic mechanism, the deviations will be pointed out.

Meiosis in a Typical Sexual Ovule

In meiosis, a megaspore mother cell differentiates from the second layer of the nucellus, undergoing meiosis and giving rise to a linear tetrad of haploid megaspores. The three megaspores nearest to the micropyle will degenerate. The remaining megaspore will undergo three successive mitotic divisions, which will originate an eight-nucleate embryo sac. This sac represents the celled female gametophyte and will include an egg, two synergids, two polar nuclei in the so-called central cell, and three antipodals. The fates and functions of all these components are described in all elementary textbooks and, for brevity, will not be reviewed here.

Apospory

Apospory is considered to be the most common mechanism of apomixis. Two parallel processes go on in the same ovule. In one of them, a normal megaspore mother cell starts to undergo meiosis. It may or may not complete a tetrad of haploid megaspores, but this is irrelevant in terms of the final product, because the sexual embryo sac, if formed, aborts before reaching maturity.

At the same time, and in parallel, one or more somatic cells in the ovule enlarges to resemble a megaspore mother cell. The nuclei undergo one or more mitotic divisions, giving origin to one or more aposporic sacs, which may show various degrees of organization. They have no meiotic derivation; all their components are diploid and include one diploid maternal unreduced egg cell which will divide in due time into an embryo, and two diploid maternal unreduced polar nuclei, which will somehow, as will be seen later, give rise to an endosperm. Antipodals are normally absent.

Diplospory

There are only a few diplosporous species among agricultural crops. The first stage of the process is represented by a megaspore mother cell in the ovule, which does not undergo meiosis at all. Its nucleus divides mitotically, giving rise to a two-nucleate, unreduced megaspore. Further nuclear division and final sac organization may vary by species.

Adventitious Embryony

This type of apomixis is a normal way of reproduction in *Citrus* species, but a rare event in other higher plants. Starting points are the somatic cells of ovules, integuments, or ovary walls. Initial cells develop as zygotes. Later on divisions occur and the pre-embryo differentiates directly into a typical embryo, without formation of an embryo sac. Formation of embryo sacs, however, is not always ruled out; where an endosperm develops, it must arise from fertilization of polar nuclei in normal embryo sacs.

Parthenogenesis

No systematic occurrence of parthenogenesis has been recorded, but it seems to be an occasional event in all sexual species, where (as in cotton and corn) it is genetically controlled.

It starts from a reduced egg cell, derived from a normal meiosis. No sexual fusion follows, however; the reduced egg cell develops without fertilization into a haploid embryo, and this into a haploid plant, usually sterile.

Androgenesis

This is an occasional event. Meiosis of megaspore mother cells takes place regularly, giving rise to a normal embryo sac. In due time, a sperm cell may enter the egg cell sac and develop into a haploid embryo, without fusing of the nuclei.

This type of apomixis offers little problem or opportunity in terms of breeding. Genetic control has been reported in some lines of corn.

Semigamy

This occurs rarely, and only in some species (i.e., Pima cotton). A normal, haploid sexual embryo sac is regularly formed. When a pollen tube discharges the two sperm nuclei into the embryo sac, one of them penetrates the egg cell, but does not fuse with the egg nucleus, and develops independently into a haploid cell mass. The same thing happens with the egg nucleus. The end result is a heterogeneous haploid embryo.

Endosperm Development and Pseudogamy

Reference has been made to embryo development in the various types of apomixis. Nothing has been said yet about endosperm development. There is an element of surprise in it, in that very often no apomictic embryo development takes place without sexual endosperm development.

For ease of illustration, reference will be made to embryo and endosperm development in most sexual plants where there is parallel development of a sexual diploid embryo and a sexual triploid endosperm, the latter deriving from the fusion of a diploid polar nucleus of the central cell with a haploid sperm nucleus of a sperm cell (double fertilization). The resulting 2:3 embryo–endosperm balance of ploidy is critical to the development of both.

In the case of adventitious embryony, an apomictic diploid embryo (maternal, without any sexual fusion behind it) only develops if there has been preliminary formation of a sexual triploid endosperm, originating from the fusion of a diploid central cell, meiotically derived, with a haploid sperm cell. The final embryo–endosperm balance of ploidy (2:3) is the same as in the case of true sexual reproduction, but in the embryo both sets of chromosomes are maternal.

In apospory, an apomictic diploid embryo, completely maternal, develops close to a sexually derived pentaploid endosperm, which originates from the fusion of a tetraploid central cell with a haploid sperm cell. The resulting embryo–endosperm balance of ploidy is 2:5.

In both adventitious embryony and apospory, even if embryos originate apomictically, their development requires an act of pollination and, successively, fertilization of the central cell by a sperm cell. This complex procedure is called pseudogamy, because fertilization does not concern the egg cell. Pseudogamy is widespread among aposporous apomicts, which agrees with the fact that apospory is a facultative type of apomixis, so that in all occurrences of sexual reproduction there is also a need for pollination.

In diplospory (distinct from the two other mechanisms of apomixis) embryo and endosperm develop independently and apomictically; an apomictic diploid embryo is accompanied by an apomictic tetraploid endosperm originating from the fusion of the two polar nuclei. This is a case of autonomous endospermy, where sperm nuclei play no role (no pseudogamy). The result in terms of embryo–endosperm balance of ploidy is 2:4, with totally maternal embryo and endosperm chromosome sets.

Genetics of Apomixis

Genetic research is difficult to carry out on facultative apomicts. The best material is represented by completely sexual plants that can be found in obligate apomictic species. Crosses between these plants and obligate apomicts (acting respectively as females and males) can be statistically analyzed for their fit to ratios, and have proven that the character “mode of reproduction” is genetically inherited, as summarized below.

Most Aposporous Species

In general, the mode of reproduction is controlled by two unlinked loci, with the alleles determining apomixis mostly recessive. Epistasis may or may not be present.

Sorbus: Presents an A genome, influencing apospory in polyploids, and a B genome, which interact as follows:

- AA = *S. aria* sexual.
- BB = *S. aucuparia* sexual.
- AAAA = *S. rupicola* obligate apomict.
- AAAB = *S. minima* obligate apomict.
- AAB = *S. arranensis* obligate apomict.
- AABB = *S. intermedia* facultative apomict.
- ABB = *S. pinnatifida* mostly sexual.

Pennisetum: A/a locus, A allele causes apospory. B/b locus, B allele overrides A, giving sexual reproduction.

Example:

- Aabb apomictic.
- AaBb sexual, due to epistatic B.
- aabb sexual, due to lack of A.

Panicum: Two complementary loci. Conditions for apomixis are one recessive homozygote with the other heterozygote.

Example:

- aaBb, Aabb are apomictic.
- AABB, AABb, AaBb, aaBB and AAbb are sexual.

Potentilla: Shows a complex determination influenced by ploidy. In some cases, there is two-locus recessive control of apomixis.

Example:

- AABB, AaBb, AABb and AaBB are sexual.
- aaBB and aaBb are aposporous and sexual, giving tetraploid sexual.
- AAbb and Aabb are parthenogenetic and meiotic, giving haploids or diploids.
- aabb are apomictic.

Diplosporous and Some Aposporous

Taraxacum: Diploids are always sexual. Polyploids show irregular meiosis.

Breeding of Apomicts

Apomicts are mostly heterozygous. In fact, a continuous apomictic reproduction keeps "frozen" a reservoir of potential variability and, possibly, whatever heterosis may be associated with heterozygosity.

If, however, a breeding program is to be applied to an apomictic species, a break of apomictic barriers is needed. A passage through the process of meiosis has to take place, with consequent segregation, recombination and sexual fusion. The result will be a release of fresh variability, giving the breeder the opportunity to select progressive recombinants. An eventual return to apomixis will permit the exploitation of all the advantages associated with it.

When proposing breeding schemes, a distinction has to be made between obligate and facultative apomicts.

Obligate Apomicts

Hybridization is only possible if some sexual plants are available. In the case of truly obligate apomixis, however, hybridization can occur with closely related species.

Due to heterozygous parentage, the F_1 generation will segregate, as well as for mode of reproduction. The F_1 offspring will include:

- Variable apomictic plants. One can select the best apomictic genotypes, possibly heterotic, and reproduce the selected true-breeding clones.
- Variable sexual plants. These may be homozygous for mode of reproduction, although heterozygous for many other traits, and may be selfed to generate further variability. In the case of heterozygosity for mode of reproduction, they will keep segregating into apomicts and sexual plants at each generation.

In general, it must be observed that the presence of apomixis is no guarantee of satisfactory seed setting. An example is represented by those apomictic pseudogamous species requiring a pollination that may be made impossible by the presence of incompatibility factors.

Facultative Apomicts

Breeding procedures are essentially the same as for obligate apomicts, but more complicated. In fact, a selection of progressive apomicts is no guarantee of apomictic reproduction. A tentative remedy may be the selection of parents from highly apomictic lines. It is necessary to carry out a preliminary determination of stability for each line for many generations before releasing it as a cultivar.

Induction of Apomixis

Apomixis may be induced by mutagenesis. Examples of induction by irradiation are known in grain sorghum, and by treatment of seed with diethylsulphate and thermal neutrons in pearl millet. The best chance of induction seems to be with sexual species that include apomicts among their wild relatives.

Apomixis and Environment

Some environmental factors may affect the expression of the mode of reproduction. The best known case is represented by *Dicanthium aristatum*, where long days (over 14 h) favor sexual embryo sacs, while short days (under 14 h) favor apomictic embryo sacs, in an infra-ovular competition between the two types.

Even better known, of course, are the environmental effects on pollen and seed setting of pseudogamous apomicts.

Advantages and Disadvantages of Apomictic Reproduction

The following summarizes the material presented above.

Advantages:

- Reproduction is assured even when pollination is absent (not when pseudogamy is present, however).
- Apomixis is equivalent to clonal reproduction by seed, with all the advantages of the latter, such as dispersal, dormancy, and virus-free reproduction, with the true-breeding associated with clonal reproduction.
- The cost of meiosis is avoided; even unfit gametes and zygotes survive.

Disadvantages:

- Disadvantageous (non-lethal) mutations may accumulate, because they are not eliminated by the meiotic sieve.
- There is an absence of recombination and, therefore, no variability.
- There is narrow adaptability of apomictic species.
- Breeding is problematic.

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Effects of Environment on Flowering and Seed Development

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Flower and seed development is responsive to a range of environmental conditions. This response must be understood if plant breeders and seed growers want to produce seed successfully.

Although seed production occurs during the last developmental stage of the plant (senescence, death), its outcome is heavily conditioned by the impact of the environment on the preceding stages.

Plants and Environment

Crop plants differ widely in their response to environment. In other words, a large genotype x environment interaction is observed. Furthermore, the same genotype reacts differently according to the stage of its development. The most important environmental factors are light and temperature.

Basically, light is sensed by the plant in two different ways, each of which requires the presence of a special pigment:

- Harnessing of light's physical energy and its transformation into chemical energy for future use occurs by means of a pigment called chlorophyll.
- Sensing of the light's quality (wavelength), duration and periodicity, is accomplished by another pigment called phytochrome, a protein with a non-protein chromophore. Its localization is very wide (leaves, stem, apex, roots, seed cotyledons). At the cellular level, phytochrome is also ubiquitous, with the exception of the nucleus.

Phytochrome exists in two forms. P_r is biologically inactive, and shows a maximum photoreception at a wavelength of 767 nm. Upon absorption of light, it is transformed into P_{fr} , the biologically active form, which has a maximum, but not exclusive, light absorption at 720 nm. Although the two peaks of absorption are different, the whole spectrum of absorption of the two phytochrome forms shows some overlapping, and the two forms in fact coexist in relative amounts that are a function of the light wavelength. Therefore, each wavelength brings about a peculiar balance between P_r and P_{fr} . This may translate into a particular balance of hormones, which, in turn, affects a tissue or organ in a particular way.

P_{fr} , the active form, is partially destroyed (hydrolyzed, involved in protease denaturation), partially bound somewhere in the cell, and, in darkness, partially reverted into P_r . No reversion is known to take place, however, in monocots or in centrospermae dicots.

Such a beautifully engineered mechanism for measuring the relative length of night and day only makes sense if the relative length is variable. Of course, when passing from the equator to the poles, the relative length of day and night is increasingly variable according to the season of the year. Thanks to the mechanism described above, plants can sense this variation and flower when it is most appropriate.

To be precise, the P_{fr}/P_r ratio is not a perfect tool for measuring the relative length of day and night. In fact, the reversion rate of P_{fr} to P_r does not remain the same throughout the night. Thus the phytochrome system is not an hourglass. Furthermore, artificial night breaks may have different effects according to the hour of the night. Thus, it must be assumed that plants also respond to some sort of endogenous circadian rhythm.

In terms of day length response, plant species are classified as short-day if they flower when the day is shorter than a threshold value, or long-day if they flower when the day is longer than the threshold value. In either case, the response can be absolute or facultative, depending on whether the departure from an optimal photoperiod prevents flowering or merely delays it. Day neutral species also exist. But, most importantly, there is a wide variation within a given species in reaction to day length or photoperiod. This permits a species (for example, soybean) to colonize a range of habitats, from the tropics to 50 degrees north latitude.

Temperature influences the growth rate and the number of seeds brought to maturity. For example, barley produces a maximum number of seeds at about 25° C. Tropical crops are characterized by optimum temperatures above 30° C.

But, most importantly, plants are differently affected by the environment according to their stage of development, as specified below.

Influence of Environment on Different Phases of Plant Development

Germination

Germination begins the life cycle. It usually requires suitable temperature and moisture. Factors of dormancy may also intervene. Dormancy is often

determined by seed coat hardness, after-ripening requirements, day length, and low temperature.

Juvenile Phase

This phase lasts from seedling emergence to floral bud formation. Here, the rate of leaf emergence is a function of temperature, but the total length of the phase is a function of day length (in day length responsive plants). Thus, at a given temperature, the number of initiated leaves, nodes and internodes, and therefore plant size, are also a function of day length: late flowering plants are taller. Soil moisture and fertility also play a role.

Reproductive Phase

This phase lasts from the initiation of flowering to fertilization.

In determinate species, or genotypes, leaf initiation stops when flowering initiation occurs (for example, corn, a determinate species, stops initiating leaves after the tassel is initiated). Of course, growth of initiated leaves continues during flowering.

In indeterminate species, leaf initiation continues after flowering initiation. Generally, however, leaf initiation stops when the last flower forms.

Some winter annuals and biennials may require vernalization to flower. Vernalization is the need for low temperature exposure in order to achieve the ability to flower, or simply to shorten the time to flowering. Vernalization *per se*, however, does not confer the ability to flower, but simply makes plants more responsive to day length stimuli.

The length of time between flower initiation and fertilization is determined by day length in some species. In long-day cereals, for instance (wheat, barley, oat) this phase is shortened by long-days.

Ripening Phase

This is the time interval between fertilization and seed maturity. Sub-intervals can be identified as follows: (i) a lag phase, which encompasses the period between fertilization and the start of active, linear filling of the kernel; (ii) a linear filling period, during which a massive translocation of assimilates into the seed occurs, with a rate of dry matter accumulation in the kernel that remains constant; and (iii) a final period, during which the rate of kernel filling levels off.

The lag phase is sensitive to day length and temperature (for example, short-days and warm temperatures shorten this phase in soybean, a short-day species). The linear filling period is also influenced by temperature and day length; higher temperatures tend to induce higher rates of filling and a shorter duration of the period.

Day Length

Day length sensitive plants are affected only after having attained a minimum amount of growth, i.e., after the so-called basic vegetative phase (BVP). BVP varies with species and genotype; in some wheat, BVP is completed right after seedling emergence. In rice, it can vary from 10–63 days, and its length is controlled by two genes. More generally, most species require 10–35 days to achieve a ripeness to respond.

After BVP is completed, a critical photoperiod must be assessed. For short-day plants, it is the photoperiod above which no more flower initiation occurs. For long-day plants, it is the day length below which flowering cannot occur. Knowledge of the critical photoperiod is essential to breeders to carry out more efficient breeding programs. Knowing, for instance, that a given variety of soybean (short-day species) does not flower if the day is longer than 13.5 hours enables breeders to stop flowering by applying a 14 hour day, and wait for another variety to flower and be crossed.

Species originating from lower latitudes usually display a lower critical photoperiod (for example, soybean).

The optimum photoperiod is the shortest interval to flowering. The optimum photoperiod is, however, not known for most crops. Usually, a breeder will apply a 12 hour day to short-day species and a 20–24 hour day to long-day species.

Manipulation of day length

Techniques to manipulate day length can be envisaged to control the interval to flowering. Parameters of illumination are: timing, duration, and level and quality of light.

Timing

A nighttime application of light translates into a shorter night. Under such treatment, short-day plants, that are in effect long night plants, will experience a delay in flowering. Conversely, long-day plants, that are in reality short night, will show a shorter period to flowering.

Duration

Generally, continuous light is the best way to shorten the time to flowering in long-day plants: short-days promote development of short-day plants. Artificial illumination is applied to lengthen the duration of the natural day. But before artificially extending the day, one must determine the natural effective duration of the day itself. Such a day begins as soon as the minimum light level occurs, at which time the plant becomes sensitive to the photoperiod. This level is largely a function of species and genotype within species. Some examples are:

- Soybean: 10 lux in the morning, 200 lux in the evening.
- Rice: 1 lux sufficient to delay flowering.
- Corn: if day-insensitive, is not affected by day length and light level; if day-sensitive, critical level to delay flowering varies from 11 to 54 lux.
- Potato: 20,000 lux.

Minimum light levels change not only with genotype, but also with plant age. Older plants are characterized by higher minimum levels than younger plants.

Minimum levels do not coincide with optimum levels of light. Time to flowering for soybean, for instance, becomes longer with higher levels of light.

If light level increases above a minimum, vernalization requirements can be lower, or unnecessary (broad bean).

Quality

Artificial lighting with different light quality (red vs. blue; incandescent vs. fluorescent) may also influence duration.

Other Responses to Photoperiod

- Development of flowers, favored by supplementary inductive light cycles.
- Pollen fertility, more sensitive to day length than ovule fertility.
- Relative number of staminate vs. pistillate flowers.
- Relative rate of vegetative reproduction (for example, strawberry runner formation, favored by long-days).
- Formation of storage organs (except onion bulbs) favored by short-days.

Temperature

Generally speaking, temperature has two somewhat contradictory effects. It enhances both the rate of biochemical reaction, and the rate of enzyme denaturation. Some of the main biological effects of temperature are:

- Stratification is the cold treatment of humid seed in the presence of oxygen to overcome seed dormancy. Among other effects, there is a breakdown of seed fats into sugars and starches.
- Vernalization is the exposure of plants to low temperatures (less than 10° C) for a certain period. It can occasionally be applied during a stratification treatment. Application of chilly temperatures usually lasts from 1 to 16 weeks, but can be as short as one day. In general, annuals show a facultative vernalization requirement and are always sensitive to long-days, whereas biennials show an obligate vernalization requirement and a facultative light response.
- Thermoperiodism is the promotion of growth and development by alternating cold nights and warm days. For example, formation of potato tubers, tomato fruit setting and flower initiation.

In most cases, plant growth and development respond simultaneously to both temperature and day length.

In species with limited light response, temperature becomes the primary factor determining floral initiation. Corn, for example, responds to day length for only 15–25 days from emergence in temperate regions. The effect of temperature on flowering is more important, and flowering and maturation occur after a certain number of growing degree days, i.e., the number of days during which temperature has been between 10–30° C. In rice, for example, the minimum critical temperature is 15–18° C.

Critical temperatures are more restrictive for hybridization than for floral initiation or development. Thus, cool temperatures induce self-pollination before the flower is large enough to manipulate. Also, less pollen is shed. High temperatures shorten the duration of stigma receptivity and pollen viability.

In general, there is an interaction of temperature and photoperiod, but it is highly variable, with night temperature playing some role.

Moisture

At any stage of plant development, adequate soil moisture is necessary to produce healthy, vigorous plants with good potential for seed production.

Stress can determine flower and seed abortion, or reduced seed production, especially during flowering and seed filling. Drought stress can be reduced by planting at a lower density and pruning part of the flowers. A reasonably low water supply in the soil can hasten maturity without, however, impairing seed viability.

If relative humidity is too high during hybridization, the pollen will clump and diseases will develop. If humidity is too low, the pollen may fail to germinate on the stigma. Bagging of flowers is one remedy.

Soil Fertility

Adequate soil fertility is necessary to produce healthy plants and enough seed. Excess N, however, will in most species produce excess vegetative growth, flower abortion and delayed maturation. Low fertility, on the other hand, can favor flowering in some species (for example, potato).

Maturing seed in some species has peculiar nutrient requirements. Peanut, for instance, requires a calcium-rich soil for normal pod formation.

Techniques for Synchronizing Flowering Dates

Multiple Planting Dates

This method is adopted if the flowering time of the parents is not known, or is known to be different, or to multiply the chances of carrying out many cross combinations.

Usually, the late parents are planted first, all at once. The early parents are planted later, at different intervals, keeping in mind that differences in times of planting do not reflect equivalent differences in times of flowering. For instance, a seven day difference at planting may result in a 3–10 day flowering difference, depending on temperature and day length responses.

Differential vernalization can also be used to modify intervals to flowering.

Day Length

Appropriate manipulation of day length can result in delaying or hastening of flowering, as needed.

Temperature

Cool temperatures generally delay, and warm temperatures hasten, flowering. Excessively high temperatures, however, may slow development.

Plant Density

Reduced plant density encourages tillering and branching. Tillers flower later than the main stem, thus prolonging the crossing season, but their flowers may be smaller and more difficult to manipulate.

Pruning

Removal of the growing point from the main stem promotes formation of tillers and branches that flower later. Pruning of leaves (in corn, for example) can delay flowering. Removal of flowers and seed during flowering prolongs the flowering.

Grafting

Flowering of a late genotype can be hastened by grafting onto an earlier flowering genotype that is in bloom. Potato is sometimes grafted on tomato roots to promote flowering of difficult genotypes and to increase the amount and duration of flowering.

Hormones

Flowering of plants is ultimately triggered by one or more hormones. Artificial treatment with hormones is therefore used. For example, many long-day plants can be induced to flower sooner by treating them with gibberellic acid, which can also replace vernalization treatments in *Lolium*.

Inducing short-day plants to flower is more difficult. Sometimes, success can be achieved by treating with ethylene-producing chemicals.

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Ecology Seed Germination Physiology

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Germination is the thread of life that assures survival of all plant species. The initiation of germination requires that many conditions be fulfilled. First, the seed must be viable and non-dormant; the embryo must be alive and capable of germination with no barrier to germination. Second, the seed must be planted under favorable environmental conditions such as available water, a supply of oxygen, proper temperature, and sometimes light. Most seed undergoes a specific sequence of events during germination. The major events are water imbibition, enzyme activation, initiation of embryo growth, rupture of the seed coat, and emergence of the seedling. Germination is affected by the ecological conditions prevailing in the habitat. The first habitat in which the seed finds itself is constituted by the mother plant during seed formation, development and maturation. The second is constituted by the external factors of the germinating seed's habitat.

Introduction

Germination refers to a large number of processes, including seed germination of flowering plants, the germination of spores of bacteria, fungi and ferns, and the processes in the pollen grain by which the pollen tube is produced.

In flowering plants, the seed's role is manifested by its reproductive ability. It is considered to be the thread of life that assures survival of all plant species. Seed germination remains a key to modern agriculture because of its role in stand establishment. Thus, a fundamental understanding of germination is essential for maximum crop production.

Definition of Seed Germination

Various definitions have been used to describe seed germination. To a seed physiologist, germination is defined as the emergence of a radical through the seed coat. Others define germination as the resumption of active growth by the embryo, resulting in rupture of the seed coat and the emergence of young plants (Copeland and McDonald, 1985). According to ISTA, seed germination in laboratory tests is defined as: "the emergence and develop-

ment of the seedling to a stage where the aspect of its essential structures indicates whether or not it is able to develop further into a satisfactory plant under favorable conditions in soil.”

Morphology of Germination

Germination falls into two categories, based on the fate of the cotyledons or storage organs. Neither appears to be related to seed structure. The two types of seed germination are illustrated by the germination of pea and bean seed. Although these seeds have similar structures, their germination patterns are different.

Hypogeal Germination

Hypogeal germination is characteristic of faba bean seed, pea seed, all grasses, and many other species. During germination, the cotyledon or comparable storage organs remain beneath the soil, while the plumule pushes upward and emerges above the ground. In hypogeal germination, the epicotyle is the rapidly elongating structure.

Epigeal Germination

Epigeal germination is characteristic of bean, pine, and many other seed species. During germination, the cotyledons are raised above the soil surface, where they continue to provide support to the growing points.

Factors Affecting Germination

Seed Maturity

The seed of many species is able to germinate long before physiological maturity. Smooth brome grass seed is capable of germination only a few days after fertilization (Grabe, 1956).

Viability and Life Span of Seeds

When seed is in a state of desiccation, it is fairly resistant to extreme external conditions. As a result, seed can retain its ability to germinate (viability) for considerable periods. The length of time is greatly variable and is determined by genetic factors. However, environmental factors and storage conditions have a great effect on the life span of any given seed. In general, seed viability is best retained under conditions in which the metabolic activity of the seed is greatly reduced (low temperatures and a high concentra-

tion of CO₂). In addition, other factors are of great importance, particularly those which determine seed dormancy (Mayer and Poljakoff-Mayber, 1982).

Environmental Factors

Seed must be placed in environmental conditions favorable to germination. The main conditions required are:

- An adequate supply of water.
- Suitable temperature.
- Suitable composition of atmospheric gases.
- Light for certain seed species.

The requirements for these conditions vary according to the variety and species. It is determined by hereditary factors and by the prevailing conditions during seed formation and development.

Water

Water is a basic requirement for seed germination. The uptake of water by the seed is the first process which occurs during seed germination. Water is essential for enzyme activation and for the breakdown, translocation, and use of reserve storage material.

Field capacity moisture is about optimum for seed germination in soil; however, seed germination may occur at soil moistures close to the permanent wilting point. Moisture content should reach a certain point before the seed begins to germinate. Rice seed begins to germinate at a moisture content of 26.5 percent (fresh weight), corn at 30.5 percent, sugar beet at 31 percent, and soybean at 50 percent (Hunter and Erikson, 1952).

Seed germination may be inhibited by high moisture levels. For example, germination of dwarf bean seed was reported to be inhibited when moisture content was increased from 20 to 40 percent (Ensor, 1967). Germination of sugar beet seed (Snyder, 1975) and many other species (Heydecker *et al.*, 1969) is also known to be retarded by excess moisture.

Gases

The process of germination is related to living cells and requires an expenditure of energy. The energy requirement for these living cells is usually sustained by oxidation processes in the presence (respiration) or absence (fermentation) of oxygen.

These processes involve an exchange of gases, with an output of CO₂ in both cases and, for respiration, the uptake of O₂.

Air is composed of about 20 percent O₂, and 0.03 percent CO₂. Germination of most seed is retarded if the CO₂ concentration is higher than 0.03 percent, or if the O₂ concentration is substantially below 20 percent. However, seed of rice and other aquatic plants can germinate underwater, where oxygen is present only in low concentrations (Copeland and McDonald, 1985). Although the O₂ concentration of air is the best for the germination of most seed species, some actually germinate better at O₂ concentrations different from that of air.

Germination of Bermuda grass and cat tail seed is improved by O₂ concentrations below that of air (Morinaga, 1926). On the other hand, carrot, curly dock, sunflower, cocklebur, and various cereals germinate better under higher O₂ concentrations (Morinaga, 1926; Albaum *et al.*, 1942).

Temperature

Different seed germinates within different temperature ranges. At very high and very low temperatures the germination of all seed is prevented.

The seed response to temperature depends on plant species, variety, region and the length of time from harvest. Temperatures between 15–30° C are considered optimum for the germination of most seed. Temperatures below or above the optimum may result in delaying germination. The optimum temperature is the temperature giving the greatest germination percentage within the shortest period of time. The minimal and maximal temperatures are the lowest and highest temperatures at which germination will occur.

For optimum germination, seed of some species requires daily fluctuation of temperatures. Kentucky bluegrass seed germinates better under alternating temperature conditions (Harrington, 1923). It seems that the need for fluctuating temperatures during germination is associated with dormancy, but alternating temperatures also accelerate germination of non-dormant seed. In cases where alternating temperatures are needed, the range between high and low seems to be more important than the actual temperature.

Light

The seed of most cultivated plant species usually germinates equally well in the dark or light. While moisture, oxygen, and favorable temperature are essential for the germination of all seed, certain species also require light. The seed light response of several hundred species has been studied to determine those whose germination was promoted by light, darkness, or indifference to light. Almost half of the species studied responded to light.

Light enhances the germination of freshly harvested seed of some species, such as lettuce (Agrawal, 1966). Seed germination is influenced by both light intensity and light quality.

Light intensities of 1,080 to 2,160 lux (100 to 200 foot candles) are probably adequate for the germination of most seed species. The greatest promotion of germination occurs in the red area (660–700 nm) with a peak at 660 nm. Wave lengths above 700 nm (far red), below 290 nm (ultra violet) and blue light (440 nm) will inhibit seed germination. It has been found that by exposing imbibed seed to alternating red and far red light, it is possible to alternately promote or inhibit seed germination. Photo reversible germination in seed appears to be due to a single coupled chemical reaction. The pigment responsible for absorbing light energy in seed during germination is phytochrome, which is proteinacious in nature, with a blue color. Phytochrome exists in two forms: one which absorbs red light (Pr) and another which absorbs far red light (Pfr). Red light (660 nm) exposure converts phytochrome into the physiologically active, far red absorbing form, and germination proceeds. Exposure to far red (730 nm) reconverts phytochrome into the physiologically inactive form (Pr), and germination is blocked.

Light sensitivity during seed germination depends on many factors, including species and variety, seed age, period of inhibition, inhibition temperature, stratification, and temperature (Toole *et al.*, 1963).

Events of Seed Germination

Most seed undergoes a specific sequence of events during germination. The major events are: water imbibition, enzyme activation, initiation of embryo growth, rupture of the seed coat and emergence of the seedling (Copeland and McDonald, 1985).

Water Imbibition

The first process that occurs during seed germination is the uptake of water by the seed. The extent to which water imbibition occurs is determined by the following factors: composition of the seed, seed coat permeability, and water availability. Imbibition is a physical process and is not dependent on metabolic energy, but is related to the physical properties of the colloids in the seed tissue.

Imbibition occurs in both dead and live seed. Protein is the principal component of the seed responsible for the imbibition of water. However, other

seed components also contribute to imbibition. The mucilage of various seeds contributes to swelling, as do parts of the cellulose and pectin substances located in the cell walls. On the other hand, starch molecules have little impact on swelling, even when large quantities of starch are present.

Water entry into the seed is influenced by the nature of the seed coat (or pericarp). The permeability of the seed coat is highly variable among species. Denny (1917) showed that the presence of lipids, tannins, and pectin substances in the seed coat contributes to its water impermeability. The ability of seed to imbibe water is dependent on cell water potential, and is a result of three forces: cell matrix potential, cell osmotic concentration, and cell turgor pressure. The soil in which the seed is planted also has its own water potential. The physical properties of the soil determine retention and conductivity of water.

The degree of seed-soil contact is also important for water imbibition. The greater the contact of the seed with the soil, the greater the amount of water imbibed by the seed.

Enzyme Activation

Dry seed is characterized by a very low rate of metabolism, which is attributed to low moisture content (5–10 percent). A marked change in seed metabolism occurs as soon as the seed begins to imbibe. During germination, a triphasic pattern of water uptake has been demonstrated. Enzyme activation begins during phases I and II of water uptake. The seed undergoes many processes essential for germination during phase II of water uptake. The process of enzyme activation during phase II serves to break down stored tissue, aid in the transfer of nutrient from storage tissue (cotyledons or endosperm) to the growing points, and trigger chemical reactions for the synthesis of new material. The early events that occur during the enzyme activation phase include the synthesis of enzymes such as amylase, ribonuclease, and phosphatase. The synthesis of these enzymes is mediated by gibberellic acid. Other hydrolytic enzymes, such as ATPase, proteases, lipase, and peroxidase are increased during enzyme activation. The enzyme activation phase is one of the most important phases for the preparation of seed for embryonic axis elongation. Enzymes that break down carbohydrates, proteins, lipids, and phosphorus-containing compounds are activated first during phase II. In order to supply the embryonic axis with the energy required for growth, storage compounds in the seed must be hydrolyzed to soluble forms, translocated from storage tissues to the embryo, and transformed to energy molecules that can be utilized by the embryonic axis.

Initiation of Embryo Growth

Studies have been conducted on the changes (seed germination patterns of monocot seeds) that occur during germination. There is a marked decrease in the dry weight of the endosperm, and an increase in the dry weight of the embryonic axis in the first 120 hours of germination.

In part, these changes are a reflection of the decrease in total nitrogen and insoluble protein of the endosperm, and subsequent translocation of these compounds to the growing axis. Similar changes would be anticipated due to the hydrolyzation of endosperm starch to maltose and then glucose, which is then enzymatically altered to sucrose and translocated to the axis (Ingle. *et al.*, 1964).

In dicot seed, the cotyledon of cowpea (*Vigna sesquipedalis*) decreases in dry weight as the hypocotyl and the epicotyl increase. Soluble carbohydrates, soluble nitrogen, and nucleic acid phosphorus levels of the cotyledons are decreased, and found in the emerging embryonic organs of the roots, hypocotyl, and plumule (Oota *et al.*, 1953).

Protrusion of the Radical

The protrusion of the radical from the seed coat can be accomplished through either cell division or cell elongation. In most cases (as in lettuce) cell elongation precedes cell division (Haber and Luippold, 1960), while in cherry seed, cell division precedes cell elongation (Pollock and Olney, 1963). In pine seed, cell division and elongation occur simultaneously (Berlyn, 1972).

Seedling Establishment

The seedling is considered to be established when it begins water uptake and photosynthesis. As the seedling takes up water and manufactures most of its own food, it gradually becomes independent of the storage tissues. Then the germination process is complete.

The Ecology of Germination

The effect of different factors on the germination of seed has been discussed above. An attempt will now be made to relate these factors to the behavior of seed in its natural habitat. Different factors regulate the germination of seed. Some are internal, and others are external, environmental factors. Any of these different factors can determine whether or not a given seed will germinate in a certain place. Seed germination is affected by the eco-

logical conditions prevailing in a given habitat. The micro-climatic conditions prevailing in the immediate vicinity of the seed are more critical than the overall climatic conditions. The first habitat in which the seed finds itself is constituted by the mother plant during seed formation, development and maturation. The second is constituted by the external factors in the habitat.

Effect of the Mother Plant

Prevailing factors during seed development and maturation can affect the germination behavior of the seed after harvesting. Among these factors are: the position of the seed on the parent plant, photoperiod, and thermoperiod. The seed position has a marked effect on the germination behavior of the seed after harvesting; celery (*Arium graveolens*) seed derived from the primary umbel was less dormant than that derived from tertiary and quaternary umbels, and somewhat heavier (Thomas *et al.*, 1979). The seed of *Chenopodium album* plants exposed to long-days had lower germination than seed from plants exposed to short-days. The photoperiod under which the parent plants of *Amaranthus retroflexus* seed were grown affects post-harvest dormancy (Kigle *et al.*, 1977). Besides the photoperiod effect, the thermoperiod also plays a role in the dormancy of seeds. Seed from parent plants grown under long (16 h) days had lower germination under no light conditions, and responded less rapidly at 30° C to short periods of exposure to light than seed derived from plants grown in short (8 h) days (Kigel *et al.*, 1979). These data indicate that parent plants, via some phytochrome, mediate mechanisms that influence subsequent germination behavior.

External Factors in the Habitat of the Germinating Seed

Water

Moisture content may vary between different soil types.

The moisture content of a given soil can also vary with climatic conditions during different seasons of the year and according to the plant cover. The availability of water at a given time is determined by: osmotic factors, binding of water by soil colloids, capillary forces, and soil composition and texture. It is also determined by competition with other organisms requiring water in the soil. The moisture content of the soil may show seasonal periodicity.

In some regions, high moisture content is associated with high temperatures (when summer rain is followed by dry or very cold winters with frozen soil). In other regions high moisture content is associated with low temperatures (winter rain followed by hot, dry summers).

Temperature

Soil temperatures vary greatly and show both seasonal and diurnal change. The extent of soil temperature change depends on the type of the soil (light or heavy, with or without leaf litter), and climatic conditions. Other factors that play a role in determining soil temperature are: soil structure and texture, amount of water in the soil, conditions for evaporation of water from the soil, and plant cover. The upper layers of the soil usually show wide fluctuations. As soil depth increases, soil conditions become more and more constant throughout the year.

Gases

Oxygen, carbon dioxide, and nitrogen are the three major gases normally present in soil. In soil containing high organic matter and many microorganisms, the oxygen content may be much lower, and carbon dioxide very much higher than in the air. In water-logged soil (especially in heavy soil) the oxygen content of the gaseous phase may drop far below normal atmospheric levels.

Light

Light is usually abundant on the surface of the soil. In light-textured soil, light penetrates for a short distance into the soil. However, its intensity falls off rapidly. In heavy soil, light hardly penetrates at all. Under a vegetation cover, light intensity will fall off rapidly, and its spectra composition is liable to change due to differential absorption and reflection under the canopy.

Biotic factors

Seed interacts with animals and other plants in its natural habitat. The interaction with other plants may be due to stimulators, inhibitors or modification of the micro-habitat. Animals may affect the germination behavior of seed due to distribution to other habitats or by seed softening in the digestive system. Man can also be a contributor to biotic factors; both accidental and planned fires can also affect germination behavior. (Mayer and Poljak-off-Mayber, 1982)

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Seed Dormancy

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The failure of seed to germinate is broadly termed *dormancy* and is a widespread phenomenon. Different types of seed dormancy are recognized according to their manner of origin. The interaction of morphology and physiology is the basis for the various types of seed dormancy. The two major types of dormancy are referred to as *primary* and *secondary*.

Primary (exogenous, endogenous, and combination) dormancy includes conditions that exist within the seed to prevent germination when the seed matures on the mother plant and immediately afterward. Different methods are used to overcome primary dormancy. These methods are: scarification (mechanical and chemical), temperature treatment (stratification and after-ripening during storage), light treatment, and treatment with growth regulators and other chemicals.

Secondary dormancy refers to the dormancy that develops within the seed after it is removed from the mother plant if subjected to adverse conditions.

Introduction

One of the interesting and important problems encountered in seed physiology is the failure of viable seed to germinate when supplied with water and oxygen at temperatures recognized as favorable for germination. The ability of seed to delay germination until the time and place are right is an important survival mechanism in plants. In plants, dormancy is not a condition restricted to seed, but a property displayed by other plant organs such as tubers, bulbs, corms, and terminal buds, as well as the mitospores and meiospores of almost all plants.

Pollock and Toole (1966) use the term dormancy to cover both conditions when there is some internal block to germination and when unfavorable environmental conditions prevail. Copeland and McDonald (1985) use the term *quiescence* to describe a state of arrested development due the absence of suitable germination conditions. They define dormancy as a state in which seed is prevented from germination even under environmental conditions normally favorable for germination.

Types of Dormancy

Different types of seed dormancy are recognized, according to origin. Unfortunately, different terminologies are used by various authorities, which can give rise to some confusion (Bewely and Black, 1978). Nikolaeva (1969) suggests a detailed classification:

- Properties of the outer layers of the embryo.
- Underdevelopment of the embryo.
- Physiological condition of the embryo.
- Combinations of the above.

Copeland and McDonald (1985) classify dormancy into two major types: primary and secondary dormancy. The following general description of the various types of dormancy follows their classification.

Primary Dormancy

Primary dormancy is the most common type of dormancy and takes two major forms: exogenous and endogenous dormancy.

Exogenous dormancy

Exogenous dormancy is a form of primary dormancy in which essential germination requirements (e.g. water, light, and temperature) are not available to the embryo so that it fails to germinate. Generally, this form of dormancy is related to physical properties of the seed coat including: impermeability to water, low permeability to gases, and mechanical restriction.

Impermeability of seed coat to water: This appears to be one of the simplest but most effective means of delaying germination, and of spreading the production of seedlings over time (Villiers, 1972). The impermeability to water is typical of many species of a number of families (e.g. *Fabaceae*, *Chenopodiaceae*, *Liliaceae*) and is caused by both genetic and environmental factors (Copeland and McDonald, 1985). Genetic control of seed impermeability was reported in some alfalfa varieties (Dexter, 1965), bean varieties and crimson clover lines (Benett, 1959).

The environment also influences the impermeability of the seed coat to water, but little is known about the nature of this influence. Apparently, several complex environmental interactions (weather and soil conditions) during seed development and ripening contribute to the seed coat's impermeability to water. Agriculturally, seed that exhibits dormancy via this mechanism is termed *hard seed*. The impermeability to water may be due to the structure

of the testa, which in many cases prevents water uptake, apparently due to a layer of palisade-like cells which are thick-walled on their outer surface with an external layer of a waxy, cuticular substance. Deposits of cutin, suberin, or lignin are common in the teguments of many legume seeds and other hard coated seed species. Cutin deposits have been reported in the nucellar layers of watermelon seed (Thorton, 1968).

The testa of *Leguminosae* seed are not made up of uniform tissue but contain hilum, strophiole and micropyle. The strophiole does not conduct water unless certain conditions have been experienced by the seed; it then becomes the site of water entry (Ballard, 1976). In other seed, water impermeability may be related to the structure of the hilum. The hilum of certain leguminous seed operates as a hydroscopically activated valve, allowing water loss without permitting water uptake (Hyde, 1954). The hilum fissure opens rapidly when the seed is in dry air, and closes rapidly in moist air.

Low permeability of seed coat to gases: In many seed species, the seed coat is selectively permeable, permitting water to enter but not oxygen. The nucellar membrane of *Cucubita pepo* (Brown, 1940), and the endocarp of coffee "seeds" (Huxley, 1965) are known to restrict the entry of oxygen. In addition, the inner membrane of *Cucubita pepo* seed is less permeable to oxygen than to carbon dioxide.

The classical example of seed coat impermeability to oxygen is provided by *Xanthium*. Crocker (1906) showed that the upper and lower seed in the fruit differ in their germination capacity. The upper seed require pure oxygen to give 100 percent germination at 21° C, while the lower seed require only 6 percent oxygen for full germination. However, more recent data indicate that the small, upper seeds of *Xanthium* contain greater amounts of inhibitors than the larger, lower seeds.

It has been suggested that the upper seeds require more oxygen to oxidize and deactivate the inhibitor before 100 percent germination is achieved (Porter and Weiring, 1974). The coats of apple seeds have been shown to restrict oxygen permeability during imbibition at 20° C, but permeability increases during imbibition at 4° C. Thus, there may be a temperature-oxygen interaction.

Mechanical restriction to embryo growth: The coats of many seeds are made up of very hard, tough tissues, which clearly offer mechanical resistance to the growth of the embryo. Therefore, if the embryo cannot develop enough thrust during imbibition to overcome this mechanical barrier it remains ungerminated. This type of dormancy is found in the seed of wa-

ter plantain (*Alisma plantago*), pigweed (*Amaranthus retroflexus*), peach (*Prunus persica*), and cherry (*Prunus*).

Seed coats are often the source of inhibiting substances, and may interfere with leaching of inhibitors or restrict water flow. Removal of the seed coat will solve the problem (Copeland and McDonald, 1985). The mechanical resistance of the seed coat is of significance in dormancy only in relation to the thrust developed by the embryo. This relation has been studied by Esashi and Leopold (1968) in *Xanthium pennsylvanicum* seed. They conclude that the testa of the upper (smaller), dormant seed are, in fact, less resistant than the testa of the non-dormant seed, but the embryos of the dormant seed cannot develop enough thrust to rupture the seed coat.

Endogenous dormancy

This type of dormancy is the most prevalent, and is due mainly to the inherent properties of the seed. For example, the seed may have an excess amount of inhibitor that must be reduced or removed prior to germination. Only physiological changes such as rudimentary embryo maturation, response to growth regulators, exposure to light, changes in temperature, and endogenous rhythms will break this type of dormancy in seed.

The duration of endogenous dormancy can be influenced by environmental conditions during seed development and maturation. Seed of *Impatiens balsamina* (touch-me-not) plants that has been adequately watered and well-supplied with nitrogen has less dormancy than seed from deficient plants. The germination requirements of *Latuca sativa* (lettuce) seed have been shown to change when grown under different environmental conditions (Koller, 1962). The stage of seed maturity has been shown to have little effect on the duration of endogenous dormancy in Kentucky bluegrass seed at harvest. The degree of dormancy in this seed was directly associated with the moisture content of the seed at harvesting time. The higher the seed moisture, the greater the degree of dormancy (Delouche, 1958).

Rudiment Embryo Dormancy: The seed of some species is morphologically immature when the dispersal unit is shed from the mother plant. Immature embryos are relatively small, and in some cases poorly differentiated, and must grow and develop to be ready for germination. Further embryo maturation occurs following seed dispersal and may last a few days to several months. The embryo of holly (*Ilex opaca*) seed is an undifferentiated mass of cells when the seed is shed from mother plant, but during subsequent maturation the cells develop a well-defined structure (Ive, 1923). Embryos of cherry seed increase in weight, size, length of leaf primordium, and in oxygen uptake by the embryonic axis after being shed from the mother plant (Pollock and Olney, 1959).

Physiological dormancy: This type of dormancy in higher plants is believed to be regulated by the balance between endogenous growth inhibitors and promoters. Therefore, dormancy may be considered a result of the absence of growth promoters, the presence of growth inhibitors, or a combination of both. The levels of these compounds are controlled by environmental conditions such as temperature and light. Khan (1971) gave the inhibitor-promoter concept a great push by suggesting the participation of three hormones in the control of seed dormancy and ascribing a particular function to each hormone. In his model, Khan suggested that gibberellins must be present for seed germination to occur and only an inhibitor can prevent this expression. He also suggested that cytokinins play a permissive role by selectively antagonizing the inhibitors when they are present.

Cytokinins have no effect on the breaking of dormancy if inhibitors are not physiologically active. In this case, breaking dormancy is governed by gibberellins. Many substances present in seed help to determine whether a seed will be dormant. Many compounds that can induce dormancy through their influence on osmotic and metabolic inhibition have been isolated.

Osmotic inhibition: Substances possessing high osmotic potential can inhibit the germination of seed. Sugar and salt compounds in sufficient concentration may compete with seed for water, and as a result the seed never becomes fully imbibed and thus remains ungerminated. However, this seed will germinate when removed from the osmotic-inhibiting conditions. Many of these substances are located in the fruit and fruit walls surrounding the seed. The inorganic substances in the fruit ball of sugar beet are responsible for the inhibition of germination. The concentration of electrolytes from sugar beet fruit ball extracts retarded the growth of wheat seedlings (Snyder *et al.*, 1965). Tomato juice completely inhibits the germination of garden cress (*Lepidium sativum*) seed, even when diluted in a proportion of 1:25 (Meyer *et al.*, 1960).

Seed germination in most fleshy fruits is delayed while the seed is still in the fruit itself. Thus, the osmotic potential of such fruit juices undoubtedly contributes to seed dormancy.

Metabolic inhibition: Certain compounds present in seed inhibit specific metabolic pathways. Cyanide, found in apple and peach seed, is an example of this type of inhibitor. Such compounds inhibit seed germination through their effect on respiration. Phenolic compounds can also inhibit seed germination, and because of their widespread occurrence, have been regarded as natural germination inhibitors. The first dormancy-inducing inhibitor found was coumarin. It is widely distributed and rapidly metabolized in seed, and is considered a natural germination inhibitor.

Coumarin derivatives (the glycosides of lactone) or substituted coumarins have also been found in many fruits, supporting the role of coumarin as a natural seed inhibitor. It is suspected that coumarin inhibits seed germination through its interference with respiration and oxidative phosphorylation, and, indirectly, through the availability of energy through its effect on phosphorous metabolism (Copeland and McDonald, 1985).

Comforth *et al.* (1966) discovered another extremely active inhibitor. The compound was initially named dormin because of its dormancy-inducing properties. In 1967, when chemical characterization revealed that dormin had the same chemical structure as abscisic acid, the name was changed to abscisic acid (ABA). Results of many studies have shown that ABA is a naturally occurring endogenous hormone in a wide range of seed, and is active at very low concentrations.

However, reports have also shown that levels of endogenous ABA are reduced following stratification of dormant ash, rose, and other seed. In apple seed, ABA has been shown to be localized in the testa, and is translocated to the embryo during imbibition to induce dormancy (Lewak and Rudnicki, 1977). The exact mechanism for ABA inhibition of seed germination is not clearly understood. Ho and Vamer (1976) reported that ABA may inhibit the synthesis of enzymes that are important in early stages of germination, perhaps by inhibiting the translation of these enzymes from mRNA. Many studies have shown that ABA can completely or partially reverse the primitive effect of either gibberellins or cytokinins. It has been reported that as ABA levels decrease during the stratification of sugar maple seed, the gibberellins and cytokinin levels increase (Van staden *et al.*, 1972; Webb *et al.*, 1973). Thus, an inverse relationship between inhibitors and promoters occurs during dormancy breaking (after-ripening process).

Combinations of dormancy types

The types of dormancy described are by no means mutually exclusive, and more than one mechanism for the imposition of dormancy may be used by seed. McDonald and Khan (1977) report that Indian rice grass (*Orizopsis hymenoides*) seed possesses both seed coat (exogenous) and physiological (endogenous) dormancy. Seed coat dormancy in this species can be effectively removed by scarification with sulfuric acid. However, addition of exogenous GA3 further enhances germination of freshly harvested seed to 70 percent. Seed that has been after-ripened in dry storage is not as responsive to exogenous GA3 treatment, presumably because the seed has had enough time to synthesize this compound.

Secondary Dormancy

Seed which can germinate after having been shed from the mother plant may be induced to become dormant by being kept under unfavorable environmental conditions. A study of winter barley and spring wheat indicates that secondary dormancy can be induced by:

- Exposure of dry barley seed to temperatures between 50–90° C.
- Seven day storage of winter barley at high moisture content at 20° C.
- One day storage of spring wheat at high moisture content in airtight containers.
- Placement of seed under water and in darkness for 1–3 days at 20° C.

Induction of secondary dormancy was possible one and a half months after physiological maturity of the seeds. However, the induction of this dormancy decreased as the time between physiological maturity and treatment increased. In general, this state of secondary dormancy can be removed by methods used to remove primary dormancy such as chilling, illumination, gibberellin, and storage at 20° C.

Two suggestions have been proposed to explain the mechanism of secondary dormancy:

- The imposition of a block at crucial points in the metabolic sequence leading to germination.
- Unfavorable balance of growth-promoting versus growth-inhibiting substances (Copeland and McDonald, 1985).

Methods of Overcoming Dormancy

In natural conditions one or more of the following factors may operate to overcome seed dormancy:

- Light.
- Temperature.
- Aging.
- Changes in the covering structure (Bewely and Black, 1982).

In the laboratory, various methods have been to break seed dormancy. Some of the simple and widely used methods are described below.

Scarification

Any mechanical or chemical treatment that weakens the seed coat is known as scarification. This method is used when dormancy is due to the physical properties of the seed coat (exogenous dormancy).

Mechanical scarification

Seeds are either rubbed by sand paper or mechanically scarified. Care should be taken not to cause any damage to the axis of the seed of, for example, green gram (*Phaseolus aurea*). Absorption of water by the seed is accomplished by piercing the seed coat with a needle in, for example bitter melon (*Momordia charantia*), and oat. In rubber (*Hevea* spp.) the seed coat has to be removed completely. Brief immersion in boiling water is an effective method of breaking the hardness of the seed coat in legumes. Care should be taken in deciding treatment duration, since with some crops, such as lentil, soaking for more than one minute was found to be injurious to germinability (Agrawal, 1987).

Other techniques such as drastic temperature change, vigorous shaking of seed (impacting), or exposure to radio frequencies alter seed coat integrity, thus permitting penetration of both water and gases.

Chemical scarification

Chemicals may also be used to cause degradation of the seed coat. Soaking hard-coated seed in concentrated or diluted sulfuric acid removes seed impermeability (cotton seed). Seed must be thoroughly washed and dried after acid treatment, as reduction in germination may occur from even slight over scarification with sulfuric acid.

Recent techniques include the use of selective seed coat enzymes such as pectinase and cellulase to degrade the seed coat. Since many seed coats contain water-insoluble compounds that retard water entry into the seed, organic solvents such as acetone and alcohol have been used to dissolve and remove these compounds and permit water entry into the seed (Copeland and McDonald, 1985).

Temperature Treatment

When dormancy is due to endogenous factors (embryo development or presence of inhibitors), seed is subjected to stratification, i.e., incubation of seed at low temperatures (0–5° C) over a moist substratum for 3–10 days (to break the dormancy) before placing it at optimum temperature for germination. Examples include cherry (*Prunus*) and mustard (*Brassica campestris*) seed.

Prolonged stratification (2–6 months at 5–10° C) is required for a number of *Rosaceae* spp. to break the dormancy (Agrawal, 1987). It is well known that physiological changes occur in imbibed seed exposed to low temperatures (stratified seed). The embryonic axis of stratified cherry seed increases

in cell number, total length, and dry weight (Onley and Pollock, 1960). A shift in hormonal levels also occurs in stratified seed. The ABA level drops during stratification of apple, walnut, and hazelnut seed. In some seed, the addition of exogenous gibberellins can substitute for the stratification requirement (Pinefield, 1968), implicating this hormone as a promoting agent.

The stratification requirement of a particular seed lot also depends on the age of the seed. Freshly harvested seed of Chinese maple (*Acer truncatum*), which requires a two-month stratification, germinated well after one year in storage (Ackerman, 1957). For most cereals, storage of dormant seed for one to two months at 15–20° C (after-ripening during storage) is sufficient to allow maximum germination. Most seed with an after-ripening during storage requirement is responsive to stratification.

Treatment with gibberellins can also remove the dormancy block in most cereals. Since gibberellins and stratification can replace dry storage after-ripening treatment, this process appears to be essential to allow the embryos to synthesize gibberellins, again suggesting that a shift from inhibitor to promoter must occur before germination.

Light Treatment

Some seed does not germinate in the dark. Therefore continuous or periodic exposure to light can be essential to break endogenous dormancy. Three well-known species whose dormancy is broken by exposure to red light (670 nm) are lettuce, birch, and Virginia pine (Copeland and McDonald, 1985).

Treatment with Growth Regulators and Other Chemicals

Since endogenous dormancy may be due to the presence of germination inhibitors, application of low levels of growth regulators may break dormancy. Different groups of chemicals have been reported to break dormancy. GA3 and kinetins are the most widely used chemicals. GA3 has been found the most effective in breaking dormancy in many cases. The seed of *Perilla* spp. is either soaked in a solution of 100 ppm GA3 and 10–50 ppm kinetin before transfer to a moistened substrate, or germinated in the substrate moistened with a GA3 or kinetin solution.

Among other chemicals, potassium nitrate and thiourea are widely used to break dormancy. Potassium nitrate breaks dormancy of light-requiring seed in the dark. Potassium nitrate (0.2 percent) has been found to be effective in breaking dormancy of oat and barley seed. Thiourea (3–9.5 percent)

breaks dormancy of both light- and chill-requiring seed. In lettuce seed, the effective concentration is 10^{-2} to 10^{-3} M. Thiourea has also been found to stimulate the germination of gladiolus seed (Agrawal, 1987).

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Section III
Genetic Aspects

Genetic Structure in Natural Populations

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Introduction

Plant populations are not random assemblages of genotypes, but are structured in space and time according to various parameters. Genetic structure stems from the joint action of mutation, migration, selection, and drift, which are in turn influenced by eco-geographical and biological factors. Factors affecting the breeding system and gene flow are important to the genetic structure of plant populations.

Genetic make-up is passed down through the generations by reproduction, while gene flow determines the post-reproductive patterns of gene dispersion within and between populations.

Because plants are immobile, their genetic structure implies spatial structure as well as the physical distribution of individual plants. The distribution of genotypes is not random (Levin, 1977), but the result of two opposing forces (Spieth, 1979):

- Selection, which tends to produce a distribution pattern with parallel environmental heterogeneity.
- Gene flow, which tends to produce a uniform distribution.

The magnitude of gene flow depends on the breeding system, longevity, seed and pollen dispersal mechanisms, and the population density of the species (McNeilly and Antonovics, 1968; Price and Waser, 1979).

Selection is a ubiquitous feature of a natural population. It alters gene and genotype frequency and acts in concert with migration, dispersion, and other processes—such as plant density, environmental heterogeneity and generation length—to create genetic structure. The magnitude of selection is usually higher than gene flow. Moreover, selection varies in ways that cannot be anticipated by the plant population. As a result, it is local and idiosyncratic in terms of how it alters genetic structure; it cannot generally be used to predict the distribution of genetic variation within or between populations.

Lack of information on the genetic structure of plant populations is a serious problem, since any understanding of speciation, adaptation, or genetic

change must take into account genetic patterns and the processes by which they are modified (Bradshaw, 1972; Antonovics, 1976). A well-known genetic structure and the factors that influence it are extremely important to seed production and conservation (Lorenzetti and Porceddu, 1976). Considerable genetic change can occur during seed multiplication, usually due to selection rather than gene flow (Snaydon, 1978).

Factors Affecting Genetic Structure

The main factors affecting the genetic structure of populations are summarized in Table 1. Since genetic structure is dependent in part on genetic variation within a population, a summary of how these factors affect genetic variation within populations is also provided. Because factors influence each other, and vary over time, populations may differ in the degree to which they demonstrate predictable genetic effects. Different combinations of characteristics may result in similar genetic structures in different species (Jain, 1976b).

Table 1. Factors affecting the genetic structure of populations and their possible effects.

	Genetic variation within	within	Genetic structure between
Mode of Reproduction			
Breeding system			
Apomixis	Low	Homogeneous	Founder effects, drift
Autogamous	Medium homozygosity	Reduces heterozygosity	High diversity due to drift; low gene flow
Allogamy	High heterozygosity	Increases population size	Reduces divergence due to increase gene flow
Floral morphology			
Hermaphroditic	Moderate	Depends on breeding system	
Dichogamous	Potentially high	Usually predominant in outbreeding	
Heterostylous	High	Outcrossing genetic structure depends on sex ratio	
Gene flow			
Pollination mechanism			
Small bee	Low	Limited pollen movement; increased differentiation	
General entomophily	Low		
Large bee	Low	Reduced subdivision	
Butterfly/moth	Low		
Bird/bat	Low	Long distance pollen dispersal	
Wind	High	Large population size	

continued

	Genetic variation within		Genetic structure	
	within	within	between	
Seed dispersal				
Gravity	Medium	Reduce population size	Promoted differentiation	
Explosive/capsule	Medium	Depends	Could prevent divergence	
Wind	High	Increase population size	Could prevent divergence	
Animal-attached	Low	May reduce clumping and family structure	Regularly promotes homogeneity	
Animal-ingested	Medium	May reduce clumping and family structure	Regularly promotes homogeneity	
Seed dormancy				
Present	Increases potential genetic variation		Retards loss of alleles, buffer	
Generation length				
Life cycle				
Annual	Low homozygosity	Increases susceptibility to drift, smaller population size	Increases chances of subdivision	
Long-life	High	Retards loss of variation, increases mating opportunity	Increases chances of migration and divergence	
Timing of reproduction				
Monocarpic		Reduces flower density and mating possibility	Promotes drift and divergence between populations	
Polycarpic		Increases population size, reduces probability of subdivision	May inhibit divergence	
Pattern of distribution				
Geographical range				
Endemic	High	Differentiation	Small, local population divergence due to drift and isolation	
Narrow, regional	Moderate			
Widespread	Low			
Population size				
Large and stable	High	Potentially subdivided	Between drift and migration, small population, more drift	
Small and stable	Lower	Homogeneous	Between drift and migration, small population more drift	
Fluctuating size	Low due to drift	Homogeneous due to loss of variation in periods of small size		
Population density				
High		Reduce pollen flow and increase subdivision		
Low		May increase pollen flow (increase population size) or decrease pollinator visits (decrease population size)	Long distance flow increases homogeneity	

continued

	Genetic variation within	within	Genetic structure between
Spatial population distribution			
Patchy		Affects pollination behavior; reduces gene flow and population size, gen. subdivision	Increased isolation reduces gene flow and enhances differentiation
Uniform		Promotes gene flow and reduces subdivision	Promotes migration and homogeneity
Population shape		Subdivision is increased in linear habitats	Divergence enhanced in linear arrays of populations
Inter-specific Hybridization	High		
Karyotype Variation and Recombination System	Higher chromosome number, increases recombination	High chromosome frequency	
Environmental Variation	Higher environmental variation, higher genetic variation	Small, depends on gene flow	High diversity
Phenotypic Plasticity	High variation	Higher diversity	Inhibits population differentiation

Mode of Reproduction

“Mating systems indeed hold a key place in the study of plant populations....In this connection, four points need to be stressed. First, reliable estimates of mating system parameters are a fundamental prerequisite of plant studies. Second, we need to go beyond the simple dichotomy of inbreeders vs. outbreeders by making allowance for the complexity of the diverse mating systems of different plant species....Third, mating systems are flexible in space and time, and such variation is likely to have profound effects on the genetic structure of populations....Fourth, variation in mating systems is subject to genetic control, and mating systems themselves evolve.” (Brown, 1979).

The mode of reproduction is the major factor affecting the genetic structure of a population. It includes (i) the breeding system, which is the formal passage of genetic information down the generations; and (ii) the floral morphology, which affects the amount of information and the probability of generational heredity. The breeding system is more important. The next section will focus on breeding system characteristics and their effect on (i) genetic variation; (ii) colonizing ability; and (iii) evolution.

Breeding System

Characteristics of breeding systems

Three main types of breeding system are usually recognized: outbreeding, inbreeding and apomixis, though these three systems are only points along a continuum.

Outbreeding increases the incidence of both genetic recombination and gene flow, furthering the maintenance of genetic variability, and increasing the potential for evolutionary change (Rollins, 1967). However, some authors (e.g. Cruden, 1977; Lloyd, 1984; Levin, 1986) have stressed the disadvantages of outbreeding, such as the cost of pollen production and the uncertainty of seed production.

By contrast, inbreeding restricts genetic recombination and gene flow, leading to homozygosity and homogeneity within populations, especially among annuals and pioneer species (Rollins, 1967).

In apomixis, reproduction is asexual, and progeny have exactly the same genetic constitution as the maternal parent. The offspring from a given parent can be heterozygous and genetically homogeneous. Apomixis is much less common among plants than either inbreeding or outbreeding.

Inbreeding has adaptive advantages under certain conditions (Antonovics, 1968; Hillel *et al.*, 1973a and 1973b). For example, reduced genetic recombination can: (i) maintain specific adaptive gene combinations; (ii) expose recessive genes, thus ridding the population of harmful characteristics; (iii) provide greater certainty of fertilization and seed production in adverse environments; and (iv) allow plants to reproduce in isolation. The main disadvantage of inbreeding is that it reduces recombination, thus preventing the construction of novel recombination, and hence restricting evolutionary potential (Table 2). The tendency to homozygosity and genetic homogeneity within inbreeding populations is disadvantageous under certain conditions. The disadvantages of reduced recombination by inbreeding are partially offset, in some species, by a greater number of chromosomes and by higher chiasma frequency (Stebbins, 1950), both of which increase recombination.

Outcrossing generally reduces correlation between uniting gametes, thus increasing population size and reducing subdivision. It also increases pollen movement and gene flow.

Table 2. Theoretical consequences of inbreeding (from Jain, 1976a).

- 1) High level of homozygosity; lowered load due to sheltered recessives.
- 2) Role of selection among homozygotes; allelic substitution more rapid; fewer polymorphic loci and fewer alleles/locus; closer local adaptation.
- 3) Greater selection among homozygotes; allelic substitution more rapid; closer local adaptation.
- 4) Lower recombination; multilocus associations giving biotypes within populations.
- 5) Occasional hybridization among biotypes and segregation leading to release of new variability.
- 6) Greater reproductive economy (smaller cost of pollination); "ineffective" principle of colonization.
- 7) Founder effects during colonization; intercolony diversity; smaller pollen flow within and among populations or races.

Breeding systems and genetic variation

Breeding systems are an important factor determining the amount of genetic variation within populations (Allard, 1975; Schoen, 1988). However, other factors, such as selection (Hayman, 1953; Hayman and Mather, 1953; Allard *et al.*, 1968; Karlin, 1968), seem to be even more important. In general, it has been assumed that genetic variation within populations is reduced by inbreeding (Rollins, 1967; Zangerl and Bazzaz, 1984; Levin, 1986), while genetic difference between populations is increased by inbreeding (Allard and Hansche, 1964; Hillel *et al.*, 1973a; Hamrick, 1983). However, when parallel sampling is carried out on species with contrasting breeding systems, comparable levels of genetic variation are found (Kannenbergh and Allard, 1967; Hillel *et al.*, 1973a; Helgadottir and Snaydon, 1986). In spite of many studies, there is no definitive answer as to whether inbred species are genetically less variable than outbred species (Jain, 1976a).

Complete inbreeding rarely occurs within a population or species. A mixed mating system is more frequent. Even a very small percentage of outcrossing can generate considerable recombination and heterozygosity (Hamrick and Allard, 1972; Schaal, 1975; Levin and Kerster, 1974; Adams and Allard, 1982). For example, in subterranean clover, which is considered a very strongly self-fertilizing species (Morley, 1961), small amounts of outcrossing occur and may be of long-term evolutionary significance (Marshall and Broué, 1973). As a consequence, the genetic structure of the mixed mating population resembles that of outcrosses.

In populations that are vegetatively propagated, variability in clones is due only to environmental factors, except for eventual mutations. Propagated populations have a large potential genetic variability, which becomes a real variability only if it is passed on by seed.

Colonizing ability

Self-compatibility offers advantages for colonizing species (Price and Jain, 1981; Jain, 1983), since a single individual can provide the basis for a new population (Baker, 1955 and 1967; Stebbins, 1957 and 1958; Antonovics, 1968). Enhanced certainty of reproduction under adverse conditions (Rollins, 1967; Jain, 1975a) is also advantageous in pioneer conditions. There is evidence of the breakdown of self-incompatibility mechanisms during colonization (Lloyd, 1980; Jain, 1983; Innes and Hermanutz, 1988). On the other hand, there are reasons to believe that crossbreeding would be advantageous for plants colonizing a new environment. For example, novel gene combinations are more likely to be generated by outbreeding (Jain, 1976a and 1979; Foin and Jain, 1977). There is no consensus on the extent of the advantages of inbreeding in colonizing species (Ehrendorfer, 1959 and 1965; Stebbins, 1965), or on why inbreeding seems more common in marginal, peripheral and disturbed habitats (Vasek, 1968 and 1971).

Breeding system and evolution

Since the time of Charles Darwin (Darwin, 1876), breeding systems have been thought to influence evolution (Kahler *et al.*, 1975; Jain, 1976b; Lloyd, 1980; Lande and Schemske, 1985; Schemske and Lande, 1985). Many authors (e.g. East, 1940; Lewis, 1942; Stebbins, 1950; 1970 and 1974; Baker, 1959; Fryxell, 1957; Grant, 1963 and 1971; Wyatt, 1983 and 1988) think that outbreeding is the ancestral breeding system for angiosperms, and that inbreeding is a derived condition.

The mating system of plants is not entirely stable. It can be modified by environmental conditions (Campbell and Abbott, 1976; Clegg, 1980; Rick, 1988; Hedrick, 1990), and can change in response to selection (e.g. Kahler *et al.*, 1975; Taylor *et al.*, 1979; Kreitner and Sorensen, 1985; Rick, 1988; Wyatt, 1988) so that populations within the same species may have different degrees of inbreeding.

Some species have a very low percentage of outcrossing; for example *Trifolium subterraneum* has an outcrossing percentage between 0.00–0.22, with an average of 0.07–0.15. Practical experience has shown that outcrossing can be ignored in multiplication schemes, assuming proper certification procedures. Nevertheless, the low rate of outcrossing could be of considerable long-term evolutionary significance (Marshall and Broué, 1973).

Floral Morphology

Floral morphology can give an indication of the breeding system, the genetic structure of a population, and whether a plant species is completely self-fertilizing. The rate of hybridization depends on floral morphology.

Plant hermaphrodites can have any breeding system, but only hermaphrodites can self-pollinate. Dichogamous and heterostylous species are usually cross-fertilizing, and thus may have less pronounced population differentiation. The rate of hybridization depends on sex ratio and amount of pollen production.

Gene Flow

Cultivars of outbreeding species are susceptible to pollen drift from adjacent populations during multiplication. Rigid rules governing isolation distances are therefore laid down in most countries. Nevertheless, these rules are often insufficient, since genetic change is also caused by other factors.

Pollination mechanism

The agents responsible for pollen dispersal have a great effect on the genetic structure of a population. The amount of pollen transported, and the distance it travels, are functions of the agent.

Actual pollen flow is known for only a few natural populations (Schaal, 1980; Handel, 1983). Usually, the larger the distance of pollen movement, the lower the genetic differentiation and variability between populations. Insect-pollinated species are less variable, since insects visit the same kind of flowers on the same day (honeybees may visit the flowers of many plant species during the year, but not the flowers of more than one species during the same foraging flight). Some plant species have evolved jointly with bee species, so that one species of plant is pollinated by only one species of bee, which, in turn, is attracted only to flowers of that species of plant.

The agents responsible for pollen dispersal can be grouped according to their specialization and pollen movement (Table 1).

Seed dispersal

Seed dispersal is analogous to pollen dispersal, since the effect on population size and variability is a function of the variance in dispersal distance. Low seed dispersal—for example, dispersal by gravity—can be partially compensated by frequent seed generation. Different modes of seed dispersal (Table 1) have different dispersal distances according to the given situation. For example, wind dispersal depends on wind speed and frequency, seed size and shape. Seed dispersal also depends on population size, as will be seen below.

Seed dormancy

Seed dormancy acts as a buffer to genetic differentiation. Plant species with seed dormancy accumulate seeds in a seed bank, which is a reserve of several genotypes selected in different years under different environmental conditions. Dormancy thus increases population size, retards loss of alleles and increases potential genetic variation.

Generation Length

Phenology

The genetic structure of populations is affected by flowering time, which can isolate individuals even better than self-fertilization or physical isolation. Synchronous flowering retards differentiation, increasing gene flow between individuals.

Life cycle

The life cycle positively affects population size. A long life cycle has the same effect on population as seed dormancy.

Timing of reproduction

Monocarpy reduces flower population density and mating events. It promotes drift and divergence between populations (i.e. population differentiation).

Pattern of Distribution

The main effect of the distribution pattern is on gene flow—especially pollen movement—and thus on the genetic structure of plant populations.

Geographical range

The geographical range is influenced, at least partially, by the dispersal ability of the species. Small, localized species have limited gene flow and higher drift, increasing differentiation within populations and reducing genetic differentiation between populations. Historical factors and habitat heterogeneity play a significant role in species distribution.

Population size

Population size can be stable or unstable. Stable populations can be either large or small. In large, stable populations, differentiation depends on gene flow. In small, stable populations migration is more effective in alternating gene frequency, but they are more susceptible to drift and fixation.

In fluctuating populations the structure depends on how long the period of small size lasts, because small populations reduce allelic diversity. Reduction in heterozygosity depends on how quickly the original population size is restored. Therefore, fluctuating populations may have a random drift (Motro and Thomson, 1982).

Population density

High population density reduces pollen movement, resulting in higher differentiation within population-increasing subdivisions. Low population density has no uniform relation with pollen movement, which is related to pollen vectors. Population size increases with higher pollen movement. Low density can increase long distance flow and homogeneity, or the pollinator might not visit low density populations, being more attracted to areas with high flower density, thus decreasing population size.

Spatial population distribution

Patchy distribution can have many effects on pollinator behavior, either restricting movement within a patch or promoting movement among patches. Limited dispersal between patches promotes differentiation. Uniform distribution promotes gene flow, reduces subdivisions within populations, and promotes migration and homogeneity between populations.

Competition

Competition, especially for scarce resources, may generate intense selection pressures, large selection differentials and rapid change in the genetic structure of populations.

Table 3. Reproductive isolation mechanisms (from Haldley and Openshaw, 1980).

	PRE-FERTILIZATION
External barriers	
I. Physical isolation	
II. Physiological isolation	
A. Barriers between the parental species	
1. Ecological isolation	
2. Seasonal isolation	
3. Mechanical isolation	
Internal barriers	
4. Prevention of fertilization	
	POST-FERTILIZATION
B. Barriers in the hybrids	
1. Hybrid invariability or weakness	
2. Failure of flowering in the hybrid	
3. Hybrid sterility	
4. Inviability and weakness of F ₂ and later generations	

Inter-specific Hybridization

The process of speciation leads to the development of reproductive barriers between populations, and maintains the integrity of species by restricting the flow of genes from one species to another. Barriers, such as pre- and post-fertilization, internal and external, prevent gene flow and maintain genetically isolated species (Table 3).

When barriers are overcome, individuals from separate populations are brought together and crossed to produce fertile hybrids that allow the free flow of genes from one population to the other. The two populations should not be considered distinct species, according to the definition of a biological species. During an inter-specific hybridization, three phenomena can occur (Briggs and Knowles, 1967):

- One or a few genes are transported from one species to another.
- New character(s) expression(s) are achieved.
- New allopolyploid species are produced.

Karyotype Variation and Recombination System

A greater number of chromosomes and a higher chiasma frequency both increase recombination and sometimes make up for the disadvantages of reduced recombination due to other factors, such as inbreeding.

Environmental Variation

Changes in environmental conditions may influence the genetic structure of plant populations, particularly in pastures.

Cultivars are exposed to extensive changes in environmental conditions during reproduction and use. The breeder may select under one set of conditions (e.g. spaced plants at one location), seed may then be multiplied under another set of conditions (e.g. wide row spacing at another location), and the cultivar may be used agriculturally under yet another set of conditions (e.g. in a dense sward, at low soil fertility, mixed with other species, or at a variety of locations). With each change the cultivar is susceptible to genetic change. Therefore, it is extremely important that each produce its own seed.

Climatic conditions vary greatly over time, on a scale of minutes or millennia. Variations can be either cyclic (e.g. diurnal and seasonal) or random (e.g. day-to-day and year-to-year). Soil conditions are less variable than climatic conditions, but still vary considerably between seasons (Raupach,

1951a and 1951b; Gupta and Rorison, 1975). These changes may influence the genetic composition of populations. Changes in response to climatic factors, soil factors, and disease have been detected over successive multiplication cycles.

Phenotypic Plasticity

Any spatial heterogeneity that occurs in an individual plant, or within its life span, must be tolerated by that individual (Sultan, 1987). Phenotypic plasticity is the ability of a single genotype to produce more than one morphological type, or physiological state, in response to change in environmental conditions (Bradshaw, 1965). Since phenotypic plasticity allows plants to respond rapidly to environmental change, it is more common in colonizing plants, which encounter many different conditions (Wu and Jain, 1978).

Although plasticity is classified as non-genetic, it is under genetic control (Bradshaw, 1965; Jain 1979 and 1983) and is subject to natural selection. Extreme phenotypic plasticity is supposed to inhibit population differentiation, since organisms with broad plasticity can tolerate environmental changes, which protects the genetic structure from the effects of selection. However, phenotypic plasticity may sometimes maintain genetic variance, since different genotypes may coexist.

Inbreeders usually have greater phenotypic plasticity than outbreeders (Table 3).

Changes During Seed Multiplication

Changes in the genetic structure of populations occur during seed multiplication as a result of:

- Differences in establishment.
- Differences in reproductive capacity.
- Differences in environmental response.
- Gene flow.

Seed multiplication generally favors genotypes that are short-lived, establish rapidly and produce the most seed. It is by no means certain that genetic change that occurs during multiplication is harmful.

Conclusion

The breeding system is the main factor affecting the genetic structure of populations. Nevertheless, there are many factors that may interfere with and modify a breeding system. Generally, the genetic structure of a population is a consequence of all the effects acting upon it, as well as neighboring populations. Seed production and maintenance are important to (i) create a population with a well-known and specific genetic structure; and (ii) maintain genetic structure. It is therefore essential to have adequate knowledge of the factors affecting the genetic structure of plant populations.

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Varietal Development in Autogamous Crops

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Varietal development in autogamous crops can be accomplished through the application of many methods, depending on the materials available and the resources available to the breeder, as well as his or her personal preferences and beliefs. In dealing with natural populations one either develops pure lines, or, through mass selection, produces cultivars consisting of a mixture of selected plants. For artificial populations the methods used are either F_1 hybrids, when heterosis or other advantages is shown, or the backcross method, when the objective is to add a trait to an otherwise desirable variety. If selection is to be practiced in the segregating generations, then the pedigree method of selection, which relies on detailed records of the individual plants selected and their offspring, is usually used. The single seed descent method is used when the material must quickly reach a high degree of homozygosity, followed by extensive testing of the resulting lines. The bulk method of breeding is employed when the goal is to take advantage of the effects of natural selection on the segregating populations for a few generations, followed by the pedigree method of selection. Multiline varieties, usually mixtures of isogenic lines differing in the specific genes for resistance to some pathogen, are used when a disease is the limiting factor for the successful growth of a crop in a region. The diallel selective mating system relies on large numbers of parents, which are crossed in as many combinations as possible, after which the resulting F_1 hybrids are either used for pure line selection or intercrossed to produce further populations. The resulting populations are either used for pure line selection or for the production, through selection and intermating, of new populations for further selection and crossing.

Introduction

China and India alone add to the world one person per second to feed, clothe and shelter, while the area available for agriculture is, rather than growing, shrinking. If we are to face this challenge successfully we must of necessity produce more plant products per unit area, even from areas which are currently considered marginal for agricultural production. The task of producing more per unit area is accomplished through the combination of

better agricultural practices (crop growing practices and application of agricultural chemicals) and better adapted plant genotypes, i.e. better cultivars. It is generally accepted that the genotype accounts for almost 50 percent of the yield increases that have been obtained in several crops in the last 50 years (Duvick, 1984).

The task of producing better cultivars falls to that branch of science known as plant breeding, which is defined as the art and science of producing new plant forms for financial gain and enjoyment.

In this paper we will describe the methods used for producing new cultivars for the self-pollinating cultivated plants upon which humankind depends for survival.

Classification of Methods

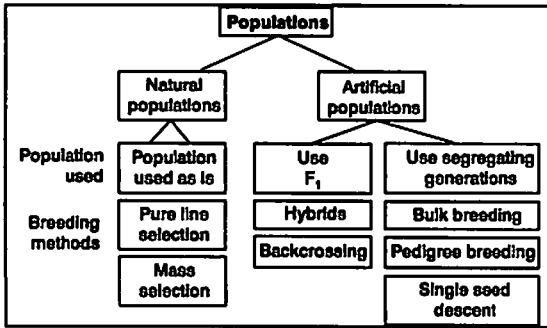


Figure 1. Classification of the various methods used for the improvement of self-pollinating plants.

For plant breeding purposes cultivated plants are traditionally divided according to their system of breeding, i.e. allogamous or autogamous, and the methods used for their improvement are described as methods primarily suited to one or the other group. Based on this criterion methods can be classified as shown in Figure 1.

An alternative classification depends on the end product, i.e. the kind of cultivar produced, irrespective of the breeding system of the crop species, as shown in Figure 2. In this paper we will follow the traditional method of describing the breeding methods but will also remark on the cases where a method used primarily for self-pollinating species is at some point used in the breeding effort of a cross-pollinating species.

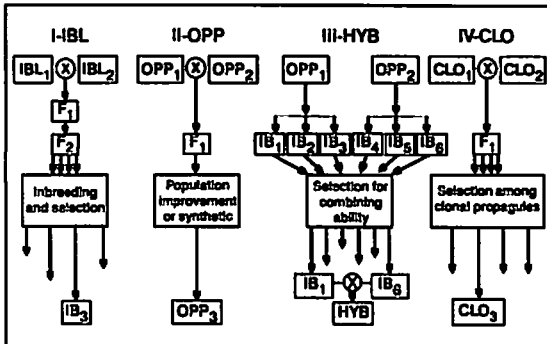


Figure 2. Populations and breeding methods (Simmonds, 1979). IBL and IB=inbred line; OPP=open-pollinated population; Hyb=hybrid; Clo=clonal population.

The methods used for the improvement of self-pollinating species, of which wheat and barley are the prime examples, can be subdivided according to whether artificial or natural populations are used.

Natural Populations

If populations are used as they are found in nature, it is imperative that they possess genetic variability so that artificial selection can be practiced successfully. Provided variability exists, one can either endeavor to produce pure lines, or, through mass selection, produce a population that can be grown on a commercial basis.

With pure line selection (Fig. 3) a sufficiently large number of plants from the population are grown at distances permitting the expression of the genetic potential of the plants and selection on an individual plant basis. Those plants that, primarily on a visual basis, are considered by the breeder as superior, are harvested individually, and their progeny are planted in the observation field in a single row. There are as many rows as plants selected, plus a sufficiently large number of check rows, the check usually being the best local variety. This practice is repeated for a few years until all rows considered inferior to the check are rejected. Those rows that have been selected are evaluated in trials with sufficient replication in time and space in comparison with the local check. The row that performs better than the check is

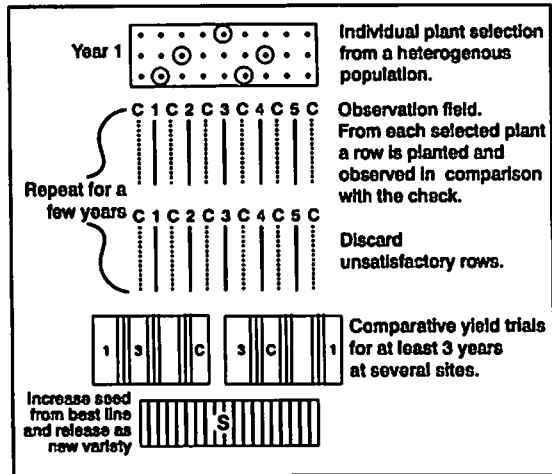


Figure 3. Pure line selection.

multiplied as a new variety. In this method the new variety is composed of the descendants of a single plant that was selected at the beginning of the breeding program.

With mass selection, plants from the population are planted as described above, but the progeny of all selected plants are bulked and planted in the field together (Fig. 4) and the process is

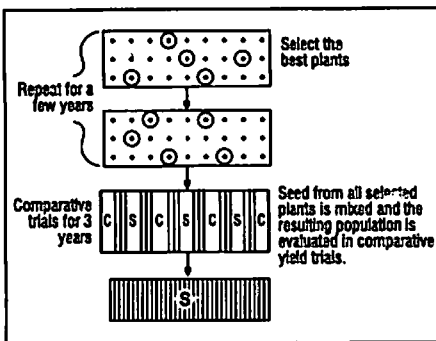


Figure 4. Mass selection in self-pollinating crops.

repeated until no further selection can be practiced among the remaining plants. The bulk is then evaluated in regional trials in comparison with the local check, and if the bulk outperforms the check it is multiplied and distributed as a new variety. The variety thus produced is composed of the offspring of all selected plants.

Mass selection has been used for the improvement of landraces and local varieties, and by those charged with maintaining the characteristics of existing varieties. Pure line selection can be used on material imported from some other area that now shows its cryptic variation in the new environment and must be cleaned up.

Artificial Populations

These populations are usually the result of deliberate crossing and can either be used commercially as they are (in the form of F_1 hybrids), in backcrossing, or in artificial selection of the segregating generations resulting from them. In the latter case the methods used for selection are the pedigree method of selection, bulk breeding and the single seed descent method.

The Choice of Parents

In starting a breeding program based on crossing and selection, the breeder must very carefully and clearly outline his or her objectives and then select suitable parents for crossing. In choosing these objectives, the breeder must be realistic and guided by the experience of others who have gone before, which teaches that improvements come in small increments and that what counts is a steady flow.

The choice of parents is very important in that between the two of them they must combine the traits we want to have in the new variety. Usually one of the parents is a good and well-adapted variety which must be made even better by adding desirable traits from another parent. This other parent must express the traits we want to add to the old variety in a degree greater than what we expect to have in the new variety. Sometimes we just choose two good varieties and cross them, hoping that in their descendants we will be able to find an even better variety, i.e. cross the best with the best and hope for the best.

F₁ Hybrids

These products will be described under the methods for the improvement of cross-pollinating species where they have found their best application and commercial exploitation.

The Backcross Method

This method is used when we have a variety that is lacking in one trait, which is usually controlled by one or very few genes, but is otherwise quite acceptable. This variety is called the *recurrent parent* and the parent which is lacking the trait from the accepted variety is called the *donor parent*. The recurrent parent is crossed as the female to the donor, and the resulting hybrid is backcrossed as the male parent to the recurrent parent five to seven times (Fig. 5). In each successive backcross care must be taken that the plants used possess the trait being transferred from the donor to the recurrent parent. This necessitates an additional generation of selfing if the trait being transferred is controlled by a recessive gene.

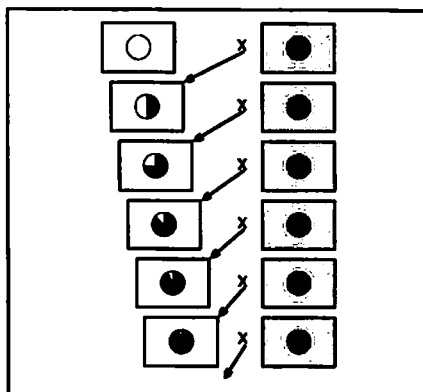


Figure 5. The backcrossing procedure. The black circle represents the recurrent parent.

With each successive backcross—provided certain conditions hold—the genetic material from the donor is halved in such a way that by the seventh backcross the resulting material is almost the same as the recurrent parent, with the addition of the trait obtained from the donor parent.

The method can be used in cross-pollinating species and also for the transfer of polygenically controlled traits, but in both these cases large numbers of individuals must be used in each successive backcross.

The chief advantages of the method are its speed and predictability, in the sense that the end product is known and there is no need for additional testing. The method has been used extensively for the transfer of genes conferring resistance to disease conditioned by single genes, as is the case of cereal rusts. The method has also been used in the development of multiline varieties (see below).

The Pedigree Method of Selection

In this method the hybrid resulting from the cross of two parents is advanced to the F_2 generation. The plants are space planted in the field, and selection is practiced, mainly visually, for traits with high heritability values. Each selected plant is harvested separately and its offspring planted in a field in a row in the following generation (F_3). Selection is now practiced within each row, and usually the best plants of the best rows are harvested separately (Fig. 6).

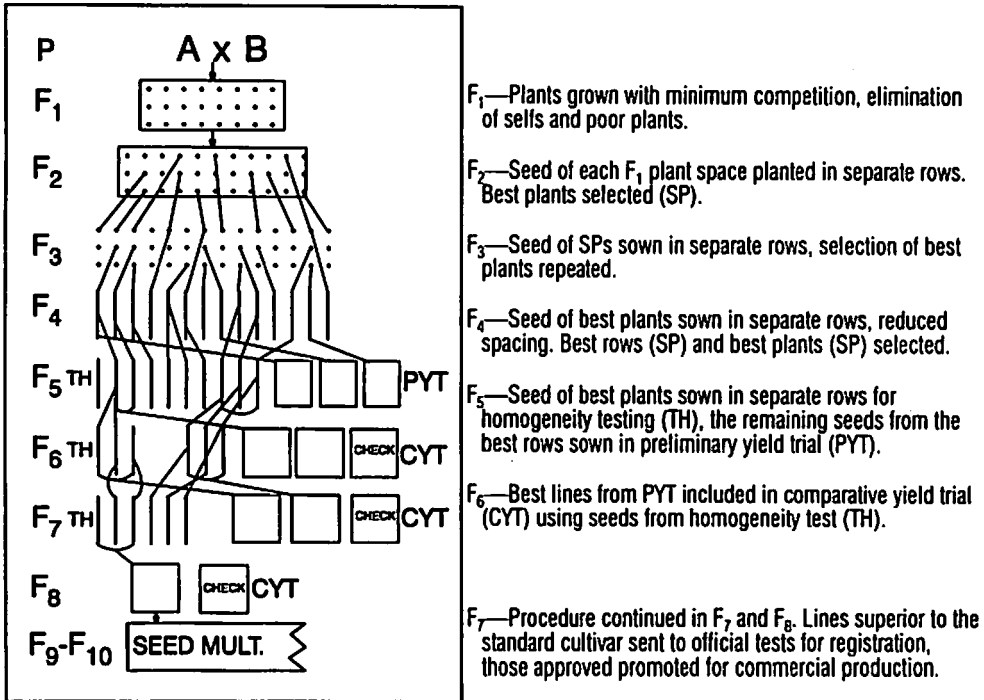


Figure 6. The pedigree method of selection (From Borojevic, 1990).

The descendants of each selected plant (now in the F_4) are planted in separate rows but at reduced distances and then again the best plants and the best rows are selected. In the following generation (F_5), the seed from the best plants is planted in separate rows for homogeneity testing while the remaining seed of each selected plant is used for preliminary yield testing.

The number of replications in these trials depends on the amount of seed available. In the F_6 the best lines from the preliminary yield testing are subjected to comparative yield trials using seed from the homogeneity part of the field. These trials must have sufficient replication and be carried out under conditions similar to those used by the commercial farmers. The

same procedure is used for two additional generations, and if lines superior to the best local cultivar are obtained they are submitted for evaluation by the authority in charge of licensing and registering new cultivars.

During the successive generations of selfing and selection, detailed records are kept of the pedigree of the plants and the lines selected so that no duplicate lines are carried along in the program.

This method constitutes perhaps *the* method used by all breeders of self-pollinating crops, and perhaps exists in as many variations as there are breeders, but the salient features of the method are those described here. The method requires a lot of manpower and equipment.

It can be seen that the development of a new variety by this method requires a minimum of ten generations, which in a crop with only one generation per year means ten years. This is a long time, and many ways have been found to speed up the process, such as growing two or more generations per year by going to the opposite hemisphere, or utilizing nurseries near the equator.

The advantages of this method are:

- If selection is effective, all inferior genotypes are discarded before they reach the stage of expensive yield evaluation.
- Because selection is practiced over many seasons there is the possibility that all existing variability will be expressed and exploited.
- The relationships of the families selected are known, with the result that it is possible to maximize genetic variability on which selection operates.

The drawbacks of the method are:

- Because of the inbreeding and the lack of opportunity for further crossing, the chances for further genetic recombination are reduced.
- Because only two parents are used the genetic material is limited.
- Usually the method results in the gradual collection of linked groups of genes that do not permit recombination and the formation of new gene combinations.

Bulk Breeding

Some breeders think that it is almost impossible to select for the most economically important traits in the first segregating generations. Furthermore, since in these generations plants are spaced widely, their yield and behavior

may not be the same as when they are planted in the farmer's field under the usual conditions. For these reasons, some breeders prefer to exploit the effects of natural selection by seeding in bulk from F_2 to F_4 , harvesting the whole plot in bulk without practicing any selection, and then reseeding (Fig. 7). This practice is repeated for a number of generations that varies according to the breeder and the degree of relatedness of the parents used in making the cross. When homozygosity has increased, usually past F_5 , then the plants are space planted and selection begins, utilizing the pedigree method.

This method can be used when the breeder handles many populations and natural selection is effective in removing undesirable plants.

There are variations of the method whereby the breeder assists natural selection by rejecting, during the first segregating generations, those plants that are visually inferior.

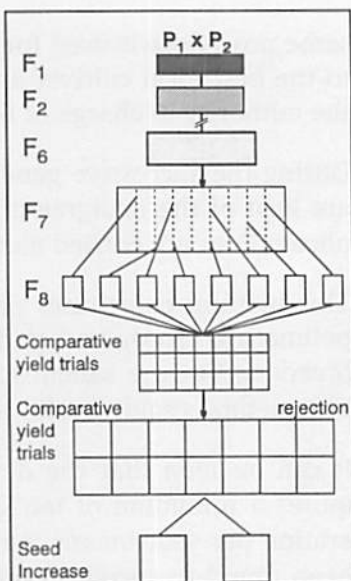


Figure 7. Bulk breeding method.

Multiline Varieties

These varieties are usually a mixture of isogenic or near-isogenic lines which differ from one another by being resistant to different races of the same pathogen, usually an airborne fungus. The lines are the result of the application of a backcrossing program in which a good recurrent parent is crossed to many different sources of resistance for different races of the same pathogen (Fig. 8).

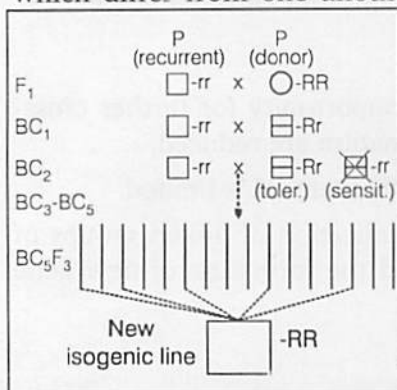


Figure 8. The transfer of resistance from donor to recipient through backcrossing. This procedure is used to add different genes for resistance to the same genotype.

The cultivar grown in the field is a mixture of the different lines in proportions bearing some relation to the prevalence of the various pathogen races. The proportions do not remain constant from year to year but may change as the pathogen changes.

This variety is difficult to produce and maintain, but it may represent the only way

to deal with situations when a disease is the limiting factor for a crop in a certain area.

The chief advantages of multiline varieties are their longer useful life relative to monogenotypic varieties and the reduction in the rate of growth of the pathogen and the disease that the pathogen causes.

Single Seed Descent

Many breeders believe that it is almost impossible to select for quantitative traits in the first segregating generations due to the masking effects of the environment and the presence of heterosis, both of which make it difficult to recognize and select any superior plants present in the population.

To overcome these problems, a large number of lines must be carried through until homozygosity has reached an acceptable level and heterosis has been reduced, and then these lines must be evaluated in comparative yield trials. This is done by taking at random one or two seeds from each F_2 plant, planting them close to each other in the following generation and repeating the process until F_5 or F_6 (Fig. 9). Following this last generation the lines are evaluated in trials replicated in time and space.

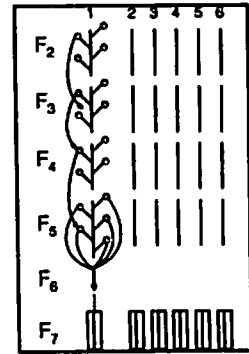


Figure 9. Single seed descent method.

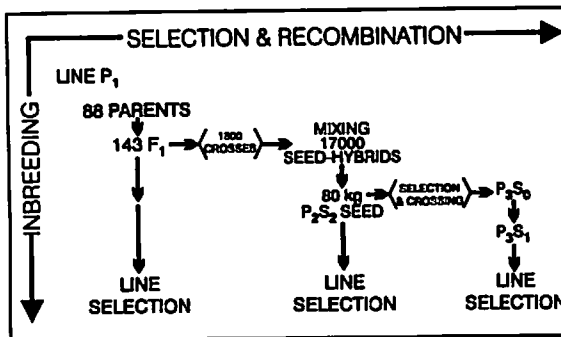


Figure 10. Simplified rendering of the diallel selective mating system.

The main object of this method is for the lines to reach an acceptable level of homozygosity as quickly as possible. To this end the seed is planted in thick stands, and all other conditions are manipulated in such a way as to induce the plants to produce seed as soon as possible. Under these conditions it is possible in cereals to have up to three generations per year and thus reach F_6 in two years.

This method has been used successfully in a wide variety of species, and sometimes one wonders why it is not used more extensively.

The Diallel Selective Mating System

In this system (Jensen, 1988), a group of carefully selected parents is crossed in some form of diallel scheme, i.e. either a full or partial diallel, depending on the number of parents chosen. Once the F_1 hybrids are produced, they can be used as any other F_1 and, through pedigree selection, pure lines or intercrosses can be developed to produce a mixture of seed from all resulting hybrids, which can in turn be used for pure line selection or for selection followed by intercrossing to produce yet another population on which to practice pedigree selection. A simplified form of this method is given in Figure 10.

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Varietal Development in Allogamous Crops

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Allogamous species require special methods for the production of the kind of cultivars required for their commercial exploitation. These cultivars can be F_1 hybrids, synthetic varieties, or various open-pollinating populations. The production of F_1 hybrid cultivars relies on a specialized methodology for the production of inbred parents, the identification of heterotic pairs, and the production of hybrid seed with a commercially acceptable cost. Synthetic varieties are used mainly in forage species, and are usually produced by the application of the polycross procedure, which ensures panmixis and adequate evaluation of the various parent combinations. Open-pollinating populations are usually used as sources of parents for hybrids, and as genetic reservoirs for the maintenance of genetic variability, rather than as cultivars per se. The main methods used for the production of these cultivars are described below, with comments as to their usefulness.

Introduction

Several of the plants essential for human survival are allogamous, or cross-pollinating, i.e. they require pollen from other plants of the same species growing in the vicinity in order to set seed. This requirement for foreign pollen has led to the development of special methods for the improvement of allogamous species.

The best example of a cultivated allogamous species is maize. Most of the methods described in this paper have been developed by maize breeders, to whom we owe a great scientific debt.

The varieties used in allogamous species are F_1 hybrids, synthetics, or open-pollinating populations. The latter can be used as is, crossed to form population hybrids or as sources of inbred lines for F_1 hybrid production.

F_1 Hybrids

F_1 hybrids are F_1 populations which are used "as is" for commercial purposes. These hybrids are the result of deliberate crossing of dissimilar parents which are pure, inbred lines. The primary aim of these hybrids is to

exploit the phenomenon of heterosis that is observed when certain parents are crossed and, in some cases, the quick combination of resistance to pathogens, without following the much lengthier route of selecting in the segregating generations.

For various reasons, heterosis cannot be easily fixed in pure lines. Hence the need for F_1 hybrids, even though commercial considerations also enter into the decision to produce hybrid or pure line varieties.

There are four steps through which the breeder must proceed to produce hybrids at an acceptable commercial cost. These are:

1. Production of inbred parents.
2. Evaluation of the combining ability of the inbred lines.
3. Determination of those combinations that produce heterotic hybrids, without making all the crosses, if possible.
4. Commercial production of hybrid seed.

Producing the Inbred Lines

This is accomplished by continuous self-fertilization and selection starting from a population with genetic variability. Individual plants are selected and then self-pollinated. Their progeny is planted in separate rows and the whole process is repeated for at least seven generations. During this inbreeding phase selection is practiced for vigorous plants so that the cost of production is kept low.

Pure lines can also be obtained from old pure lines by incorporating new traits through backcrossing or crossing and pedigree selection. Other schemes for inbred line production have been proposed, such as gamete selection, but have not found commercial acceptance.

Determining the Combining Ability of Inbred Lines

Every year many pure lines are produced, but only a very tiny fraction will take part in commercially successful hybrids. Yet all possible combinations must somehow be evaluated. This number is quite high: $n(n-1)/2$, where n is the number of lines. With even moderately high values of n , the number of possible crosses can be impossibly high. To avoid this, many breeders resort to top-crossing, i.e. crossing a group of lines to a common tester of wide genetic base, thus making n crosses instead, a more manageable number. By making all possible crosses one is able to determine both the general (the mean performance of all crosses in which a particular line has taken

part) and the specific (the performance of one particular cross in which the line has taken part) combining the abilities of the lines. The top-crossing method generally provides an estimate of the general combining ability of the lines.

Another way to reduce the number of crosses is to cross the available lines to those lines which are already parents of successful commercial hybrids. If the hybrid to be produced is a three-way hybrid, then the lines are crossed to the single-cross hybrid that is involved in a successful commercial hybrid.

The problem of ascertaining the value of a particular cross without actually making it remains, and will be with us for some time to come, despite claims of impending success from molecular biologists.

Production of Hybrid Seed

To produce an F_1 hybrid one needs at least two parents that, when crossed, produce a heterotic hybrid, plus a mechanism for emasculation of the parent to be used as the mother.

Emasculation mechanisms: Emasculation can be performed either mechanically or through genetic means such as male sterility, incompatibility, sex mechanisms or through chemical substances which act as gametocytes.

Mechanical emasculation: This can be done manually by carefully removing the anthers using forceps or, if the plant species has separate male and female inflorescences, by cutting off the entire male inflorescence, as is done with maize. Emasculation with forceps is carried out only in species in which the hybrid seed can be sold at high prices.

Incompatibility: Incompatibility ensures that the female parent cannot be pollinated by its own pollen, and there is no need for emasculation. The female parent is maintained through sibbing, bud pollination or by using high temperatures to temporarily inactivate the incompatibility mechanism.

Gametocytes: These are chemical substances acting on the male gametes, which they usually destroy without affecting the female gametes. They are usually sprayed on the parent to be used as female at some suitable growth stage which must be determined for each species.

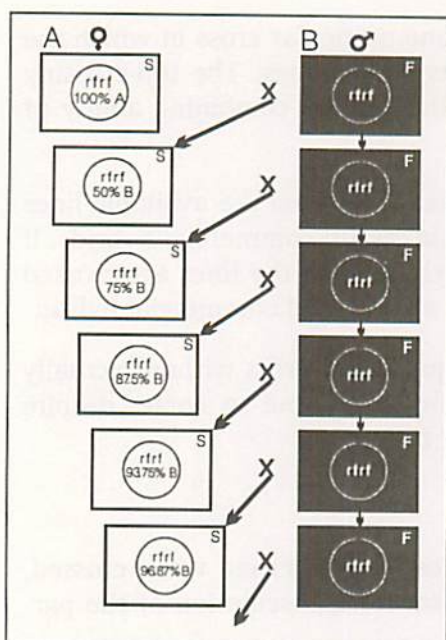


Figure 1. The conversion of a male fertile line (right) to a cytoplasmically male sterile line through backcrossing.

Male sterility: Male sterility can be due to nuclear genes, cytoplasmic genes or a combination of both. All types of sterility can either be found in most plant populations, if one looks hard enough, or created by artificial means, i.e. mutagens. Once a male sterile line has been obtained it can be used for the conversion of other lines to male sterility. At present this is done through extensive backcrossing (Fig. 1). The line to be converted is used as the male parent and should not have fertility restoration genes. It is crossed to a cytoplasmatic male sterile line and then backcrossed to the resulting hybrid several times. A minimum of seven backcrosses is required to transfer the nuclear genotype of the fertile line into a sterile cytoplasm and thus transform it into a male sterile line.

In addition to the two lines which are used as the parents of a hybrid, a maintainer line is also required to maintain the cytoplasmatic male sterile line. The maintainer line must have the same nuclear genotype as the male sterile line but a normal cytoplasm and must lack fertility restorer genes. By crossing the male sterile line to the maintainer one produces a seed supply of the male sterile line.

Genic male sterility is due to recessive nuclear genes that are present in low frequencies in almost all populations of plants. If genic male sterility is involved then the whole hybrid seed production process is as shown in Figure 2.

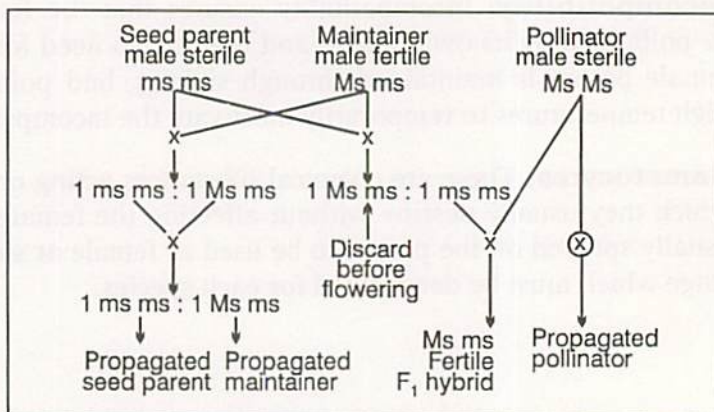


Figure 2. The production of hybrid seed and the maintenance of a genically male sterile line (from Borojevic 1990).

Kinds of Hybrids

Hybrids are usually characterized by the number of lines that take part in their formation.

Single cross hybrids: These have two parents and are usually depicted as $A \times B$, where A and B are the two parents. These hybrids are characterized by a high degree of uniformity.

Double cross hybrids: These are the results of crossing four parents and are depicted as $(A \times B) \times (C \times D)$. Their production requires different approaches depending on whether the species is used for its seed or for other parts, for example, roots.

Three-way hybrids: They are depicted as $(A \times B) \times C$ and require three parents. In this case the female parent of the final product is a single-cross hybrid.

Modified single-cross hybrids: These are depicted as $(A \times A') \times B$, where A and A' are similar lines and B is a dissimilar line. This kind of hybrid is usually utilized in order to reduce the cost of hybrid seed production, since the seed to be sold to the farmer is produced on a productive single-cross hybrid.

The actual production of hybrids in the field is a complicated procedure requiring careful isolation of all lines and fields used in production, plus a very strict quality control process both with respect to field operations and during seed processing.

Synthetics

When the cost of producing hybrid seed is too high, and the area under cultivation too small or near the fringe of the crop area, hybrid seed production is not justifiable. Instead, synthetic varieties, which are defined as those varieties that are produced by crossing in all possible combinations of selected genotypes of known combining ability, are warranted.

Most synthetic varieties are produced in forage plants. We will describe, therefore, the methodology used in this large class of plants (Fig. 3).

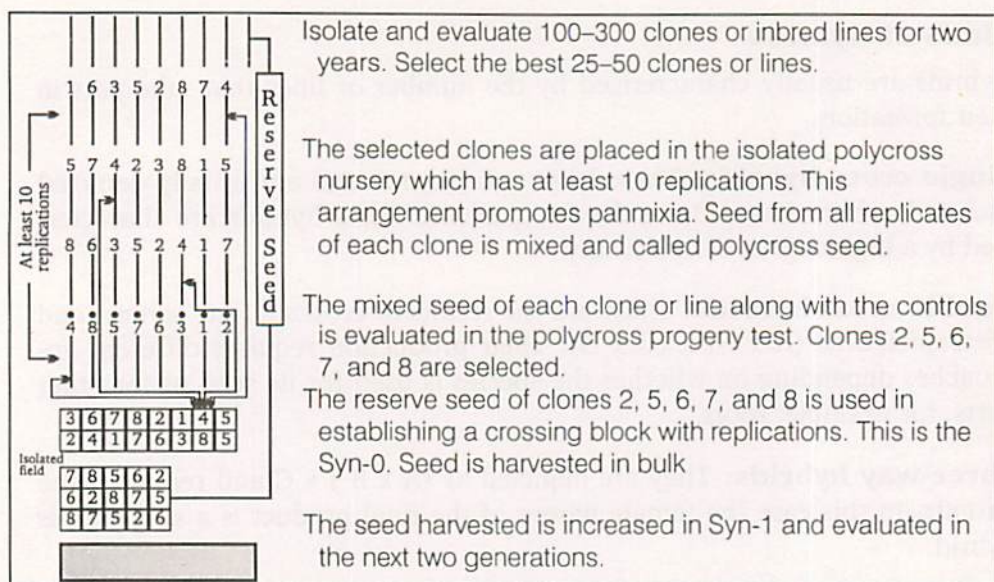


Figure 3. The production of a synthetic variety in a forage species through the polycross method.

To produce a synthetic variety the following steps are required:

1. Create populations with sufficient genetic variability to be used as sources for the isolation of lines or clones.
2. Based on visual criteria, select the best plants and create lines or clones from them.
3. Evaluate from 200–300 clones or inbred lines for a period of one to two years using the usual methodology. Harvest and store seed from the best 25–50 clones or lines.
4. Evaluate the combining ability of the lines or clones using one of the accepted methods. For forage grasses, a special method has been developed known as polycross for the evaluation of combining ability. A description of this method follows. Establish an isolated polycross nursery with at least ten replications and small plots. The purpose of this arrangement is to promote panmixis among all clones in the trial. Harvest each line or clone separately and mix the seed from each replicate of each clone or line from all replicates, taking equal amounts of seed from each replicate. The seed of each line or clone, together with the appropriate controls, is evaluated in comparative yield trials in what is known as the polycross progeny test. Select the best polycross based on all criteria, if possible.

5. Using seed from storage of the lines which were identified in the superior polycross, establish a crossing block with sufficient replication. All seed from this trial which must be isolated is harvested in bulk and constitutes the Syn-0 generation of the synthetic variety.
6. Increase the seed supply by growing the Syn-0 in isolation to obtain Syn-1 and evaluate it for a generation or two before releasing it to farmers for commercial purposes. Some breeders prefer to release Syn-1 seed to farmers.

Synthetic varieties have rendered a valuable service to forage production both in legumes and grasses.

Recurrent Selection

This term is used to describe any procedure used with the aim of concentrating desirable alleles from a population of plants into a small number of individuals. This is accomplished by selecting in each generation the best individuals, and intercrossing them to form a population that is then used again for selection of the best individuals. The method has the following objectives:

- Increase mean population yield by increasing the frequency of the desirable alleles.
- Maintain sufficient genetic variability so that continuous improvement can be maintained.

In its general form the method is as follows:

1. Certain plants from the source population are selfed, and at the same time evaluated for the trait under improvement. This evaluation can be done in various ways such as on the basis of phenotype or various kinds of test crosses.
2. Select the best plants and increase their seed by using the reserve seed from their selfing.
3. Cross the selected plants in all possible combinations either manually or through some other suitable method.
4. Take equal numbers of seed from each cross, mix them and form a new population that will be the source population for the next cycle of recurrent selection.

There are several methods of recurrent selection, which can be classified in many ways. We will present here the traditional scheme which is used for teaching purposes (Fig. 4).

The importance of the starting population cannot be stressed too strongly. There must be sufficient genetic variability in this population for the trait under improvement. These populations are created by intercrossing many parents, which express the trait under improvement at an acceptable level.

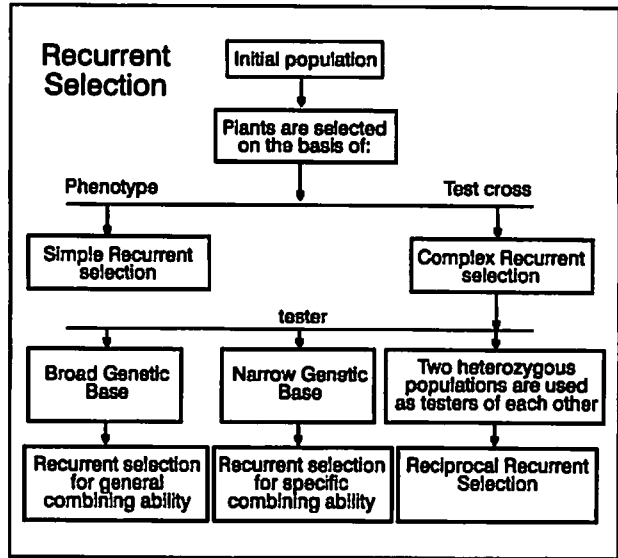
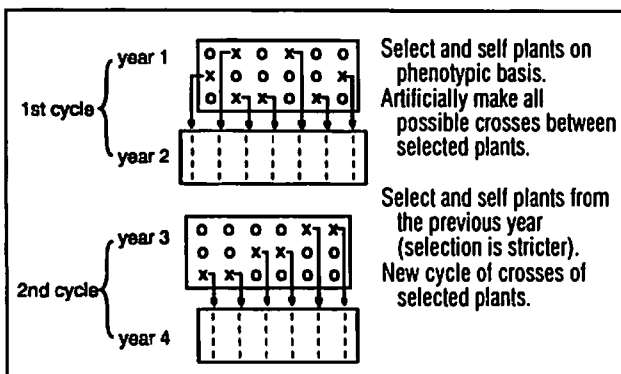


Figure 4. The traditional classification of the methods of recurrent selection.

The problem of selecting individual plants from the starting population is a complex one, and can be approached either by gridding or by using honeycomb designs to minimize the effects of the environment and allow full expression of the genetic potential of the individual plant.

Phenotypic Recurrent Selection

This is the simplest form of recurrent selection, and relies on the phenotype for the selection of the plants. Starting from a heterogeneous population (Fig. 5), individual plants are selected on the basis of their phenotype or the phenotype of their progeny. These plants are selfed and in the following



season all possible crosses are made between the selected plants. From each cross an equal number of seeds is taken and mixed to form the starting population for the next cycle of selection. Selection is continued for as long as there is genetic variance.

Figure 5. Phenotypic recurrent selection.

This method can be used successfully for traits with high heritability.

Recurrent Selection for General Combining Ability

The starting population must have sufficient genetic variability for the trait under improvement.

From the initial population, a number of plants are selected which seem to have the desired trait. These plants are selfed and crossed as males to the tester, which must have a broad genetic base (Fig. 6). The resulting hybrids are evaluated in experiments with sufficient replication. On the basis of the comparative trials the best hybrids are selected. The selfed seed of the parents of the selected hybrids is used to make all possible crosses among them. From each cross an equal number of seed is taken and then mixed to form the starting population for the next cycle of selection. Sometimes the population itself can be used as the tester of inbred lines developed from it.

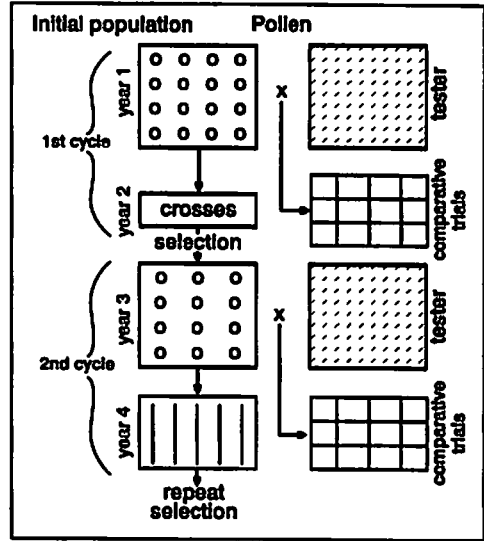


Figure 6. Recurrent selection for general combining ability.

This method has been effective in improving the combining ability of populations.

Recurrent Selection for Specific Combining Ability

The procedure is the same as for selection for general combining ability. The difference lies in the tester, which has a narrow genetic base and is usually an inbred line. Since both parents are selected, the genetic gain and the genetic variability among the families are expected to be greater.

This method has been used successfully in maize.

Modified Ear to Row Method

This method has been used mainly in maize. Starting from a heterogeneous population, a number of plants is selected such that together with the necessary controls the square of a number is obtained, e.g. 196 or 225. From each selected plant an ear is harvested. From each plant and from each control a row is planted using the triple lattice design, which has three repli-

cates. Each replicate is planted at a different location.

In the main location, which must be isolated from pollen from other populations, for each four rows from the ears of selected plants two rows are planted with a mixture of seed from all selected plants in equal proportions. Before flowering, the male inflorescence is cut off from the rows of selected plants and the controls. This will facilitate genetic recombination among the plants resulting in a polycross. In each row we mark the best five plants and at harvest we keep the five ears of each row separate.

On the basis of the yield in all three locations, the best 20 percent of rows are selected. The five ears kept separate from each selected row will constitute the starting material for the next cycle of selection. The seed from each ear is planted in a separate row in this next cycle.

This method has been successful in increasing the yield of maize.

Reciprocal Recurrent Selection

This method, used when the simultaneous improvement of two populations is desired, is summarized in Figure 7.

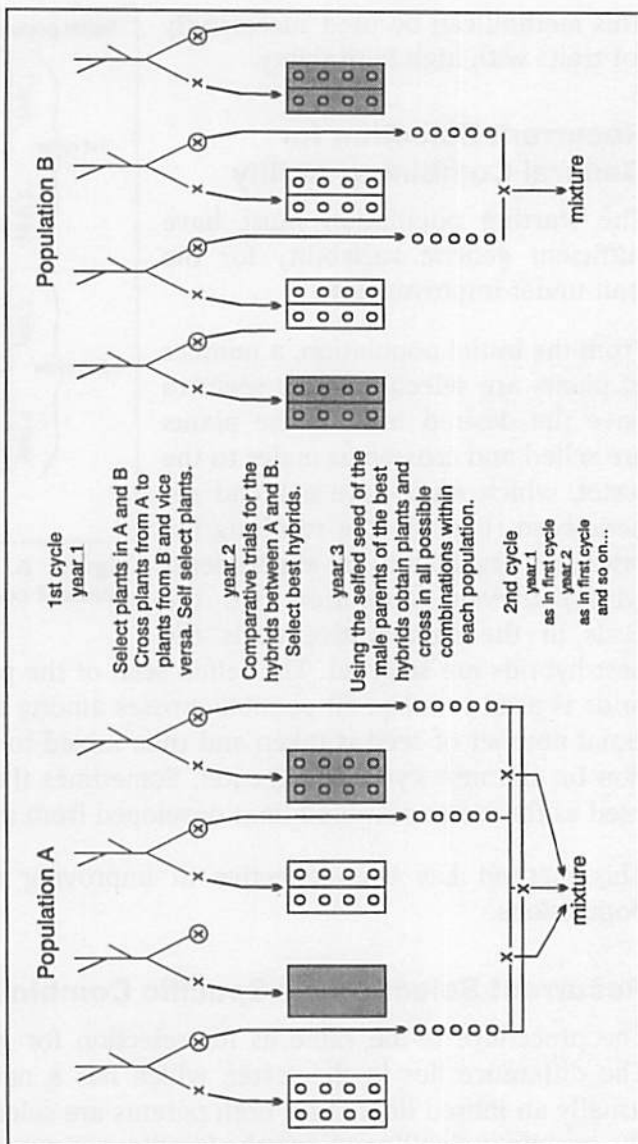


Figure 7. Reciprocal recurrent selection.

Start with two populations, A and B, which must be genetically heterogeneous and as genetically different from each other as possible.

Year 1: Two hundred plants are selected from each population and selfed. All selected plants from population A are crossed as males, each to a different sample of 4–6 plants from population B. The same procedure is followed in population B. The selfed seed is kept in reserve.

Year 2: For each population all hybrids produced in year 1 are evaluated separately in replicated trials. There are at best 200 such hybrids per population. On the basis of the results of these trials the ten best hybrids are selected.

Year 3: For each population we separately plant the selfed seed of the parents of the selected 10 hybrids, and make all possible crosses among them using four to five plants from each parent. Equal numbers of seed are taken from each cross and mixed to form the starting population for the next cycle.

The procedure is repeated for as long as there is genetic variability in the populations. The method has been modified in many ways. If in the species under improvement crosses can be made easily, each cycle can be reduced to two years by making the crosses at the same time that the hybrids are being evaluated.

The products of this procedure can be used “as is” for the production of the population hybrid, or as sources for the production of improved inbred lines which are then used to produce single or double cross hybrids.

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Variety Evaluation

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Introduction

New varieties have to be evaluated before they are released to farmers for commercial use. Different approaches to variety evaluation exist.

In small or initial stage seed programs, the breeder evaluates new material at the breeding station and in different ecological zones. These trials are required to obtain reliable information on the agronomic value of new experimental varieties. Varieties are compared under different management levels. Usually no attention is paid to distinctness, uniformity and stability of new varieties, and—when the number of varieties increases—the program encounters problems with regard to recognition of the different varieties.

In comprehensive seed programs, the final evaluation is usually carried out by a separate varietal evaluation agency. Varieties from different breeders are objectively compared with existing varieties at a large number of locations with a wide range of soil and climatic conditions. The evaluation includes assessment of distinctness, uniformity and stability.

Variety Evaluation Agency

In many countries, the variety evaluation agency is an independent governmental organization, charged with the final evaluation of new varieties before release. This agency's farm must be centrally located and represent the conditions in major growing areas. Many agencies have substations.

The test results (VCU and DUS) are reviewed by the variety release committee, which is usually composed of six to eight members representing organizations involved in the seed industry, such as breeding institutes, seed multiplication organizations, seed firms, extension services and farmer organizations. The committee reviews the results of VCU and DUS tests and makes recommendations for release and withdrawal.

In many countries, variety lists are prepared each year. The aim of such lists is to inform the farmer about the morphological and agronomic characteristics of commercial varieties, including the latest releases. Information on

cultural practices is often included as well. The list also indicates those varieties eligible for certification. Variety lists can either be advisory or restrictive; in the latter case, seed trade is limited to the listed varieties.

Performance Trials (VCU)

Performance trials aim to compare the agricultural value of new varieties with those of existing commercial varieties, and to identify those that are superior in certain ecological zones. Varieties with wide adaptability are also identified. Varieties are commonly tested in three subsequent years. However, there are shortcuts, and a superior variety should be released as quickly as possible.

Variety trials in different agro-ecological zones are carried out by breeding stations, universities, agricultural schools and training centers, and also in farmers' fields under the supervision of the varietal evaluation agency.

Preparation of Samples

The varieties to be tested may come from different breeding institutions. To minimize the risk of differences in agricultural performance due to different pre-planting treatments, seed for all locations should be prepared by the varietal evaluation agency. If treatment is needed, the varietal evaluation agency should treat all seed before it is dispatched for planting.

Experimental Setup

Performance trials require the selection of appropriate statistical designs as well as the proper size, shape and number of replicates. The choice of experimental design depends mainly on the number of varieties to be tested. For a small number of varieties, a randomized block design can be used. If the number of varieties is large, more sophisticated experimental designs must be used, such as Latin squares or lattices.

When yield is being assessed under different management practices, such as various nitrogen levels or different cultural practices, a factorial or split plot design can be used. Statistical textbooks give complete explanations of these methods.

Field Observation and Scoring

For cereals and grain legumes, the most important character to assess is the yield of grain per hectare (taking into account the moisture content of the crop). For pasture and forage crops, the total dry matter is the key charac-

ter; the different quantities of dry matter produced at different times throughout the year are valuable agronomic data. In grasses, grain yield is of interest for seed production. Data are also gathered for other important agronomic characteristics.

Table 1 shows the characteristics recorded for wheat. The same type of observations are made for other crops, but each crop has a number of specific characters to be recorded.

Table 1. Characters recorded for wheat.

Characters		
Frost resistance	Resistance to diseases and pests	Lodging
Germination capacity	Stripe or yellow rust	Drought resistance
Vigor	Stem or black rust	Tolerance to:
Tillering	Leaf or brown rust	sprouting
Heading date	Loose smut (<i>Ustilago</i>)	shattering
Number of heads	Covered smut or bunt (<i>Tilletia</i>)	
Lodging	<i>Septoria</i>	
Days to maturity	<i>Fusarium</i>	
Kernel weight		
Hardness (particle size index test, near infrared reflectance spectroscopy, barley pearler)		
Protein content		
Milling quality		
Baking quality		
Suitability for bread, macaroni		

Special Tests

Special laboratory tests may also have to be carried out, depending on the purpose for which the crop is intended. For wheat milling properties, dough quality and baking quality are determined. For barley, beer-making quality is assessed. Other crops have their own special tests. The variety evaluation agency does not carry out all such tests, but is assisted by other specialized organizations.

Statistical Analysis and Reporting

Extensive analysis is usually carried out only on the yield data. Each experimental design has its own method of statistical analysis. The method chosen, however, must allow analysis to be carried out at a number of locations within a given year, as well as over different years. Since variety evaluation is a continuous process, with new, promising varieties entering the experiments every year, and other varieties withdrawn upon completion of the three-year performance trial cycle, the method must be flexible. Computers are often required.

To make the description of yield more meaningful, it is often expressed as a percentage of the yield of the control variety. This is done because a variety's yield varies at different locations in the same year, and in different years at the same location.

Many characters are expressed on a 1–9 scale, with the higher number indicating the more desirable state, or by abbreviations, representing the status of a character (for example: la = late maturing, me = medium maturing, er = early maturing).

Table 2 gives an example of a table that summarizes the results of pea performance trials.

Table 2. Results of performance trials in pea.

	Blue pea			White pea	
	Finale	Maxi	Solara	Belinda	Miranda
1. Length of straw	4	4	6	4.5	5
2. Strength of straw	7	7	8	7.5	7
3. Leafiness	5.5	5	3	6	6
4. Earliness of beginning of flowering	7	7	7	8	7.5
5. Shortness of flowering period	8	8	5.5	7	7
6. Earliness of ripening	7.5	7.5	6.5	8	8
7. Height of insertion of the pods	8	8	8	8	8
8. Size of the seed	7.5	7.5	6.5	6	8
9. Roundness of the seed	8	8	8	8	8
10. Color of the seed	7.5	7.5	7.5	7	7
11. Appearance, attractiveness	8	8	8	8	8
12. Consumption quality as dry pea	8	8	-	-	-
13. Resistance to top yellows	8	8	9	7	7
14. Resistance to wilt disease	10	10	10	10	10
15. Resistance to pea blight or leaf spot	8	8	-	-	7
16. Resistance to marsh spot	8	-	-	-	-
17. Resistance to bad weather	8	8	9	8	8
18. Resistance to early browning virus	5	-	-	-	-
19. Suitability for mechanical harvest	6	6	8	6	6
20. Suitability as nurse crop	7	7	9	7	7

DUS Tests

The second test is the test for distinctness, uniformity and stability. DUS tests are conducted to establish whether or not a variety is sufficiently distinct from all other varieties, sufficiently uniform and stable. Based on DUS tests, a variety description is produced that is used for varietal recognition in the seed production program (field inspection, roguing, etc.) and granting proprietary rights.

Distinctness

This quality is essential because a new variety must be different from an existing variety. Each variety must be recognizable not only in field inspection, but also for seed growers and farmers. For granting proprietary rights (Plant Breeder's Rights), the variety must be clearly recognizable.

Uniformity

To guarantee constant quality, and also for field inspection, a variety must be as uniform as possible. The degree of uniformity depends on the mode of reproduction. Varieties of self-pollinating crops are more uniform than varieties of cross-pollinating crops. In a highly mechanized agricultural system in a region that is agriculturally and climatologically homogeneous, a high degree of uniformity may be desirable, but under other conditions, a certain degree of variability may be advantageous.

Stability

During the various stages of seed multiplication, from breeder seed to certified seed, the variety should not lose its distinctive characters. The genetic make-up should remain the same as near as possible. Varieties of self-pollinating species are more stable than varieties of cross-pollinating species; hybrids are not stable, and new hybrid seed must be produced each year for farmers.

Categories of Varieties

Three categories of varieties can be distinguished:

- Varieties of vegetatively propagated species.
- Varieties of self-pollinating species.
- Varieties of cross-pollinating species.

Varieties of vegetatively propagated species are all alike. Varieties of self-pollinating species are homozygous and all plants are more or less alike; not much variation and segregation occurs. The majority of variation results from environmental conditions. Many self-pollinating species have a small percentage of outcrossing, resulting in variation.

Varieties of self-pollinating and vegetatively propagated species are often not difficult to describe.

In varieties of cross-pollinating species, the individuals are not alike and the population has a certain equilibrium (with regard to gene frequency). Varietal description is more difficult and often has to rely upon segregation percentages.

Characteristics for Varietal Description

Any characteristic (morphological, physiological, cytological, chemical or other) may be used as long as it serves the purpose of distinguishing between variety and species. Obviously, the more easily observable characteristics are preferred (i.e., clear morphological differences). Moreover, the less the character is influenced by environmental conditions the better. Either quantitative or qualitative characteristics may be used to distinguish varieties.

Quantitative

(i) have continuous variation; (ii) can be measured numerically or metrically; (iii) are influenced by the environment; and (iv) are controlled by many genes. Examples of quantitative characters are: plant height, days to flowering, and days to maturity. Many characters are scored on a 1–9 scale. Table 3 shows examples of scoring for three characters.

Table 3. Examples of scoring for three characters.

Character	Score				
	1	3	5	7	9
Presence of anthocyanin	absent				present
Intensity of anthocyanin color	absent or very weak	weak	medium	strong	very strong
Plant growth	erect		intermediate		prostrate

Qualitative

(i) have a discontinuous variation; (ii) are measured visually in discrete classes; (iii) are affected little or not at all by the environment; and (iv) are usually controlled by a single or very few major genes. For example, clear qualitative characters are: flower color, testa color, seed shape, and the growth habit in faba bean.

Further details on characteristics can be found in the Guidelines for the Conduct of Tests for Distinctness, Uniformity and Stability. These guidelines are available for many species and can be obtained from the International Union for the Protection of New Varieties of Plants (UPOV), Geneva, Switzerland. The Plant Genetic Institute (IPGRI) has developed guidelines which describe characters that should be used for characterization of accessions. These characters can also be used for the description of varieties. Guidelines are available from IPGRI, Rome, Italy. Furthermore, the Organization for Economic Cooperation and Development (OECD, 1971a; 1971b), Ulvinen *et al.* (1973), Hervey-Murray (1980), and Milatz (1970) describe characteristics in detail.

UPOV has also prepared a general introduction for the use of its guidelines for the Conduct of Tests for Distinctness, Uniformity and Stability. This guideline (UPOV, 1979) provides essential information for varietal description work and has been extensively used to prepare this paper.

Experimental Setup of DUS Tests

In advanced seed programs, special experiments are planted for the description of varieties. The DUS tests are carried out on the varietal evaluation agency's farm. The farm's location should be representative of the major growing areas, because environmental conditions can influence the expression of varietal characters.

Distinctness, uniformity, and stability are studied on individual plants sown in small plots. A wide range of existing varieties is usually available for comparison. During the growing period, careful observations are made on individual plants; after the crop has matured, ears, heads, and other parts, as well as seed, are elaborately scored.

DUS tests are often difficult, requiring considerable skill and time. More than 40 different characters may be recorded in DUS tests.

DUS tests usually have an unconventional setup:

- Only two replicates are planted.
- There are often two different environmental locations.
- Seed should not be treated.
- Seed is hand planted (to avoid risk of admixture).
- Hand weeding is done (herbicides may influence character expression).
- Similar varieties are grouped.
- Tests are usually carried out over two years.
- A wide range of existing varieties is included.
- In addition to plots, single ear rows or single plant progeny are planted.

Scoring

Qualitative characteristics are usually scored as consecutive numbers, starting with 1 with no upper limit. Each state is different (distinct) from the others. The smaller, the lesser or the lower should be assigned the number 1. For example:

- Ear shape in barley: tapering (1), parallel (2), fusiform (3).
- Color of grain in wheat: white (1), pale red (2), dark red (3), brown (4), black (5).

- Awn length compared to grain (barley): shorter (1), equal (2), longer (3).
- Rachilla hair type (barley): short (1), long (2).

Quantitative characteristics are often scored on a 1–9 scale, where 1 indicates the lowest state of the character (very short for plant height, very weak for glaucosity, etc.) and the 9 the highest state (very tall, very strong). Scoring is done according to one of the following scales:

- 1 (absent) and 9 (present).
- 3 (weak) and 7 (strong).
- 3 (weak), 5 (medium) and 7 (strong).
- 1 (very weak), 3 (weak), 5 (medium), 7 (strong) and 9 (very strong).

Quantitative characters are also often measured and compared with the standard variety; data may be transferred to a 1–9 scale. (Days to heading: measure number of days to 50 percent heading on a plot basis and transfer to a 1–9 scale.)

To be able to score the status of a characteristic correctly it is essential that all known varieties are included in the experiment. It is useful to always refer to standard varieties.

Distinctness

Two varieties are distinct if the difference:

- Has been determined in at least one testing place.
- Is clear.
- Is consistent.

For qualitative characteristics, the difference between two varieties is clear if the state of the characteristic falls into two different classes.

For quantitative characteristics, if measured, the difference is considered clear if it occurs with 1 percent probability (LSD test). The difference is consistent if it occurs with the same sign in two consecutive, or two out of three, growing seasons. If visually observed, consistent differences in pair comparison are required.

Uniformity (homogeneity)

Tolerance levels, which differ according to breeding system and mutability of the variety, are as follows for vegetatively propagated crops and truly self-pollinating varieties:

Sample size	Maximum number of off-types
less than 5	0
6–35	1
36–82	2
83–137	3

For mainly self-pollinating varieties, the tolerances are doubled.

For cross-pollinating varieties, no fixed tolerances can be determined, but the variation is compared with comparable (known) varieties.

For measured characteristics, the standard deviation is used as the criterion for comparison. A variety is considered not to be homogeneous if its variance exceeds 1.6 times the average of the variance of the varieties used for comparison.

Visually assessed characteristics are handled in the same way; the number of off-types should not be significantly more than those in comparable varieties (5 percent error).

Stability

A variety which is uniform is also considered to be stable. Since it is not possible to test stability in the two growing seasons that are required for DUS tests, the inclusion of material from different multiplication cycles within the two consecutive testing years is recommended.

Ear Row Progeny

A special method often used in DUS tests is the observation of the progeny of single ears. Progeny of 50 to 100 ears are grown in rows (ear rows) and carefully observed. Any lack of uniformity is thus easily detected.

Reporting

The final report is often brief, listing only the major differences between similar varieties, as well as stating that the variety is sufficiently distinct, uniform and stable. More elaborate descriptions are always available upon request.

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Different Methods of Variety Maintenance

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Introduction

New and superior varieties developed by plant breeders should be multiplied and made available to farmers in the shortest possible time. To supply farmers every year with certified seed, each year a new multiplication (from breeder to certified seed) should be started. The objective of a variety maintenance program is to produce new lots of breeder seed at regular intervals, which are the basis for further multiplication.

Due to the fact that deterioration will inevitably take place during multiplication, new starting material cannot just be taken from any seed field. A special procedure which aims at high genetic purity and freedom from seed-borne disease is required.

In many developing seed programs variety maintenance is neither done by the breeder nor by the seed producer. Consequently, genetic purity is often low, and obvious contaminants are present in certified seed fields.

Deterioration

Three types of deterioration can be distinguished:

- Genetic contamination.
- Mechanical contamination.
- Pathological contamination.

Genetic contamination results from cross-pollination and spontaneous mutations. Some wheat and barley varieties will have a higher outcrossing percentage in semiarid areas (up to 10 percent). The rate of spontaneous mutation is generally low, but, considering the large number of genes, may have an influence. The rate of spontaneous mutations increases significantly after storage for several years. Since mutations are usually micro-mutations and recessive, they are difficult to detect.

Moreover, natural selection always occurs, and may result in a genetic shift if sufficient care is not taken to produce the seed in an environment where the variety is adaptable, or if a selection pressure is applied that was not intended by the breeder.

Mechanical contamination results from a wide variety of sources but is mainly due to insufficient cleanliness of fields (volunteers) and equipment (planter, combine, transport vehicle, bags, store, seed cleaning equipment), and inadequate measures to combat such contamination (field selection, physical isolation, equipment cleanliness).

Pathological contamination occurs through infection with disease.

Maintenance for Self-pollinating Crops

Natural populations of most autogamous cultivars are made up of different, mostly homozygous, genotypes. Experience indicates residual heterozygosity in almost every autogamous cultivar. Generally, strictly self-pollinating crops have very low outcrossing percentages, and using recommended isolation distances will reduce outcrossing to a great extent.

For varieties of such self-pollinating species the following techniques can be used to produce breeder seed.

Methods

Produce enough breeder seed for the lifetime of the variety

The most simple method is to produce a large amount of breeder seed that can be used for several years (Julen, 1983). This is a good method if good storage facilities are available, but only possible with crops that are grown on a restricted area or with crops that have a large multiplication rate (tomato, tobacco). In many European countries this is done because varieties have a rather short commercial lifetime. This approach is described by Bouwman (1992) for pea and faba bean.

Unfortunately, for many field crops, it is impossible to store sufficient seed for the lifetime of the variety, because areas are large and multiplication rates (wheat, 25–40; bean, pea, soybean, 20–30; potato, 6–10) are not high enough. Furthermore, the material is too bulky, and sometimes cannot be stored for long periods.

Produce breeder seed every fifth year

Every fifth year a new lot of breeder seed is produced. This breeder seed is used to start the multiplication cycle for a number of years, before a new lot of breeder seed is produced.

Use basic seed as source for breeder seed without selection

A very simple method of producing breeder seed is to set aside a part of the

basic seed harvested and use it as breeder seed for the next cycle. Obviously this is not a very good approach, because hardly any special control is possible.

Use Basic Seed as Source for Breeder Seed with Negative Mass Selection

A small improvement is to use a small part of the basic seed field for the production of breeder seed. Obvious contaminants are removed by negative mass selection. Plants which do not conform to the variety characteristics are rogued and the crop is bulk harvested. This method is not regarded as an efficient method, because genetic contamination is usually not easily seen.

Another slight improvement is that a small part of the breeder seed is kept apart and planted separately to produce the next generation of breeder seed. The breeder seed can be space planted and careful observation is possible.

Selection

Ears of wheat and barley are selected, threshed together and planted in a spaced planting arrangement. Individual plants are carefully observed and rogued out if necessary. Seed is bulked up to constitute the breeder seed.

Ear-to-row or Plant-to-row Selection for Self-pollinating Crops

The best approach to breeder seed production is based on an ear-to-row or plant-to-row procedure.

Single ears or single plants typical of the variety are selected and harvested separately. The seeds of each ear or plant are carefully observed, and any ear (or plant) that produces one or more deviant seeds is discarded. The seeds of selected ears (or plants) are planted in ear rows (or plant rows). Each ear (or plant) is planted into a row of 1–3 m. The ear rows are periodically examined throughout the growing season. For wheat, at least five inspections should be made: (i) early growth stage; (ii) before ear emergence; (iii) at ear emergence; (iv) at dough stage; and (v) at maturity. The varietal characteristics are clearly expressed at these stages. Selected ear rows are bulked up to constitute the breeder seed, which is used to initiate the seed production cycle (pre-basic seed, basic seed, certified seed).

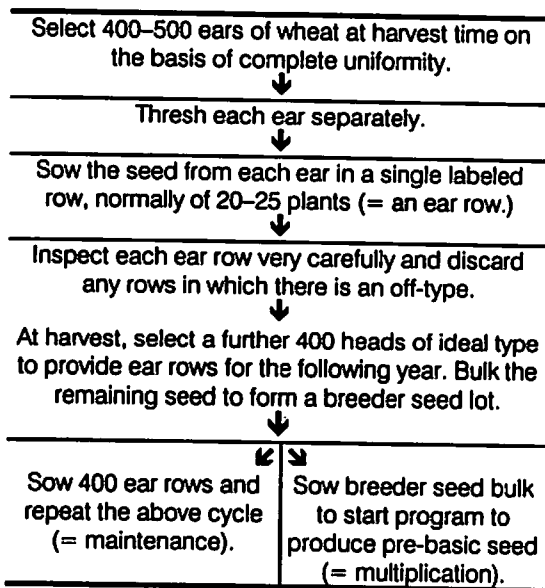


Figure 1. Maintenance scheme for wheat.

uniform and stable—the number of ear rows sown each year can be considerably reduced.

Many modifications of this scheme are possible:

- The ear row progeny can often be kept separate for an additional generation before they are bulk harvested. Suspect progeny can be discarded before bulking. The wheat variety maintenance schema used by a private firm in the Netherlands is shown in Figure 2. Selected ear rows are planted as separate ear row progeny and the bulk harvest is grown for an additional year (small multiplication), before it is bulked up as breeder seed.
- In exceptional cases individual ear row progeny are kept separate for a second year. This approach ensures that no genetic contamination is present in the bulk harvest material that is used to produce—through one or two further generations—the certified seed (Aalders-du Bois, 1989). This is usually considered the best method; the longer the lines are kept separate the greater the uniformity that can be achieved.

This scheme is illustrated in Figure 1, using wheat as an example. It concerns a small variety, since the number of initial ears is only 400–500. Often many more ears (1,500–6,000) are used.

The number of ears used for maintenance can be determined by the number of generations, the multiplication ratio, and the quantity of certified seed required. Oka (1975) suggests that the population suitable for maintenance of seed production of commercial self-pollinating crops should be 300. Theoretically—if the variety is known to be uni-

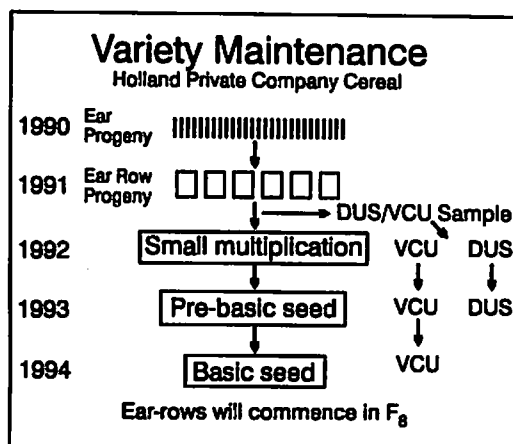
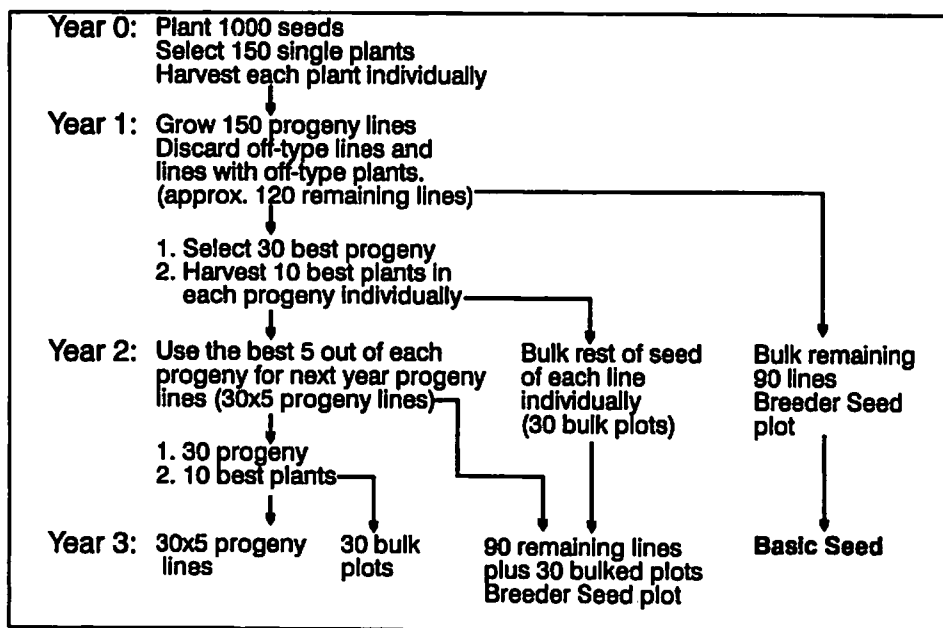


Figure 2. Maintenance scheme for wheat (modification 1).

- In Figure 3, the maintenance for *Phaseolus vulgaris* is described (Drijfhout, 1981). The principle is the same, but extreme care is taken to select the 150 plants which will be used to initiate the next year's maintenance cycle. (i) Of the 150 progeny lines, lines with off-types and the least promising lines are not used (usually 20 percent are rejected). Among the 120 remaining progeny, the 30 best are used; in each progeny the 10 best plants are chosen (on the basis of production expectation) and the five best plants among each of the 10 are then chosen on the basis of highest number of seeds. These 5 x 30 plants will be used to produce the next year's progeny rows. The remaining seed of each of the 30 progeny is harvested per progeny and planted as 30 bulk plots. (ii) The remaining 90 pedigrees are bulked (the breeder seed plot), to be used for further multiplication.



1. Average cross-fertilization in phaseolus is 1.4 percent.
2. Isolation by strips of other crops like maize, sunflower; tall and dense crops are preferred.
3. In the 150 progeny lines all lines with off-types are rejected as well as the least promising lines; total 20 percent discarded.
4. The 10 best plants of each of the 30 selected progeny lines are chosen on the basis of production expectations; the five best plants among each of the 10 are chosen on the basis of highest number of seeds.
5. Bulk plots are rogued (negative mass selection). Bulk plots are usually planted in the same field as the progeny lines to make comparison possible, where deviations are found the progeny and the mass plots can be discarded.

Figure 3. Variety maintenance of *Phaseolus vulgaris* (Drijfhout, 1981).

Bulk plots are rogued (negative mass selection). Bulk plots are usually planted in the same field as the progeny lines, to make comparison possible. Where deviations are found, the progeny and the mass plots can be discarded.

Ears or Plants as the Basis for Maintenance?

For certain crops (wheat, barley) the maintenance program can be based on either selected ears or selected plants. The disadvantages of using plants are:

- Since only a limited number of plants are used to obtain sufficient breeder seed, the genetic base of the variety might become too narrow. However, according to genetic population considerations for self-pollinating crops, this does not easily occur, since 80 plants are considered a minimum, for example, in wheat (C. Hellingman, personal communication).
- Secondary tillers may have smaller seed, resulting in less uniform and less viable plants.
- Selection between plots is often more difficult than between rows.

Material for the Next Breeder Seed Generation

Before (bulk) harvesting, ears for the next breeder seed generation should be selected (bundles of 50 ears, cut with 20–30 cm of stem). Each bundle should be labeled and prepared for threshing. The remaining field will then be bulk harvested.

Maintenance for Cross-pollinating Crops

Several approaches are possible for cross-pollinating species, where the risk of genetic contamination through pollen of other varieties is obviously much larger. Consequently, larger isolation distances are necessary. The methods used are more or less the same as those described above.

Negative Mass Selection

Part of the basic seed field is given special attention, and will produce the next generation of breeder seed.

Ear-to-row and Plant-to-progeny Selection

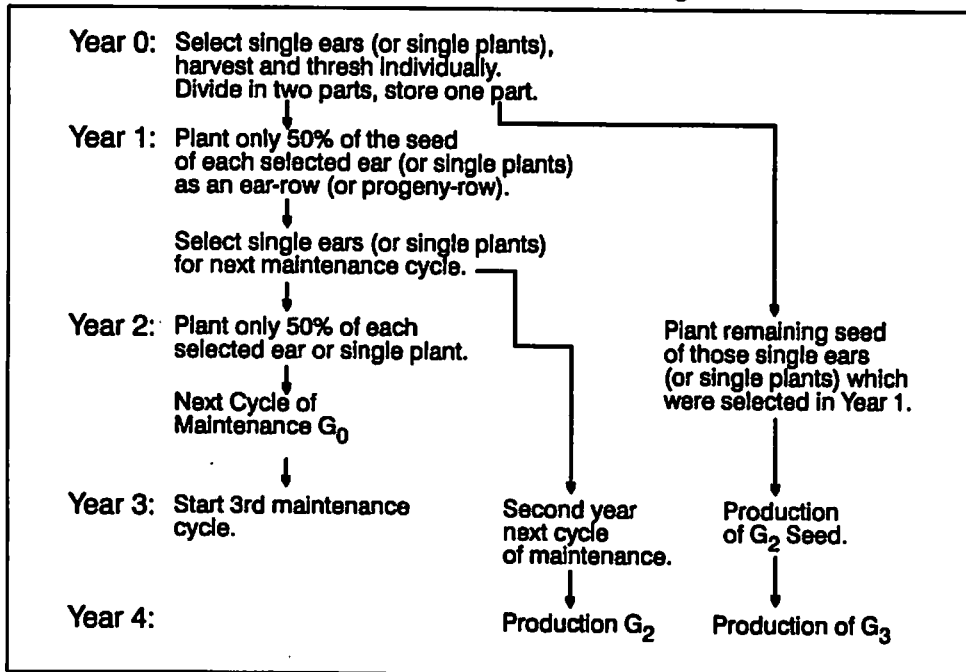
Select single plants and grow the progeny in single rows. For cross-pollinating crops, roguing before flowering is important to ensure that off-

type plants do not contribute to the pollen that will fertilize. Any row with off-types detected after flowering will not be bulked with the breeder seed, but its pollen has contributed to the “pollen cloud,” and possibly to fertilization.

The approach described above does not exclude off-type pollen grains as participants in pollination and fertilization. A better method is the rest seed method.

Rest Seed Method

This method is basically an ear-to-row (or plant-to-row) approach. In year 0 single ears (or plants) are selected, individually harvested and threshed. Only a small part of each progeny is planted as ear rows; the remaining seed of each single ear is stored. In year 1 the small part of the seed of each single ear is planted as a progeny row, and careful selection is carried out before and after flowering and until harvest. All off-types are removed; only selected ear rows are harvested, and the seed carefully observed. The rest seed of year 0 ears that produced the best progeny is used for planting in year 2. In year 2 strict selection is carried out before flowering, and deviating progeny are removed. Seed is not harvested on plants which show off-types after flowering. This process is illustrated in Figure 4.



G_0 is the breeder seed field; G_1 is the harvest from the G_0 field, etc.

Figure 4. Variety maintenance of open-pollinating species (Nötzel, personal communication).

This procedure will lead to a very acceptable degree of varietal purity, but the need to carry out a progeny test causes a delay of one season in the multiplication program.

Agronomic Precaution

Cropping History

Maintenance plots should be grown on clean, fertile land, if possible at a research station in the region for which the variety is recommended. The land must be suitably prepared, and the plots isolated to avoid genetic contamination. Maintenance should be carried out on land which has not produced the same crop species for at least two years.

Planting and Inspection

Maintenance plots may be sown by hand or with small-scale plot equipment, since production areas are usually small. Machine planting is preferred because it leads to more even emergence and easier selection. The rows should be sufficiently spaced to permit easy passage and inspection.

Inspection should be done on several occasions, and rows with off-types marked and discarded. The remaining ear rows should conform to the description of the variety.

To achieve a high multiplication factor a 10 percent lower seed rate is recommended for early generations, and optimal agricultural practices should be used. To facilitate roguing every 2–3 m, one row should be left unplanted. It is recommended that early generations be produced at different locations to reduce risk of losing the complete crop.

Isolation

Strictly self-pollinating species need only a few meters of isolation; insect pollinating species need larger isolation distances, and wind pollinating species much larger. The prevalent wind direction should always be taken into account.

The breeder seed plot should be surrounded by the next generation (pre-basic seed) to avoid crossing and mechanical mixing at harvest. The earliest generations should be planted first, and then the later stages.

Another method is to plant several crops in one field separated by alternating the crops and/or making use of different planting times. Isolation is by

strips of other crops such as maize or sunflower; tall and dense crops are preferred. If crops are similar it is advisable to alternate crops with different ploidy levels: bread wheat, then barley, then durum wheat, then oats, then triticale.

Harvesting and Cleaning

Harvesting and threshing can be done entirely by hand, or by hand-harvesting and threshing by small-scale portable or stationary threshers.

Cleaning should be done by hand or small-scale cleaners. Treatment can also be carried out with a small laboratory treater.

The surrounding stages should be harvested first, then the breeder seed. This will reduce changes of mechanical mixing.

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New Developments in Varietal Identification

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Introduction

Many changes in plant breeding practices, and in the commercial exploitation of the plant breeding products, the varieties, have taken place in recent years. The single most important change concerns attempts to patent varieties in more crops and in more countries than before. If patents are granted and are to be enforced, the variety they protect must be distinct from all others, and this should be provable in a court of law, if necessary.

Traditionally, and until about twenty years ago, varieties were identified on the basis of their morphological characteristics, either in the form of the growing plant or of the end product that reached the market place. More recently, protein polymorphism, revealed through starch gel or other kinds of electrophoresis, has also been used for varietal identification. The method is quite reproducible but suffers from the fact that only a small part of the genome is analyzed, since only a fraction of the gene is transcribed and translated at any given time in the life of the plant. Furthermore, several other factors, such as the age and the state of the plants and the environment, may affect the protein profile of a given genotype. To avoid these problems one needs markers that are not influenced by the factors mentioned above, and that are constant for a given genotype. Such markers are DNA-based molecular markers, which depend upon differences in the DNA sequences of the cells of the organism.

With the inevitable narrowing of the genetic base of cultivated plants, and the proliferation of private firms in the breeding business, it becomes more and more necessary to rely on molecular markers for variety identification. In this paper we describe the developments in varietal identification since the advent of protein electrophoresis, concentrating on RFLP (Restriction Fragment Length Polymorphism) and RAPD (Random Amplified Polymorphic DNA) analysis.

Restriction Fragment Length Polymorphism

This method provides the means for developing large numbers of neutral and co-dominant genetic markers (Tanksley *et al.*, 1989) by relying on the DNA fragments that are produced when the DNA of an organism is cut by restriction enzymes that recognize specific DNA sequences. The resulting fragments are electrophoretically separated on agarose or polyacrylamide gels, followed by a Southern transfer to bind the DNA onto nylon membranes. To detect any differences in the sequence of the DNA, one has to be able to detect differences in the length of the fragments. This is accomplished by hybridizing the digested DNA on membranes with labeled probes that are homologous to the sequences in question. Visualization of the hybridization event occurs, in the case of radioactive labeling, through blackening of an x-ray film, and with non-radioactive labeling, through a coloring reaction on the membrane, or light emitted blackening of an x-ray film. Variation that is found following this method is called Restriction Fragment Length Polymorphism.

A detailed description of the method (Fig. 1) follows (Potter and Jones, 1991).

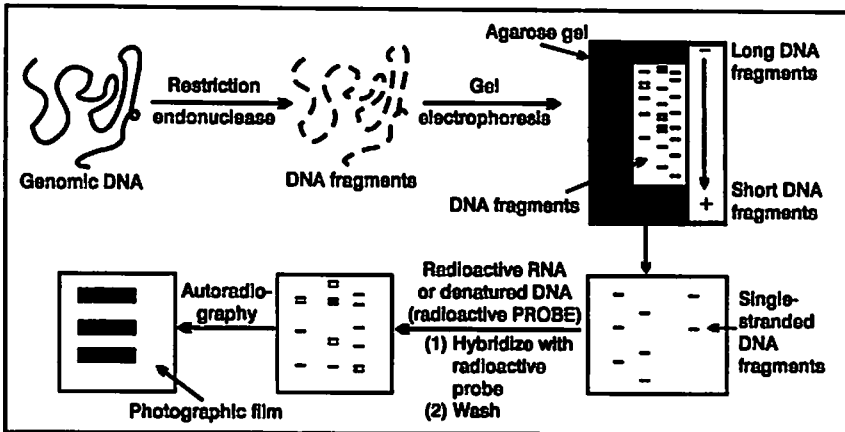


Figure 1. The various steps in the RFLP method.

DNA Extraction

Extracting DNA from plant cells is more tedious than from animal cells because of the cell wall. Plant material is ground in liquid nitrogen with various other agents. (Since there is no need for highly purified DNA, the cesium chloride centrifugation step is omitted.) The extraction method used

must be such that large numbers of samples can be processed quickly, yet it must provide reproducible results. One such method is described by Delaporta *et al.* (1983). The quest for even faster and simpler methods is going on as these lines are being written.

The concentration and quality of the DNA that has been isolated is determined by spectrophotometry, fluorometry or in comparison with DNA standards on concentration gels.

Restriction

The restriction enzymes recognize and cut specific DNA sequences that are found in the DNA; different enzymes recognize different sequences. The distribution of the sequences recognized by these enzymes is essentially random throughout the genome. The shorter the sequence, the more often it is found along the DNA and, consequently, if an enzyme that recognizes it is used, more DNA fragments will be produced, thus allowing for a finer resolution. The electrophoretic facilities must be taken into consideration when deciding which kind of a restriction enzyme to use. The usual compromise is to use enzymes that recognize six base pair sequences; these enzymes are called six-cutters.

A hypothetical example using two different enzymes on the same DNA that was altered by deletion or base changes is shown in Figure 2. These enzymes have different recognition sites that do not coincide.

Complete digestion of the DNA is essential for resolvable RFLP patterns. To avoid problems it is recommended that the buffer system provided by the enzyme supplier be used.

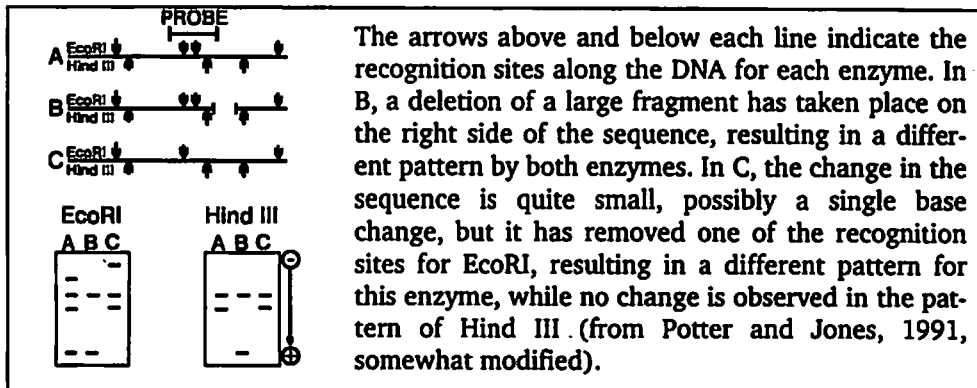


Figure 2. Schematic representation of the use of two different restriction enzymes and the patterns they produce when used on the same stretch of DNA which has been altered.

Electrophoresis

The use of six-cutters results in fragments ranging in size from 200–20,000 bp, which provides sufficient resolution of the genome and allows the use of agarose gels. If finer resolution is desired, restriction enzymes which recognize and cut four base sequences (four-cutters) are used, and the resulting fragments are separated on a polyacrylamide gel that can resolve even single base differences. Polyacrylamide gels cannot effectively resolve fragments greater than 1,500–2,000 bases. Most people use enzymes that recognize six base sequences and agarose gels.

Gels of 0.8 percent agarose are useful for most cases when using six-cutter enzymes. This concentration can be lowered to 0.4 percent for fragments up to 30 Kb, while higher concentrations will help in distinguishing smaller fragments.

Electrophoresis is carried out on agarose gels in buffers near neutral pH at voltages below 5 V/cm. At the end of electrophoresis the gel can be stained with ethidium bromide at a concentration of 0.5 µg/ml, and viewed under UV light of short wavelength.

Southern Blotting

The fragments on the gels cannot be visualized, but must be transferred on a solid support membrane or filter, which is usually made of nylon and then probed with radioactive, labeled probes that hybridize to specific sequences. This procedure is called Southern blotting. The filters are baked at 80° C or UV irradiated so that the DNA is irreversibly bound on them. These filters can be stored for great lengths of time.

There are several methods, which rely on electric current, capillary action, or vacuum, for transferring the DNA onto the filters, the latter two methods being the most common.

Probes hybridized to nylon membranes can be washed off by different procedures. They can then be hybridized more than once, with a different probe each time, thus yielding more than one pattern.

Probe Preparation and Labeling

The probes can be either fragments of nuclear DNA (genomic clones) or cDNA clones, i.e. DNA that was synthesized from a molecule of mRNA through the use of enzyme reverse transcriptase. For genomic clones, total genome DNA is restricted with a specific enzyme (e.g. PSTI) to obtain fragments of suitable length. Of the fragments that are produced and sepa-

rated by electrophoresis, only those in the region of 500–2,000 bp in length are usually retained and cloned. A collection of such clones is called a library, and usually contains several thousand different DNA sequences.

The DNA to be studied is usually digested using more than one restriction enzyme. Each restriction enzyme produces a different pattern, which may help in identification. Many are commercially available in kit form. Genomic clones will not help detect small changes, but they cover a larger portion of the genome, while cDNA clones offer finer resolution. This necessitates the use of many more cDNA probes than genomic clone probes.

To label the probes, DNA must be synthesized in the presence of a radioactive isotope, usually ^{32}P , using oligolabelling, which relies on special enzymes and random hexanucleotides as primers. The clones are usually separated from the vector before labeling so that non-specific hybridization is reduced. The cloned fragments are separated from the vector on a gel that is then stained with ethidium bromide. The segment of the gel containing the desired fragment is cut out and boiled in a small amount of distilled water and stored at -20°C . Non-radioactive reporter molecules such as biotin can also be used and are visualized by staining. Since the stain cannot be washed off, a filter on which such a molecule has been used cannot be used again. The former approach is preferred, as it allows the filters to be used in several cycles of hybridization. Commercial kits are available for labeling.

Hybridization

This is the process whereby the labeled probe binds to complementary DNA on the nylon filter. Hybridization occurs between single strand molecules of DNA. So the DNA is denatured on the gel before being transferred to the filter. The probe is also denatured before being applied to the filter for a sufficient period of time for hybridization to occur. Non-specific hybridization is washed off by increasing the temperature, or through other means.

To avoid background signals, the target DNA on the membranes is pre-hybridized with non-homologous random DNA from fish sperm under special conditions. This DNA binds to repetitive sequences which are then unavailable to the radioactive probe. The radioactive probe is then added in solution and, usually after overnight hybridization, is removed and reused.

The probe that has bound to the fragments is visualized by autoradiography. This is done by placing an x-ray sensitive film over the filter for 1–2 days, and then removing the film and developing it. The positions of the labeled fragments show on the film as darkened areas. After washing the probe off, the filter can be re-probed with other probes.

Applications of the Method

The method as described above or with some modification has been used on many plant species, for instance in *Malus*, *Prunus* and *Rubus* by Nybom *et al.* (1990), potato by Gorg *et al.* (1992), and *Picea* by Bobola *et al.* (1992) with varying degrees of success. In most cases it has resulted in clear identification of cultivars. The usefulness of this method is that in theory, an unlimited number of loci can be analyzed, upon which cultivars can be identified.

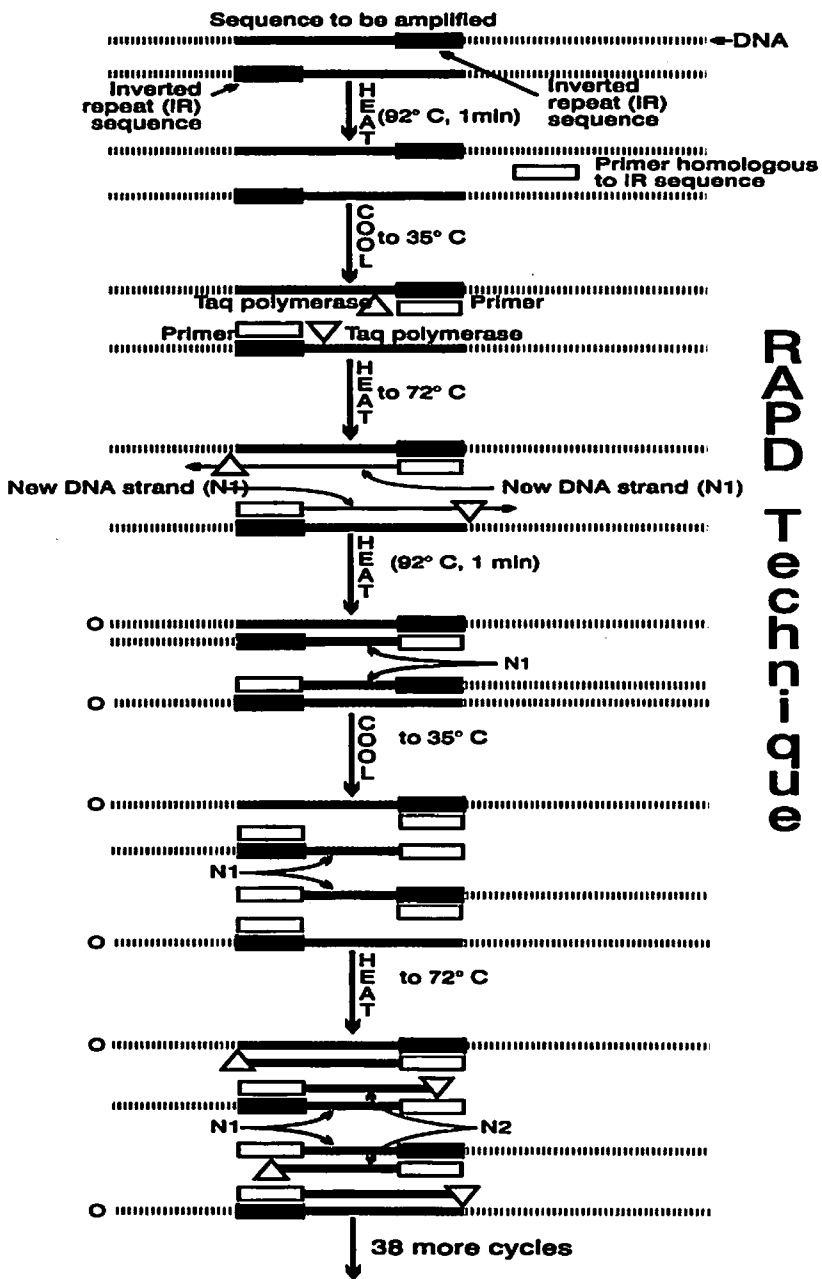
Of course the question of cost arises. One tries to minimize costs by taking into account the number of different enzymes and probes to use and the number of informative bands each probe produces. In sugar beet, for instance (Hallden and Tuveesson, 1991), three different enzymes and 69 probes were sufficient to group 37 different lines according to their known pedigree relationships. In potato (Potter and Jones, 1991), just one combination of one enzyme and one probe resulted in the identification of eleven varieties.

Random Amplified Polymorphic DNA

The RFLP analysis described above requires radioactivity labeled probes to visualize the different fragments generated by the use of the restriction enzymes. This makes it cumbersome. Recently, Random Amplified Polymorphic DNA analysis has become available (Welsh and McClelland, 1990; Williams *et al.*, 1990), which does not require DNA hybridization for the visualization of the bands, but relies instead on selective DNA amplification, electrophoresis, ethidium bromide staining and UV illumination.

The Principles

This method relies on single or paired decamer primers of random sequence that amplify target DNA by Polymerase Chain Reaction (PCR). Fragments of DNA are generated by PCR amplification, if the target sites for the primer occur to within approximately 200–2000 bases of each other in inverted configuration on opposite DNA strands (Fig. 3). These fragments are separated through agarose electrophoresis, stained with ethidium bromide and observed under UV light.



RAPD Technique

Figure 3. Schematic representation of selective DNA amplification by random primers using a thermal cycling procedure.

The polymorphism between different genotypes arises through (Williams *et al.*, 1990):

- Nucleotide changes that prevent amplification by introducing a mismatch at one priming site.
- Deletion of a priming site.
- Insertions that render priming sites too distant to support amplification.
- Insertions or deletions that alter the size of the amplified product.

The number and the size of the fragments generated depend on the primers used and the target DNA.

The Method

Genomic DNA preparation: DNA is usually extracted from fresh plant material of young plantlets using a method such as the one described in the RFLP section. Its concentration must be estimated by using mini-gel methods in comparison with lambda DNA of known concentration, spectroscopy or fluorometry. The tendency is now towards using crude extracts of DNA. One method proposed by Yoshimura *et al.* (1992) relies on DNA obtained by grinding fresh plant material in 100 µl of TE buffer using a mortar and pestle. These authors claim that this crude extract, without any further treatment, gives resolvable patterns with some primers. Deragon and Landry (1992) have presented a method relying on non-destructive sampling via small leaf discs (5 mm in diameter), which are then enzymatically digested to provide template DNA for up to 20 cycles of PCR. The resulting amplification products are equivalent to those obtained from CsCl purified DNA. Up to 120 plants can be treated in two days, according to the authors, who have tested the method successfully with eight major crops. No doubt more methods for quick DNA extraction will be devised. For any method of extraction to be used in plant breeding, and for varietal identification where large numbers of samples are used, it must be quick and inexpensive.

The primers: These are usually 9–10 bases long, have upwards of 50 percent G+C content and lack self-complementary ends. They are usually bought in kits from various manufacturers or can be synthesized by the investigator if he or she has a DNA synthesizer and the expertise. Not all primers amplify informative fragments. It is the practice, therefore, to screen several primers and retain only those that provide PCR amplification products that help distinguish one variety from another.

DNA amplification: This is carried out with the help of a specialized instrument, called a thermal cycler, which has the capacity to raise or lower the temperature quickly or maintain a set temperature.

Usually, 10–20 ng of genomic DNA or 1 μ l of crude extract is used in conjunction with a small quantity of primer (0.2–0.8 μ l), a buffer mixture usually supplied ready-made by the supplier of the Taq polymerase, one or more units of Taq polymerase or some other appropriate enzyme, and appropriate quantities of dATP, dCTP, dGTP and dTTP. The PCR buffer is usually modified to increase the MgCl₂ content, and the final volume of the mixture is adjusted to a final volume, usually 25–50 μ l. The PCR mixture may be overlaid with a drop of mineral oil to avoid evaporation, and placed in the thermal cycler, where the reaction is allowed to run 40–45 times. A representative cycle consists of one minute at 94° C, 1 minute at 36° C and 2 minutes at 72° C, followed by several minutes at 72° C, depending on the instructions of the instrument manufacturer. The procedure can take up to four hours, but it can be reduced to 2.5 h. with no adverse effects on the quality of the banding pattern (Yu and Pauls, 1992). The samples are then kept at low temperatures (4–15° C). What happens during the above procedure (Fig. 3)? By raising the temperature to 94° C, the target DNA is denatured and rendered single stranded. When the temperature is dropped to 36° C, the primers anneal to the homologous DNA sequences on each strand. Because the primers are short they will anneal at many regions of the genome. In some cases, however, a second sequence, homologous to the primer but in inverted configuration, occurs on the opposite strand within 2,000 bases downstream. With primers ten bases long, this inverted arrangement of the sequence is expected to occur in the angiosperm genome 1–10 times, depending on the size of the genome (Weeden, 1991).

When the temperature is raised to 72° C, the primers extend DNA synthesis using the target DNA as a template. Most of the genome where the primers annealed, and which is outside of the sequence flanked by the inverted arrangement described above, will be replicated once. The sequence, however, enclosed by the inverted arrangement, will be replicated twice. Thus most parts of the target DNA will be replicated once for each cycle of the reaction, while the sequence within the inverted arrangement will be replicated twice. With each additional cycle, therefore, the number of copies of this sequence is doubled, i.e. it increases exponentially while for the rest of the genome the increase is linear. Thus, if we let the replication process run 30 times, only 1–10 segments of the genome will be replicated 2³⁰ times, while most of the genome will be replicated only 30 times. Thus, for the sequence enclosed by the inverted arrangement of the sequence homologous to the primer, there will be sufficient quantities of DNA to be visualized after electrophoresis and ethidium bromide staining.

PCR product electrophoresis: The whole amount of the reaction is loaded on a 1–2 percent (wt/vol) agarose gel and electrophoretically sepa-

rated with 0.5X Tris-borate EDTA buffer. Electrophoresis is carried out at 30–35 V for five hours. Some researchers prefer to add ethidium bromide to the gel, but this practice is not generally recommended. Instead, staining by ethidium bromide is carried out after electrophoresis.

For the determination of the molecular weights of the DNA fragments, λ X174 DNA cut with HaeIII is also loaded on the gel. The gels are examined under UV light (312 nm) and are photographed so that they can be analyzed in detail. The speed of electrophoresis can be increased.

Gel analysis: Based on the total number of different bands observed in the accessions under study and those present or absent from a particular accession, several similarity indices can be computed to determine the relationship of the accessions. Computers can be used for image analysis of the gels and the computation of the indices. For varietal identification there must be bands that are exclusive to the individual varieties.

Advantages of RAPD Analysis

There are several advantages of RAPD analysis over that of RFLP:

- **Speed.** According to Welsh *et al.* (1991), 120 individuals can be analyzed in a 36 hour period as compared to one week using RFLP analysis. Sixty lanes of data can be handled in a single day, one-gel experiment.
- **Smaller amounts of DNA.** The amount of DNA required for RAPD analysis (10–50 ng) can be from 200 to 1,000 times less in comparison to that of the RFLP analysis (2–10 μ g of purified DNA). This advantage can be used for the analysis of plant parts, single seeds, etc.
- **The method is capable of detecting single base pair changes in genomic DNA** (Williams *et al.*, 1990; Klein-Lankhorst *et al.*, 1991) and provides markers over the whole genome.

The simultaneous use of pairs of primers can result in the appearance of additional polymorphisms not observed when only one primer is used (Klein-Lankhorst *et al.*, 1991).

One disadvantage of the RAPD analysis relative to RFLP is that most of the polymorphisms are dominant and cannot therefore be used to distinguish heterozygotes with accuracy. This disadvantage is offset by the larger number of polymorphisms generated by the RAPD analysis. Nevertheless, RAPD analysis was used with success in determining the parentage of maize hybrids (Welsh *et al.*, 1990).

Applications of the Method

In deciding to apply the method on a particular species using a particular instrument, it may be necessary to experiment first to determine the optimal conditions. The differences may be due to the instruments (most likely), or to the differences in the purity of the template DNA and the primers (Klein-Lankhorst *et al.*, 1991).

One also must be in a position to calculate the number of different primers required to detect polymorphism between two genotypes differing by a certain number of base pairs. Such calculations have been performed in tomato by Klein-Lankhorst *et al.* (1991), who arrived at the conclusion that 17 different primers used alone and in pairs are sufficient to detect such a polymorphism between two genotypes differing only in a 10 cM genomic region. This translates to approximately 7 Mb.

Some of the species in which the method has been successfully applied for genotype differentiation purposes are: *Theobroma* (Wilde *et al.*, 1992), wheat (He *et al.*, 1992; Devos and Gale, 1992), *Arachis* (Lanham *et al.*, 1992), and *Brassica oleracea* (Hu and Quiros, 1991).

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Section IV

Seed Production Aspects

General Agronomic Aspects of Seed Production

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Farmers must use proper production techniques to achieve high quality and high yield. Special methods and precautions are needed. The seed farmer must be diligent and familiar with the crop to produce seed of sufficient quantity, high varietal purity, and free of pests, disease and weeds.

Selection of Fields and Crop Rotation

Seed fields must be capable of high productivity, and be suitable for the best cultural practices (soil preparation, irrigation, etc.). Different crops have specific soil requirements. Fields carrying soil-borne diseases must be avoided.

Isolation requirements for the specific crop and seed category must be met. Seed fields should be located away from similar crops to avoid contamination from pests and disease. Wild species, similar to the seed crop, should not be too near seed fields, especially in cross-pollinating crops. Examples include sunflower in Central America and sugar beet in the Mediterranean, where these crops originated.

Crop rotation is necessary to avoid volunteers, which may grow from seed shed in previous seasons and persist despite drought, cold and cultivation. The actual length of time required between two seed crops of the same species depends on the dormancy period, the persistence of volunteer seed, rainfall, and other climatic and cultural factors. Two cropping seasons is a desirable interval for cereals, more time is required for forage and other crops such as sugar beet (Thomson, 1979). If the cultivar is the same as the first crop, and is grown from certified seed, growing a seed crop on the same field for two successive seasons is permitted.

Crop rotation is also an effective method—in some cases the only method—for controlling difficult diseases such as *Verticillium* spp., *Phytophthora* spp., eelworm, etc. (Galanopoulou, 1992).

A seed crop should follow a crop which deters weeds or permits intensive weeding. For example, wild oat is an annual weed with a life cycle and growth habit similar to a temperate cereal, so it is a very undesirable weed in cereal seed crops (Thomson, 1979). Some seed is shed and remains in the soil to germinate in subsequent years, while other seed is harvested with the cereal crop and may be sown in another field. Long seed dormancy (more than three years in deep soil) makes wild oat one of the most persistent weeds (Galanopoulou, 1992). Wild oat plants can be eradicated in row crops by inter-row cultivation, or by rotation with dense pastures that are grazed or cut periodically. *Abutilon theophrasti* is a very dangerous weed for cotton seed, as it belongs to the same botanical family. The most efficient method of control is crop rotation.

Field Preparation

Soil preparation should start early to ensure that suitable tilth, weed control and soil preparation are completed by the sowing date. For small seed, a fine tilth is necessary for contact between the seed and soil particles to ensure seed germination and seedling emergence. Suitable seed bed preparation is especially important for spring crops to avoid soil moisture loss. Appropriate field preparation contributes to uniform sowing depth and plant emergence, which are essential for seed production (Thomson, 1979).

Fertilization

Fertilization of seed crops should be based on practices recommended for common cultivation, modified where necessary. The value of a high quality seed crop justifies additional expenditure. In cereals, for example, a continuous supply of soil nutrients, especially nitrogen, is necessary after flowering, because seed development is dependent on the photosynthetic process of the remaining green tissue, especially the flag leaf.

Phosphate and potassium are generally more important for seed crops than for common crops, but can be applied in full at sowing time. Phosphate enhances crop maturity, a critical factor for some seed crops such as cotton. Nutrient deficiency may lead to weak seedlings. Potassium is related to chlorophyll production and, consequently, to photosynthesis, as well as to the translocation of carbohydrates in the storage parts of the plant (such as seed). It also affects cell division, seed development, and increases plant drought resistance—a very valuable factor, especially for seed crops under dry conditions. Some crops (mainly tobacco, cotton and to a lesser extent maize, potato and barley) have a higher need for potassium than others (Galanopoulou, 1992).

Excessive nitrogen fertilization should be avoided, especially in crops and varieties susceptible to lodging such as cereals, maize, cotton, etc. Heavy nitrogen application is likely to encourage foliar disease, cause excessive vegetative growth instead of reproductive development, and may lead to reduced seed yield and late maturity. On the other hand, nitrogen deficiency may cause the crop to mature early, leading to seed deterioration.

Excessive vegetative growth due to heavy nitrogen application is very harmful in dense stands because it increases competition for available light, water and soil nutrients. Under these conditions, early fruit shedding in cotton increases, resulting in crop lateness and yield decrease. In cereals, nitrogen increases the number of tillers, many of which die off, and the survivors are weak and do not produce seed to their full potential (Thomson, 1979).

Lodging and excessive vegetative growth produce a sheltered, humid environment, unfavorable to pollination but favorable to infestation by fungi. Such an environment is unsuitable for the development and ripening of seed, and causes deterioration in seed quality. In cereals, lodging causes reduced light to the lower parts of the plant, thus delaying tillering until it is too late to produce ripe seed, while seed from photosynthesized material from the main stem is completely developed (Thomson, 1979).

Lodging can also create harvesting difficulties, resulting in yield loss. Only late lodging in cereals and grasses can be beneficial, by protecting the plants from wind, thus reducing the shedding of ripe seed.

Sowing

Nowadays sowing by machine is common all over the world, but it is not essential for the production of quality seed. Seed is usually sown in rows rather than broadcast, because this method requires less seed, facilitates uniform seed distribution, allows mechanization of cultural practices—especially weed control—and provides paths for roguing and crop inspection. Most crop species are sown directly into the field. Tobacco is sown in a nursery, and the young plants are transplanted by machine or by hand. Rice can either be sown in the field, or by transplanting young plants from the nursery. Direct sowing of rice can be in flooded or dry soil. Transplantation gives a higher return of harvested seed per kilogram of seed planted.

Prevention of Mechanical Contamination

There is always a risk of mechanical contamination of seed and consequent loss of varietal purity, especially during sowing and harvesting. Tractor

wheels, fertilizer distributors and other implements can bring in unwanted seed from other fields. Bags should be thoroughly cleaned of foreign seed before they are filled with new seed. New bags are preferred. The highest risk of contamination by foreign seed is by sowing and harvesting machines, especially if they have been used previously for sowing other varieties or related species. Machinery must be thoroughly cleaned before it enters the field. As few machines as possible should be used for sowing all the seed fields of a certain variety.

Special Precautions

Seed companies should supply a given seed farmer with one variety of one seed category for all fields. Farmers should not be allowed to cultivate other varieties. Unwitting variety contamination will thus be avoided. Seed must arrive on the farm in sealed bags bearing the official tags from the certification authority. The tags should indicate the variety and seed category and should be checked before the bags are opened, especially when dealing with parents of a hybrid (Goulas, 1992). The seed should be ready for sowing, i.e. treated with insecticide, fungicide, etc. However, *Rhizobium* inoculation of seed for leguminous crops can be done at sowing, as bacteria cannot survive for many days after they have been mixed with seed. Inoculation of the seed is not necessary if the same crop has recently been grown in the field. For F₁ hybrid seed production, male parent and female parent seed should be dyed different colors so they can be distinguished (Thomson, 1979). After sowing, one of the tags should be attached to the seed field marker and the other kept for inspection.

Field Design of Sowing (Especially for Hybrid Seed Production)

For the production of F₁ seed, the two parents have to be sown in the same field in a predetermined number of rows. In the case of sugar beet, polysperm pollen parent seed is mixed with monosperm female parent seed. This mixture is sown in the same rows, and the harvested seed is separated mechanically.

The proportion and row arrangement of pollinator and seed-bearing parents are recommended by the breeder and/or the seed company, and should be clearly shown on the field layout. The rows should be labeled, and the field design must be precisely followed. The presence of an agronomist from the seed company is necessary in most cases.

In all field crops, including self-pollinating, a field plan should be made showing the position of the field and the exact area of the field where the seed was planted.

Sowing Date

Sowing dates depend mainly on temperature—which must be between the accepted limits for the crop—and on soil moisture or the availability of water, whether from rainfall or irrigation. In temperate regions the most critical factor is usually temperature, which should not be below the minimum limit for seed germination and seedling emergence. This is important for spring crops, and for those with a long life cycle that are sensitive to low temperature. Cotton, for example, is a crop with an approximate life cycle of six months. In marginal areas with a restricted growing season, this six month requirement cannot be met. On the other hand, the minimum temperature for seed cotton germination is 15° C. Thus, the choice of sowing date is of great importance. For common cultivation, the earliest possible sowing date is usually recommended, as it results in high yield, despite the often lower percentage of plant emergence. However, for seed multiplication, the risks of early sowing should be avoided, especially for early generations where limited quantities of seed are available (Galanopoulou *et al.*, 1980).

The breeder or seed company usually suggests the sowing date. Careful date selection is of great importance, as it affects plant growth and development as well as the harvesting date. The interval between sowing and harvesting depends mainly on the sum of temperature units, otherwise known as growing degree days. With crops such as maize, which vary in the length of time they are on the field, the sowing date depends upon the hybrid.

The sowing date further depends on the requirement for day length and low temperature for flowering initiation. Most improved varieties are neutral to day length, but some winter cereal varieties cannot bloom if they are sown during spring in temperate regions, because their low temperature requirement is not fulfilled. Similarly, if soybean varieties that are released for higher latitudes are grown for seed production at lower latitudes—where temperature is higher and the photoperiod during summer shorter—the date of flowering and maturity of the crop will be advanced. Thus, there must be an adjustment of the sowing date (Kontas, 1989).

In order to synchronize flowering for F₁ hybrid seed production (such as maize and sunflower), the breeder recommends different sowing dates for

male and female rows. For sunflower, it is sometimes suggested that a seed field should be sown thirty days later than other fields in the same area to achieve time isolation and avoid undesired cross-fertilization (Xanthopoulos, 1993).

For crops that are normally grown for their vegetative parts (such as vegetables), and for biennial species such as sugar beet, other factors have to be considered to establish the best sowing date. Sugar beet is grown for its root as an annual spring crop in temperate regions. The switch from the vegetative to the flowering phase requires a period of low temperature, and if the low temperature requirement is fulfilled, inflorescence may occur in the first year, which is undesirable for root production but very useful for seed production. In the latter case, seed may be sown in the field (in temperate regions) during late autumn, or in the greenhouse at low temperature for transplanting to the field in early spring (Goulas, 1992).

Seed Rate and Row Distance

The optimum rate for seed production maximizes the ratio of harvested seed to sown seed per unit area. When multiplying breeder seed, or even pre-basic seed, the ratio of harvested seed to sown seed may be more important than actual crop yield. For this reason the sowing rate may be significantly lower than for common cultivation. This is also the case for the rapid increase of a new variety. The appropriate seed rate ensures the optimum plant population for seed production. Extensive research has been done on the definition of an optimum plant population, on spacing of plants within rows, and on row spacing. Growth habits, ecological conditions, cultural practices and type of mechanization greatly affect the optimum plant population, even among varieties of the same species. Mechanization of cultural practices and the need to maximize yield per unit area have led to considerable increases of plant population in most crops. Recommending plant population and row spacing for different crops is beyond the scope of this paper.

In some cases, wider row spacing is necessary to facilitate field inspection and roguing. High plant populations should be avoided, especially for seed production, as they lead to a dense leaf canopy which prohibits light induction and aeration to the lower parts of the plant, resulting in prevention of pollination by insects, seed deterioration, fruit shedding and incomplete seed development.

On the other hand, low seed rates may lead to plant populations unable to withstand competition from weeds. Low populations may also lead to nu-

merous lateral branches or tillers, so that the flowering period is extended and seed does not ripen uniformly on all parts of the plant.

Generally, the plant population for seed multiplication is only slightly lower than the recommended population for the particular variety and area.

Irrigation

The amount of water, time of irrigation, and irrigation system should meet the particular demands of a crop, but consideration must be given to seed production. Sprinkler irrigation should be avoided during flowering and seed filling, because it may encourage foliar and seed-borne diseases and can prevent pollination by interfering with stigma receptivity and insect pollination. Flooding is not recommended, except for rice, as it may result in excessive humidity in the lower parts of the plant, which adversely affects seed health and normal development. Surface water, which is preferred for seed production, should not previously flow through another crop, because of the risk of introducing foreign seed (Thomson, 1979).

Water should be supplied during all stages of crop development, especially seed filling, to ensure normal development of the greatest possible number of seeds. At this stage the plant should not be under any form of stress. During the vegetative period, excessive water may lead to competition between vegetative and reproductive plant growth. Water should be avoided during late ripening to minimize harvest difficulties, fruit and seed rot, and seed germination before harvest.

To synchronize flowering in some F_1 hybrid seed production, different irrigation dates are adopted for male and female parents.

Plant Protection

Weeds

Weeds are objectionable in all crops, mainly because they compete for ecological factors, impede cultivation and harvesting, and harbor pests and disease. In seed crops they are particularly undesirable, because if they ripen at the same time, their seed is harvested with and may contaminate the crop. Examples include wild oats in cereals, wild red rice in rice, and *Xanthium* spp. and *Abutilon theophrasti* in cotton. Regulations of the European Community and national laws describe the upper limit of weed seed allowed in a given seed crop.

In seed crops, other crop species are regarded as weeds. They are particularly objectionable if they can cross-pollinate the seed crop. Examples include *Gossypium barbadense* in *G. hirsutum* (upland cotton), wild beet in *Beta vulgaris*, and wild types and *Helianthus tuberosus* in cultivated *Helianthus annuus*. The presence of such plants is not allowed in seed crops. According to seed production regulations, they should be removed by roguing before flowering.

Standard methods of weed control are applicable to seed crops, and should be used with care and attention. The most effective method is sowing absolutely clean seed on a clean seed bed. Roguing of foreign plants is an additional and effective method.

Diseases and Pests

The incidence of pests and disease in crops is affected by their presence in the soil and climate. This must be considered when selecting fields for seed multiplication.

All standard methods for pest and disease control are applicable in seed crops, and the high value of the crop justifies special measures against birds and rodents, as well as additional insecticides. For sunflowers, special measures should be taken against birds, which are an important factor in seed loss, especially in areas with surface water and isolated fields.

Seed field isolation is necessary not only for protection against foreign pollen but also against some air-borne or insect-borne disease. Loose smut in cereals is caused by a fungus (*Ustilago tritici*) which produces its spores at flowering, and these are carried by wind, infesting neighboring crops. Many viruses are transmitted by aphids and can cause severe infestation.

According to national and EU regulations for seed production, seed fields should be free from destructive and contaminating pests and disease. For some fields the percentage of infestation should not exceed a certain upper limit. Oil crops, for example, should be free from *Sclerotinia* spp. and cotton seed fields from *Platyedra gossypiella* (pinkworm). Seed infestation in the latter should not exceed 1 percent for certification of the seed lot. Similarly, cotton seed produced in areas infested with the fungus *Glomerella gossypii* is restricted by law from entering an area or country free from that disease, such as Greece.

Field Inspection and Roguing

Field inspection and roguing are very important in seed multiplication to ensure that seed lots have high genetic purity, physical purity and are free from seed-borne disease and noxious weeds.

Field Inspection

Seed fields need to be carefully inspected during the growing season to maintain the standard of the variety. This is particularly necessary for cross-pollinating species during the pollinating period to avoid undesirable crossing. Fields used to produce breeder seed are inspected by the breeder; basic seed fields are inspected by the breeder or his/her authorized representative. The latter should be trained by the breeder and be well-acquainted with the morphological characteristics of the variety. Fields used to produce certified seed are usually inspected by authorized inspectors.

Seed crops may be inspected frequently during the growing season, but at least one inspection must be timed to assess trueness to cultivar. With most crops this is during flowering or immediately before anthesis (Kelly, 1988).

The number of inspections depends on the species and cultivar. Bread wheat, as well as many open-pollinating species, generally requires only one inspection. Hybrids and inbred lines require four to five; at least one inspection must be carried out during vegetative growth to check morphological and physiological characters; one or two is carried out during flowering to remove plants which may show variation in inflorescence characters; and one is done during seed ripening to remove diseased plants and noxious weeds.

Steps during each inspection are:

- Check field details to ensure that the field is located as indicated in the application.
- Check the number of varieties (for a given species, each farm can grow only one variety of basic seed or two of certified seed).
- Check the category of seed used (the seed growers must show the official tag from the seed bag).
- Check that isolation distances are as required by regulation.
- Check that the previous crop was a different species (it can be the same species only if the seed grower grew the same variety the previous year).

Variety identity and purity must conform to regulations. Inspection is carried out by walking through the field, following a route which allows the

entire area to be covered. Inspection should be carried out on limited areas, or sample areas of at least 100 m². Off-type counts in the sample area are then related to the population estimate to determine the cultivar purity for the crop. At the end of each inspection a report is written including the decision to approve or discard the seed field.

If approved, recommendations which will improve the quality of the seed and increase yield may be given.

Roguing

Many varieties which are produced from seed tend to show genetic change over generations. During seed production control must be exercised to keep varietal variation within acceptable limits. This is achieved by inspecting the crop and removing those plants which do not conform to the characteristics of the variety (roguing).

Roguing may be carried out for any of the following reasons (George, 1980):

- Variation of morphological type within a crop. This is greater in crops which are predominantly cross-pollinating if the varieties are synthetic (forage species, cabbage, melon and onion), than in self-pollinating crops (wheat, barley, oat, soybean, lettuce, pea and tomato). Today, varieties of self-pollinating crops are generally pure lines and are therefore uniform and stable. The F₁ hybrids are extremely uniform but, of course, not stable, and their seed should not be harvested to be grown in the next generation.
- The presence of plants of other varieties of the same species or wild plants that will produce hybrids.
- Deviations from the normal type due to mutation.
- Mixture of seed of other varieties during planting, production, harvest or processing.
- Volunteer plants from vegetative organs or from seed which remained dormant from the previous crops.

Harvest

In many species the seed is capable of germinating a few weeks after the formation of the embryo. However, this does not mean that harvesting can start at that time. In fact, at this stage a loss of yield would result, because the seed is still in the filling stage; seed vigor is low due to the incomplete accumulation of storage nutrients. Moreover, harvesting is almost impossi-

ble because of the high moisture content. Even when physiologically mature (the seed reaches physiological maturity at the end of the filling period, when its weight does not further increase) it is difficult to harvest because of the high seed moisture (sorghum 26–30 percent, wheat 28–34 percent, maize 30–35 percent, sunflower 35–42 percent).

After physiological maturity the moisture content starts to decrease, approaching a level (12–13 percent) that allows harvesting and ensures good storage. However, sometimes seed germination on the mother plant is risked. This occurs when there is lack of the embryo dormancy mechanism.

Harvesting Date

The best time for harvest is difficult to determine for crops that have a very long flowering period (sugar beet, soybean, onion, and many forage legumes), because when the seed of the first flowers is ready to be harvested, the seed of the last flowers may still be in the filling phase. A compromise is needed between quantity and quality. Harvesting should start as soon as the risk of loss becomes high, even if part of the seed has not yet reached the desired moisture level.

Maturity has an enormous influence on the physical quality of the seed, processing requirements, storability, and seedling vigor.

The harvesting date is usually indicated by moisture content and appearance. The best moisture level for harvesting depends on the facilities available to the seed grower and the seed company. If the fruits can be artificially dried and threshed mechanically, harvesting can safely begin when the seed has attained physiological maturity. However, at this stage the seed is easily damaged because of high moisture content.

In maize, the proper time for harvesting is when the moisture in the seed is 25 percent or less (FAO, 1982). In sorghum and pearl millet, physiological maturity is reached at a seed moisture of 25 percent. Panicles should be dried till the moisture of the seed is approximately 12 percent, and then threshed and stored (Chopra, 1982). Wheat, barley and oat can normally be harvested when moisture is 20 percent or less.

Appearance (change of color) of the plants or inflorescences can also indicate harvesting date.

Harvesting Systems (Especially for Hybrid Seed Production)

There are basically two harvesting systems: (i) the plants may be cut and allowed to dry in the field or dried by other means before threshing; and (ii) the seed may be removed immediately from the plants for further processing (Kelly, 1989).

Whichever method is chosen, simple hand tools may be used when production is small scale. Where seed crops are cut mechanically and threshed later, the choice is between a binder and reaper or a special windrower. Crops which have been stacked or windrowed can be collected from the field and taken to a stationary thresher.

For larger areas, direct combine harvesting is the best system, with the advantage of obtaining the seed immediately if conditions are right.

In hybrid seed production when two parents are involved, the plants of the maintainer male line or of the restorer male line are harvested first and moved to a distant place. The whole field is then inspected, and broken or lodged male parents and diseased fruit are removed from the seed rows. The hybrid seed on the female parent is then harvested.

In many developing countries harvesting is done manually, the advantage of which is that laborers can be trained to identify and remove leftover off-types or diseased fruit.

Harvesting of hybrid seed maize may be done by mechanical pickers or picker-shellers (Poehlman, 1979). With the mechanical picker the ears are delivered to field sorting tables, or to the processing plant, for sorting. Husks are removed and undesirable ears discarded. With the picker-sheller, sorting and removal of undesirable ears is not possible. Damage may occur during shelling if the moisture of the grain exceeds 20 percent. Seed is dried at temperatures of 40–45° C. The temperature needs to be carefully controlled, since higher temperatures may reduce germination. After drying and shelling, the seed is cleaned, sized, treated, and bagged.

Prevention of Mechanical Contamination

The main points to consider to avoid mechanical contamination are:

- Grow only 1–2 varieties of the same species per farm.
- For self-pollinating species (barley, wheat), leave strips (8–10m wide) between adjacent crops. Cross-pollinating species must be isolated as regulations requires.

- Great care should be exercised when cleaning the thresher or the combine, as well as trailers or any other means of seed transportation.
- During storage, each variety should be kept separately.

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Seed Production of Cereals

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Introduction

To produce the grain needed to feed a country, a handful of breeder seed has to be multiplied until there is enough seed to plant large areas. Several generations of seed growing are often involved, and extreme care must be taken in early generations to produce later generations of high quality.

Producing seed and producing a commercial crop can involve two completely different technologies. A flowering leaf is considered low quality animal fodder, but flowering is a prerequisite for seed production. Hybrid seed production involves the growing of parent plants, with the female detasseled (maize) or the male sterile. Commercial maize production does not involve any detasseling.

Every farmer is not necessarily a good seed grower, and in many countries farmers are thoroughly screened before they can become seed growers. In general, seed producers should be more quality than quantity conscious. They are often distinguished from commercial crop producers by their attitude towards contamination—not only of the crop in the field, but also of planting and harvesting equipment, transport, storage facilities, etc.

Fortunately, in cereals, seed and commercial crop production are similar. Moreover, the majority of cereals are self-pollinating. These two factors diminish the number of problems. However, due to the similarity between the two production methods, grain can easily be mistaken for seed, and mixing of grain and seed in processing plants, where both commodities are processed, can easily occur.

Since a poor seed crop can always be sold as grain, a seed grower might not be willing to give the crop the special care and attention it needs. The price difference between seed and grain also plays an important role. Is the extra premium for seed sufficient to cover the farmer's extra effort?

In this paper we will use the term *breeder seed* for the very early generations. *Certified seed* is the end product of the seed production program. *Basic seed* generations are intermediate stages.

Land Selection

Seed should be produced in areas where: (i) the variety is adapted (to prevent genetic shift); (ii) climatic and soil conditions are optimal (to avoid losing the crop due to hazards such as frost and drought); and (iii) the variety can be economically produced. This is extremely important for the early generations.

The land selected for basic seed growing should have been free from cereals for at least two years. Exceptions are often made if the land has been under the same variety.

When growing certified seed, no cereal crop should have been grown in the previous year, and only other cereal species are allowed in the year previous to that. Preceding cropping for cereals includes clean fallow, pulses, vegetables, and industrial crops. Other cereal and forage crops should be avoided. Fields heavily infested with noxious weeds, or with weeds with seed difficult to remove upon processing, should be avoided. Wild oat is the prime example. Too many seed production fields in too many countries are infested with wild oat, a noxious weed which is very difficult to eradicate. Campaign-style measures must be taken to control this weed.

When selecting fields for seed multiplication, ease of access should also be considered. Easy-to-reach fields are more easily managed and inspected than fields that are far away, and in cases of emergency, quick action can be taken. Thus, the essential prerequisites for seed fields are easy access and a clean field in the correct agro-climatic zone.

Planting and Crop Care

After the land has been selected for seed production, the seed is planted. Seed bed preparation is the same for seed crops as for commercial crops. Slightly less than the optimum amount of nitrogen should be used; a crop that lodges badly cannot be inspected and will not be approved.

When planting, one should remember that the maximum yield per hectare is not as important as the maximum multiplication rate. The more rapid the increase, the sooner a country will benefit from an improved variety. To achieve rapid multiplication, the seed rate is usually decreased. The initial amount of breeder seed should be as large as possible.

Some rows should be left unplanted for the farmer to walk through when he or she rogues the crop. The unplanted rows should be the same as the tractor tire width to minimize damage when the crop is sprayed.

To avoid contamination, not only the fields, but the planting drills and the vans and trailers used to transport the seed to the field should be clean.

To summarize, low seed rates coupled with high initial amounts of breeder seed increase the multiplication rate significantly. Cultural practices are the same as for a commercial crop.

Isolation

The early generations (breeder seed) should be located in the middle of a field planted to the same variety. The early generations can be planted at two different locations to lessen the risk of losing a complete generation.

For basic and certified seed, minimum isolation distances are usually prescribed, depending on generation and crop. Since cereals are self-pollinating, it is usually sufficient to leave a small strip of land between fields to avoid mechanical admixtures. A physical barrier, such as a ditch between two fields, is preferred. For wheat seed production in India, an isolation distance of at least three meters is required between fields of the same variety (not conforming to standard). But in the Netherlands, a 0.5 m strip is required for separation of certified seed, and a 1 m strip for basic seed.

If a certain species or variety has a relatively high percentage of outcrossing, larger isolation distances may be required.

Fields planted with smut-susceptible varieties should have larger isolation distances. In Morocco, a 150 m isolation distance is required if the loose smut infection is beyond 0.5 percent. In the Netherlands, only fields within 80 m are considered during field inspection.

There are often regulations concerning the number of varieties that can be planted on one farm. In Tunisia, only one wheat variety can be multiplied per farm. In the Netherlands, two varieties of self-pollinating crops may be grown on a multiplication farm.

To summarize, isolation distances have only limited importance for self-pollinating cereals. Only with loose smut infection do they become very important. Growing only one variety per farm reduces the risk of admixture.

Roguing

Roguing is carried out to maintain the varietal purity of a crop, and is an important aspect of seed production. When roguing, one must be careful

not to attempt selection; the genetic make-up of the variety should remain the same. For this reason, roguing in the early generations should be carried out by the breeder or under the supervision of the breeder.

In addition to roguing all plants that do not conform to the variety description, noxious weeds (wild oat, wild barley, *Vicia spp.*, *Raphanus raphanistrum*, *Sinapis arvensis*, *Astragalus spp.*, etc.) and plants of crops and weeds that are difficult to remove during processing (wheat in barley, barley in wheat, etc.) should be removed from the field. Roguing may need to be carried out several times. All tillers and roots should be removed. Off-types should be removed before they have flowered.

Plants with seed-borne diseases should also be rogued. However, once the plant has headed, *Ustilago* has already done its devastating work, and roguing does not help. Therefore, the certification service often does not allow removal of smut-infected plants. Reports from India indicate that loose smut plants head earlier and can thus be rogued before flowering.

To summarize, roguing is one of the most important aspects of seed crop care.

Harvesting

Precautions are no less stringent at the end of the season when the crop is harvested. The cleanliness of the combine-harvester is extremely important. Early generations are harvested by hand or by plot combine and do not constitute many problems. It is advisable, however, to harvest the surrounding area (which was planted with the same variety) first, then clean the plot combine before harvesting the early-generation multiplications. The plot combine should be cleaned between the harvest of different varieties.

Basic and certified seed must be harvested with commercial combine harvesters, which are often difficult to clean. The parts of the machine which may still harbor contaminating seed, even after thorough cleaning, are described by Feistrlizer (1975). Before harvesting, the combine should be properly cleaned by running it empty for several minutes at high and low speeds. The first 500 kg should be discarded because of contamination still present in the combine harvester.

During harvesting, the straw and harvested seed should be checked at regular intervals, and the combine should be adjusted if necessary. Proper adjustment is essential to avoid damaging a large proportion of the seed crop.

After the seed has been harvested, vehicles and trailers used to transport the seed should also be thoroughly inspected. Trailers used for bulk transport, elevators, conveyers, etc., may easily harbor contamination. Cleanliness during harvest is extremely important for seed production fields.

Processing and Storage

Processing

Commercial grain processing should be separate from seed processing. A separate processing plant for the cleaning and treating of basic seed (with different lines for different species) is ideal. The cleaning of the seed processing machines (and the cleanliness of the plant) is very important. The cleaning machines should be thoroughly cleaned, not only between different species, but also between different varieties. All sieves, cylinders and decks should be disassembled for thorough cleaning. Compressed air, brushes, etc. should be available throughout the plant to properly clean the sieves and insides of all machines. Vacuum cleaners are indispensable. The machines should be run for several hours at full speed to discharge hidden grain.

The following sequence should be used for processing seed of the same variety: certified seed, basic seed, pre-basic seed, and certified seed, with the early generations in the middle.

Every processing plant should have a complete set of hand sieves, a small air-sieve cleaner and an indented cylinder to determine the proper processing requirements. This is particularly important when a new variety is processed. A small seed testing facility to permanently monitor seed quality is also useful.

Seed Treatment

Seed treatment is a powerful tool for controlling fungi and pests. Incorrect application can, however, significantly reduce germination and vigor.

Because the early generations of seed are a particularly expensive commodity, the loss of an early generation will have serious consequences for the supply of certified seed. Therefore, the most effective chemicals are always recommended for these early generations, even if these chemicals are expensive. For certified seed, where the volume becomes very large, it is often too expensive to treat all seed with the highest quality chemicals. However, where renewal rates are low, a very positive effect is expected from treatment of certified seed.

Since many chemicals contain toxic substances, it is important to add a warning color to the seed treatment compound. The more dirty-looking the color the better. This will prevent people from eating the seed. It is also advisable to add a warning on the tag such as, "Treated seed, not fit for human consumption."

Storage

After the seed has been processed, it often has to be stored. Poor storage can easily destroy the quality of the seed. Measures previously taken are worthless without good storage (Gregg, 1984).

Breeder Seed Production

The growing of the very early generations involves a different approach than has been discussed here. Breeders are usually responsible for these generations, not seed growers.

Aspects of Seed Quality Control

One of the differences between seed and grain growing is the interference of the quality control authority. All operations may be checked by this agency, and the field may even be rejected as not suitable for seed. The seed grower often has to carry out roguing to make sure that the crop will be up to standard.

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Seed Production of Grain Legumes

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Only a limited number of improved grain legume cultivars has been released throughout West Asia and North Africa. Legumes are considered secondary crops in the farming system. Breeding work to improve these crops has only recently begun, and experience is lacking.

Successful seed production of grain legumes requires special care in crop management. Proper seedbed preparation is essential to ensure a good crop stand and harvest mechanization. Seed dressing and crop spraying with fungicides are important to prevent disease infestation. Seed inoculation is required for good nodulation.

Major constraints to grain legume seed production programs include: crop mechanization, lack of trained personnel, low crop value and the susceptibility of these crops to a large variety of diseases and insects.

Introduction

Seed has been an important agricultural commodity since the early days of crop domestication. Quality seed of improved cultivars is an important element in the progress of agriculture.

The success of a crop breeding program depends on production and maintenance of the genetics of the improved cultivars to be planted by farmers. In most countries the West Asia and North African (WANA) region, the seed legume industry is not well-developed, however, substantial progress has recently been achieved in countries such as Turkey, Egypt, India, Pakistan and Iran.

Compared to cereals, only a few improved legume cultivars have been released by the national programs in WANA. This is because legume crops are considered only secondary to the farming system; they have limited demand, and breeding efforts have only begun recently.

The legume crops discussed in this paper are the food and forage legumes produced for their grain. Self-pollinating crops such as lentil, chickpea, pea and vetch, and cross-pollinating crops such as faba bean, are included.

Development and Release of Legume Cultivars

The majority of grain legumes are self-pollinating, with outcrossing in lentil and chickpea between 1–2 percent. Cultivar development and release procedures do not differ in principle from those for other self-pollinating crops such as cereals. Faba bean, on the other hand, has a high level of outcrossing, reaching 40 percent.

Traditional breeding methods include: the introduction of finished material; segregating material followed by pure line selection; hybridization, usually followed by pedigree, bulk, and back-cross methods of selection; and, recently, the single seed descent method of selection for specific breeding purposes. Growing the segregating material in off-season nurseries usually shortens generational advancement, and F_5 or F_6 seed can be obtained within two years.

When the pure lines are developed, they undergo evaluation under different environments for a number of years. During this evaluation, several lines will be discarded, while the most promising for yield and other specific traits will be maintained. One or two superior lines will undergo verification trials before final adoption and release as a new cultivar.

Needless to say, the newly released cultivar should carry desirable traits, have good resistance to prevailing diseases, and have high yield and quality compared to the local cultivar (or to the most recently released cultivar under cultivation).

Identification of Cultivars

To successfully identify new cultivars, it is important to understand the most important traits in a crop.

There are two main types of chickpea: *desi*, with small angular, light-colored seed; and *kabuli*, with large, ram-shaped and beige seed (Singh, 1986). It is easy to differentiate between the two types, but it is more difficult to differentiate between cultivars within each type. Several traits are considered in the identification of chickpea cultivars. Important traits are: growth habit, stem color and thickness, plant pigmentation, plant hairiness, leaf color and size, seed weight, flowering and maturity date, marker genes such as simple leaf, resistance or tolerance to biotic and abiotic stress, and protein content (IBPGR, 1985).

In lentil, traits considered in cultivar identification include: anthocyanin pigmentation of stem, leaf and pod pubescence, leaflet size, testa pigmen-

tation, flower color, seed size, cotyledon color, protein content, and resistant or tolerance to biotic and a biotic stress (IBPGR, 1985).

Because of its high outcrossing percentage, the description of faba bean varieties is more difficult than for lentil and chickpea. Descriptions of the varieties could include: seed testa color, hilum color, flower color, seed weight (small, medium or large), pod length, and number of seed in the ovary. The quantitative traits are described by means of averages and standard deviations (Erskine *et al.*, 1986).

Management for Grain Legume Seed Production

Seedbed Preparation and Seeding

Management practices for seed production are the same as for crop production, except for the rate of seeding, which is reduced by half. This ensures the production of good and plump seed, and also allows for better field inspection.

The land selected should not have been planted with the same legume crop in the previous season. To avoid volunteers, a non-leguminous crop in the previous year is preferred. Soil should be well-prepared, and if mechanical harvesting is intended, then rolling, following seeding by seed drill, is recommended to level the surface.

Phosphorous should be applied to soil with low P_2O_5 content, and seed inoculation with the proper rhizobium is recommended prior to planting. Weed control should be carried out using the recommended pre-emergence or post-emergence herbicides, or by mechanical cultivation or hand weeding. The distance between rows should be between 30–40 cm to allow cultivation, roguing and field inspection.

The soil should not have any history of disease infestation such as *Ascochyta* blight and *Fusarium* wilt. Fields with a history of *Orobanche* infestation should be avoided.

Since several diseases, such as *Ascochyta* blight and *Fusarium* wilt, which infect grain legumes, are transmitted by seed, it is advisable to treat the seed with chemicals before planting.

Harvesting should be done with care to avoid seed mixture.

Cultivar Isolation

For chickpea and lentil, a 20 m minimum isolation distance between cultivars should be maintained to increase foundation seed, and 10 m for certified seed (Erskine *et al.*, 1986). It is important that faba bean cultivars are well-isolated to avoid any outcrossing; thus a 200 m isolation distance between cultivars is recommended. Brassica plants can be used to isolate fields, because they attract bees and reduce cross-pollination. This isolation form is practical when producing small quantities of seed (Ibrahim, 1987).

Off-types

Off-types should be eliminated. For lentil, however, the maximum permitted percentage is 0.10 for foundation seed and 0.20 for certified seed (Singh, 1986; Erskine, 1986).

Seed Standards

Standards for foundation and certified lentil and chickpea seed have been recommended by the Indian Research Institute. They are presented in Table 1, while those for faba bean are presented in Table 2 (Singh, 1986; Erskine, 1986; Khalil, 1988).

Table 1. Suggested standard for lentil and chickpea seed (from Singh, 1986 and Erskine, 1986, after P.K. Agrawal, India).

Factor	Lentil		Chickpea	
	Foundation	Certified	Foundation	Certified
Pure seed (min. %)	98.0	98.0	98.0	98.0
Other crop seed (max. %)	0.1	0.2	2.0	2.0
Weed seed (max. %)	0.1	0.2	0.0	0.0
Germination including hard seed (min. %)	75.0	75.0	85.0	85.0
Moisture (%)	9.0	9.0	9.0	9.0

Table 2. Suggested standards for some crops in Egypt (Khalil, 1988).

Crop	Min. %	Max. other crop seed %	Max. weed seed %	Min. germ. %	Maximum of other varieties (seed)			
					foundation	registered	certified	commercial
Wheat	95	0.5	1.0	85	0.1	0.5	2.0	3.0
Barley	93	0.5	1.0	85	0.1	0.5	2.0	3.0
Rice	95	-	0.3	85	0.1	0.3	1.0	2.0
Maize	95	-	-	85	0.1	0.5	3.0	5.0
Faba bean	93	0.3	1.5	85	-	-	-	-
Soybean	90	0.5	0.5	75	0.1	0.5	1.0	2.0
Lentil	90	0.5	0.5	85	-	-	-	-
Chickpea	90	0.5	0.5	85	-	-	-	-

Wheat: not more than 3 percent of the seed should pass through a 2.5 mm diameter hole.

Maize: moisture content should not be more than 15 percent.

Rice: moisture content should not exceed 15 percent during Oct. and Nov. and does not contain red rice seed.

Field Inspection

Field inspection should be carried out at flowering or shortly after. Two inspections are recommended. Several traits are considered to ensure that no off-types are present. Some of these traits were mentioned in the previous section. As mentioned, the maximum off-type percentage allowed in lentil and chickpeas is 0.1 percent for foundation and 0.2 percent for certified seed. Laboratory tests are also carried out later to ensure the purity of the samples. If the sample meets the requirement, the seed will be certified.

Constraints to Grain Legume Seed Production

Recently, more improved legume cultivars have been released in the WANA region, and the number is steadily increasing. These cultivars were released in some European Mediterranean countries about a decade ago.

The production of improved grain legume seed throughout the region still faces many difficulties. The differences between legumes and cereals causes many problems, as does the limited knowledge and experience compared to cereals. The major constraints are discussed below.

Crop Mechanization

Legume mechanization is not as successful as cereal mechanization, mainly due to crop stature. Even though seeding mechanization (seed drills) and chemical weed control has been successful in many countries in the region, harvesting mechanization is still a constraint to improved seed production.

Considerable success has been achieved with chickpea, faba bean and pea, although success has been limited with lentil. This is mainly due to the cultivars used, and the management practices followed by farmers. Improvement of the harvesting machines is also needed.

Lack of Trained Personnel

Because of the limited importance of legumes, technicians trained in grain legumes are very few throughout the region. National programs and international institutions such as ICARDA place more emphasis on training and upgrading the capabilities of the national scientists in the production and management of legumes.

Low Value of Legumes

The value of legumes is still low compared to cereals. In addition, the cost of producing legume seed is much higher than for cereals or other crops.

Therefore, farmers growing certified legume seed should be guaranteed a price at the start of the season. This price should be fair, and based on a study of the production cost of legume seed.

Specific Problems Related to the Crops

When grown for the production of certified seed, legume crops have more problems, such as disease infestation, than other crops. Chickpea is usually infected by *Ascochyta rabiei* or Fusarium wilt (*Fusarium oxysporum* f. sp. *ciceri*), which is a serious problem in the Mediterranean. Both are seed-transmitted. Lentil is also susceptible to *Ascochyta lentis* and to *Fusarium oxysporum* f. sp. *lenti*. Seed treatment is needed before planting, and field inspection should be carefully carried out to ensure healthy plants.

Faba bean is a partially outcrossing crop and requires bees for pollination. Separation from seed production fields of different varieties is important to avoid outcrossing.

Several viruses infect faba bean, so this crop must be planted where aphids and beetle vectors are less abundant. Vigorous spraying with insecticide is also required. *Ascochyta fabae*, a major fungal disease, is seed-transmitted, while *Fusarium* species can be seed-transmitted. Both require weekly application of fungicides (Erskine *et al.*, 1988; Khalil and Abdul-Kader, 1988).

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Seed Production of Industrial Crops Including Maize

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Industrial crop plants are either self-pollinating (cotton, soybean, tobacco) or cross-pollinating (maize, sugar beet, sunflower). The prevailing variety type for self-pollinating crops is a *line*, while the corresponding for cross-pollinating crops is a *hybrid*.

Seed production is the term used to describe the procedure followed to make varieties—products of plant breeding—available to the farmer in the form of certified seed. The general scheme for instructional purposes is viewed as a two phase operation: variety maintenance and certified seed production.

Maize Seed Production

Introduction

Maize has long been the favorite crop for plant breeders and geneticists, mainly because of its unique flowering morphology which, along with the pronounced expression of heterosis, led to the development of the conventional breeding methodology currently used and its application in the form of the single-cross hybrids.

Reproduction

Maize is a monoecious cross-pollinating species in which all male flowers are borne in a terminal panicle (tassel), while the female inflorescence (ear) is placed on a side node in the middle of the stem (culm). This unique morphology makes either selfing or crossing easy, and thus maize was the first important crop to lend itself to large-scale hybridization because of easy and effective emasculation.

Each male flower has three anthers, and each tassel produces a huge amount of pollen. The ear is well-covered by bracts, and the long and slender styles (silks) gradually appear on the top of the ear within 3–5 days. Pollen shedding starts 2–3 days before the silk appears and continues for several days, depending on the weather. Pollen is air-borne and usually maintains its viability for 18–24 hours. Fertilization takes place within 12–18 hours after pollination.

Varieties

Hybrids are the prevailing variety type in our modern agriculture, while synthetics are also available.

Hybrids

The seed sown by farmers is the product of a cross between two parents.

Single-cross

Two inbreds, A x B. In some cases the inbred line A can be of some broader genetic basis (i.e. a cross between sister lines). This is referred to as a modified single-cross hybrid, usually designated as "A" x B.

Three-way cross

Designated as (A x B) x C. Three inbred lines are involved, the seed plant being the single-cross (A x B).

Double-cross

Four inbreds are involved (A x B) x (C x D), the final product being the cross between two single-crosses.

Variety Cross and Population Cross

In both cases, parents are of broad genetic basis (varieties or populations) designated as Var "A" x Var "B" or Pop "A" x Pop "B". These two types are important for breeding purposes rather than farming.

Synthetics

Product of the random cross between a number (5–9) of inbred lines.

Cryptic Hybrids

Cross between lines with some degree of inbreeding (S2, S3 etc.).

Variety Maintenance

Hybrid varieties are maintained through the maintenance of their parental components, which are in most cases (single, three-way, double-cross and synthetic) homozygous inbred lines.

Parental Stock

The breeder maintains the parental or foundation stock for each inbred line. This stock consists of a small quantity of seed produced by the breeder in the nursery under strictly controlled conditions.

Parental seed is planted in the nursery, and a number of plants, depending on the size of the operation, are self-pollinating. All these plants have to be verified as being true-to-type. The verification procedure involves an ear-to-row planting of control seed from each self-pollinated ear, while the rest of the seed is kept as remnant. The remnant seed of the rows which are true-to-type is then bulked to form breeder seed. A sufficient quantity of breeder seed is kept in cold storage to provide the nucleus for future multiplication. Depending on storage conditions, this stock can be kept as long as 5–10 years before renewal from the parental stock becomes necessary. The ear-to-row procedure is usually carried out in a winter nursery.

The general scheme for hybrid certified seed production is as follows:

Seed Class	Single-cross Lines		Three-way Cross Lines		
	Breeder	A ↓	B ↓	A ↓	B ↓
Pre-basic	A ↓	B ↓	A ↓	B ↓	C ↓
Basic	A ↓	B ↓	(A x B) ↓		C ↓
Certified	(A x B)		(A x B) x C		

Seed Class	Double-cross Lines			
	Breeder	A ↓	B ↓	C ↓
Pre-basic	A ↓	B ↓	C ↓	D ↓
Basic	(A x B) ↓		(C x D) ↓	
Certified	(A x B) x (C x D)			

Breeder Seed

The small quantity of breeder seed produced is increased in an inbred increase nursery. Plants are either selfed or sibbed. Selfing maintains the line in its pure form but sibbing is usually preferred to prevent excessive loss of vigor.

Pre-basic Seed

This is produced in an isolated block by natural random sibbing. The purity of the seed harvested is verified by post-control, although any outcrosses (off-types) in an inbred line are detected and rogued out before flowering.

Basic Seed

This is produced from pre-basic seed. It may be either an inbred line or a cross between sister lines (modified single-cross). Basic seed is produced in a well-isolated block. For three-way or double-cross hybrids the production of the single-cross or crosses is done in an isolated crossing block.

Certified Seed

Certified seed is produced from basic seed in an isolated crossing field. The multiplication factor is roughly 1:100; the actual figure depends on the type of hybrid and/or the inbred lines used. For some inbred lines used in the production of modern single-cross hybrids the multiplication factor can be as high as 1:200. Normally, seed production of three-way and double-cross hybrids has a higher multiplication factor than single-crosses.

The production of adequate amounts of certified hybrid seed requires several years of advanced planning. Starting with breeder seed, the process takes four seasons. It is common practice to keep fairly large reserve stocks of the various seed classes in cold storage as a safeguard against crop failure and/or unexpected market demands.

Cultural Practices

The area for certified seed production is carefully chosen on the basis of land productivity and the farmer's ability to carry out the required field operations. Special care is taken to ensure isolation from unwanted pollen.

Field Plan

Before planting the isolation crossing field, the ratio of seed plant rows (female or mother) to pollinator rows (male) has to be determined. The

choice of the inbred line to be used as the female plant depends on its production ability, while the male should be a good pollen producer.

The planting ratio (female:male) is usually 2:1 or 3:1. A ratio of 4:1 is sometimes used, although it is not recommended. The most common field patterns (male-female rows) used are 2:4:2:4:2 and 2:6:2:6.

Planting Time

Planting time should be carefully decided. Since we are dealing with inbred seed that is of less vigor compared to hybrid seed, a very early planting could result in poor crop establishment because of unexpected low temperature. A late planting might result in flowering under extremely hot summer temperature. In order to ensure the best synchronization of pollen shedding and silk appearance, it is sometimes necessary to stagger the planting time of the female and male rows. The lag time is often only a few days (3–5). Sometimes even the two male rows are planted with a lag time of few days. In this case, the 2:4:2:4:2 row ratio is used, and the second male row is planted as soon as the first has emerged.

Field Inspection

Field inspection is carried out several times from the early stages until before flowering to rogue out any off-types from both male and female rows. If more than 0.1 percent off-types are found upon official inspection, the crop cannot be certified.

Pollen Control

This is the most important and crucial stage of hybrid seed production:

- **Isolation:** The minimum isolation distance is 800 m for fields and blocks where inbred lines are increased or single-cross basic seed is produced. This distance can be modified depending on local conditions such as the existence of physical barriers, use of border rows, etc. For certified seed production the isolation distance is usually 200 m.
- **Detasseling:** The emasculation procedure of the female rows is accomplished by tassel removal, unless a cytoplasmic male sterile line is used. Complete detasseling of all female plants is necessary to avoid unwanted sibbing and ensure 100 percent crossing. Tassels are removed early, before pollen is mature. Attention should be paid to inbreds which shed pollen while the tassel is not fully expanded (a part of the tassel is still covered by the top leaves). Care is also taken to avoid any leaf removal with the tassel, since data have indicated that a yield decrease of 4, 9 and 16 percent is expected with the removal of one, two or three leaves,

respectively. Tassels are pulled out by hand in an operation which could last 1–5 weeks, depending on the crop condition, but it usually is a one to two week operation. During this period tassels are removed at least every other day, though sometimes field conditions require that detasseling is carried out daily. The following rules are usually applied. One person detassels one female row at a time. The work of 6–8 persons is checked by one person, who does not detassel. During the fourth pass each plant must be checked and late-flowering plants must be removed. Within 8–10 days 98 percent detasseling must be accomplished. The field is checked every other day until all silk is dry.

- In large-scale operations the crew sits on a platform that is driven through the field. Detasseling can also be done with specific cutting machines that are adjustable to tassel height to avoid leaf removal.
- Field inspection is carried out during the detasseling period by official authorities to verify complete and effective emasculation. A tassel is considered flowering when a small piece (5 cm) of the main branch is shedding pollen and/or secondary branches are shedding pollen. No more than 0.5 percent of flowering plants are allowed in female rows.
- The costly and tedious detasseling operation can be avoided by using a cytoplasmic male sterility (CMS) inbred line as the female parent. Such lines were generally used prior to 1970, using the T-cytoplasm, which proved susceptible to *Helminthosporium maydis* and forced seed production to go back to conventional detasseling. Since then, new sterile sources have been identified and CMS lines are again used for commercial seed production on a large scale.
- Genetic engineering has developed pollen sterility that is expected to become available in the near future.
- In the case of CMS lines, inbred line A should be male sterile without fertility restorer genes. Such a line is indicated as A(CMS)rr and requires a maintainer A(NC)rr, where NC indicates the normal cytoplasm and rr the lack of fertility restorer genes. In the production of hybrid certified seed, the pollinator line should carry fertility restorer genes (RR); such a line is designated as B (NC)RR. Thus the single-cross hybrid is finally produced from the cross A(CMS)rr x B(NC)RR, while the seed increase of the female inbred line “A” implies the cross A(CMS)rr x A(NC)rr. In the same manner, production of a three-way hybrid implies production of the single-cross female parent A(CMS)rr x B(NC)rr, and the certified hybrid seed from the cross AB(CMS)rr x C(NC)RR. If pollinator lines B or C carry no restorer genes, such certified seed has to be planted in the farmer’s field as a mixture with certified seed produced in the conventional manner.

Removal of Male Rows

This is done as soon as fertilization has been accomplished and well before harvesting. This operation results in yield increase of the female rows and in ensuring the purity of hybrid seed during harvesting.

Harvesting

Seed moisture of approximately 18 percent is preferred. Harvesting is a two-way operation. Ears are hand or machine picked and brought into the seed house where, after husking, they pass through a conveyor belt for inspection and removal of any diseased and off-type ears. Shelling is done at low speed to avoid any mechanical damage.

Processing

The only specific point to be mentioned here is sizing, where seed is classified according to shape, such as round (R) or flat (F) and to size, such as large (L), medium (M) small (S). The final seed classification is: RL, RM, RS, FL, FM, FS.

Crop Husbandry

For fertilization, irrigation, disease and weed control, the best practices applicable to ordinary farming are followed. Special attention is given to irrigation so the seed crop does not suffer water stress during flowering and pollination. Planting density is 75,000 plant/ha (50,000 female, 25,000 male).

Maintenance of Populations and OP Varieties

These are maintained by using balanced composite seed from selected families. The balanced composites are multiplied by random intercrossing in an isolation block. Sometimes, to assure balanced intercrossing and additional family selection, the modified ear-to-row procedure is applied.

Cotton Seed Production

Introduction

Cotton is the world's leading textile fiber. Besides its fiber, cotton provides seed, which is a rich source of protein and oil for human and animal consumption. This paper focuses on *Gossypium hirsutum* L. (American upland), which represents approximately 90 percent of world cotton production. Although it originated in tropical and subtropical regions, more than 80 percent is now grown between latitudes 20–42°. Each cotton cultivar is usually a mixture of homozygous strains derived from the same cross (multiline) in order to show sufficient homeostasis (stability) under a range of ecological conditions.

There is a remarkably useful heterosis in intra-specific as well as inter-specific cotton crosses (*G. hirsutum* x *G. barbadense*), but there are still problems for commercial seed production, which have so far prevented exploitation of heterosis on a large scale. Consequently, the following data concern seed production of homozygous varieties.

Seed Development and Morphology

Practically speaking, cotton is a self-pollinating plant, with outcrossing between 0–10 percent according to flower morphology, the presence of insects—especially honeybees—and environmental conditions. The pollen is heavy enough to be diffused by wind. Lack of pollination, incomplete development of zygote, or failure of endosperm development leads to undeveloped seed (motes) which adversely affect seed quality.

Cotton bolls have 4–5 lobes with 8–12 ovules and 6–10 seeds on each lobe. The boll maturation period, usually 45–65 days, is influenced by genotype, environmental condition, the age of the plant and cultural practices. Fertilized ovules develop rapidly at the beginning, and in approximately 18 days the seed obtains its final length, while final weight is achieved a few days before boll opening. The seed is pear-shaped, its length approximately 9 mm, and the weight of 1,000 seeds is between 100–130 g.

The parts of the seed are: perisperm, embryo, and traces of endosperm. The embryo is differentiated into radicle and shoot. Two folded cotyledons, for storage of protein and oil, fill most of the space inside the seed. There are black spotted glands in the embryo, filled with the alkaloid gossypol, which

is poisonous. Oil and protein content each exceed 20 percent. Due to high oil content, the cotton seed is extremely sensitive to storage at high temperature and humidity. The seed is covered by fibers (lint and fuzz). The fiber to seed weight ratio is approximately 1:2.

Under satisfactory conditions, seed production is more than 1,000 kg/ha, the sowing rate is approximately 40 kg/hectare, and the seed multiplication factor is approximately 1:25.

Seed Categories

According to the European Union scheme, seed categories for cotton are: breeder, pre-basic, basic, certified first generation, and certified second generation.

The area needed for seed production depends mainly on the cultivated area of a certain variety. It usually takes four years to multiply breeder seed to seed sold to farmers (certified second generation).

Breeder Seed

After a new variety has proven satisfactory in all tests and has entered the national list, it can be multiplied for general use. It is well-known that although cotton is considered a self-pollinating crop, no cotton type is ever perfectly homozygous with regard to all characters, so a certain amount of segregation is bound to take place, giving rise to new types on which natural selection may act. Moreover, seed mixing may occur during handling, as well as contamination or outcrossing in the field. Thus, to avoid degeneration, the removal of off-types is needed as long as the variety is cultivated.

Seed multiplication of early generations should be conducted in conjunction with breeding. The two fundamental principles which should govern any project for seed multiplication are: (i) avoid mixing and degeneration; and (ii) multiply seed as quickly as possible (Christidis and Harrison, 1955).

In Greece, breeder seed is produced every year from the breeding plot, an area of 0.2–0.3 ha surrounded by later generations of the same variety. A micro-trial with 4–6 replications is carried out in which the best lines of the variety are evaluated under normal plant population conditions.

Each row in the breeding plot is sown with seed derived from hand self-pollinated bolls from one selected plant of the previous year. Single plants are spaced 40 cm apart, with rows spaced 100 cm apart, to facilitate phenotypic evaluation and maximize seed yield.

Breeder seed is derived every year from the bulk harvest of the best plants from the breeding plot, based on single plant selection and progeny testing (pedigree method). Selection criteria are based on the desired morphological and physiological plant characteristics to be maintained or improved. Phenotypic selection in the field takes place in two stages. The first occurs as flowering begins. The second is at harvest, when plant phenotype is fully expressed, to permit accurate plant selection. Each selected plant is then harvested, ginned separately and evaluated according to its yield, earliness, technological fiber and seed characters. This is followed by a comparison of the evaluation data with mother genetic material.

Following this system, new breeder seed is produced every year from the best plants of the breeding plot. Part of the seed from each of the best plants is sown in separate rows in the breeding plot the next year, so that selection work will be continued. This intensive and continuous selection between and within lines not only prevents contamination but leads to further variety improvement. The breeding work is completed with breeder seed production; during the following stages of seed multiplication the focus is on maintaining varietal purity.

Other Seed Categories

Breeder seed is sown at a low seed rate, surrounding the breeding plot and the micro-trial. The plot, called the nucleus plot, is approximately 1 ha, and produces pre-basic seed. Pre-basic seed is sown in fields (more than one for safety) of approximately 40 ha, and produces basic seed. From basic seed, the first generation of certified seed is produced, and from the latter, the second generation of certified seed, which is released to the farmers. This certified seed should not be essentially different from that produced in the breeding plot four years earlier.

For a variety cultivated on 150,000 ha, the area needed for the production of sufficient certified second generation seed (including 20 percent reserve stock) is 6–7,000 ha.

Environmental Requirements

The general environmental requirements for cotton seed production are discussed in the paper "Abiotic Stresses on Seed Production." Some specific requirements are presented below.

Temperature

Temperatures below 15° C contribute little to cotton growth and development, and temperatures below 10° C during emergence have detrimental effects. High day temperature usually decreases seed weight and oil content, which indirectly decreases the germination ability of the seed. Optimum temperatures for plant growth and development are considered to be between 30–33° C. Cotton cannot produce well when growing degree days are less than 2,200 heat units (temperature above 10° C).

Water supply

Sufficient water, especially from flowering initiation to boll maturation, is essential. Under drought stress, seed weight and oil content decrease. On the other hand, excessive soil moisture or atmospheric humidity may be detrimental, especially late in the growing season, as they promote boll and seed rotting and prevent cotton maturation and harvest.

Light

Insufficient light greatly restricts cotton growth, therefore high plant populations should be avoided for seed multiplication.

Soil

Cotton has low soil condition requirements. However, good drainage, structure and tilth are essential. Optimum pH varies between 7–8.

Cultural Practices

Advanced cultural practices for cotton cultivation should also be used for seed multiplication. General considerations to be followed by seed growers are listed, as well as considerations for seed growers (see also: “General Agronomic Aspects of Seed Production”).

Fertilization

Excessive nitrogen fertilization should be avoided, as it leads to undesired vegetative growth, susceptibility to pests and disease, lateness of production and decrease of seed oil content.

Date of Sowing

The crop should be sown when soil temperature reaches 14–15° C. Early sowing should be avoided, especially for early seed categories when limited

quantities of seed are available. Sowing machines should be thoroughly cleaned before use to avoid mechanical contamination.

Defoliation

Defoliants should not be applied before the boll is 50 percent open, otherwise late bolls will not mature normally, and will be lighter and produce inferior lint and seed quality.

Harvest

Harvesting machines must be well-cleaned and free from seed of other cotton varieties. Seed cotton should be picked with no more than 12 percent moisture content, otherwise seed quality will deteriorate, especially if it remains unginninged for many days. Degree and speed of deterioration depend on the amount of moisture. The second picking is not generally used for seed, as it usually consists of undeveloped bolls.

Processing and Seed Treatments

Ginning of Seed Cotton

Ginning of seed cotton should follow harvest as soon as possible, especially when seed is damp. Special gin factories containing equipment for seed cleaning are used. To maintain varietal purity, the gin factory should not be used for other varieties unless it is thoroughly cleaned. For breeder seed production, small roller gins are used, while for pre-basic seed a small-scale industrial ginnery located at the breeding institute is often used.

Seed Drying

If seed cotton is transported directly from the field to the gin and dried before ginning, seed deterioration will be largely avoided, even when picked wet. If the seed is or becomes wet after ginning, drying may prevent loss of vitality if carried out quickly.

Seed Delinting

In Greece, as in many parts of the world, acid delinted seed (with sulfuric acid) is generally used, but double mechanically delinted seed (with special saw-gins) is also used.

Seed Disinfecting

To avoid or reduce damage by various disease organisms, seed lots are treated with a fungicide before sowing. In countries infested with larvae of pink bollworm (*Pectinophora gossypiella*), seed should be disinfected with the right insecticide before planting.

Germination Tests

The seed company should test the viability of the seed before and after acid delinting, as well as after treatment of the seed. A final germination test is carried out by the certification authority.

Reserve Stock and Storage

A 20 percent stock from each seed category should be reserved for emergencies. All seed lots, until their final use, should be stored under satisfactory conditions, especially regarding humidity, temperature, insects and rodents.

Legal Framework for the Cotton Seed Industry

Greece is the principle EU cotton producer, and seed production is in accordance with EU regulations and directives from the Greek National Legislation.

According to law, all cotton seed categories are produced under the responsibility of the breeder or his/her representative. For state-bred cultivars (roughly 80 percent of the cultivated area) the plant breeding station (Cotton and Industrial Plants Institute) is considered the breeder. Seed categories up to basic seed are produced under the control of the plant breeding station, either on its farms or on approved private farms. Other seed categories are produced mainly on private farms under the responsibility of the seed company, to which the Institute transfers its rights.

Producing seed for marketing in Greece is permitted when: (i) the seed company has a license from the Ministry of Agriculture; (ii) the variety is listed in the National Catalogue; and (iii) the seed company has submitted a declaration for control and certification to the certification authority. The certification authority for oleaginous plants and fibers is the Hellenic Cotton Board, officially designated by the Ministry of Agriculture.

Control is carried out in: (i) fields of seed growers; (ii) establishments used by seed companies; (iii) post-control trials carried out by the State Institute of Variety Control for Cultivated Plants; (iv) laboratories of the seed control certification and authority; and (v) all stages of seed marketing.

Terms of Seed Production and Marketing

The process involves the following:

1. Application for approval of seed lots for multiplication.
2. Submission of a declaration by the seed grower. The declaration for seed farms must be submitted no later than the 31st of January, while additional information, as well as the multiplication contracts, are submitted after crop establishment (through the end of May).
3. Control of seed farms. The requirements for acceptance of seed fields, in accordance with Appendix I of the 66/402/EEC Directive are:
 - The previous crop should have been of the same variety, another species, or the field should have been fallow.
 - The seed field must have the correct varietal identity and purity, and conform to the description of the variety given by the breeder. The characteristics regarding distinctness, uniformity and stability are stated in Commission Directive 72/180/EEC of 14 April 1972. For pre-basic and basic seed, no foreign plants (another variety or species, weeds excluded) are allowed in the seed field, while for other seed categories one plant per 10 m² is acceptable.
 - Minimum isolation distances are 400 m for basic seed and 200 m for certified seed (distances may be different in case of barriers).
 - Micro-organisms (e.g. pinkworm) that have deleterious effects on seed are accepted up to a certain limit.
 - Weeds which are transferred by cotton seed (e.g. *Xanthium* spp. and *Abutilon theophrasti*) are taken into account during field inspection.
 - General remarks concerning plant population density, plant growth, cultural practices, etc., that may affect seed quality, are recorded.

Two official inspections of the seed farm should be carried out, one during reproductive development before seed maturation, and the second before harvest.

Detailed observations are made by the inspector-agronomist in randomly selected rectangles of 1 or 2 rows (10 m²). The total number of rectangles varies from 5 to 15 depending on the size of the field.

Certification Requirements for Produced Seed

These requirements are in accordance with Appendix II of the 66/402/EEC Directive:

- Minimum varietal purity, which is checked only in the field.
- Seed germination ability should be above 80 percent for all seed categories.
- Physical purity should be at least 98 percent.
- The number of weed seeds from *Xanthium*, *Avena*, *Cuscuta* and *Oribanche* in a 1 kg sample should be zero (other weeds are considered where necessary).
- Micro-organisms which have a deleterious effect on seed are accepted up to a certain limit. For example, seed infestation by pinkworm should not exceed 1 percent.
- Seed moisture must not exceed 12 percent.

All lots for seed multiplication or marketing are sampled by the certification service. A certificate is issued for all approved lots. Every seed bag is officially sealed and carries one exterior and one interior label (white for basic seed, blue for certified first generation and red for certified second generation), showing the seed category, name of variety, country of production, net weight, seed company, certification authority, etc.

The above requirements are in accordance with the General Technical Regulations of the EEC (Official Journal L 195/42, 26.07.1985) and the directive for seed marketing of oil and fiber crops (69/208/EEC of the Council, 30.06.1969).

Sugar Beet Seed Production

Introduction

The evolution of sugar beet from a fodder crop to an important, highly improved source of sugar has been unusually rapid. Along with the development of the modern monogerm triploid hybrid varieties, it is another major achievement of plant breeding. Variety maintenance and multiplication for sugar beet are more difficult than for maize, with special problems related to the biannual nature of the crop and its greater genetic complexity.

Sugar beet as a root crop for sugar production is an annual, while for seed production it is biannual.

The Seed Plant and its Requirements

Growth Habits

After emergence, true leaves start developing in a spiral arrangement, and when there are about 15 leaves the root begins to swell to become the sugar storage organ. If a seedling receives a period of cold followed by growth during lengthening days and increasing temperature, it changes habit from a root plant into a seed plant, a procedure known as bolting. The cold treatment for effective induction (vernalization) is 5–10° C for a period ranging from 2–3weeks to 2–3months, depending on the genotype (easy, medium, hard bolters). Plants will bolt if seedlings have at least four true leaves when undergoing cold treatment, and then grow during long days and warm temperature. Plants can be devernalized if they are grown at high temperatures (above 15° C) immediately after the cold treatment.

Induced plants produce a rosette of new leaves, and as the days lengthen (after about six weeks of growth) the apex begins to elongate, thus initiating the development of the floral axis (bolting). Stems grow very rapidly, flowering two weeks later. Second and third order flower-bearing branches develop. Flowers are in clusters (multigerm) or single (monogerm), with five stamens and feathered stigma. Copious supplies of yellow pollen are mainly wind distributed, and pollination is general. The flowering period is usually two weeks long. High temperatures and rains during this period do not favor fertilization. After fertilization the seed grows and is ready for harvesting in about six weeks. Dry weather during harvest is preferred.

Choice of Seed Production Location

The prevailing weather conditions of the seed production area must fulfill the seed plant growth requirements and, furthermore, be far away from the root crop area for phytosanitary reasons. Early fall temperatures favoring plant growth, winter temperatures to secure induction (without extreme lows), a warm spring (below 15° C) and dry summer summarize favorable weather conditions. These conditions are found in parts of north and central Greece. Under such conditions, the seed crop planting period is late August to mid September, with harvesting concluded by the following July. The variety type (easy, hard bolter, etc.) is also taken into account when choosing the appropriate production area.

Seed Growing Methods

Direct

The crop is sown directly in the seed production field from late August to mid September, and by the end of November plants have grown sufficiently to survive the winter and to be effectively induced. Under this method, plants grow in the same field for a period of 11 months (August–July).

Indirect

The crop is sown from mid to late August in a nursery, and small plantlets with a root diameter of 0.5–2.0 cm, known as stecklings, are produced by November. Stecklings are either over-wintered in the nursery, or lifted and put in cold storage. Transplanting to the seed production field can be done from late November to mid December, or from late February to early March. The indirect method with spring transplanting is preferred in areas where freezing winter temperatures prevail. The indirect method with either over-wintering in the nursery or November/December transplanting facilitates seed growers in their choice of rotation, allowing the seed crop to follow not only small grains but maize, potato, cotton, etc.

Variety Types

Hybrids

The prevailing modern sugar beet varieties are monogerm top-cross hybrids, mainly triploids and some diploids. This variety type implies a seed plant (female) component and a pollinator (male) component.

Mother component

This is a diploid monogerm male sterile single-cross hybrid designated as A x B. Line A is monogerm CMS, and the corresponding "0" type line is required for its maintenance. It can either be inbred or have some broader genetic basis. Line B is an inbred, like A, monogerm and "0" type.

Male component

The pollinator is an OP line, usually multigerm and tetraploid; diploids are less frequently used. Monogerm pollinators and/or inbreds are not excluded, but to our knowledge they are not used.

Multigerm hybrid

This can be produced in the manner described before. The only difference is that the mother component is multigerm. They are of no practical importance.

Synthetics

These are mainly diploid, anisoploid or tetraploid multigerm, with some monogerm as well. Synthetics are produced from free intercross between selected OP lines, which can be diploids or tetraploids for diploid or tetraploid synthetics respectively. For anisoploid synthetics, diploid and tetraploid OP lines in a 1:4 ratio are freely intercrossed, producing a mixture of triploids (50 percent), tetraploids (30 percent) and diploids (20 percent).

Variety Maintenance

Parental or Foundation Seed Stocks

These are maintained by the breeder under strictly controlled conditions. The nature of the sugar beet plant, as compared to maize, requires strict isolation, either under glass or tents made from pollen-proof fabric. The cytoplasm male sterile line A is seed increased by paired crosses A(CMS) x A "0" while lines A"0" and B"0" are increased by selfing. Line C is seed increased by sibbing.

Breeder seed is produced in the manner described for maize.

The general scheme for certified seed production for the prevailing top-cross hybrids is as described for the maize three-way cross, and thus only some specific points will be discussed below.

The multiplication factor for certified seed is about 1:500. There are two patterns for planting the isolation field, mixed and pure (strips). Seed from the female component (A x B) is mixed with that of the male component (C) in a 75–80 : 20–25 percent ratio. The seed is bulk harvested and monogerm seed (the hybrid seed) is separated from the multigerm (pollinator) during processing. In pure (strip) planting, female and male rows alternate in a 2:1 or 3:1 ratio, which results in a field planting pattern of 2:4:2:4:2 or 2:6:2:6:2:6:2:6:2 male to female rows. Male rows are removed after pollination and destroyed.

Field Inspection

Field inspection is carried out several times from the early stages until before flowering and is aimed at roguing any off-types from the male and female rows. Female rows are mainly checked for monogermity and male sterility, and both components are checked for any other off-type plants (i.e. fodder or table beet).

Pollen Control

Sources of pollen contamination are: (i) table and fodder beets; (ii) related species or subspecies like *Beta maritima*; (iii) volunteer beets (annual, weed beets); (iv) bolters from an ordinary beet crop; and (v) certified seed production fields of different hybrids. Minimum isolation distances are 300 m from seed fields cropped with different hybrids, and 600 m from other pollen sources.

Topping

Topping is done to secure the best synchronization in flowering between female and male rows. Furthermore, topping at the right stage results in yield increases.

Harvesting

Harvesting is a two-way operation, cutting followed by threshing. Specific cutting machines are used that separate the cut plants and deliver them to the center or the side of the machine in a swath that is laid on the stubble left by the cut plants. After a few days the seed is threshed using a combine harvester fitted with a pickup reel.

Processing

Sizing is an important aspect of processing. Monogerm naked seed should be 3.5–4.5 mm. During processing, any seed larger than 4.5 mm is re-

moved, and is usually multigerm. Seed smaller than 3.5 mm (2.5–3.5) is used for pelleting.

Crop Husbandry

A planting density of 50–60,000 plant/ha is the only specific point to be mentioned.

Removal of Male Rows

This is done, as has been mentioned, soon after fertilization to avoid field contamination and development of weed beets. Seed fields are checked in the following years for volunteers.

Post Control

Besides the regular seed quality tests, confirmation of 100 percent crossing is performed, in the case of triploid hybrids, through chromosome counting. Another test for identifying outcrosses, using hypocotyl color and stem elongation (annual beets), is carried out in glass houses. Samples are grown at a temperature of 20° C under continuous light of high intensity. Outcrosses can be recognized six weeks after sowing.

Sunflower Seed Production

Introduction

After soybean, sunflower (*Helianthus annuus* L.) is the most popular crop for oil production. Oil seed content varies from 40–50 percent. Seed weight (1,000 seed) is 40–200 g, and the seed multiplication factor varies between 20–60.

Practically speaking, sunflower is a cross-fertilizing plant, pollinated mainly by bees. Every composite flower includes 700–3,000 flowers arranged in homocentric circles. Flowering proceeds from the circumference to the center of the flower and is completed in 5–10 days.

Cultivated varieties were synthetics up to the mid Seventies. Since the discovery by Leclerg of cytoplasm male sterility in 1969, and of restorer genes

for pollen fertility in F_1 , the exploitation of heterosis has led to the domination of hybrids in most countries. Therefore, hybrid sunflower seed production is discussed in this paper.

Seed Class and Variety Maintenance

Most sunflower hybrids are single-crosses between two inbred lines, designated as A, the female parent of the cross with cytoplasm male sterility, and R, the male parent with fertility restorer genes. Inbred A lines are maintained through fertilization with pollen from B lines (maintainers) which are identical to A lines but with fertile pollen.

Seed classes are the same as for maize. Breeder seed is the foundation seed of A, B and R inbred lines. The breeder seed of each line is multiplied to produce pre-basic seed, which will result in basic seed. Certified seed is the F_1 of A x R, which is sown by the farmers. Variety maintenance of A, B and R lines, up to the production of basic seed, is the responsibility of the breeder, while hybrid seed production (certified seed) is usually carried out either by the breeder or the seed company to which the breeder's rights have been transferred.

The variety maintenance scheme is similar to that described for maize. Pre-basic and basic seed of the inbred lines A and B are produced in successive years (isolation 8 km from other sunflower crops). The row proportion for A and B lines depends on the requirements of seed production. Off-types are rogued out. During the first year, 1,000 A line plants are usually hand pollinating by 1,000 B line plants (1,000 combinations), which are selfed at the same time (flowers are covered before flowering by paper or cloth bags). Next year the progeny of each A plant are sown in a separate row and the best (10–15) AB combinations are selected. In the third year the parents of the best combinations are sown to produce basic seed from A plants. In some cases, B inbred lines are multiplied separately in isolated fields.

The multiplication of R lines from breeder to basic seed is carried out on a separate farm. Pre-basic seed is produced from approximately 500 selfed flowers that belong to selected plants conforming to the description given by the breeder. During the second year of multiplication some seed from each selected head is sown in separate rows on a properly isolated farm (at least 6 km away from any other sunflower crop) in order to produce basic seed of the R line. Off-type rows and plants are rogued out before flowering. Two plants from each row are selfed to produce seed for the next cycle of variety maintenance. The rest of the plants are open-pollinated to pro-

duce basic seed. Usually, R inbred lines have plants with many branches, and flower for a long period.

Certified Seed Production

The scheme for hybrid sunflower seed production is similar to that for maize. Control and certification follows the system described for cotton. The following should be considered for certified seed production.

Isolation

To prevent pollination contamination, seed production fields should be well-isolated, especially in Central America, where sunflower originates, and where many wild types exist. However, isolation is also a critical factor in Europe, as volunteer sunflower plants from previous years may contaminate the hybrid purity. In most countries the minimum isolation distance is 4,800 m from commercial sunflower crops, and 3,200 m from other seed multiplication fields. In some cases a time isolation system is also suggested (see "Agronomic Aspects on Seed Production").

Sowing

Different sowing dates for the two parents, and manipulation of irrigation may facilitate the flowering synchronization of male and female parents.

Plant density for seed production is usually lower than for commercial crops, and generally does not exceed 60,000 plant/ha. Recommended row distances are 60–100 cm. The proportion of female to male rows varies from 2:1 up to 7:1, according to the recommendation of the breeder.

Cultural Practice

Special care for crop protection should be taken. Birds can seriously damage seed production fields.

Roguing

Roguing is very important to control and remove all plants not representing the inbred line. Identification of off-types is based on the detailed description provided by the breeder. Plants with dark brown anthers should be discarded from the female rows, as this color is related to pollen fertility, in contrast to the yellow color of male sterile anthers. Characteristics and minimum standards for inspection are given in the 72/180/EEC Commission directive.

The critical time for off-type identification is the flowering period; most can also be distinguished by their morphological characters before flowering.

Pollination

The presence of beehives is essential for pollination. The number of hives should increase during the first seven days to an optimum level of 0.5–2.5 hives/hectare. More hives may induce the bees to visit other pollen sources, which leads to seed contamination.

Harvesting

Thorough cleaning of the harvest machine is very important. Seed moisture should not exceed 11.5 percent. Under adverse environmental conditions, defoliation at the proper time is advised.

Harvesting starts with the male rows—if they have not been removed after pollination—followed by the female rows. Preferably, the female rows are harvested with another machine.

Storage

Seed moisture content must be 9–11 percent. For all oil seed storage conditions, especially with regard to temperature, humidity and aeration are very important. Rodents control is critical, since they can cause serious damage.

Seed Production of Soybean

Introduction

Soybean (*Glycine max* (L) Merrill) is the world's leading oil crop (30 percent of total plant oil production). Seed oil content is approximately 21 percent. Soybean is a self-pollinating plant, and cultivated varieties are homozygous, multiplied by standard methods.

The fruit is the classical leguminous pod, and the seed is of various colors (light yellow, oil green, brown, red) and shapes (egg-shape or round), with a typical scar. The seed multiplication factor is between 24–40.

Soybean is a short-day and short-photoperiod plant, with temperature also affecting life cycle (see "Abiotic Stresses on Seed Production"). Each variety, according to the photoperiod response and the thermoperiod, is adapted to a geographical strip of 160–240 km.

Seed Class, Seed Production, Control and Certification of Seed

EU regulations and directives for the soybean seed industry are the same as those described for cotton, as are seed categories. Breeder seed is produced from selected plants.

The minimum seed germination percentage is 80, the minimum specific purity is 98 percent, with a zero tolerance for foreign seed. Varietal purity must be 100 percent for breeder, 99 percent for pre-basic, 97 percent for basic and 95 percent for certified first and second generation seed.

Roguing off-types should be done during two periods, i.e., the first during full flowering to remove plants with different flower color, and the second after defoliation when differences in earliness and fuzz of the pods and other parts are most easily observed. Minimum plant characteristics to be assessed during inspection are described in the 72/180/EEC Commission Directive (see cotton).

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Seed Production of Pasture Plants

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Introduction

Improved forage plants must also be good seed producers, otherwise their seed will never be economically available to farmers, and most of the scientific and economic effort put into developing better varieties would vanish. The full potential of species and varieties is achieved by the choice of environment and the adoption of well-tailored management practices.

In some regions, forage seed crops with increased yield could replace crops that are no longer economically advantageous (Hides and Desroche, 1989), and at the same time make seed of local varieties available to farmers.

The Most Important Temperate Grasses and Legumes

Of the 10,000 grass species present in the world, only 40 are of actual interest for sowing pastures (Hartley, 1964). Some 20 of these, originating in Eurasia, are of interest in temperate regions. The most important are *Lolium perenne*, *L. multiflorum*, *Festuca pratensis*, *F. arundinacea*, *Dactylis glomerata*, *Phleum pratense*, *Bromus inermis*, *Phalaris aquatica* and *Poa pratensis* (Knight, 1983; Kelly, 1988; Hampton, 1991).

Over 4,000 legume species can be found in temperate regions. The most important are *Medicago sativa*, *Trifolium repens*, *T. pratense*, *T. subterraneum* and *Lotus maizeiculatus*. In peculiar edaphic and climatic conditions many other species are useful for certain applications or as prospective sources of germplasm (Mathison, 1983; Kelly, 1988; Hampton, 1991).

Choice of environment and appropriate agronomic practices can only be based on an extensive understanding of plant genetics and physiology, but up to now genetics and physiology of seed production have not received the attention they require. Most importantly, genetic knowledge of seed production has never been utilized in breeding programs because breeding efforts have focused on the improvement of herbage production. Therefore, as far as seed production is concerned, forage varieties remain wild types, perhaps improved a little by the automatic selection imposed by subsequent

generations of seed harvesting which favor genotypes with higher seed retention, bigger seed, more uniform seed maturity, etc.

The Choice of Suitable Environment

Knowledge of the specific requirements of different species and the phases of their reproductive growth dictates the choice of environment best suited for seed production. After induction and initiation requirements are satisfied, seed production of fodder plants requires moderate and well-distributed rainfall during the vegetative growth period in spring and early summer, a spell of fine weather at pollination, followed by a gentle rain during the filling of the seed and a dry and sunny period for ripening and harvesting. These conditions are well met in the western valleys of the northwestern USA, where favorable weather at harvest, combined with plentiful natural precipitation during the spring and early summer, provides a unique competitive advantage in forage seed production.

In many instances seed cannot be economically produced in the area of largest use. Production becomes, therefore, a national or international problem. Varieties adapted to one area for use as forage must at the same time be capable of producing profitable seed yields in another. In the USA for example, birdsfoot trefoil seed is commercially produced predominantly in Michigan, Wisconsin and Minnesota. In these regions, day length is well above the critical photoperiod when the plants break dormancy in the spring and reach the flowering stage. Therefore, high seed yield in these areas is encouraged by a short period of profuse flowering following cool temperatures. In the southern area of adaptation, birdsfoot trefoil begins to flower earlier, when day length is shorter, because plants are exposed to air and soil temperatures conducive to plant growth. It follows that flowering is not as profuse in southern regions and occurs over a longer period. This affects natural reseeding and, consequently, the long-term persistence of stands (Beuselinck and McGraw, 1988).

The problem of seed multiplication is particularly acute for grass varieties used in the driest areas and for legume varieties in more humid areas. In some environments, e.g. typical Mediterranean, grass seed production is severely limited because of a combination of high temperature, strong wind and water deficit during the filling period. In winter cereals, e.g. wheat, the difficulties have been overcome by breeding very early varieties able to escape drought. With grasses, this approach is impossible because early varieties have severe productivity limitations. Consequently, seed of varieties adapted to difficult environments must be produced elsewhere or with higher inputs, i.e. under irrigation.

However, this is difficult to put into practice. For example, seed of varieties of *L. perenne*, *D. glomerata* and *F. arundinacea* bred at our Institute have never been produced on a large scale because: (i) our farmers are not familiar with the special techniques required by a grass seed crop; (ii) forage seed yield is not competitive with other irrigated crops; (iii) in northern Europe, plants of these varieties do not survive the winter; and (iv) in Oregon their seed yield is not competitive with that of other varieties.

For legumes, dry regions favor insect pollination and allow better water supply control, which is a means of favorably distributing relative resources within the plant (Mansat, 1988; Marshall and Hides, 1988). White clover seed production is low and erratic in northern European countries, which mainly use seed of New Zealand varieties. Attempts have been made to multiply northern European varieties in Mediterranean Europe under irrigation or in New Zealand. Varieties of white clover and lucerne bred in Germany and in Finland have been successfully multiplied in California and Oregon. The occurrence of genetic shift and of genetic contamination must be considered when multiplying varieties outside the adaptation areas.

The greatest variability in seed production found in most forage species, even when cropped in a suitable environment, is due to climate variability. An examination of macroenvironmental factors during anthesis of perennial rye grass shows that minimum temperature accounts for 70 percent of the variance observed in the number of seed/m² (Hampton and Hebblethwaite, 1983). An extensive review of the effect of environmental condition at anthesis is given by Hill (1980).

Management

A proper knowledge of plant physiology and morphology promotes a better understanding of the management required to consistently produce high-yielding seed crops (Clifford, 1987). Bearing this in mind, agronomic aspects that have a major role in determining seed yield are discussed below.

Time of Sowing

Great care should be taken in preparing the seed bed in order to produce a fine tilth for sowing. Sowing must be carried out when the soil surface is still humid enough to promote germination; the seed needs to be well-covered and the soil rolled to achieve maximum soil-seed contact.

Grass seed crops should be sown at a time (end of summer) that allows optimum tiller density to be obtained in time for vernalization during the win-

ter (Talamucci and Falcinelli, 1977), and for maximum fertile tiller production the following spring. Nevertheless, the best time for sowing is spring (March–April), since weed control by agronomic practices during the longer seed bed preparation period (summer, autumn and winter) is more effective. With end-of-summer sowing, timothy, cocksfoot and meadow fescue should be sown not later than the end of August due to their slow rate of establishment and development. Italian rye grass and hybrid rye grass can be sown in September.

Method of Sowing

The method of sowing depends on the species; cocksfoot, timothy and tall fescue give the best results only when drilled in rows (30–60 cm wide), while perennial rye grass, Italian rye grass and meadow fescue give the best results when they are broadcast or sown in very narrow rows (20–30 cm wide).

In the past, the standard width drill was 60 cm, but in more recent times a width of 40–50 cm has been found to be equally satisfactory. A width more than 70 cm is considered wasteful in terms of optimum ground cover. Large seeded forages should be sown at 2–4 cm or 1–2.5 cm for light land and heavy land, respectively.

Table 1. Seed rates and row spacing for forage seed crops.

Specie	Seeding rates (kg/ha)	Row spacing (cm)
<i>Dactylis glomerata</i>	4–5	40–60
<i>Festuca arundinacea</i>	2–4	40–60
<i>Festuca pratensis</i>	5–7	30–40
<i>Lolium multiflorum</i>	4–7	20–40
<i>Lolium perenne</i>	5–7	35–45
<i>Phleum pratense</i>	5–12	35–40
<i>Phalaris tuberosa</i>	5–10	50–70
<i>Phalaris truncata</i>	10–15	50–70
<i>Medicago sativa</i>	1–3	50–70
<i>Trifolium repens</i>	2–3	20–30
<i>Trifolium pratense</i>	1–3	50–80

Seeding Rates

The seed rate should be flexible, depending mainly on seed bed condition and row spacing. The better the seed bed preparation and the wider the row spacing, the lower the seed rate should be. High rates result in many seedlings which compete with one another, resulting in many weak plants. The best density for obtaining higher seed yield is intra-row plant spacing between 10–20 cm, depending on species and variety. Seeding rates and row spacing of the most common forage species are given in Table 1.

Optimization of Density of Inflorescences

The number of inflorescences per unit area is the most important component of seed potential, both in grasses and legumes (Marshall, 1985; Evans *et al.*, 1986; Bullitta *et al.*, 1989). Management practices are required that optimize flower induction in as many shoots as possible by light interception from leaves and meet the cold requirement. The space available to the plant should ensure the most beneficial reproductive response (Clifford, 1987).

For these reasons, plant density in seed crops has to be lower than in forage crops. Optimum plant density varies greatly as a result of species, cultivar, environment, and agronomic technique.

Decreasing the number of plants per unit area increases seed yield until it reaches its maximum, and then it tends to decrease. The right balance between the number of plants per unit area, maximum floral induction and ovule development per plant has to be determined. After the first seed crop, the yield of perennial forage plants tends to decrease due to tillering of the mother plants or their death (Lovato and Montanari, 1991).

Several agronomic techniques, such as fall cutting, grazing or burning are aimed at achieving an optimum number of reproductive tillers per unit area in grasses (Chilcote and Youngberg, 1980; Ensign *et al.*, 1983; Wiesner *et al.*, 1987). Grasses respond favorably to autumn/early winter grazing, since it encourages tillering and removes excess growth. On the contrary, delayed spring grazing or haymaking markedly depresses yield as a consequence of the removal of initiated tillers.

The proper timing of herbage removal is also important in legumes for increasing seed yield. Removal of surplus growth by topping or haying, rather than grazing, is recommended in white and red clovers and lucerne. In western Oregon and the midwestern states of the USA, the first crop of red clover is generally harvested for hay and the second crop for seed. However, in the arid western states, where seed production is the primary reason for growing red clover, mowing after the beginning of flowering usually results in reduced seed yield (Rincker and Rampton, 1985). In other legumes, such as annual clovers, winter/early spring grazing is the common practice. Sub-clover takes advantage of frequent and moderately severe grazing up to the time of flowering.

Use of Fertilizers

Grasses generally show a positive seed yield response to nitrogen application. The effect of nitrogen is mainly related to an increase in the number of

fertile tillers per unit area, seeds per spikelet and seeds/m² (Hebblethwaite and Ivins, 1977, 1978; Hebblethwaite *et al.*, 1980; Nordestgaard, 1986), but not, as already discussed, to an increase in seed set (Marshall and Ludlam, 1989).

The level of application should not be high (80–120 kg/ha/year is recommended according to soil fertility), since it may cause increased lodging and excessive production of vegetative tillers, which are not ideal for pollination, seed set or development.

Many papers deal with the time of nitrogen application. In most seed production areas two alternatives are followed depending on the species: (i) an autumn application, eventually followed by another early application in spring; or (ii) an early spring application. The autumn application is mainly aimed at increasing the number of reproductive tillers, while the spring application increases the number of flowers per inflorescence. Late application promotes the growth of vegetative tillers. In Mediterranean areas the spring application of nitrogen should normally be carried out at the end of the winter.

In dry areas phosphorus is also important for grass seed production (Talamucci, 1988). Phosphorus is a key element for legume growth, but in white clover a general decline in harvestable seed is found as the available soil phosphorus level increases in moisture retentive soils (Clifford, 1985). In our Institute, the application of readily available nitrogen on lucerne at flowering was inconclusive (Falcinelli, personal communication).

Irrigation

Seed production, as well as herbage productivity, can be enormously increased by irrigation, particularly in the dry season or in areas with a typical Mediterranean climate, where soil moisture is likely to be limiting during inflorescence emergence and seed formation in grasses, and during the period of plant regrowth after defoliation in clovers. This is particularly crucial in late flowering varieties. Irrigation is also recommended when the crop is to be established in early autumn in order to promote prompt germination and emergence.

Irrigation requirements depend on soil texture and depth, natural precipitation, evaporation, temperature, length of the growing season and cropping practices. Highest seed yields are obtained when irrigation practices prevent severe plant stress and promote slow, continuous growth through the entire production period without excessive stimulation of vegetative growth.

Irrigation just before anthesis can also give a major lift to seed yield in dry seasons and on shallower soils. During anthesis irrigation should not be practiced, since it may interfere with pollination. Irrigation after seed set is also undesirable, because it is likely to cause severe lodging such as when high levels of N fertilizer are applied.

Seed weight is increased by irrigation because it is associated with a longer seed filling period (White, 1990).

Chemical Control of Plant Growth

The use of growth regulators has opened new prospects to seed production. Yield increases 100 percent or more above the control have been reported for both grasses and legumes (Hampton and Hebblethwaite, 1985; Hampton, 1986; Hebblethwaite, 1987; Rijckaert, 1991). The use of chemicals (chlormequat chloride, paclobutrazol, flurprimidol and ancymidol), the main effects of which include reduced lodging and more uniform ripening, makes it possible to obtain for grasses what breeders obtain via genetic manipulation in wheat, rice and barley.

The mode of action of these chemicals is not well understood. Further studies are probably required to clarify the effective role that growth regulators play in assimilation and transfer of assimilates, and on the effect of residues in the soil. The increasing concern about pollution urges great caution in the use of chemicals.

Weed Control

Weeds reduce yield and quality of the seed lot for subsequent sowing. Yield losses, which can reach 100 percent, result from weed competition with the growing crop and from separation losses at harvest and in seed cleaning.

Weeds are generally controlled by herbicides that have been largely derived for other crops, since the chemical industry considers forage seed crops minor crops that do not merit research (Oswald and Hagggar, 1980). It is likely that this trend will continue in the future. Other control measures include traditional agronomic practices such as rotation or deep plowing, but not all are applicable or convenient on all crops, and in many cases herbicide spraying is the most effective and cheapest way to control weeds.

Perhaps future research will lead to a different solution of the problem through the breeding of cultivars that inhibit the growth of other species. At the moment, the problem of freedom from weeds appears to be far from a solution that does not include the use of herbicides.

Pest Control

Pests can affect seed yield, either reducing or destroying the seed, and seed quality, resulting in reduced seed vigor. In comparison to pest attacks on herbage crops, little has been published on pest attacks on seed crops (Labruy  r  , 1980). It has been estimated that about one tenth of the lucerne and clover seed crop is lost annually in the USA just as a consequence of disease attack (Graham *et al.*, 1972; Leath, 1985). Sometimes epidemics that completely destroy the crop occur (Urbahns, 1920; Hardison, 1963; Labruy  r  , 1980; Meyer, 1982).

In some seed production areas farmers are advised to spray. In others, pest control is limited by low economic return (Freeman, 1985; FNAMS, 1986; Welty, 1992). The use of sprays appears unwise because of ground and water contamination, the effect on non-target organisms, pesticide label restrictions, and added production costs. However, if breeding for disease resistance is applied systematically to major pathogens of cool season forages grown for seed, long lasting and environmentally safe control of pests can be achieved (Welty, 1992).

It has recently been discovered that *Acremonium* endophytes have a profound effect on some grass species (Fletcher *et al.*, 1990). Endophyte-infected grasses may show, along with negative effects on animals, enhanced growth and resistance to drought, pests and disease. What is more relevant to this paper is that endophyte-infected tall fescue plants were reported to have much higher relative fitness for seed production than uninfected plants (Rice *et al.*, 1990). However, this was not found in another study (Siegel *et al.*, 1985).

Pollination

Pollination is a key factor in seed production of temperate forages, which are generally cross-pollinating. In wind-pollinating grasses, good pollination is often prevented by rain, wind and lodging, which may cause a 60 percent seed yield decrease (Hebblethwaite *et al.*, 1980).

In legumes, fertilization is mainly promoted by foraging bees. Plant physiological processes which affect nectar secretion and thereby bee visitation must be carefully studied (Clifford, 1987). The development of cultivars with florets that trip easily has been indicated as a means of increasing seed yield in lucerne (Knapp and Teuber, 1990).

Abundance of effective pollinators is an agronomic practice which must be pursued. Studies conducted in California on lucerne and ladino clover indi-

cate that 4–7 bees per hectare are necessary to obtain good results (Marble *et al.*, 1970; Muller, 1981).

Harvest

Because of the range in ripeness, lodging, shattering/pod dehiscence and harvest losses, not all the seed that is produced can be harvested. Harvest losses ranging between 20–75 percent are often reported both in grasses and legumes (Foster *et al.*, 1962; Talamucci and Falcinelli, 1977; Andersen and Andersen, 1980; Clifford and McCartin, 1985; Meijer, 1985; Simon, 1987; Horeman, 1989; Elgersma, 1990b; Hampton, 1991).

The optimum time for harvesting is a compromise between the time when the moisture content of the majority of the seed is still high and the time when an excessive amount of seed has been shed (Simon, 1987). In legumes, the use of chemicals which desiccate the aerial parts of the plant, allowing direct combining, is rather common. The time of application is very important, because too early application can negatively affect seed quality. Because of increasing pollution concern, this type of chemical is not encouraged.

The mechanization of herbage seed harvesting has been at the initiative of seed growers, field advisors and other organizations rather than by experimentation. The main objective of seed growers is to find the best harvesting time in order to achieve maximum seed yield; in all cases this is strongly related to the proposed method of harvesting. There are two basic methods. The first is to cut the crop with a mower, leaving it in swaths to dry and then be threshed. The second is to harvest the seed crop by direct combining.

Swathing

This method is normally used for harvesting broadcast grass and legume crops. The crop is cut at an early stage of maturity, the seed matures in the swath and is then picked up and threshed by a combine. Cutting too late for swathing will lead to losses due to shattering.

Direct combining

Compared with the previous method, this has the advantage of saving time and labor; if the harvest is carried out earlier than the optimum harvesting time, there could be serious physical damage to the seed because of its high moisture content and soft endosperm. If, on the other hand, it is delayed because of bad weather, the crop could be over-ripe, and seed shattering could cause significant losses. To reduce the latter possibility, double com-

binning can be used: the combine is first used with the widest drum at a low speed so that only the ripe seed is removed, and after a period of conditioning, the windrow is picked up again and threshed normally.

Seed Drying for Storage

Most grass seed crops are harvested when the moisture content of the seed is 15–30 percent; sometimes the moisture content of the seed is higher than 30 percent. These conditions are often necessary to avoid seed loss due to shattering, but they cannot be maintained afterwards because seed vitality could be seriously injured if the seed was stored at these moisture levels. A few hours after storage, high moisture content causes heat that is sufficient to injure the germination vigor or germination capacity of the seed. In addition to this, at certain temperatures damage is also caused by fungi and insect attack. If the objective is to store seed for a short period (about 12 months) then the moisture content of the seed should not be higher than 12–13 percent.

Artificial drying is a common practice in more humid areas, but is also important in dry areas to maximize seed yield. The higher the moisture content of the seed, the lower the drying temperature has to be and, as a result, the longer the time required to reach the desired condition.

Conclusions and Prospects for Seed Yield Improvements

Forage seed must be produced in high quantities and, at least in the poorest environments, at a low price. It follows that forage species must be bred specifically for seed production using an approach which takes into account the choice of the basic material, the environment in which the breeding work has to be done and the characters that must be improved.

In forage legumes, seed yield is positively correlated with forage yield. For grasses, the two characters seem to be negatively correlated but evidence is far from conclusive. If the crop is properly managed, a potentially high seed yield can be obtained with all varieties. However, it is not always possible to obtain high seed yield and high forage yield from the same crop.

At present, most forage varieties are synthetics based on a number of clones, which should leave intact the reproductive capacity of the populations. However, since the seed potential of the basic clones is almost never taken into account during the breeding process, and reliable criteria to predict seed yield have not been found (Rincker *et al.*, 1988), the choice of

new varieties for seed yield must be carefully evaluated before starting seed multiplication.

Four regions of the world produce nearly all the temperate grass seed that is marketed: the USA (Oregon), the EU (Denmark, UK, France, The Netherlands), New Zealand and Australia (Hides and Desroches, 1989). All these countries have strong grassland activities and a long history in forage plant breeding. Scientific information on seed production of grassland species is mainly related to the environmental conditions of these countries. This creates limitations on the development and improvement of the seed industry in and for more difficult environments. It follows that, as Talamucci and Choulet (1989) have indicated, an extensive varietal uniformity exists in the world, which does not allow the environmental requirements of new and less favored environments, e.g. Mediterranean, to be met. In these environments, when using the genetic resources of forage species, emphasis has been on collecting, introducing and evaluating, with very little emphasis on creating varieties, and almost none on producing seed.

Agronomic research in the traditional seed production areas has obtained very important results, and for each crop and environment the production strategy is well-defined. In order to obtain new, significant improvements, research has to play a key role. It should be directed at optimizing: (i) the number of inflorescences per unit area; (ii) seed setting and development; (iii) synchronization of flowering and ripening; (iv) seed retention; and (v) weed and pest control.

Seed setting and development and seed retention should have high priority. Of course the best results can only be achieved by a close cooperation between agronomists, breeders and plant physiologists.

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Seed Production of Vegetable Crops

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Introduction

Royal Sluis is a private Dutch seed company dealing with breeding, production and selling vegetable and ornamental seed. Additional services—coating, priming, pelleting, and packaging for the hobby market—are carried out separately from core activities. Electrophoresis testing to determine true hybrids was recently added to the list of services.

This paper focuses mainly on those vegetable seed production activities that cannot be carried out effectively without a production manual and specific crop information. Quality throughout the production process is of main concern.

Practical seed production problems for cauliflower are presented as an example.

Seed Production Manual

In the manual the seed production process is described in detail for each crop and variety. To minimize risks, a diagram exists for all production locations, for each crop. There is a general timetable for each crop, covering the complete production process, including locations in the northern and southern hemispheres.

It is most important that quality standards for seed be maintained.

Producing seed involves a number of steps. Sowing and nursing are set apart because of the growing numbers of specialized people handling these operations. Seed production is mostly carried out by contract growers. Optimal conditions must be created for pollination and seed setting. Specialized skills are required to handle seed maturation, ripening, and harvesting. Threshing can damage seed quality if not carried out correctly.

If the moisture content is appropriate, the seed lot will be cleaned immediately, otherwise the seed is dried first. Upgrading of the seed is often needed. Seed samples are taken to assess quality (including health), as well

as for other purposes. Seed is sometimes frozen to kill insects, then sized, upgraded, treated with fungicide, packed and labeled. Seed treatment is often carried out before delivery.

Regulations require a form describing the product for delivery.

Seed can vary greatly in size and shape. For mechanical sowing, a uniform size is required. This can be accomplished through calibration and/or sizing. Seed can also be coated, primed, or pelleted. Chemicals and/or herbicides can be added to the seed. Figure 1 summarizes the various product forms.

Superfrax: This is the brand name for graded precision seed with very high germination, vigor and uniformity. Superfrax is therefore a good basis for a uniform product.

Split Pill: Seeds are coated and as soon as they get moist, the pill splits open, allowing maximum access for moisture and oxygen. Coated seed is particularly suited for precision sowing because of its shape and specific gravity. Together with the use of high quality seed, split pill is the best process for indoor sowing on both soilblocks and plugs.

Split Pill Special: In this process, split pill is combined with pregerminated seed. The result is improved uniformity and germination, together with a perfect sowing ability. Recommended for indoor sowing.

Sanokote: In this process the coating contains fungicides and an insecticide. These chemicals protect the crop against early attacks of soilborne disease and pests. It is the safest seed treatment because the chemicals do not come in direct contact with the seed. Sanokote also provides regular spacing and a more uniform emergence. Especially suitable for outdoor precision sowing, not for soilblocks and plugs.

Splitkote: This is a special process for outdoor precision drilling. The result is a very high germination rate under all kinds of climatic conditions. In combination with an excellent sowing ability, splitkote is a very successful process for many crops.

Figure 1. Treatments and costing processes for vegetable crops.

If a good seed production manual is followed, faults are fewer, even uncommon. It is furthermore important to evaluate all seed production operations, and modify the operations (and the manual) accordingly.

Crop-Specific Information

To start seed production of a new variety, product information and the results of all trials must be combined into a variety-specific information system. This requires the genealogy and history of the parent lines as well as available information about the hybrid. Detailed seed production informa-

tion is also added. From these files a catalogue is prepared for potential client-growers.

Seed production of each variety has its own specific bottlenecks, which tend to change over the years.

Since different types of male sterility are used for hybrid production, the multiplication of parent lines is complicated. When male sterility is absolute, no additional precautions need be taken. In many cases, however, male sterility is not complete, resulting in a certain percentage of selfing. Selecting the seed that resulted from selfing requires careful attention.

Quality in the Seed Production Process

The production process is a critical operation for a seed company. It is difficult to estimate, plan for and obtain the right amount of seed of the desired quality at the right time. Yet this is necessary to minimize risks and avoid over-cropping. This can be best realized when the total seed production process is of high quality.

A high quality seed production process will result in fewer losses such as process disruptions, product refusals, and inspection costs. High quality also means reliable forecasts with less stock, and fewer occurrences of “not available.”

Cauliflower Seed Production

Problems in cauliflower seed production are presented here to demonstrate the importance of the methods discussed above.

Developmental variations that create differences during flowering can be minimized by selecting plants at the generative phase and placing them under cold conditions. Synchronization of the male sterile and female is accomplished by field experiments to determine the exact flowering periods of the parent lines. Because active bees are important for good cauliflower pollination and seed formation, and bees in poor condition are not active, healthy beehives must be maintained. Birds can result in seed losses as high as 10–15 percent. Because cauliflower seed fields are large the problem is hard to solve, but bird nets or repellents can be successful.

Abiotic Stresses on Seed Production

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Introduction

Each crop has its own set of environmental conditions under which it grows most efficiently. Generally, crops are not profitable unless they are adapted to the region in which they are produced. Otherwise, high yield can be obtained only by high energy inputs during crop production, which generally increase production costs and environmental pollution.

Only the most favorable regions should be selected for seed production, and if such regions cannot be found in a certain country where the crop is cultivated, seed production in another country may have to be considered. The planting of high-vigor seed is justified for all crops to ensure adequate plant populations across the wide range of field conditions that occur during emergence.

The three groups of factors that determine whether a crop can be grown economically in a given region (and therefore determine its distribution) are: climate, soil, and socioeconomic conditions (Wolfe and Kipps, 1959).

Environmental and biological stresses (otherwise abiotic and biotic stresses), are the main factors limiting crop production throughout the world. Hence, breeding for tolerance to such stresses, while maintaining high yield and other desirable agronomic characteristics, is one of the most important functions of plant breeders.

This paper considers abiotic stresses due to climate and soil that affect seed production. A brief examination of the factors affecting plant growth and development, with emphasis on seed production, is also provided.

General Factors Affecting Plant Growth and Development

Plant growth and development are influenced by internal factors such as genotype and plant growth regulators, and external factors which interact with the genotype. The most influential of the environmental factors on plant growth and development are climate—especially light, temperature

and water supply—and soil. There are other climatic factors such as wind, but generally they are of less importance. All abiotic factors are interrelated in their affect on plants, and their influence depends mainly on the plant growth stage.

For successful seed production, the plant, before entering the reproductive phase, should have completed sufficient vegetative growth and development to bear as many seeds as possible. It then requires a long enough growing period to complete its reproductive growth and development. Reproductive development starts when apical meristems begin producing inflorescences, and stops when plants have produced ripe seed and die. Flower development includes the following stages: (i) induction of reproduction; (ii) initiation of bud meristem development; (iii) morphological development of flowers; and (iv) blooming. For seed production the following stages are necessary: (i) pollination and fertilization; (ii) ovule development to seed; and (iii) safe harvest.

Abiotic Factors Affecting Seed Production

For crops grown for seed production, normal agricultural conditions are suitable, but the most favorable location should be used in order to obtain the highest possible multiplication rate. There may, however, be difficulties in securing adequate field isolation (Thomson, 1979).

Conditions which favor crops grown for their vegetative parts, e.g. cool temperature and rainfall, do not always promote seed production. Dry weather (especially during the harvest period), prolonged sunshine, and warm conditions during plant growth and development are generally the ideal conditions for seed production. For such crops, commercial seed production is not often carried out in the cropping area. There is a tendency for seed production to be in dry regions. For example, in Europe, herbage is a more important crop in the north than in the south, but there is proportionately more seed produced in the warmer and drier Mediterranean countries (Thomson, 1979). Similarly, a large proportion of the of sugar beet seed used in Central Europe is produced in Greece, where favorable conditions prevail, especially for crops maturing during summer.

Rainfall and relative humidity generally do not favor pollination, and inhibit the movement of bees for pollination. Moreover, the plant should not be under water stress during pollination, because the receptivity of stigma is adversely affected. Also, high temperatures during summer midday hours prevent germination and proper function of the pollen grain. In Greece, favorable conditions for pollination usually prevail during morning hours.

For seed development, sufficient photosynthetic products, with a maximum proportion translocated to developing seeds, are necessary. Photosynthesis depends on a sufficiency of water, light, and nutrients. In some plants with indeterminate flowering periods such as cotton, the plant continues vegetative growth throughout the reproductive phase, especially when environmental conditions are favorable (Galanopoulou, 1992).

Stress on Seed Production from Abiotic Factors

Light Stress

Day length and light intensity have a strong influence on plant growth and development. Seed of most grasses is light sensitive, and light is necessary for its germination (Thomson, 1979). Other seed requires an absence of light for germination. Generally, the absence of sufficient light increases the rate at which stems elongate.

Light is necessary for photosynthesis, and is therefore required by green plants for the manufacture of food. If photosynthesis is restricted by insufficient light duration and intensity, crop growth and development are hampered. In temperate species, low radiation enhances and prolongs vegetative rather than reproductive growth. For example, cloudy days expand the vegetative growth of cotton and delay flowering, thus affecting crop earliness and yield. Duration of sunshine from the vegetative phase to flowering was found to be closely related to yield in cotton grown in Greece (Chlichlias *et al.*, 1977). Cloudy and wet weather does not provide suitable conditions for pollination, ripening and drying of seed.

In many plants flowering is influenced by day length. If plants experience the wrong day length during the vegetative stage, they remain vegetative indefinitely. This effect of light on plants is known as photoperiodism. Long-day plants require a comparatively long day for flowering, and their vegetative growth increases when the days are short. Wheat, oat, red clover and most species originating from temperate regions are among the long-day plants. The short-day plants, such as maize, soybean, and sorghum, as well as most plants of tropical and subtropical origin, achieve their vegetative growth when the days are long, and flower and produce seed when the days are short. The location chosen for seed multiplication must have the appropriate day length at the right time of the year (Thomson, 1979).

Photoperiodism may preclude the possibility of cultivating a variety in an environment different from that in which it was released. Due to photoperiodism, most soybean varieties become earlier as they move from a higher to

a lower latitude, and vice versa. Exposure of sugar beet seedlings to continuous light causes induction to flowering during the first instead of the second year. In maize, the number of leaves and the time from emergence to flowering increase almost linearly alongside increasing photoperiod. This is important for describing cultivar adaptation (Bonhomme *et al.*, 1991). Plant breeders of many crops have succeeded in overcoming the barrier of geographical plant expansion due to photoperiodism by creating varieties indifferent to day length. For example, cotton was originally a short-day crop.

Inadequate light interception at the lower parts of the plant due to high population density may cause fruit shedding in cotton and incomplete seed development in many crops (Galanopoulou *et al.*, 1980).

Stresses from High and Low Temperature

Temperature is the main environmental factor that determines a crop belt. Knowledge of the three growth temperatures—minimum, maximum and optimum—for each species and variety of plant, and for each growth stage, is essential for successful crop growth and seed production. Stresses from minimum and maximum temperatures occur very often, especially with crops grown in the marginal areas of their particular belt (Galanopoulou, 1992).

Plant sensitivity to cold stress is usually higher during germination and flowering. The temperature at sowing time, particularly in the soil, affects stand establishment. Cotton seed, for example, is very sensitive, requiring a minimum soil temperature of 15° C, while temperatures below 10° C cause severe damage, and temperatures below 5° C—even for a short period after germination begins—have a detrimental effect on seedling emergence, survival and subsequent development (Christiansen, 1964). Field establishment of *Phaseolus* and maize is very slow below 10° C, while seed germination of cereals, especially rye, may start at just above zero (Thomson, 1979). Certain crop species and varieties have a chilling requirement for seed germination and flowering; this phenomenon is called vernalization. Winter wheat cultivars have a low temperature requirement at an early stage of growth for flowering induction. If these cultivars are sown in temperate regions, such as Greece in the warm days of spring, the plants remain vegetative and no ears are formed. Similarly, exposure of sugar beet seedlings to low temperatures causes flowering during the first year of life, which is useful in seed production.

Weather colder than a crop requires generally reduces the speed of germination, emergence and plant growth, and increases susceptibility to disease.

Rapid increase of temperature after the cold period may increase plant damage. Temperatures below optimum during the vegetative phase, in correlation with high water supply, postpone initiation of the reproductive phase, resulting in undesired crop lateness. High temperature and dry weather accelerate the entrance of plants into the reproductive phase before there is sufficient vegetative growth to bear enough reproductive organs.

Flowering, pollination, seed setting and ripening are favored by warm weather at the appropriate time. Plants are very sensitive to cold stress during flowering, but a temperature that is too high may inhibit pollination, development of ovules and fruits, and cause shedding of flower buds or young fruit. Heat and drought stress during the filling period in cereals cause shriveled seed of poor quality. Cotton plants exposed to daytime temperatures of 30°, 35° or 40° C during the fruiting period accumulated 47, 5.7 and less than 1 percent, respectively, of their biomass as bolls (Reddy *et al.*, 1992a). Four times as many fruiting branches were produced at 30°/22° C (day/night temperature) as at 20°/12° C, and all flower buds were abscised from plants grown at 40°/32° C (Reddy, *et al.*, 1992b).

High temperatures, especially accompanied by wind, may lead to plant dehydration, wilting, and increased evapotranspiration. Another adverse consequence is the decrease of the plant's photosynthetic capacity.

Perhaps of greater importance than minimum and maximum temperature is the growing degree day value (GDD), which corresponds to the sum of heat units during the growing period. GDD is more important for long life crops grown in marginal regions where the growing season is shorter than the life cycle of the plant. For example, in some areas of northern Greece such as Macedonia, GDD (temperature above 10° C) is often less than 2,200 (which is considered the minimum for successful cotton growing), causing considerable reduction in yield and quality. Early crop sowing is necessary to overcome these detrimental effects (Chlichlias *et al.*, 1977), but for seed propagation, these areas should be avoided.

Night temperatures below 20° C significantly reduce oil and protein content as well as germination ability in cotton seed (Gipson and Joham, 1969). Seed that matures at 11–15° C has a slower germination rate than that which matures at 21–27° C. Plants grown from seed developed under low night temperatures show a slow growth rate and lower yields (Quinsenberry and Gipson, 1974).

Stress from Rainfall and Water Supply

Ideally, a seed crop requires dry weather, especially during the reproductive phase, along with sufficient water supply.

Adequate soil moisture is necessary for seed germination and seedling emergence. For germination, most crop seed should absorb water up to 26–75 percent of its dry weight. Oil seed needs more water in order to germinate, e.g. cotton seed must absorb water up to twice its weight or more. Dry soil may delay germination for days or weeks, leading to poor stand and undesired crop lateness. On the other hand, high soil moisture excludes oxygen and consequently prevents germination, resulting in seed rotting. Cold weather increases the stress from excessive soil moisture. Saturated soil does not permit germination, as there is no oxygen for seed respiration. Extreme soil moisture also reduces or prevents the function of useful soil micro-organisms, such as those involved in nitrogen fixation, due to decreased respiration. Ample rainfall or other water supply is generally favorable during the vegetative phase.

Excessive drought and soil moisture cause similar effects, as is the case with high and sub-optimal temperatures.

Flowering, pollination and seed setting are favored by a moderate atmospheric relative humidity in combination with sufficient soil moisture. Rainfall and sprinkler irrigation are not desirable during these stages for cross-pollinating plants, especially those in which pollen is transferred by insects, such as sunflower and clover. For insect-pollinated flowers, especially those with a short flowering period, bad weather may lead to complete failure of fruit and seed production. Very dry weather and high temperatures prevent receptivity of stigma and pollen germination. For most crops flowering is the most sensitive (critical) period to drought stress. Water deficit at pollination decreases kernel set in maize, as seed development fails after fertilization. With severe water deficiency, complete kernel loss may be observed (Schussler and Westgate, 1991).

Drought stress during the reproductive phase can cause reduction of seed yield and quality. In oil seed, water deficiency during seed development affects subsequent seedling growth, and can pose problems in the succeeding crop establishment. Drought stress often occurs in soybean production areas during the critical period of seed formation and filling, causing reduction in yield and seed quality. Variable seed quality in soybean can be attributed to the time of drought stress and pod position. Drought stress in soybean has no effect on seed germination or vigor, unless the stress is severe enough to produce shriveled, flat, or undeveloped seed (Smiciclas *et al.*, 1992).

Dry atmospheric conditions are necessary for late seed ripening and harvest. Regions with early rainfall during harvest should be avoided for seed production. Wet conditions are likely to cause seed rotting, seed shedding, fungi proliferation, and harvesting problems. If seed moisture is too high, artificial drying becomes necessary after harvest. Safflower seed deteriorates and becomes black in color with wet weather after flowering.

Wet weather may cause pre-harvest sprouting in wheat, especially in white seedcoat spring wheat, which, contrary to red winter wheat, has traditionally been associated with pre-harvest sprouting (McCaing and Delauw, 1992). Wet weather or excessive water late in the season may cause plant re-growth, in competition with the useful reproductive organs on the plant.

Wind Stress

Strong wind causes mechanical and physiological damage. During the reproductive phase it can cause severe crop loss through lodging, shattering and shedding of seed. In order to avoid losses of seed cotton due to strong wind after boll opening, storm-proof varieties have been released, in which the seed is firmly attached to the capsules. Wind and high temperatures increase evapotranspiration and may bring about early maturity leading to severe yield loss, especially in cereals.

Wind does, however, have some favorable effects. Light wind is necessary for cross-pollinating plants such as sugar beet. It accelerates defoliation as well as the opening of late cotton bolls, and decreases the moisture content of seed before harvest. It reduces excessive humidity inside the plant canopy, protecting the plant from disease.

Stress from Soil Abiotic Factors

Acceptable soil conditions for seed production are generally easy to maintain or modify.

The soil of seed fields should be of the highest yield capacity for seed production. Each crop's specific requirements for soil fertility, water capacity, drainage, alkalinity, acidity, salinity, etc., should be considered when choosing the proper field for seed production.

Heavy soil is usually slow to warm up. This can delay emergence, plant growth and seed maturity. The soil should facilitate unimpeded root establishment and penetration.

Very fertile soil may encourage excessive vegetative growth, as in forage crops. Thus, medium fertility soil is preferable for seed production.

Many crops such as cotton, and most plants originating from dry regions, are tolerant to salinity. Some, such as barley and safflower, are more tolerant to alkaline soil, and others, such as *Lupinus*, to acidity. High soil salinity reduces seed yield as well as oil and protein content, even with relatively tolerant plants such as safflower. Soil acidity and alkalinity also interfere with mineral absorption by plants, and therefore may lead to nutrient deficiency.

Soil should be able to provide adequate minerals to plants, otherwise it should be supplemented by fertilizers. The amount and time of fertilizer application depend on individual crop requirements. Maize absorbs more than 50 percent of its nitrogen after flowering, and if this demand is not satisfied plant growth and development are retarded. For seed production, high nitrogen fertilization should be avoided, because it leads to excessive vegetative growth, lodging and late crop maturity. For oil seed, high nitrogen fertilization increases seed protein content but decreases oil content, which is usually correlated with seed germination. Phosphate-deficient seed generally produces poor seedlings and its use should be avoided (see "General Agronomic Aspects of Seed Production"). In order to reduce production costs and environmental pollution, modern agriculture tends to use varieties with minimum nitrogen fertilization requirements.

Some soils are deficient in certain minor elements that are of particular importance to normal seed development. Accordingly, they should be supplemented through the soil or by foliar application. In pea, for example, there is a seed defect called "hollow heart" which is due to boron deficiency (Thomson, 1979).

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Diseases in Vegetables during Seed Production and Conservation

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Introduction

People all over the world are dependent on crop products for their existence. Crops are considered healthy when they are capable of carrying out their physiological functions to the best of their genetic potential, resulting in fruit and seed production. Any biotic or abiotic disturbance that interferes with these physiological functions and affects yield and seed production is called a disease.

Almost all cultivated vegetable crops in Jordan, except for a few species, are subject to attack by disease and pests, resulting in slight yield losses, severe losses, and sometimes, when an epiphytic disease occurs, the loss of the whole crop. Roughly 16 percent of the world's crop production is lost every year due to plant disease. This means that an estimated 550 million tons of crop products are lost due to disease each year, valued at 4 billion US dollars.

Importance of Vegetable Diseases

Vegetable diseases are important to humans because they damage plants and plant products, especially fruit and seed. Disease causes reduction in the quality of vegetable products and reduces their marketability. Disease is one of the main factors causing economic loss to farmers, resulting in increased prices. The kind of plants grown in a geographical area may be limited because particular plant species are susceptible to a particular disease. Vegetable diseases may also play a role in determining the kinds of agricultural industries, or the level of employment in an area, by affecting the kind of produce available for canning and processing.

The damage to vegetable products caused by pathogens arising from diseased seed varies. Such damages are:

- Failure of germination.
- Damping-off.

- Root rot.
- Foot rot.
- Seedling blight.
- Stem rot.
- Flower deformations.
- Fruit rot.
- Wilt.
- Infection of green parts, which affects fruit and seed production.

Specific Vegetable Diseases in Jordan

The number of diseases affecting vegetable crops is very large. Almost all vegetables, except a few species, are subject to one or more diseases. These diseases are caused by a variety of pathogens such as viruses, bacteria, mycoplasma, Rickettsia, fungi, nematode and parasitic plants. The majority of plant diseases are caused by fungi, so we will deal with them, concentrating those that are economically important.

The most devastating vegetable diseases are seed-borne pathogens. More than 1,000 diseases are known to occur in about 300 crops species grown in the world. The economically important diseases in Jordan are summarized in Table 1

Many diseases start in the field while plants and fruit are still developing, or affect fruit and seed during maturation before harvest. Such infections continue to develop after harvest in storage with other pathogens.

Table 1. Fungal seed-borne diseases of vegetable crops in Jordan.

Group	Disease	Causal Agent
Solanaceae plants	Early blight	<i>Alternaria solani</i>
	Leaf mold	<i>Cladosporium herbaru m</i>
	Stem canker	<i>Didymella lycopersici</i>
	Wilt	<i>Fusarium oxysporium</i>
	Anthracnose	<i>Glomerella cingulata</i>
	Late blight	<i>Phytophthora infestans</i>
	Damping off	<i>Pythium aphanidermatum</i>
	Seedling blight	<i>Rizoctonia solani</i>
	Leaf spot	<i>Septoria lycopersici</i>
	Wilt	<i>Verticillium dahliae</i>
	Black scurf	<i>Rizoctonia solani</i>
	Dry rot	<i>Fusarium solani</i>
	Leaf spot	<i>Cercospora capsici</i>
	Anthracnose	<i>Colletotrichum capsici</i>
Pepper blight	<i>Phytophthora capsici</i>	
Crucifer plants	Leaf spot	<i>Alternaria brassicae</i>
	Leaf spot	<i>Alternaria raphani</i>
	Black ring spot	<i>Mycosphaerella brassicolae</i>
	Downy mildew	<i>Peronospora parasitica</i>
	Black leg	<i>Phoma lingam</i>
	Club root	<i>Plasmodiophora brassicae</i>
	Stem rot	<i>Sclerotinia sclerotiorum</i>
Legume plants	Leaf spot	<i>Alternaria phaseolorum</i>
	Anthracnose	<i>Colletotrichum lindemuthianum</i>
	Wilt	<i>Fusarium oxysporium f.sp. phaseoli</i>
	Root rot	<i>Fusarium solani f.sp. phaseoli</i>
	Charcoal rot	<i>Macrophomina phaseoli</i>
	Seedling blight	<i>Rhizoctonia solani</i>
	Stem rot	<i>Sclerotinia sclerotiorum</i>
	Ascochyta blight	<i>Ascochyta fabae, A. rabeih</i>
Cucurbitaceae plants	Anthracnose	<i>Colletotrichum laginarium</i>
	Wilt	<i>Fusarium oxysporium f.sp. cucumerinum</i>
Other vegetable plants	Lettuce downy mildew	<i>Bremisa lactucae</i>
	Lettuce leaf spot	<i>Septoria lactucae</i>
	Onion purple blotch	<i>Alternaria porri</i>
	Onion neck rot	<i>Botrytis allii</i>
	Onion downy mildew	<i>Peronospora destructor</i>
	Carrot leaf blight	<i>Alternaria dauci</i>
	Carrot white rot	<i>Sclerotinia sclerotiotum</i>
	Beet leaf spot	<i>Cercospora beticola</i>

Seed Health Testing

There are different methods for seed health testing (Neergaard, 1977; Diekmann, 1992). Special tests such as indicator plant test serology and the phage-plague test detect a particular pathogenic species, and are mostly used for viruses, and to a lesser extent for bacteria and fungi. General tests

such as the blotter test and the agar plate test reveal a wide range of fungal and bacterial pathogens. In both cases a sufficient sample is required to ensure reliable results. Some of the methods which are used widely in seed health laboratories are:

- Direct inspection.
- Seed washing test.
- Blotter method.
- Seed extraction.
- Embryo test method.
- Agar plate method.
- Growing-on test.
- Indicator test.
- Serological test.
- Phage-plaque method.

Seed health status can be correctly evaluated only by a thorough knowledge of the biology of the pathogen, and of the methods used for their detection and identification. Therefore, coordinated research projects on seed-borne vegetable diseases must be established to conduct research on the various aspects of healthy seed production.

Significance of Seed Health Schemes

The main objectives of producing healthy seed are:

- To reduce crop yield losses.
- To avoid development of disease epiphytotics.
- To prevent the spread of pathogens into areas where they do not occur.
- To meet standards set in certification schemes.
- To prevent discoloration and shriveling of seed.
- To prevent losses in germination.
- To prevent biochemical changes in seed.
- To prevent toxin production.

How We Produce Healthy Seed

There are a number of methods that can be followed to ensure a reliable production of healthy seed:

- Selection of areas with low disease occurrence.
- Field inspection and rouging.
- Fungicide spraying on infected plants.
- Laboratory seed health testing.
- Seed treatment by fumigation, heat, and seed dressing.
- Production of resistant varieties.
- Use of biological control measures/integrated pest management.
- Crop management and sanitation.
- Seed certification.
- Quarantine measures.

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Seed-borne Diseases in Seed Production

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Introduction

Most crops are subject to attack by a variety of pests, i.e. viruses, bacteria, fungi, nematodes, insects, weeds, etc. Hereafter, the term *pest* will be used when referring to any of the organisms mentioned above, including viruses. The term *pathogen* is used for anything able to cause a disease, i.e. viruses, bacteria, fungi, and to some extent nematodes. The vast majority of plant diseases are caused by fungal pathogens. In most cases crops tolerate the attacks to a certain extent, or react with slight yield losses only. Sometimes, however, attacks result in severe yield loss and even wipe out a crop.

In general, plant diseases are recognized by certain symptoms. Characteristic disease symptoms include: (i) yellowing; (ii) necrosis; (iii) stunting; (iv) wilt; and (v) root rot. Similar symptoms, however, may also be caused by other factors than pathogens, for example, mineral deficiency is often expressed by yellowing; pollution (acid rain) may cause necrosis in many plants; and lack of water results in wilting of the plants.

In some cases, the structure of the pathogen itself is a clear sign of disease, e.g. teliospores of rust, conidia of mildew, etc. Any part of the plant is subject to disease, which may occur at any stage: seed, seedling, growing plant, etc. The infection may be systemic, that is, invading the whole plant, or restricted to the attacked parts of the plant. There are various ways of pathogen dissemination: (i) wind (mildew, rust); (ii) water (nematodes); (iii) plants (viruses through *Cuscuta*); (iv) insects (viruses); (v) man or man-made tools (nematodes); (vi) seed (bunt, smut); and (vii) cuttings (viruses).

It is generally accepted that a disseminated pathogen does not necessarily cause a disease. If that is the case, one should use the term *transmitted*. For example, spores of barley powdery mildew (*Erysiphe graminis*) are easily disseminated by wind. Transmission of this pathogen occurs, however, only if the spores meet with a susceptible barley plant, and if the conditions are favorable for infection. The same holds true for pathogens that are disseminated by seed. For example, if wheat seed is contaminated with *Tilletia contraversa* (i.e. spores of this pathogen are carried on the seed surface), this pathogen will be disseminated with the seed wherever it is planted; it is a

seed-borne pathogen. We would use the term transmission only if an infection actually takes place. If this seed is planted in Egypt, most likely the pathogen will not be transmitted, because the environmental conditions are not favorable for the germination of the spores. For some pathogens, transmission by seed is the most important, or even the only means of transmission. An example of an exclusively seed-transmitted pathogen is *Ustilago nuda*. *Tilletia* spp. are potentially seed-transmitted, the other possibility being through soil.

We must distinguish between seed pests and diseases, where the seed is the victim (e.g. storage pathogens like *Aspergillus flavus*) and storage pests like grain weevils, where seed is the vehicle of pest and pathogen dissemination (e.g. plant diseases like bunt, smut, etc.). For some diseases the seed is both victim and vehicle for dissemination, e.g. *Ascochyta rabiei*, where infected seed has lower germinability. Usually, adequate precautions are taken for storage pests and pathogens, because they may quickly destroy a seed lot, and because they are easily detected. Optimum storage conditions help in suppressing this group. In this paper we deal only with seed-borne diseases in the narrow sense.

Seed-borne disease can be viewed from several vantage points:

- The farmer, unless he is reusing his own seed, is concerned only if yield loss results from seed-borne disease.
- The producer of certified seed has to expect rejection of a seed lot if a certain level of infection or contamination is exceeded.
- In the international transfer of seed, a complete zero-tolerance is required for specific pathogens, because they might not occur in all countries (quarantine).

It is difficult to obtain reliable data on the economic importance of seed-borne disease. In some crops (cereals, soybean) about half of the losses due to disease are attributed to seed-borne pathogens. We have to distinguish two types of seed infection:

- The pathogen is present on or in the seed, and will only attack the seedlings derived from this seed. This reflects a relatively simple relationship between the percentage of infected seed, the disease incidence in the field, and crop loss (for example, loose smut, *Ustilago nuda*).
- The pathogen is present on or in the seed, but may also infect the plant by other means during vegetation. In this case the relationship between seed infection and crop loss is much more complicated, since it depends largely on conditions during vegetation (for example, *Ascochyta* blight *Ascochyta* spp).

The number of seed-borne diseases is very large. In the literature, some 500 different plant species are reported to be hosts for at least one of the more than 1,300 different pathogens that are potentially seed-transmitted. There are probably only a few species that are not hosts for one or more seed-transmitted pathogens. Cereals and legumes are among the crops with a large incidence of seed-borne disease.

Examples of Important Seed-borne Diseases

Wheat

The most important seed-transmitted pathogens are bunt and smut fungi, *Fusarium*, *Septoria*, *Drechslera* or "*Helminthosporium*," and *Anguina*. Of the bunt fungi, in northern Africa only *Tilletia foetida* and *T. caries* are prevalent. *T. contraversa* requires low temperatures for infection, it occurs mainly in areas that are covered by snow in winter. *T. indica* has a different mode of infection, it requires free water during flowering. Because of this specific requirement it is restricted to India, Pakistan, Afghanistan and parts of Mexico.

Loose smut in wheat is caused by *Ustilago tritici*, and can result in significant yield losses, generally if seed treatment is not exercised. In some areas flag smut (*Urocystis tritici*) is prevalent.

A wide range of *Fusarium* species is pathogenic on wheat. Most if not all are potentially seed-transmitted. Which species prevails depend largely on climatic conditions. In large parts of Europe, *Fusarium nivale* causes considerable yield losses. In northern Africa, *F. graminearum* (scab) and *F. culmorum* (foot and root rot) are probably predominant. Potentially important "*Helminthosporium*" species are *H. sativum*, which cause spot blotch, and *H. tritici-repentis*, which cause tan spot.

The wheat nematode *Anguina tritici*, which causes "ear cockle," is widespread in most wheat growing areas, but rarely causes major yield losses. The bacterial stripe disease, or black chaff disease, caused by *Xanthomonas campestris* pv. *translucens*, may be serious, particularly under sprinkler irrigation.

Barley

Important seed-transmitted pathogens of barley are *Ustilago*, *Drechslera* or "*Helminthosporium*," *Fusarium*, *Rhynchosporium*, and barley stripe mosaic virus. Loose smut of barley (*Ustilago nuda*) is similar to loose smut of

wheat. Different symptoms and a different mode of infection are found with *Ustilago hordei*. Relatively high yield losses are caused by "Helminthosporium," namely *H. gramineum* (stripe disease), *H. teres* (net blotch) and *H. sativum* (seedling blight, foot and root rot).

Fusarium spp. are largely the same as in wheat, causing the same diseases. *Rhynchosporium secalis* causes characteristic scalding. This pathogen is seed-transmitted to a relatively small extent.

Barley stripe mosaic virus is probably the only seed-transmitted cereal virus. Compared to other virus diseases of barley, such as barley yellow dwarf, it is less important. However, since it does not occur in all barley growing areas, it is of quarantine significance.

The bacterial stripe disease, or black chaff disease, caused by *Xanthomonas campestris* pv. *translucens*, occurs on barley only when grown in high moisture areas.

Rice

The three major diseases of rice, namely blast (*Pyricularia oryzae*), brown spot (*Drechslera oryzae*), and bacterial blight (*Xanthomonas campestris* pv. *oryzae*) are seed-borne. Losses vary according to climatic condition, and may reach almost 100 percent, as was the case in 1942/43 in Bengal, India when the crop was destroyed by brown spot disease (Great Bengal Famine). Other important seed-borne fungi are *Trichoconiella padwickii* and *Fusarium moniliforme*. Also, two seed-borne nematodes may be extremely harmful to rice: *Aphelenchoides oryzae*, the white tip nematode, and *Ditylenchus angustus*, the stem nematode.

Lentil

Among legumes, lentil has the fewest diseases. Important seed-transmitted pathogens are *Ascochyta lentis*, *Fusarium* spp., and some viruses. Yield losses due to these pathogens are usually less than in other legumes.

Chickpea

The main diseases of chickpea are *Ascochyta* blight, *Fusarium* wilt and root rot. They are potentially seed-transmitted, but are also transmitted by plant debris and other means. Under certain conditions, nematodes (root-knot nematodes, *Meloidogyne* spp. and cyst nematodes, *Heterodera* spp.) could also be of importance. They might be seed-transmitted in the sense that soil, which could be infested with nematodes, is frequently mixed with seed.

Faba Bean

Disease causes many problems in faba bean. This is probably due to the fact that it requires growth conditions that are favorable to pathogen development (warm temperatures, high rainfall or irrigation). Most of the pathogens, including a number of viruses, are transmitted by seed. Among these are the already mentioned *Ascochyta* spp. and *Fusarium* spp., as well as *Botrytis fabae* and *B. cinerea*, bean yellow mosaic virus, broad bean stain virus, true broad bean mosaic virus, pea seed-borne mosaic virus, *Ditylenchus dipsaci*, and others.

The parasitic weed *Orobanche crenata* causes serious yield losses in faba bean production in northern Africa. Its tiny seeds may get mixed with faba bean seed and can thus reach into uninfected areas.

Pea

Again we have the *Ascochyta* complex, and the various *Fusarium* species. In environments with high moisture, bacterial blight caused by *Pseudomonas pisi* can be devastating.

Production of Healthy Seed

Healthy seed, i.e. seed that is free from pests, is a prerequisite for a high-yielding crop. Seed health is a component of quality seed, as are viability, vigor and purity. Whereas in seed production schemes all efforts are usually made to supply the farmers with pure seed of high germination capacity, little emphasis is put on the health aspect in its narrowest sense. There are different ways to achieve healthy seed: (i) seed production in pest-free areas; (ii) seed production under effective pest control; (iii) field inspection schemes; (iv) seed treatment; and (v) seed health testing.

Normally a combination of these methods will be used, since it is not feasible to rely on only one method. In many cases it will not be possible to select areas that are completely free from pests. In some cases, however, problems can be avoided by preferring one site over another. Production of seed potato is recommended in windy areas, where important virus vectors, namely aphids, do not play a significant role. During multiplication, effective control can be exercised against a number of pests, such as *Ascochyta* spp. (blight), *Ditylenchus dipsaci* (stem nematode), *Bruchus* spp. (bruchid weevils) and others. For viruses, there is only the possibility of vector control, mostly with insecticides. Seed-transmitted pathogens with only one generation per year, such as *Tilletia caries* and *T. foetida* (bunt) and *Ustilago* spp. (smut), cannot be controlled in the field.

Seed treatment is suitable to control a number of fungal diseases, and to some extent insects, nematodes and bacteria. Care must be taken to choose a dosage sufficient to eradicate the pathogens, but not kill the seed.

Field inspection and laboratory seed health testing are important components of seed certification schemes.

Seed Certification

Seed certification schemes usually focus on various aspects of seed quality to be tested or inspected. These include: trueness-to-variety, physical purity, content of other seed, germinability, health, and moisture. Some of these can be checked only or mostly in the field (such as trueness-to-variety), others require laboratory testing (germinability, moisture). Assessment of the health status of seed necessitates both field inspection and laboratory testing.

Certain diseases, such as loose smut of wheat and barley, can be readily detected in the field, but detection in laboratory tests is difficult or time-consuming. Other diseases are difficult to detect in the field, unless the inspector is well-trained for detection of these diseases, or they appear at a high incidence or severity. Kernel bunt (*Tilletia indica*), for example, is hard to recognize in the field, but a centrifuge wash test reveals spore contamination even at low levels. Viruses usually produce distinct symptoms in the field. However, they may also be latent, that is, infected plants may not show symptoms, but may produce infected seeds. Laboratory testing for viruses requires antisera and special equipment for serological tests.

To allow an objective evaluation of seed quality, tolerances have to be established. This will allow rejection of a field or a seed lot that does not meet the prescribed standard. Tolerances are not universal; they have to be set according to the situation in a particular area, country, or region where the seed will be planted. A seed certification scheme just starting in a country will probably set lower standards than one used by a well-established organization. Standards for seed-borne pathogens are even more variable than those for other quality aspects.

Field Inspection

Field inspectors must know which diseases may be seed-transmitted, and to what extent. In wheat, for example, a field may be severely affected by one or more of the rust diseases. With yellow rust, even glumes and seeds can be covered with rust pustules. Yet the pathogen is not transmitted by seed, and is therefore not of primary concern in seed certification, although it

may considerably reduce yield. This will affect producers of breeder seed and foundation seed, who would like to supply a particular variety in larger quantities, and of course the seed grower, because his return will be less than expected due to the yield loss. Also the percentage of shriveled seed will increase.

A general field inspection scheme is shown in Figure 1. For identification of the pest or pathogen, pictorial keys such as those listed in the references can be used. When in doubt, the help of an experienced plant pathologist/entomologist/plant protection specialist should be sought.

1	Identify the pest/pathogen, then:	2
2	The pest/pathogen is not known to be seed-transmitted	3
2*	The pest/pathogen is known to be seed-transmitted: take records and compare with standards, test seed after harvest in laboratory, in addition:	5
3	The pest/pathogen is not likely to affect yield or quality: do not do anything and hope for the best	
3*	The pest/pathogen is likely to affect yield or quality:	4
4	The causal agent is a fungus, insect, or nematode: select an appropriate pesticide and advise application at recommended dosage	
4*	The causal agent is a virus or bacterium: advise grower to rogue infected plants in order to prevent further spread (if feasible) and to spray insecticide if potential vectors are there	
5	The causal agent is a virus:	6
5*	The causal agent is not a virus:	7
6	The virus is not known to be insect transmitted: advise grower to rogue and burn infected plants (if feasible), test seeds after harvest	
6*	The virus is not known to be insect transmitted: advise grower to rogue and burn infected plants (if feasible), and to spray insecticide to prevent further spread, test seed after harvest	
7	The pathogen has only one generation per year, e.g. smut, bunt: advise grower to rogue infected plants (if feasible), test seed after harvest, treat infected seed lots	
7*	The pathogen has several generations per year, e.g. <i>Ascochyta</i> blight: advise application of appropriate pesticides at early stages of disease development, test seed after harvest, treat infected seed lots.	

Figure 1. Key for decisions concerning pests and diseases in seed increases (for field inspectors).

Sampling of inspection areas is done the same way as is field inspection for purity. The timing of the inspection is crucial; some diseases show only at certain stages, e.g. common bunt after development of ears. In some cases, neighboring fields must also be inspected (for example, when exporting to countries requesting additional declaration on the phytosanitary certificate stating that a particular “disease does not occur at the place of production

or in its immediate vicinity"). Field inspectors should be familiar with the pests and diseases endemic in their region. Cooperation with plant pathologists, entomologists and plant protection specialists should be sought. A short meeting with specialists from extension services, universities and other research institutions each year before field inspection starts will help to familiarize field inspectors with current problems, and put them in a position to advise the farmer. Field guides with pictorial keys should be available, as well as herbaria specimens of diseased plants. For the final identification, isolation of pathogens may be necessary, and for this the cooperation of a specialized laboratory is normally required.

Table 1. Examples of field standards for seed-borne disease.

Disease	Tolerance	Country
Loose smut (wheat and barley)	1 in 10,000 ears	England
Loose smut (wheat)	Found. seed: 0.1% of plants Cert. seed: 0.55 of plants	India
Loose smut (wheat and barley), common bunt, barley stripe	Basic seed: 0.02% Cert. seed (1): 0.05% Cert. seed (2): 0.1%	Morocco
Ashy stem blight (cowpea)	Found. seed: 0.1% Cert. seed: 0.2 %	India
Bean blight	0.005% of plants	USA
Pea early browning	1 plant per 100 m ²	Holland
Cowpea mosaic	Found. seed: 0.2% Cert. seed: 0.2%	India
Soybean mosaic	20 plants per line tested by ELISA	USA
Barley stripe mosaic	Zero in seed production fields	USA
Pea seed-borne mosaic	Zero in seed production fields	USA (WA and ID)

Examples of field standards from various countries are given in Table 1. However, this is not meant as a guideline, because each country has to set the required standards individually, considering the following:

- **Seed generation:** The strictest standards are required for early generations, i.e. breeder and foundation seed.
- **Available methods of control:** Control is either by seed treatment or another way. For a pathogen that can be easily controlled with a fungicide seed treatment (common bunt), the standard may be much higher than for one with no control method available (lettuce mosaic virus).
- **Epidemiology and pathogenic potential:** Countries in which environmental conditions are conducive to the development of a particular disease, e.g. bacterial leaf streak of wheat or barley, may set stricter standards for the pathogen, in this case *Xanthomonas campestris* pv. *translucens*. Generally, for pathogens with only one generation per year (monocyclic pathogens, for example *Tilletia caries* or *Ustilago nuda*) the

epidemic potential is less than for polycyclic pathogens with several generations per year (such as *Xanthomonas campestris* or *Ascochyta rabiei*). In this context, the seeding rate may also be important. A 1 percent infection in a crop for which 25,000 seeds per hectare are planted brings more initial inoculum to a field than the same percentage in a crop where 5,000 seeds per hectare are planted.

- **Other means of transmission:** If a pathogen is mostly transmitted by means other than seed, e.g. *Alternaria brassicae* in cabbage, the seed-borne inoculum is not important to the development of epidemics, and strict tolerances will not reduce the incidence (Agarwal, 1983). The situation is different with exclusively or almost exclusively seed-transmitted pathogens, such as barley stripe mosaic virus (Carroll, 1983).
- **Relation between seed inoculum and field infection:** This relates, for example, to the number of spores, bacteria, etc. per seed required to cause an infection. Whereas in ideal conditions (for the pathogen) as little as one spore can infect a plant, normally more spores are required, for example, approximately 3,000 per seed for common bunt. This is a matter of probability and varies according to varietal resistance, environmental conditions, virulence of the pathogen, and other factors.
- **Quarantine:** If seed is produced for export, the phytosanitary import regulations of the country in question have to be observed.

Laboratory Seed Health Testing

The main purpose of laboratory testing in seed production and seed certification is the evaluation of seed quality. The different aspects of seed quality that can be evaluated in the laboratory are: physical purity, germination, moisture, health, and to some extent varietal purity and vigor. The test results of all the components together give an indication of the planting value. A seed lot free from pathogens and meeting high purity standards, but with poor germination, is of low planting value, as is a lot with high purity and good germination but infected with seed-borne pathogens. The actual stand in the field also depends on a number of other factors, e.g. soil conditions, weather, planting techniques, etc. Standards for pathogens are even more variable from one country to another than those for germination or purity. It is of utmost importance that the correct method for the respective pathogens is used. If a tolerance for *Ustilago nuda* is set, there is only one possible test method, the embryo test. For *Tilletia* spp., a visual inspection can reveal bunted seed. However, the lack of bunted seed does not mean the seed sample is free from *Tilletia* spp. The presence of a spore contamination can be revealed only in the washing test.

As with field standards, laboratory standards have to be set by each seed program according to local conditions. Examples are given in Table 2. However, copying other countries' standards without considering possible differences could lead to unnecessarily strict standards, resulting in too many rejections without benefit to the national seed program or the farmers. It may also result in a situation where more stringent standards could prevent severe yield losses. Furthermore, it is important to periodically review standards.

Table 2. Examples of laboratory standards for seed-borne diseases.

Pathogen	Tolerance	Country
<i>Tilletia caries</i>	500 spores per g seed	Sweden
<i>Tilletia contraversa</i>	100 spores per g seed	Sweden
<i>Tilletia indica</i>	0.1% in found. seed	India
<i>Neovossia horrida</i> (rice)	0.5% in cert. seed 0.1% in found. seed	India
<i>Ustilago nuda</i> (wheat, barley)	1.5% in cert. seed 0.2% in basic seed	UK
<i>Ustilago nuda</i> (wheat)	0.5% in cert. seed 0.5% without treatment 2% with treatment	India
<i>Botrytis</i> spp. (linseed, sunflower)	5%	France
<i>Claviceps purpurea</i> (rye)	1 pc. per 500 g (basic seed) 3 pc. per 500 g (cert. seed)	France
<i>Colletotrichum lindemuthianum</i>	0.1% in basic seed 0.5% in cert. seed	France
<i>Phoma lingam</i> (cabbage)	Zero in 1,100 seed	Denmark
<i>Pseudomonas phaseolicola</i>	Zero in 1,000 seed (pre-basic and basic seed)	England
<i>Xanthomonas phaseoli</i>	Zero in 5 kg seed	Canada
Lettuce mosaic virus	Zero in 30,000 seedlings	USA (CA)
Pea seed-borne mosaic virus	Zero in 200 seeds/seed lot (ELISA)	USA (ID, WA)
Barley stripe mosaic virus	Zero in foundation seed (latex flocculation method)	USA (ND)
Barley stripe mosaic virus	5% (1964) 3% (1968, 200 embryos) 0% (1972, SDS disk test, USA, MT)	

Storage Pests and Their Control

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There is a wide range of storage pests. In general, any organism that reduces the quantity and/or quality of stored grain or other food is a storage pest. This includes rodents, birds, insects, mites, fungi and bacteria.

Economic Importance of Storage Pests

The losses due to storage pests, sometimes referred to as post-harvest losses, vary according to climatic condition, crop, and storage facility. Quantitative losses are estimated as high as 30 percent worldwide. Qualitative losses, such as toxins produced by fungi, losses in seed viability, etc. are more difficult to determine or even to estimate. In absolute figures the loss in food or food stuffs is much higher than in seed, but the consequence of loss in stored seed can be more drastic. If seed quantity is reduced considerably, the country concerned might have to import seed and become dependent on the world market. The varieties available might not be suitable for growing under the country's specific conditions. Varieties unfit for local conditions inevitably result in poor harvests. For these reasons the prevention of storage losses in stored seed deserves special attention.

Rodents and their Control

Rodents, mainly rats and mice, can be very destructive, not only to seed, but also to buildings, wiring (which may result in fire), etc. They can consume substantial amounts of seed, and may also damage bags, boxes and other containers. Different species of rat may differ in their susceptibility to control measures.

A very effective, though expensive way of control is to keep rodents away from stored products through rodent-proof stores. However, due to the high cost of concrete buildings and silos, many seed stocks are still easily accessible to rodents.

Traps are cheap and quite efficient if the population to be controlled is small. Biological control by predators (cats, hawks, etc.) or pathogens (causing rat diseases) depends on too many factors to be reliable.

Still the most widely used method of rodent control is poisoning. A wide range of rodenticides is available from different companies. Those that are extremely toxic to human beings have been replaced by less hazardous ones (anticoagulants). It is advisable to put the bait only in special bait-boxes, that can be checked daily. The boxes should measure 30 x 40 cm, with a height of 20 cm, with two 8 x 8 cm openings opposite each other. The boxes should be placed along the walls of the store, and remain in place for several weeks. Initially, bait should be offered without poison to lure the rats to the feeding place. Poisoned bait has to be refilled regularly. Examples for rodenticides are Klerat (ICI) or Racumin (Bayer). Poisoning of rodents can be effective in the long run only if re-infestation is prevented, e.g. by rat-proof buildings, or by rodent control campaigns in larger areas.

Insects and Mites and their Control

Insects can be very destructive to seed. Mainly those species of storage pests that develop inside the grain are important. Others, such as flour beetle (*Tribolium* spp.), mealworm, etc. are more important in stored food grain. Mites play a role mainly by transmitting the spores of storage fungi, and by causing skin irritation and allergies in persons handling infested seed.

Different species feed on cereals and legumes, and their different life cycles affect the appropriate control measures. In legumes, particularly faba bean, *Bruchus* spp. infest the seed in the field, and the insects complete their life cycle in the store. There is only one generation per year, and no re-infestation in the store. However, *Callosobruchus* spp., and *Bruchidius* spp. are typical storage pests. Eggs are laid on the dry seed, and several generations develop in the store through re-infestation.

In cereals, only the true storage pest occurs. There are more than 20 different species, most of them belonging to the *Coleoptera* (weevil, beetle, grain borer), and some to the *Lepidoptera* (grain moth, mealworm). Most frequently encountered are the lesser grain borer (*Rhizopertha dominica*) and the granary weevil (*Sitophilus granarius*). Some storage insects, such as the Khapra beetle (*Trogoderma granarium*), are considered quarantine pests in some countries.

In general, development of insects is faster at higher temperatures. About 35° C is considered the maximum for development and about 38° C the maximum for survival. There are differences between the different species, *Rhizopertha dominica* being an example of a heat tolerant species. Below 0° C most insects cannot survive for more than 2–3 weeks.

Also, a certain moisture level is required for insect development. The minimum for insect development is generally considered 10–11 percent seed humidity. The number of insects increases with increasing moisture up to a level where too many micro-organisms develop (at about 15–16 percent).

When it comes to control measures, the sources of infestation are important to know. As mentioned already, for *Bruchus* spp. infestations the source is in the field. Spraying the fields with insecticides during oviposition can prevent infestation, but is expensive unless other insects have to be controlled at the same time. Care should be taken to select insecticides that are not toxic to bees, since some plants will be flowering.

Visual inspection of seed after harvest can give an indication of the level of infestation. Infestation will be revealed by characteristic dark spots, which are the holes through which the newly hatched larvae penetrate the seed.

If high infestation levels are detected, fumigation might be used. This helps to stop larval feeding and thus retain germinability. Moreover, the source of infestation for the next season, the adults that are carried with the seed to the field, is reduced. For true storage pests, there are many sources of infestation. A very important one is contamination from stored infested seed.

Frequently, stores have hidden focal points—such as crevices, corners, spilled seed outside the store, empty sacks with some leftover seed, etc.—where a stock of insects can survive attempts at control. It is extremely important to control these areas. It seems highly desirable to detect infestation in the early stages, before the insect population increases too much. Careful inspection of the stored material at regular intervals helps.

There are various methods to detect infestations before they become clearly visible. Some of them are simple (flotation of grain), others are very sophisticated (x-ray). However, insect infestation is detected mainly by visual inspection.

Storage insects are generally controlled chemically. Unlike grain stored for food or feed purposes there is no problem with residues hazardous to warm-blooded animals. The effect of chemicals on seed viability must, however, be considered. Nevertheless, a low toxicity to humans is important, in order to protect the workers. A high and long-lasting effectiveness against a wide range of insects is required. Last but not least, the chemical should be inexpensive.

We have to distinguish between insecticides that kill the insect population and have a longer-lasting effect (pyrethroids, organo-phosphorous insecti-

cides) and those which only kill the insects and have no residual effect (fumigants). These types are not true alternatives, but should be used to complement one another and in combination with storage sanitation.

Protective Insecticides

There is a wide range of products on the market. Table 1 gives some examples, but is not exhaustive. Insecticides may be applied in different ways. Dusting does not require much equipment; a powder is either mixed with the seed, applied in layers (sandwich method), or dusted over stacks. The latter can prevent only re-infestation (e.g. after fumigation), since most powder insecticides do not penetrate the stacks to control internal infestation.

Table 1. Insecticides for control of storage pests in buildings (formulations for spraying, dusting, fogging, etc.).

Active Ingredient	Trade Name	Manufacturer
Bromophos	Nexion	Shell
Cyfluthrin	Baythroid, Solfac	Bayer
Dichlorvos	Nuvan, Nogos, Mafu, Vapona	Ciba Geigy, Bayer, Shell
Fenitrothion	Folithion, Sumithion	Bayer, Sumitomo
Jodfenphos	Nuvanol N	Ciba Geigy
Lindane	Lindagrain	Rhone Poulenc
Malathion	Malathion, Cythion	American Cyanamid
Methoprene (growth Regulator)	Diacon	Gustafson
Phoxim	Baythion	Bayer
Pirimiphos-methyl	Actellic, Actellifog	ICI
Tetrachlorvinphos	Gardona	Shell

For spraying, either a suspension (solid insecticide suspended in water, usually a wettable powder) or a solution (liquid insecticide diluted in water) can be used. They can be applied with knapsack sprayers. For suspensions, care should be taken that the particles remain suspended and do not sink to the bottom of the sprayer. This can be achieved by special stirring devices or by shaking the container frequently. Fogging is a technique used especially in stores. The droplets are much finer than in spraying. Special equipment is required. Evaporation can be used in special cases to control flying insects (moths). This technique requires volatile insecticides, and well-closed stores.

Fumigation

In many countries, fumigation is a routine treatment, carried out mainly against storage insects (grain weevil, bruchid; Table 2). In general, the procedure is to apply a volatile insecticide in a confined area (silo, warehouse, or fumigation chamber). For effective fumigation, air-tight sealing is essential. The main advantage of fumigation is that all insect stages, including eggs, larvae and pupae, are controlled.

Table 2. Fumigants for control of storage pests.

Active Ingredient	Trade Name	Manufacturer
Ethylene dibromide (EDB)	Dowfume	Dow Chemicals
Methyl bromide (MB)	Dowfume MC, Haltox	Dow Chemicals, Degesch
Magnesium phosphide	Magnophos, Magtoxin	Degesch
Aluminum phosphide, phosphine (PH ₃)	Phostoxin	Degesch

Two chemicals are widely used: phosphine and methyl bromide. Others are dichlorvos, carbon dioxide, ethylene oxide, and HCN.

Phosphine: This chemical is available in a solid form (0.6 g pellets, 3 g tablets). The active ingredient is aluminum phosphide, mixed with ammonium carbonate and paraffin (trade name: Phostoxin). After exposure to the atmosphere, the pellets decompose and release the active substance, hydrogen phosphide (PH₃), which has the same specific weight as air, and is thus evenly distributed in the fumigated material or chamber.

Phosphine is able to penetrate bags, cartons, boxes, and other containers. The recommended dosage depends on the temperature. Sometimes an extra day is recommended when tablets are used. Tablets or pellets are placed on cardboard, spaced sufficiently apart to prevent spontaneous ignition. A powdery inert material remains after fumigation. This should be buried. The major advantages of Phostoxin are that it does not accumulate in seed, does not affect flavor or germination, and is easy to handle.

Methyl bromide: Above 5.6° C, methyl bromide is in the gas phase, and is available in cylinders similar to those used for cooking gas. Since it is odorless, other gases such as chloropicrin are sometimes added to facilitate detection of leaks. Because methyl bromide is 3.5 times heavier than air, care has to be taken that it is properly distributed within the goods to be fumigated, for example, by using a fan.

The recommended dosage is 24 g/m³ for 24 h at 10–19° C, and 16 g/m³ above 20° C. Fumigation with methyl bromide is not advised above 25° C. Special safety measures are required, since methyl bromide is absorbed through the skin. It tends to accumulate in commodities, therefore repeated fumigation with methyl bromide should be avoided.

Equipment

Gas-proof plastic sheets with at least 50 cm overlap, firmly pressed to the ground with sand, iron bars, or other weights, are frequently used. Gas escape results in reduced insecticide effect and is a hazard to users. A cement floor is necessary to prevent gas escape through the soil.

Care must be taken that the fumigation area is properly aerated, fans may help. If a store's doors and windows can be hermetically sealed, fumigation of the entire store is possible.

Most stores, however, allow gas to escape through other openings. Silos are usually good fumigation facilities. When large quantities must be fumigated within a short time, a vacuum fumigation chamber is appropriate. These chambers are available in sizes between 1–50 m³, and sometimes as a plant of up to 6 x 50 m³, equipped with fans, pumps, and other equipment. The insecticides used are methyl bromide or ethylene oxide.

Safety

Face masks with proper canisters should be used, especially during the aeration process. When handling Phostoxin, cotton gloves should be worn. Gas concentration can be checked with a halide gas detector for methyl bromide and with a tube detector (Draeger) for Phostoxin. A warning sign should be clearly visible to prevent people from inadvertently removing plastic sheets or entering a building under fumigation.

The use of fans is recommended for even distribution within the fumigated space. Methyl bromide leaves a residue in the fumigated seed, which accumulates when fumigation is repeated. For this reason it is also more hazardous for operators.

Methyl bromide is a gas at temperatures above 6° C, whereas aluminum phosphide comes in solid tablets or pellets, and is thus easier to handle. The concentration to be used depends on exposure time and temperature. At temperatures below 12° C no fumigation should be executed. For methyl bromide, the product of concentration and time required to kill all stages of insects is constant at a given temperature. That means a high concentration

requires a short exposure time, and a low concentration requires a longer exposure time. At 12–19° C the recommended dosage is 24 g/m³ for 24 hr, and above 20° C, 16 g/m³ for 24 hr. Fumigation with aluminum phosphide takes longer; the recommended exposure time is given in Table 3.

Table 3. Exposure time for aluminum phosphide, depending on temperature.

Temperature	12–15° C	16–20° C	21–25° C	above 26° C
Number of days	7	6	5	4

The recommended dosage is 2.5 g phosphine per m³, generated, for example, by 2.5 Phostoxin tablets or 3 g or 12 Phostoxin pellets of 0.6 g (Bond, 1984).

A more recent development is the storage of grain and seed in a controlled atmosphere. This method is well-known from the storage of fruit, particularly apples. The air in stores is replaced by an inert atmosphere that is less than 1 percent oxygen, about 9 percent carbon dioxide, and the balance nitrogen. This inert gas is generated by the combustion of air and propane in special devices. In this environment no storage pest can survive. The seed retains its viability for years, thus this method is far superior to storage in normal air. However, it is quite expensive. In addition to the equipment for changing the atmosphere, it is absolutely necessary to have airtight stores in order to retain the inert atmosphere over the whole storage period.

Special Case: Potato Tuber Moth

This pest attacks potato plants in the field as well as seed potatoes in the store. The damage in the field is minor and does not usually result in severe yield loss. The main problem with this pest is loss of tuber quality for consumption. The insect has several generations per year, depending on the temperature. Stores, with their constantly high temperatures, offer ideal conditions. Control of the pest in the seed store breaks its cycle and prevents planting of infested tubers, and thus helps to control the pest in the field. For effective pest control in the store a combination of different measures is required:

- Only uninfected tubers are stored.
- Fumigation is carried out if there is infestation.
- Doors and windows are protected with insect-proof mesh to prevent moths from invading the store.
- Reusable containers (bags, etc.) are treated with malathion.
- Traps (pheromone traps, light traps) are installed for monitoring infestation; to some extent they will reduce the population.

- As with the control of other storage pests, cleanliness of the stores and their surroundings is very important.

Fungi and Bacteria and Their Control

Here we deal only with those fungi that infect seed in the store, and not the seed-transmitted fungi, which cause plant disease in the field and are carried on or in seed. Bacteria normally do not affect stored seed, unless the moisture content is very high and the temperature has already been raised by fungal infection. Fungi require a high moisture content to grow (minimum 14 percent seed moisture) so they do not play a very important role in dry climates.

The most important genera are *Aspergillus* and *Penicillium*. These are mostly saprophytes, which means they are unable to attack living tissue. They grow on dead cells of the seed surface, where they produce toxins. They cause seed decay and may kill the embryo. Heavily infected seed should not be used for feed because of the toxins many of these fungi produce.

The best way to control storage fungi is to maintain low moisture in seed and store. Low temperatures slow down the growth of fungi. Fungicide seed treatment does not often give the expected results because of the lack of free water, which is required for many fungicides to become effective. The use of propionic acid to control storage fungi is restricted to grain that will be eaten. In summarizing the control of storage pests, it must be stressed that the best method of control is solid buildings and a rigid storage hygiene.

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Section V
Seed Processing and Quality Control

Seed Cleaning

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Introduction

In many cases, harvested seed has to be cleaned, i.e. undesirable materials such as weed seed, other crop seed, diseased, damaged and deteriorated seed, as well as inert material, must be removed. Seed cleaning can be done because seed differs in length, width, thickness, density, weight, shape, surface texture, color, affinity for liquids, and electrical conductivity.

Screen Separation

Screens separate seed based on width and thickness, as well as, to a much smaller extent, length. Screens with round openings separate based on the diameter or width of the seed, whereas a thickness separation is obtained by using oblong or slotted screens.

The size of a round screen corresponds to the diameter of the holes, and that of a slotted screen the width of its oblong holes. Screens with triangular holes are also used. Screens are usually made from iron sheeting, but mesh can also be used to clean certain crops (e.g. vegetable seed).

The screen is kept clean during the cleaning process by vibration brushes mounted under the sieves, or by rubber balls placed in a frame of wire mesh under the sieve. The balls bounce up and down during operation, pushing seed out of the holes in the sieve. Balls are preferred over brushes because seed may get stuck in the brushes, which will lead to varietal mixture. Smaller machines use a device that vibrates the sieves.

Cylinder/Disk Separation

Length separation is made by indented cylinders or disks. The indents (cells or pockets) will, depending on their length, lift those seeds that fit in the indents. Larger seed will not be lifted, or not lifted high enough to be separated. The cylinder pockets may be hemispherical (most common), cylindrical or tapered.

Air Separation

Air separators (aspirators) separate seed according to its behavior in an air-stream, which depends on the speed of the particle in free fall. This speed depends on density, shape, size and surface texture. The most important characteristic is the weight in relation to the air resistance. Certain particles (dust, chaff, empty or partly filled seeds, husks, and glumes) will be transported, whereas heavier seed will fall down through the air stream.

Separation by screens, air and cylinders are the most commonly used practices.

Cleaning Machines

In general, a combination of machines is necessary to obtain the required quality; each crop requires a specific set of machines in a specific sequence (Table 1).

Table 1. Machines used to clean different crops.

	Chickpea	Lentil	Faba bean	Cereals
Air-screen cleaner				
Pre-cleaner		*		
Fine cleaner	*	*	*	*
Indented cylinder	↓	*	↓	*
Gravity table	(*)	*	*	*
Spiral separator	↓	(*)	↓	↓
Color sorter	↓	(*)	(*)	↓
Scarifier	↓	↓	(*)	↓
Treater	(*)	(*)	(*)	*
Bagger weigher	*	*	*	*

* machine usually used

(*) optional

↓ not used

Air-screen Cleaner

The air screen cleaner is the most important seed cleaning machine in the seed plant, and is used to clean almost all seed. It uses a combination of screens and air for separation. The air screen cleaner is used for all crops: cereals, legumes, pasture and forages, vegetables, etc. In many cases this machine is sufficient to clean the crop, because it separates according to the major characteristics of the seed: width, thickness, length, shape and behavior in an air stream.

Indented Cylinder

The indented cylinder is the second most important cleaning machine. All seed plants usually have indented cylinders in the standard setup of the plant. Indented cylinders are less essential for the round seed of many food legumes. The indented cylinder separates mainly on the length of the grain, and can be used in two different ways.

Round grain application (right grading): Short impurities are lifted and thrown in the tray, whereas the longer crop seed is left in the cylinder.

Long grain application (reverse grading): The crop seed is lifted and longer impurities are left behind. The capacity of the cylinder has to be larger, because all crop seed has to be lifted. In many cases long grain application is not necessary.

For many crops the indented cylinder is used in both ways during the cleaning process, whereby two or more cylinders are installed, one after one another, to perform both applications.

Specific Gravity Separator

After the seed is cleaned by the air-screen cleaner and indented cylinders, it may be necessary to use a gravity separator. The gravity separator is able to separate seed of the same size with different density/weight.

Assuming that lower density seed is less vigorous, or lower in germination capacity, the gravity separator may thus be used to improve germination. Moreover, particles (clay, sand, and stones) of similar size to the seed can be removed with the gravity separator.

The gravity separator classifies components of a seed mixture mainly according to density or specific gravity. Seed is fed into the machine in a rather thick layer (3–5 seeds). The shaking deck will push the heavier seed uphill, because it is in contact with the deck. The airflow through the deck lifts the lighter seed and—because it is not in contact with the deck—it tends to float downhill.

Debearder/Deawner

Although not strictly a seed cleaning machine, a deawner is used to remove awns (barley, grasses) or to separate multi-seed units (grasses) before the actual cleaning process begins. The seed is rubbed by mechanical devices (rotating beater arm) until the appendages have been removed.

Spiral Separator

The spiral separator classifies seed according to its shape and ability to roll. The machine consists of sheet metal strips fitted around a central axis in the form of a spiral. The seed is fed in at the top into the inner spiral. Round seed rolls faster down the inclined flight and obtains a higher speed than flat or irregularly-shaped seed. The speed of the round seed increases, until it rolls over the edge of the inner flight into the outer flight, where it is collected. The slower moving seed does not build up enough speed to escape from the inner flight. Most spirals have multiple inner flights, arranged above each other to increase capacity. A spiral separator is used to remove damaged seed from brassica, pea, soybean, vetch, and lentil, and to remove *Galium* (cleavers) from spinach and lentil.

In general it can be said that the spiral separator is very useful to remove broken grain from a round seeded crops.

Color Separator

Discolored seed is of a lower quality and should be removed from the good seed. Since both density and dimensions are the same as the good seed, none of the previously mentioned machines can be used. By means of photocells, the color of the seed is compared with "background plates," which are chosen in such a way as to reflect the same light as the good seed. Seed that differs in color is detected by the photocells, which generate an electric impulse. This impulse is used to activate a jet of air that blows away the discolored seed. The color separator is extensively used to remove discolored seed from pea, bean, faba bean, and sunflower. It could also be used to remove black vetch seed from the much lighter lentil. This machine is, however, rather expensive, and the cost is often not worth the small increase in quality.

Picking Belt

A picking belt is a conveyor belt on which the seed is fed. The individual seeds are observed by a number of laborers who sit along the picking belt. A picking belt can be successfully used as an alternative for a color separator (off-colored seed is removed), as well as for a needle indented cylinder (insect infested seed is removed). A picking belt can basically be used to remove any seed which should not be part of the clean material.

A picking belt is also often used before drying/shelling maize cobs as part of the pre-cleaning operation.

Needle Indented Cylinder

This is a special type of indented cylinder in which the cylinder has a large number of needles. Insect infested grain will be lifted in the tray, because the needle fits in the hole made by the insect.

Belt Grader (Band Grader, Draper Mill)

A belt grader separates on the basis of ability to roll or slide. The machine consists of a turning belt and a feeder that drops the seed onto the belt. The angle and speed of the belt can be adjusted. Smooth seed slides against the direction of rotation; rough seed or particles that cannot roll easily (e.g. stalks) are conveyed upwards. The grading is dependent on shape, weight, surface structure of the seed, and on the inclination, speed, and surface of the belt. A canvas or rubber belt can be used. The belt grader is used to remove stalks from beet seed, to remove buckhorn plantain (*Plantago lanceolata*) from red clover (*Trifolium pratense*), and to clean flower seed.

Magnetic Separator (Magnetic Drum)

The magnetic separator exploits surface texture. When seed is treated with iron filings, rough seed will pick up the filings, but smooth seed will not. When such seed is passed over a revolving, magnetic drum, the seed coated with iron filings is attracted to the drum and separated from smooth, uncoated seed. To improve the effect, water can be added while mixing; in some cases water is indispensable. The greater the difference in surface texture of the components to be separated, the more effective the separation will be. A magnetic separator is used to remove *Stellaria media* (chickweed) from clover and alfalfa, *Cuscuta pentagona* (dodder) from clover, alfalfa, and red clover, and *Sinapis arvensis* (wild mustard) from brassicas.

Paddy Table (Table Separator)

Paddy (rice with glume) can be cleaned from naked caryopses with a table separator, which works on the specific gravity and surface texture of the seed, combined with the resilience. The machine is often replaced by a specific gravity separator. This machine is often successfully used to remove barley from wheat seed.

Scarifier

The purpose of a scarifier is to remove the hardness of the seed coat to improve germination. Seed is fed through a drum with sandpaper on the inside wall. A scarifier is often used with lucerne or sweet clover.

Elevators and Conveyors

During processing, seed must flow efficiently between the different machines. Mechanical seed movement is necessary to ensure maximum capacity, lowest cost, and shortest time.

Elevators are used to move seed vertically; conveyors are used to carry seed horizontally or at an angle. Maximum care should be taken when choosing elevators and conveyors because certain crops (legumes) are sensitive to mechanical damage. Screw conveyors should never be used for seed because they easily damage the seed; belt conveyors are preferred.

Pipes connecting or feeding machines should be at 45 degree angles to allow the seed to slide into the machines.

The bucket elevator lifts seed by carrying it in small buckets attached to an endless belt which moves vertically. The buckets are filled with seed at the bottom; at the top, seed is dumped via a spout into the desired machine or bin. The speed of the elevators may have to be adjusted to deal with the fragile legume seed. Other elevators, using various types of chains or links instead of a flat belt, are generally not recommended for legume seed. Still other elevators convey seed in an airstream which flows through sealed pipes; again, this may lead to damage of legume seed.

Bins

Proper use of bins in a seed plant maximizes bulk handling and flexibility in capacity, minimizes labor costs and operating delays, and reduces possibilities for contamination. The different machines in a processing line do not operate at exactly the same capacity; without holding bins, all machines must run at the capacity of the slowest. Some cleaners do not separate seed as effectively when operated below normal capacity. Holding bins absorb differences in capacity, and permit each machine to operate at its most effective rate. Every machine should be served by a bin large enough to permit efficient operation, both of itself and the previous machine. The first bin in the processing line, and the elevator serving it, should be as large as possible, to fill the bin rapidly with enough seed for several hours. Similarly, the final bin, which receives processed seed and supplies the bagger/weigher, should be large enough to hold all seed cleaned during several hours.

Air Compressor, Vacuum Cleaner and Brooms

The cleaning of the seed processing machines (and the cleanliness of the plant) is very important. The cleaning machines should all be thoroughly

cleaned not only between different species, but also between different varieties. All sieves, cylinders and decks have to be taken off and properly cleaned. Compressed air, brushes, etc. should be available at all places in the processing plant to properly clean the sieves and insides of all machines. Vacuum cleaners are also indispensable. The machines should be run for several hours at full speed to discharge hidden grains.

Computerizing Seed Processing Facilities

Microprocessor use in seed conditioning facilities is expanding. There are two main areas where microprocessors currently are used in seed processing: control of seed processing equipment adjustment and product weighing/packaging systems. We will focus on equipment adjustment control.

When the seed conditioner adjusts his equipment, he has three main goals: (i) high quality; (ii) fast conditioning; and (iii) high efficiency at low cost. Normally, the higher the quality standard, the slower the operation and the higher the cost.

During processing of a seed lot, the incoming product is constantly changing in quality. Since high quality standards are required, the amount of material discarded must be such that these standards are always met. In this way, the important goal of high quality seed is set, but speed and efficiency of the operation are compromised. To reach the other two goals, the seed processor must regularly adjust the machine to minimize the discarding of good seed with the bad.

There is a commercial microprocessor system available today for the automated monitoring and adjustment of cleaning equipment. This system is called Kamatrol and was developed by KAMAS. The system uses three sets of sensors (under the bottom screen and in the pre- and tail-aspiration channels) to gather data inside the air-screen cleaner. Using these data, the microprocessor adjusts the amount of material removed in two air channels and adjusts the feeding rate of the machine.

The operator must set the machine initially, but the microprocessor then combines initial criteria and operating data to adjust the cleaner to reach quality standards at maximum capacity. Air channel performance is monitored by sensors in the channels. Machine speed is monitored by sensors under the bottom sieve. The data are then processed by the microprocessor, which adjusts the air and the machine speed by servo motors.

Seed plant tests show that between 2–10 percent of the seed is saved from being discarded with the waste.

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Seed Fumigation

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Fumigants are insecticides which are used to combat storage pests and for quarantine purposes. They are extensively used in seed production programs. The most important application method in seed production programs is one in which stacks of seed are fumigated under fumigation sheets. Complete seed store fumigation should be abolished, because stores are never sufficiently airtight.

Fumigants have the advantage of penetrating into bags, boxes and other types of containers. The disadvantage of fumigants is that they are not persistent.

Two products, aluminum or magnesium phosphate and methyl bromide, are mainly used for fumigation. The characteristics of Phostoxin (phosphine) are: (i) excellent penetration capacity; (ii) easy handling; (iii) no influence on germination capacity; and (iv) no residue.

The advantage of methyl bromide (MB) is its quick action (12–24 hours). Furthermore, MB may be used at temperatures below 15° C. Disadvantages of MB are: (i) extreme toxicity; (ii) accumulation in the body of human beings; (iii) residue remains in the seed; (iv) poor distribution; and (v) influence on germination. Because of its disadvantages MB should not be used for seed fumigation.

As with insecticides, resistance is building up against fumigants. This is very serious, because there are no alternative chemicals. The main cause for this resistance is the fact that fumigation is usually not carried out under airtight conditions, resulting in sub-lethal dosages.

Factors determining the success of fumigation are: (i) hygienic conditions; (ii) proper dosage/exposure time; (iii) adequate sealing; and (iv) correct temperature and sufficient moisture.

The lessons learned are: (i) abolish complete store fumigation; and (ii) do not use methyl bromide.

Components of Seed Quality

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Introduction

A cultivar or variety is defined as “an assemblage of cultivated plants which is clearly distinguished by a given character (morphological, physiological, cytological, chemical or other), and which when reproduced (sexually or asexually) retains its distinguishing characters.”

Improved crop varieties are under constant development through plant breeding programs, each with one or more additional merits over the existing cultivars under crop production. New varieties are released based on better yield or product quality, tolerance to biotic and abiotic stress, response to modern inputs or suitability to farming practices. In recent years varieties have been bred for specific quality characters and need to be grown in pure stands.

Alternatively, the use of variety mixtures in a varied environment as well as for disease control may be more useful than pure stands. In a survey of literature, Wolfe (1985) stated that mixtures usually yield at least the mean of their components and sometimes exceed the highest component. Similarly, host mixtures may restrict the spread of a disease, provided that the components differ in their susceptibility (Browning and Frey, 1969).

The seed of improved varieties should be made available to farmers in the shortest possible time. This is usually done by multiplying breeder seed through a number of generations to obtain the large quantities of certified seed that are required to produce a commercial crop.

Seed is a living product that acts as a catalyst to transform the genetic improvements of plant breeders into crop production potential. To play its role, the seed supplied to farmers should be of very high quality.

Quality seed can be defined as seed of an improved variety which: (i) is high in species, varietal, and physical purity; (ii) has high germination and vigor; (iii) is free from weeds and seed-borne pests; (iv) has a low moisture content; (v) is uniform; and (vi) is properly processed for distribution to farmers.

Thus the most important components of seed quality are: (i) species purity; (ii) varietal purity; (iii) physical purity; (iv) germination; (v) vigor; (vi) seed health; (vii) moisture content; and (viii) size and uniformity. Aspects of quality also include seed treatment, packaging and labeling.

Each component is of great importance to the user under different circumstances, as poor quality in any one factor may result in reduced quality and partial or total crop failure.

Physical Purity

Farmers require seed that is uncontaminated with seed of different crop species or weeds, or inert matter (straw, soil, etc.) that may reduce the quality of their product. For example, noxious weeds may overrun cultivated fields.

It is not possible to get completely pure seed, because cleaning machines cannot usually remove all impurities. Moreover, the more impurities that are removed, the greater the amount of crop seed lost during processing. Yet cleaning is one of the most effective ways to upgrade seed quality. Considering its benefits, it is also inexpensive.

The physical purity of seed that is offered for sale is easy to check in the laboratory. This guarantees that farmers buy seed of the required species and not inert matter (stones, chaff, etc.) and dangerous weeds (wild oat) or parasitic weeds (*Orobanche*, *Cuscuta*, and *Striga*).

Physical or analytical purity is the proportion of pure seed in a certain lot and the composition of the undesirable matter. To assess purity, two tests are often used: the physical purity test and the number count test. Purity testing does not differentiate between different varieties of the same species.

Physical Purity Test

The purity test is used to determine the composition by weight of a sample, the nature of the contaminants present and, by inference, that of the seed lot it represents (ISTA, 1985). During analysis the sample is divided into three fractions—pure seed, other crop seed, and inert matter—and reported on a percentage weight basis. However, for national purposes, the sample can be divided into four fractions by separating other crop seed into cultivated crops and weeds.

Number Count Test

The physical purity test only determines the percentage of pure seed, and as a result the presence of a very small percentage of noxious weeds is greatly underestimated. For instance *Orobanche* seed is small (0.2–0.3 mm; 150,000–350,000 seeds per gram) and expressing it—under the other seed fraction—as a weight of the total sample is meaningless. To determine the degree of contamination with competitive noxious or parasitic weed seed, the number count test (to determine the numbers of other seed) is used to further evaluate seed quality.

The test actually counts the number of all other species (complete test) or of one specific species (limited test) in a sample. Because the sample used to assess physical purity (as a percentage) is too small to give a precise estimate of the number of other species present, usually a sample ten times larger than that used for the purity test is tested. The results are expressed as the number of seeds per weight of seed examined.

Germination Capacity

In addition to physically pure seed, farmers require that seed germinate and produce vigorous and “strong” plants that can withstand all biotic and abiotic stresses.

High standards of purity are of no advantage or benefit if the seed is incapable of germination. Therefore, germination, the second quality attribute, is considered important to avoid planting seed of low quality, which may lead to crop failure. Germination capacity indicates the percentage of pure seed that produces normal seedlings under optimal conditions in the laboratory test and, by inference, the field planting value in a favorable environment.

Germination capacity combined with analytical purity can be used to determine the proportion of seed which can produce normal seedlings, called the pure live seed content (PLS). The PLS is generally used to express seed quality, and can be employed to choose among different seed lots. To judge which seed is the best buy, the price paid for seed can be calculated as price per unit of PLS.

Germination capacity, assessed under laboratory conditions, will not accurately predict field establishment, because field conditions are seldom optimal and most seed lots will not reach the predicted value due to field stresses.

Seed Vigor

Vigor is not a simple measurable property like germination, but rather a quantitative character, controlled by several factors affecting the germinating seed or seedlings. Vigor is affected by mechanical damage to embryo or seed coat, environment and nutrition of the mother plant, stage of maturity at harvest, seed size, senescence, attack by pathogens, drying temperature, etc.

For example, higher temperatures speed up drying, but drying injuries may occur. This is not immediately detected by a normal germination test. However, vigor tests may show that the slowly dried seed is more vigorous than seed that is dried fast at a higher temperature. When such seed is stored under less ideal storage conditions, the fast dried seed deteriorates much more rapidly than the slowly dried seed lot.

Due to variations in seed vigor, seed lots with similar germination may respond differently when subjected to adverse field conditions. Contrary to germination, vigor indicates the capacity of seed lots to produce a good crop stand under sub-optimal field conditions.

Generally, high germination capacity is believed to be associated with high vigor, and low germinating seed must be rejected since field emergence is reduced drastically (Table 1) and cannot be compensated by increasing seed rates.

Table 1. Germination capacity and field establishment of five lots of pea seed.

Lot	Germination laboratory (%)	Establishment field (%)
1	98	91
2	93	68
3	80	39
4	71	33
5	56	16

Source: Thomson, 1979 (cited from Franck, 1979)

TeKrony and Egli (1991) stated that the effect of seed vigor on yield depends on the stage of crop harvest. There is a consistent positive correlation between seed vigor and yield in crops harvested during vegetative growth (lettuce, cabbage, turnip, carrot) or early reproductive growth (tomato, pea). However, for annual crops harvested at full reproductive maturity (seed), there is no relationship between seed vigor and yield under normal conditions, unless there is a low plant population or later than normal planting. Similarly, Khah *et al.* (1989) found that low vigor spring wheat seed produced lower yields only when it resulted in low plant populations or when planting was later than normal.

Several vigor tests have been developed to predict field establishment. Vigor tests should be rapid, cheap, easy to perform, and reproducible for objective assessment and meaningful interpretation of results. Many of the vigor tests are, however, too complicated to be carried out as a routine test in a seed laboratory. So far not a single test, whether physiological or biochemical, has proved successful even for a single species under different field conditions (Hampton and Coolbear, 1990).

The different vigor tests are:

Physical test	Seed volume, weight, size.
Physiological test	Standard germination, speed of germination, seedling evaluation, cold test, accelerated aging, controlled deterioration,
Biochemical test	Tetrazolium, conductivity, respiration.

Varietal Purity

In addition to physically pure seed that is capable of germinating and producing a good crop stand, it is important that seed is genetically pure. The genetic potential of an improved variety can only be exploited by farmers if the genetic make-up is not diluted during multiplication. Varietal purity refers to whether a variety is true-to-type, and if it still has the original genetic make-up. For pure line varieties, all plants are similar in agro-ecological, morphological, physiological, cytological and chemical characters.

Varietal or cultivar purity is an important attribute of seed quality, because it guarantees that the genetic make-up (agro-ecological performance) of the variety as defined by the breeding methodology is still present when the seed of improved varieties reaches the farming community.

Varietal purity is important for seed program activities, because it allows identification of varieties. Moreover, it makes it possible to guarantee quality when selling seed to farmers and to demand quality when buying seed from seed retailers.

Varietal purity tests establish whether or not a field or a seed lot of a variety is sufficiently pure, i.e. whether a sufficiently large percentage of seed, seedlings or mature plants conforms to the original description of the variety. It can be controlled by inspection of plants in seed multiplication fields or by examining seed or seedlings in the laboratory, or growing plants in field plots.

Field and laboratory controls should be supplemented by administrative control of the seed movement to verify the source of multiplication stocks in the seed production cycle.

Moisture Content

Most seed reaches physiological maturity at moisture contents ranging from 35–45 percent (cereals) or 45–50 percent (legumes), but the seed should be dry enough to be harvested. Furthermore, seed should be kept at moisture contents of 10–12 percent for safe storage. Quality seed should also have an acceptable moisture content to enable storage for longer periods. Since moisture content influences seed quality during harvesting, processing and storage, it should be kept low at all stages. Low moisture content makes seed liable to mechanical damage during harvesting (Table 2) and processing. Moreover, low levels of moisture content may cause germination problems such as inducing secondary dormancy.

Table 2. Effect of moisture content (MC) and cylinder speed on mechanical breakage of mung bean.

MC (percent)	Hand harvest	Mechanical harvest (cylinder speed)			
		510	1,020	1,530	2,040
15.2	1.6	2.2	3.3	2.9	5.4
11.2	1.8	16.8	32.1	44.1	59.2
7.2	1.9	19.9	39.4	52.9	70.2
LSD (0.05) = 3.0					

High moisture content at harvest damages the seed coat, whereas during storage it initiates fungal development, insect activity, heating and germination, which contribute to rapid seed deterioration (Table 3).

Table 3. Estimated maximum storage period (weeks) for faba bean.

Temperature (°C)	Seed moisture content (percent)					
	11	12	13	14	16	18
5	370	270	170	110	70	39
10	200	140	95	60	38	20
15	100	75	50	30	20	12
20	55	40	28	19	13	7
25	31	22	16	17	7	4

Source: (Ellis *et al.*, 1988, cited from Hebblethwaite *et al.*, 1983).

Faba bean seed harvested at moisture contents between 51–24 percent showed a decline in the proportion of broken seed from 19 to 2 percent, and an increase in germination from 48 to 92 percent (Sjödin *et al.*, 1972).

Moisture content tests can be carried out in the laboratory by the oven method, but portable moisture meters are available to make quick determination of moisture in seed production fields, while processing, or during storage to decide alternative measures.

Moisture content should also be taken into account when buying seed; 1 percent greater moisture in a lot of 10 tons represents 100 kg of water that could have been seed.

Health

Seed can serve as a vehicle for the dissemination of plant pathogens, which can result in disease outbreaks. Seed-transmitted pathogens include fungi, bacteria, nematodes and viruses. They can be transmitted as contaminants with seed, on the seed surface, or through seed infection (in the endosperm or embryo).

Seed health is an important factor in the control of crop disease, since infected seed is less viable, has low germination, reduced vigor and reduced yield. For example, wheat grain severely infected with karnal bunt either fails to germinate or produces a greater percentage of abnormal seedlings (Singh, 1980; Singh and Krishna, 1982).

Chemical seed treatment can be used to control both external contamination, or internal infection, or to protect young seedlings or adult plants against attack from soil or air-borne pests. A wide range of chemicals for seed treatment and equipment for application is now available for such purposes (Diekmann, 1988).

Size and Uniformity

Size is the most common difference among seeds, and generally varies from one plant to another in one field or even within the same plant. Variation from plant to plant, which results from genetic differences, inter-plant competition for light, water, and nutrients, and the effect of disease, contributes to a wide range of seed sizes within a seed lot. Similarly, seed size also varies within plants due to the location on an inflorescence, which reflects differences in flowering time (main or side tillers) and in the nutrition of the developing seeds (basal or apical flowers).

Within a lot, large seed usually results in increased and rapid emergence, although sometimes it performs more poorly than small seed (Wood *et al.*, 1977). In wheat, seed size is positively correlated with seed vigor, and

larger seed tends to produce more vigorous seedlings (Ries and Everson, 1973). But Mian and Nafziger (1992) found that seed size has little effect on emergence in soft red winter wheat. According to Wood *et al.*, 1997, Hewston also showed that large cauliflower seed with greater laboratory germination may not necessarily result in a greater percentage of field emergence (Table 4).

Table 4. The effect of seed size on germination and emergence of summer cauliflower "Meteor."

Seed diameter (mm)	Germination (%)	Field emergence (%)
1.75-2.0	72	68
2.0-2.5	81	82
2.5-3.0	88	63

Source: Wood *et al.*, 1977 (cited from Hewston, 1963).

Wood *et al.* (1977) also indicated that large seed produces larger seedlings, and the advantage persists to increase final yield, particularly for short growing season crops or where economic yield is a seed or storage organ. But differences in yield between crops from large and small seed is most likely to be maintained if certain yield components or a significant part of the stored material are produced in the early stages of growth, when the effects of seed size are greatest.

Larger seed of spring wheat produced higher yields than smaller seed under late sowing conditions (Singh and Kailasanathan, 1976), not under optimum management (Kalita and Choudhury, 1984).

Uniformity of size is more important in mechanical planting than hand planting, as precision planters require accurate and uniform seed size. Seed lots are less uniform at harvest since the proportion of impurities present is very high. Processing operations may improve uniformity of seed by removing undesirable extraneous material, if carried out with extra care. Therefore, processed seed lots are expected to be more homogeneous in terms of their size and constituents.



Conclusion

Although each component of seed quality has significance under specific conditions, it is possible to rank them in terms of relative importance (Fig. 1). Germination seems the most critical factor, followed by vigor and

health. Failure in germination may lead to total crop failure. Similarly, less vigorous seed may fail to emerge and drastically affect plant populations in a harsh environment, which reduces the overall production potential of the crop. The same is true for seed health, if the pathogen is exclusively seed-transmitted, has a rapid transmission rate, and is influenced by weather.

Purity, on the other hand, appears to be relatively less important. Seed with some varietal, species and physical admixtures will still produce a reasonable crop.

Size and uniformity are considerably less important, unless used for mechanical planting. But with widespread precision drilling in a mechanized system it is becoming increasingly important.

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Seed Certification

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Introduction

Seed certification is a quality control system to ensure that seed sold to farmers is of the indicated variety, has sufficient purity (physical and varietal), high germination capacity and is free from seed-borne disease.

Legally sanctioned certification systems operate in many countries, under various systems. In developed countries the systems are backed up by a seed act. Internationally recognized systems have been developed by the Organization for Economic Cooperation and Development (OECD, 1971c; 1971d; 1977a; 1977b) and the Association of Seed Certifying Agencies (AOSCA, 1971). They are designed to evaluate:

Eligibility of varieties: Only officially released varieties, which have been evaluated for DUS and agronomic performance, are eligible for certification.

Seed classes: Classes, as well as the maximum number of multiplications in each class, are set for each species.

Seed quality: Minimum standards are set for quality features.

Only varieties which have been officially evaluated and satisfy minimum quality standards can be marketed. Seed certification agencies exercise tight administrative control over the multiplication process, implement field inspections, test processed seed lots for various qualities, carry out labeling, and conduct post-control of approved seed.

Eligibility of Varieties

Only officially released varieties that have been evaluated for DUS and agronomic performance are eligible for certification. In developed seed programs, new varieties are evaluated for performance, distinctness, uniformity, and stability before they are released by a variety release board. In developing countries, performance is often established before release, but no attention is paid to distinctness, uniformity or stability.

Seed Classes

Seed produced under a quality control system must relate, through one or more generations, to seed obtained from the plant breeder (breeder seed). The first generation is known as basic seed (in the USA and many other countries, the term foundation seed is used). The generations between breeder and basic seed are called pre-basic seed. The generations after basic seed are called certified seed first generation, certified seed second generation, etc. (in the USA, registered seed, and certified seed). Other countries use a system whereby G_0 is breeder seed, G_3 basic seed, and R_1 , R_2 etc. are the different generations of certified seed.

Restricting the number of generations is one way to preserve quality. This is more important with cross-pollinating than with self-pollinating crops. In developing certification systems, generation controls are not often very strict.

Standards

In a certification scheme, quality standards (Table 1) have to be established for contaminants in each class. During multiplication, a small quality loss is expected, so standards are highest for early generations. Standards should not—at least in the initial stages—be too rigid, because this may lead to seed shortages. Nor should they be so low that the farmer loses confidence in the benefits of quality seed.

Table 1. Types of quality standards.

Field inspection	Seed testing	Pre- and post-control
Field preparation		
Planting		
Cultural practices		
Seed source	Varietal purity	
Varietal purity	Moisture content	Varietal purity
Noxious weeds	Physical purity	Weeds
Seed-borne disease	Germination	Seed-borne diseases
Other crop seed	Seed health	Other crop seed
Isolation distance	Vigor	
No. of generations	Other crop seeds	
Previous cropping	Viability	
Stand		
Yield estimate		
Combine cleanliness		
Post-harvest		
Store sanitation		

Initially, seed lots should be surveyed for the occurrence of seed-borne disease, weeds, etc. An example of standards for different crops is given in Table 2. As the system develops, standards must be revised as needed.

Table 2. Standards for field inspection.

Crop	Minimum isolation distance (m)			Undesirable plants (%)			Minimum number inspections	
	A-B	C ₁	C ₂ -C ₃	A-B	C ₁	C ₂ -C ₃	Field	Post-harvest
Maize open pollinated	400	200	200	0.1	0.3	1.0	2	2
Wheat	10	5	5	0.05	0.1	0.3	2	1
Barley	10	5	5	0.05	0.1	0.3	2	1
Rice	10	5	5	0.05	0.1	0.3	2	1
Sorghum open pollinated	400	200	200	0.1	0.3	1.0	2	1
Field bean	50	25	25	0.05	0.1	0.3	2	1
Pea	100	50	50	0.05	0.1	0.5	2	1
Soybean	10	5	5	0.05	0.1	0.3	2	1
Cowpea	10	5	5	0.05	0.1	0.3	2	1
Chickpea	50	25	25	0.05	0.1	0.3	2	1
Ground nut	20	10	10	0.05	0.1	0.5	2	2

Source: Wellving, 1984.

A=pre-basic, B=basic seed, C=certified seed.

Administrative Control

Seed certification agencies should keep records on contract seed growers. An application for field inspection should be sent to the agency before the start of the growing season. The application should provide details on (i) the seed grower; (ii) field; (iii) species and variety; (iv) seed origin; (v) certification class; (vi) amount of seed; and (vii) area to be planted. Seed labels (certificates) must be handed over to the certification agency.

The multiplication process is carefully monitored to verify the source and suitability of a seed lot. Generation control is strict. By keeping records of the fields used for multiplication, previous cropping problems can be avoided. Registration of seed growers helps to monitor their performance, with only dedicated growers accepted for seed growing.

In certain schemes the transport of seed from field to factory is also under the control of the certification agency.

Field Inspection

Without standardized inspection procedures, an inspector can only rely on his or her experience, and wide quality variations will occur. This is because

different inspectors will use different standards to accept or reject fields, and the standards by which one inspector accepts or rejects fields will vary over time. Lack of standardized inspection procedures causes variations in seed quality.

With standardized procedures, all inspectors uniformly assess the quality of all fields. Standardized procedures do not have to be ideal; uniformity of procedures and criteria is more important.

A field inspection consists of two steps. First, the field is observed in a "field overview" to see that it is uniform in quality. Second, a statistically determined sample of plants, called the "field inspection sample" is inspected to identify contaminants. These are counted and an occurrence rate established to determine if the field meets seed production standards.

Field Overview

To carry out the field overview, the inspector walks through the field to determine if it is uniform in quality. The inspector assesses the following: (i) variety; (ii) disease infection; (iii) weed infestation; (iv) isolation; (v) stand; (vi) yield estimate; (vii) previous cropping; and (viii) cultural practices.

The crop in the field must be of the indicated variety. A reasonable number of plants should be identified for this purpose.

The inspector should assess disease and weeds present in the field. Since weeds are often found in discrete areas, the inspection must not rely on field counts alone.

The crop must be effectively isolated from similar varieties, from the same varieties in a different multiplication cycle, and from crops which can cross-fertilize. Isolation must also include individual plants of such varieties and crops growing nearby. Isolation distances are very important for cross-pollinating crops, as well as for self-pollinating crops with a percentage of out-crossing. They are less important for strictly self-pollinating crops. For cereals, a physical barrier is sufficient.

The inspector often assesses the general stand of the crop. Weeds and admixtures that are not dangerous or difficult to remove during processing are considered. The inspector should also check on field operations, rotation, and previous cropping. A yield estimate is often made.

Between zero and two cropping seasons should elapse between two seed crops, depending on local conditions.

Threshing grounds, harvesting and planting equipment, drying floors and storage areas may be checked.

Field Inspection Sample

To determine if the field meets the standards, the actual number of each contaminant needs to be established. The inspector does not inspect the entire plant population; he or she measures field quality by inspecting the plants in a representative sample area, called the "field inspection sample." He counts the contaminants present, records the numbers, and compares them with the numbers allowed by the standards.

Statistically, a sample of three times the number of plants in which one contaminant is allowed is large enough to decide whether or not the field has met the standard. In other words, if an area in which three contaminant plants are allowed is analyzed for a certain character, an accurate picture of the field quality is obtained.

Field Counts

To ensure that the field inspection sample accurately represents field quality, it is divided into smaller areas called *field counts*. The field inspection sample is equally divided into five or six field counts. They are located in different places of the field (edge, side, center, etc.).

Walking Pattern

A specific walking pattern is followed, so that the inspector can see all parts of the field, while minimizing the distance walked and time spent. This pattern also lets the inspector locate the field counts randomly, so that the measurement of contaminants is accurate and represents the quality of the entire field.

Quality Factors to be Assessed

Contamination includes genetic contamination, physical contamination, and pathological contamination. Genetic contamination is caused by pollination by other varieties of the same crop and/or related species in the field or within the isolation distance. Physical contamination is caused by seed from: (i) other varieties of the same crop; (ii) other crops; and (iii) weeds. Pathological contamination is caused by plants of the same or other crops that carry pathogenic agents. The following factors should be considered:

Off-type plants and other varieties: These are plants of the same crop that differ in one or more characteristics. Variations which are characteristic of the variety are not considered off-types.

Inseparable seed: Seed of other crops is considered inseparable when its physical characteristics are so similar to the seed crop that it cannot be separated in processing.

Undesirable weeds: Weeds are undesirable in a seed production field if: (i) their seed is so similar to the seed crop that it is difficult to separate in processing; (ii) their growth habit is competitive; or (iii) their method of propagation makes them difficult to eradicate.

Diseased plants: Plant diseases are caused by fungi, bacteria, viruses, or nematodes. Seed may carry pathogenic agents internally, externally, or both, and transmit them from one crop generation to the next. Disease transmission is usually avoided by eliminating plants whose seed may carry pathogenic agents, and by isolating the seed crop from diseased plants.

Table 3. Standards for laboratory tests.

Species	Purity % (lowest value)			Germination % (lowest value)			Weeds number/kg highest value**				Moisture content all classes	Defect seeds % by weight (highest value)		
	A-B	C1-C3	D	A-B	C1-C3	D	A	B	C1-C3	D		A-B	C1-C3	D
Maize	99.5	99.0	98.0	90	90	90*	1	2	5	10	14.0	1.0	2.0	2.5
Millet	98.5	98.0	97.0	80	75	75*	250	500	750	2000	14.0	0.5	1.0	1.5
Rice	98.5	98.0	97.0	85	80	80	1	2	5	10	14.0	1.0	2.0	2.5
Sorghum	98.5	98.0	97.0	80	75	75*	250	500	1000	2000	14.0	0.5	1.0	1.5
Wheat	99.5	99.0	98.0	85	85	85	1	2	5	10	14.0	0.5	1.0	1.5
Bean	99.5	99.0	98.0	80	75	75	1	2	5	10	16.0	1.0	2.0	2.5
Pea	99.5	99.0	98.0	80	75	75	1	2	5	10	16.0	1.0	2.0	2.5
Soybean	99.5	99.0	98.0	80	75	75	1	2	5	10	16.0	1.0	2.0	2.5
Lucerne	99.0	98.0	97.0	75	70	60	200	250	500	1000	10.0			
Guinea grass	70.0	60.0	50.0	60	60	40	250	500	1000	2000	10.0			
Rhodes grass	70.0	60.0	50.0	60	60	40	250	500	1000	2000	10.0			
Rye grass	97.0	96.0	94.0	85	85	75	250	500	750	2000	10.0			
Groundnut	97.5	92.0	96.0	80	75	65	1	2	5	10	10.0	2.0	5.0	10.0
Sunflower	98.5	98.0	97.0	85	80	70*	5	10	50	100	10.0	1.0	2.0	4.0
Cotton	99.0	99.0	98.0	85	85	75	50	50	100	200	10.0	1.0	2.0	4.0
Kaaf	99.0	99.0	98.0	75	70	70	100	100	200	400	10.0	1.0	2.0	4.0

A=pre-basic seed, B=basic seed, C=certified seed, D=commercial seed.

* Not applicable for F₁ hybrids, ** For these weeds, nil seed is allowed in a seed lot.

Avena fatua, *Avena ludoviciana* (Wild oats), *Cuscuta* spp. (Dodder), *Rotboellia exaltata* (Mulungwe) *Xanthium pungens*.

Source: Wellving, 1984.

Seed Inspection

After the seed has been cleaned at the processing plant, samples are taken to check quality. Upon sampling, the homogeneity of the seed lot is assessed, and other aspects such as correct labeling, lot number, etc. are checked. The sample is sealed in such a way that any attempt to open it would be noticed. The sample is immediately dispatched to the seed testing station, where, according to the International Seed Testing Rules (ISTA, 1985), tests are carried out for: physical purity, germination, moisture, varietal purity and seed health. Other tests (e.g. vigor test, viability test, 1,000 grain weight test) are often carried out. An example of the standards for laboratory seed testing is given in Table 3.

Control Plots

Advanced certification schemes usually include further checks on varietal identity, genetic purity, weeds and seed-borne disease. Post-control plots are mainly a check on the certification agency's work (OECD, 1971a; 1971b). They are a useful tool to train inspectors, and act as a warning system to identify problems in multiplication fields. Results of pre-control plots are used when certifying a seed lot.

Many developing countries are not using these very important tools to improve the quality of the seed.

Certification

The act of approving a seed lot is called certification. Certification is based on the combined results of field inspection, seed testing and, if applicable, pre-control plots. If a seed lot is certified, each bag, container or package is labeled. This label is often the official certificate, and opening a bag will result in a damaged certificate.

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