Plant Breeding, Practice

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Glossary

Allele One of the alternative forms of a gene at a particular location on a chromosome (i.e., at a locus).
Haploidy The condition of a cell or nucleus containing only a single set of unpaired chromosomes.
Heterosis The superiority of the heterozygous progeny over its inbred parents.
Heterozygosity The situation in which the alleles at a locus are identical by descent.
Homozygosity The situation in which the alleles at a locus are not identical by descent.
Locus The region of DNA in a chromosome that is occupied by a particular gene determining a biochemical function.
Meiosis The process of cell division which produces daughter nuclei which have half the parental chromosome number.
Mutation A change in the DNA of either sequence, structure, or amount.

Introduction

Plant breeding has been practiced since humans first began to cultivate crops. Over the past 100 years the intensity of plant breeding has increased, and is now recognized as an intricate integration of science (or sciences) and practicality. Over recent times the increasing need to feed the world's population ever-greater demand for food, fuel, and fiber, as well as in the developed countries a balanced and healthy diet, has meant that there has been a continuing pressure to produce improved new crop cultivars. The strategies used to produce these are increasingly based on our knowledge of relevant science, particularly genetics, but involve a multidisciplinary understanding that optimizes the approaches taken.

A first requirement of any breeding program is to produce genetic variation in the characters that are to be improved. Once genetic variation is produced, it is necessary to select the desired types, which have a better expression of particular characters or combination of characters. Once identified the selected types need to be stabilized and propagated/multiplied for commercial use.

Plant breeding therefore appears to be a relatively simple process, and in many ways it is true that ideas of crop improvement are simple. However, the reality is more complex. It is possible to consider the three elements of the plant breeding processes (noted above as: to produce genetic variation, to select, and to stabilize and multiply for commercial use) in order to understand modern plant breeding. With this standpoint, it is possible to realize what is being done and what alternative techniques might play a role in future cultivar development. However, each of these elements is tailored to be appropriate to the particular type of crop, or species, being improved.

Production of Genetic Variation

Conventionally, genetic variability is exposed, naturally, through sexual crossing to exploit the very principles that Mendel explained. Two parents, that between them have the expression of desirable characters, are intercrossed, and the subsequent segregating generations are examined for plants with the desired characters in new combinations (recombinants). This process, therefore, relies on the segregation of alleles at all the relevant genetic loci, during the normal process of meiosis. At fertilization, there is a random fusion of gametes (pollen from one plant and egg from the other) to give the embryo, which develops into the seed. So by the natural process of sexual reproduction, but between plants that the breeder has deliberately chosen, we get offspring, which contain novel combinations of the alleles that were originally dispersed between the two parents. It should therefore be clear that choice of parents is critical.

Breeders generally use the process noted above to take advantage of natural variation that already exists within crop species. Genetic variation can be observed for virtually all characters of interest in plant breeding. This naturally occurring genetic, therefore heritable, variation accounts for most of the responses that have been made in plant breeding. However, reliance on this one source of variation does limit the potential for long-term progress particularly in relation to improving specific characters. In these instances, genetic variation can be produced by

1. Wide intercrossing to wild, ancestral relatives of the crop itself, which may still be able to cross sexually (albeit sometimes only with the intervention of tissue culture techniques) with the crop species and which may be indigenous in another country.
2. Induced mutagenesis. Genetic variation in all plants arises initially from rare mistakes (mutations) that occur in the replication of DNA. Variation for all characters in today's crops is the accumulation of advantageous mutations over time. The frequency with which mutations occur can be increased using specific 'mutagens,' and the subsequent variants exploited.
3. Developments in the areas of molecular biology and biotechnology. These have extended the possibilities for

introducing additional variation in the breeding process by transferring genes from different species, which would not be possible by more traditional means.

Selection among the Variation

The first difficulty is to decide which characters to select for. This may seem straightforward but in practice means trying to put in order of priority what characteristics are needed in future cultivars, and what level of expression will be necessary for each trait in say 10–12 years time. Breeders also face several practical difficulties. First, it is impossible to measure every relevant character because there are simply too many for this to be practical. Second, evaluation of some characters takes excessive time and effort, and demands more resources than are available. Breeding programs need to test large numbers of genotypes but have available only small quantities of planting material. In addition, which characters can be effectively evaluated based on small plots (often a single plant)? How does the breeder grow plants such that they display characters under conditions that resemble those that they will experience in the hands of the grower?

Some selectable characters show variation that it is easy to classify into discrete classes (i.e., controlled by major genes) such as those investigated by Mendel, and can usually be selected efficiently. However, most important breeding traits are controlled by multiple genes and so show continuous variation. With these the genotypic performance can be extensively modified by the environment in which they are grown. For example, yield is a character of interest to all breeders, but is controlled by many genes and affected by environmental factors such as fertilizer levels, husbandry, and weather.

The general breeding objectives of all crops are to increase usable yield, improve quality/nutritive value, and increase stability of these traits over different environments and years.

Usable Yield

Crude yield is not important, as it is the usable fraction that can actually be eaten, processed, etc., that is of major interest. This therefore brings in factors such as storage life, waste produced, and consumer acceptance. Therefore knowledge of the crop utilization (i.e., for direct human consumption, animal foodstuff, processing, etc.) must be gathered at the outset of the breeding program.

Quality of the Product

This includes, for food crops, nutritional quality, taste, and consumer preference. Although this can be related to usable yield, quality is also concerned with the nutritive value, calorific value, protein content, fat level, vitamin concentration, oil composition, etc.

Stability of Yield and Quality

Some cultivars do very well in some years or under some particular conditions, but their performance is not consistent. This can lead to disaster when they fail because of changes in the growing conditions, a poor year for rain, no fertilizer available, too wet a period at harvest, etc. Thus breeding for resistance/tolerance to all biotic and abiotic stresses is a major aim. The need to evaluate breeding lines over several years to test for stability is one reason that it takes a number of years from starting to breed a cultivar until its release to the grower (often 10 years or more). This means that breeders require an ability to forecast the future. So, for example, a breeder might need to assess:

- What characteristics will growers be requiring in the future?
- What will happen in terms of the emphasis for growers, for example, what subsidies will there be and what will the political situation be in the future?
- How will climate change have affected growing patterns?
- How will farming systems have changed?
- What will be the spectrum of diseases and pests?
- What will the end users require in the future?

Stabilizing and Multiplying New Cultivars

One of the most important determinants in setting the breeding strategy is the natural breeding system of the crop species. The main natural breeding systems can roughly be classified into inbreeding, cross-pollinated (outbreeding or outcrossing), and vegetative reproduction (i.e., clonally propagated). These differences in reproduction lead to the main differences in classical breeding programs, and are briefly reviewed here.

Schemes for Inbreeding Crops

Examples of inbreeding crops include wheat (*Triticum* spp.), barley (*Hordeum vulgare*), rice (*Oryza sativa*), soybean (*Glycine max*), pea (*Pisum sativum*), tobacco (*Nicotiana tabacum*), tomato (*Lycopersicon esculentum*), millet (*Panicum miliaceum*), lentil (*Lens culinaris*), flax (*Linum usitatissimum*), and chickpea (*Cicer arietinum*). Inbreeding species naturally self-pollinate, and in commercial practice are grown as true-breeding, homozygous lines. So all the individuals of a particular cultivar are genetically identical.

Each generation is produced by allowing the plants to self-pollinate in each cycle of the breeding program so that while the testing and selection process is proceeding the plants are at the same time becoming more inbred. When 'finished cultivars' are identified they will breed true from seed (homozygous). So inbred cultivars can be multiplied simply by letting them set seed in isolation from any other cultivars of the same species, so as to minimize the possibility that some pollen could pass between them causing contamination.

There are two main methods by which selection is achieved during this inbreeding process, bulk method, and pedigree method, although many breeding programs employ refinements of those described or indeed a combination of both methods.

Bulk Method

Bulk methods start with the creation of genetic variation, usually by hybridization between two parents. The F_1 and

several subsequent generations, often up to and including the F_6 generations, are grown as field populations, and the seed harvested and resown as a bulk. No conscious selection is imposed on these generations, and it is assumed that the genotypes most suited to the environment in which they are grown will produce more offspring, by selfing, and hence predominate in future generations. It is therefore critical that these populations, or 'bulks,' are grown in an environment that will be similar to that expected for the developed cultivars. At the second stage, individual plants showing desirable characteristics are selected. From each selected plant, a plot is grown and the produce from the best plots is selected, bulk harvested, for initial yield trials, and for seed multiplication.

This method is one of the simplest and least expensive inbreeding methods. It also avoids conscious selection among segregating genotypes. The disadvantage is the length of time from initial crossing until yield trials are grown. In addition, it has often been found that the natural selection acting in the early bulked generations is not always that which favors characters desirable for agriculture. Despite these disadvantages the concept of bulk breeding methods have been more recently explored by utilizing methods to accelerate the time toward homozygosity including double haploidy and single seed descent.

Pedigree Method

In a pedigree breeding scheme, single plant selection is carried out at the F_2 down to the F_6 generations. Again, the scheme begins by hybridization between chosen homozygous parental lines; F_2 populations are obtained by selfing the heterozygous F_1's. Single plants are selected by the breeder from among the segregating F_2 population. The progenies from these selected plants are grown in plots at the F_3 generation. The most desirable single plants are selected from the 'better' F_3 plots, and these are grown in plant plots again at the F_4 stage. This process is repeated until plants are near homozygous (i.e., F_6). At this stage the most productive rows are bulk harvested and used as the seed source for initial yield trials and multiplication at F_7.

In addition to being laborious (as a considerable amount of record-keeping is required) and relatively expensive, individual plant selection is often ineffective and leads to the loss of valuable genotypes before they are fully tested. However, this is a preferred method in many crops.

Schemes for 'Cross-Pollinated Crops'

Some examples of cross-pollinated crops include alfalfa (*Medicago sativa*; lucerne), rye (*Secale cereale*), herbage grasses, forage legumes, red clover (*Trifolium pratense*), some corns (*Zea mays*; maize), perennial ryegrass (*Lolium perenne*), sugar beet (*Beta vulgaris*), and oil palm (*Elaeis guineensis*).

The selection of new cultivars of cross-pollinated crop species is a process that changes the gene frequency of desirable alleles within a population of mixed genotypes while trying to retain a high degree of heterozygosity. So it is the properties of the population that are vital, not individual genotypes (as in self-pollinating crops). Instead of resulting in a cultivar for release that is a uniform genotype, the population will be a complex mixture of genotypes, which together give the desired performance.

Selection of desirable characters is usually carried out by mass selection, recurrent phenotypic selection, or selection with progeny testing, or a combination of all three. The maintenance of these cultivars is through uncontrolled (random) mating.

Mass Selection

This is a simple selection scheme that uses natural environmental conditions to alter the genotypic frequency of an open-pollinating population. Initial seed that results from a set of controlled crosses is grown under field conditions over a number of seasons. It is assumed that mating will occur at random, and so result in a population quickly moving toward equilibrium that can be maintained, as a population, for exploitation. Mass selection plots need to be isolated from other crops of this species to avoid any unwanted cross-pollination.

Recurrent Phenotypic Selection

In general, this type of scheme tends to be more effective than mass selection. A population is created by cross-pollination between two (or more) populations to create a base population. A large number of plants is grown from the base population, and a subsample of the most desirable phenotypes are selected and harvested as individual plants. These plants are then randomly mated to produce a new improved population. This process is repeated a number of times – it is a recurrent process. Populations that can be exploited as cultivars can be extracted at any stage, tested, and distributed to growers.

Developing Hybrid Cultivars

High productivity in most outpollinating crops is often thought to be related to high heterozygosity. Plant breeders have utilized this feature by developing hybrid cultivars, which can be highly heterozygous and hence express hybrid vigor or heterosis. In theory, any species might be used in hybrid production but commonly it is outbreeding species, which are somewhat tolerant to inbreeding, such as Brussels sprouts (*Brassica oleracea* var. *gemmifera*), kale (*Brassica oleracea* var. *acephala*), corn, onions (*Allium cepa*), rape (*Brassica napus*), sorghum, and tomato.

At the beginning of the twentieth century there was a general awareness, especially in the United States, that the means being used to develop new corn (maize) cultivars (mass selection and ear-row selection) were less effective than expected in producing cultivars with increased yield. Another approach was suggested from the knowledge that hybrids produced by cultivar × cultivar crosses often showed heterosis (i.e., produced yields greater than the better parent). It was then proposed that this could be exploited by manually detasseling one corn line (designated as the female parents, i.e., removing the male flowers) in plots also containing the second line, so that seeds produced on the line designated as female must have been pollinated by the pollen from the flowers of the male line.

There are hardly any agricultural crops where hybrid production has not been considered, although hybrids are exploited in relatively few crop species. The reasons behind this are the following: first that not all crops show the same

degree of heterosis, and, second, that it is not possible to find a commercial seed production system that is economically viable. Indeed if corn had not had separate male and female reproductive organs, thus allowing easy manual emasculation, hybrid cultivar development might never have developed, or would have been delayed at least 20 years, until cytoplasmic male sterile systems were available. If hybrid cultivars are to be developed from a crop, then the species must (1) show a high degree of heterosis; (2) be capable of producing inexpensive hybrid seed; (3) not easily be produced uniformly by other means; and (4) have a high premium for crop uniformity.

Hybrid cultivars have been developed in

- sorghum, onions, and other vegetables using a cytoplasmic male sterile (CMS) seed production system;
- in sugar beet and some Brassica crops (mainly Brussels sprouts, kale, and rapeseed) using CMS and self-incompatibility to produce hybrid seed;
- in tomato and potato using hand emasculation and pollination.

There are two major steps in producing hybrid cultivars: (1) develop inbred lines to be used as parents and (2) intercross these lines to identify parent combinations that give the best progeny. Inbred parental lines are developed in the same way as described above for inbreeding species, although pedigree methods are most common. After near-homozygous parents have been developed, their performance is first evaluated by test crosses, where each potential new parent is crossed to a parent of known worth and the progeny evaluated. After further parental selection in this manner, selected parents are intercrossed in different combination, and the progeny evaluated in yield trials to identify the most productive hybrid cross combination.

It is believed by many breeders (and geneticists) that the magnitude of heterosis is directly related to the degree of genetic diversity between the two parents. For this reason, it is common in most hybrid breeding programs to maintain two, or more, distinct germplasm sources (heterotic groups). Breeding and development is carried out within each source, and the different genetic sources are only combined in the actual production of new hybrid cultivars. For example, corn breeders in the United States found that they observed significant heterosis by crossing Iowa Stiff Stalk breeding lines with Lancaster germplasm. Since this discovery, these two different germplasm sources have not been intercrossed to develop new parental lines but, rather, have been kept genetically separated.

Schemes for Clonal Cultivars

Examples of clonal cultivars are: bananas (*Musa* spp.), cassava (*Manihot esculenta*), Citrus spp., potatoes (*Solanum tuberosum*), rubber trees (*Ficus elastica*), soft fruit (raspberry (*Rubus strigosus*), blackberry (*Rubus fruticosus*), and strawberry (*Fragaria*)), sugarcane (*Saccharum officinarum*), sweet potatoes (*Ipomoea batatas*), and top fruit (apples (*Malus*), pears (*Pyrus*), plums (*Prunus*), etc.). Clonal crops are often perennial, although several crop species, particularly those where the actual unit of clonal reproduction is the part of the plant that is exploited (e.g., tubers of potato and sweet potato) are treated in agriculture as annual crops. Clonal crops also include many long-lived tree crops (e.g., apple, cherry, rubber, and mango (*Mangifera indica*)), which can be productive crops for many decades after being established.

Methods of propagation are various. Rosaceous top fruits, citrus, avocado (*Persea americana*), and grape (*Vitis vinifera)* involve budding and grafting onto various rootstocks. Leafy cuttings are used for pineapple (*Ananas comosus*), sweet potato, and strawberry. Leafless stem cuttings are used in sugarcane, and lateral shoots are used for banana and palms. There is also, for a number of species, the potential for clonal reproduction via tubers (swollen stems, e.g., potatoes) bulbs, corms, etc.

In general, clonal crop species are often out-breeders that are basically intolerant to inbreeding. Individual clones are genetically heterozygous and so it is easy to exploit the presence of any heterosis.

The process of developing a clonal cultivar is, in principle, very simple. Breeders generate segregating progenies of seedlings, select the most productive genotypic combination, and simply multiply this asexually; thus there is no need for extra procedures to stabilize the genetic makeup (i.e., it relies on asexual reproduction, thus avoiding problems relating to genetic segregation arising from meiosis). Despite the apparent simplicity of clonal breeding, it should be noted that while clonal breeders have shared in some outstanding successes, it has rarely been due to such a simple process.

In the case of potato, the length of the breeding process is partly related to a slow multiplication rate. In addition, seed tubers are bulky and require large amounts of storage space. To accommodate planting material for 1 acre of potatoes will require approximately 2000 lb of seed tubers. With many other clonal species, there may be some considerable time between crossing to first selection. In apple breeding, for example, if a breeder is successful with the very first cross combination made, then it is still unlikely that a cultivar will be released (from that cross) by the time the breeder retires! Finally, many diseases and viruses are transmitted vegetatively and great care needs to be taken to ensure that breeding lines do not become infected.

New Genetic Approaches

Tissue Culture (*In Vitro*) Techniques

A variety of techniques (micropropagation, haploid production, protoplasts, embryo culture, apical culture, somatic embryogenesis, etc.) have been developed under the title of tissue culture and so just two particular examples are noted here to give an idea of the possible applications.

Haploid Production

Producing inbred (true-breeding) homozygous lines is an essential part of developing new cultivars in many crop species. These homozygous lines can be used directly as cultivars (i.e., for inbreeding crop species) or as parents to produce hybrids which are then exploited. Plant breeders in the past have used the process of selfing to achieve homozygosity, which is a time-consuming process. Therefore the possibility

of producing plants from gametic, haploid cells has been an ambition as a way to produce 'instant' inbred lines (after chromosomes of the haploids are doubled).

Haploid gametes are produced during normal meiosis and so are obvious targets for exploitation in obtaining homozygous lines. If such gametic, haploid cells can be stimulated to develop into plantlets, a haploid plant can develop, which can then be encouraged to double its chromosomes complement, to produce a completely homozygous line (a doubled haploid).

Although haploidy is a very attractive technique the natural rate of occurrence of haploid plants is low. However, it is possible to produce plants from gametic cells at a higher frequency by culturing plant tissue *in vitro*.

Although male and female sex cells might be used to give rise to haploid plants, it is the male cells (microspores or pollen) that have been exploited most successfully for the regeneration of large numbers of haploid and doubled-haploid lines. This is in part because of the ease with which pollen, as opposed to eggs, can be collected, and in part because pollen grains are produced in much greater numbers than eggs.

Other approaches to deriving haploids are also being used, and one that has been successful in some specific cases is interspecific or intergeneric crossing. With the right cross, rather than giving rise to a hybrid, such crosses give haploid plants and examples of successful crosses are *S. tuberosum* (cultivated European potato) crossed with a wild relative *Solanum phureja*; *H. vulgare* (cultivated barley) crossed with *Hordeum bulbosum*; and *Triticum aestivum* (bread wheat) crossed with *Z. mays* (corn). The results of these crosses are now leading to cultivars being widely released.

In Vitro *Multiplication*

In vitro multiplication of breeding lines has two major advantages in plant breeding programs: (1) plants propagated *in vitro* can generally be initiated and maintained in a disease-free state, and so can be used to maintain stocks of breeding lines, facilitate long-term germplasm storage, and smooth the progress of international germplasm exchange; (2) short generation times and fast growth mean that rapid increases in plant number can be achieved.

Both the above are important with clonal crops in which there is a tendency for relatively low multiplication rates (a property of their vegetative propagation) and which are particularly susceptible to viral and bacterial diseases (which tend to be multiplied and transmitted through each clonal generation). Good examples of maintaining high disease status and offering rapid plant regeneration potential include potato and strawberry.

Plant Transformation

Using plant transformation techniques makes possible the transfer of lengths of DNA (single genes, i.e., simply inherited traits) into plants, to have such 'transgenes' expressed, and to have them function successfully in their new genome. In theory, any gene can be transferred from any source into a developed cultivar or advanced breeding line, and this can be achieved in a single step. So, recombinant DNA techniques allow breeders to transfer genes between completely unrelated organisms. For example, bacterial genes can be transferred and expressed in plants. Thus plant transformation has added to the tools available to the breeder for genetic manipulation, but it does, as with all techniques, have its own limitations. Some of the limitations will diminish as the protocols are further developed; others are inherent to the basic approach. At present, recombinant DNA techniques can generally only transfer single genes. This means that they are very effective where the trait can be affected significantly by one gene, or a few genes, of large effect. This means that such genes need to have been identified and cloned. The number of such identified, desirable genes is still finite, although increasing rapidly.

Already there is a growing list of crop species that have proved successful hosts for transformation including alfalfa, apple, barley, carrot (*Daucus carota*), cauliflower (*Brassica oleracea* var. *botrytis*), celery (*Apium graveolens* var. dulce), cotton (*Gossypium*), cucumber (*Cucumis sativus*), flax, horseradish (*Armoracia lapathifolia*), lettuce (*Lactuca sativa*), corn, peas, potato, oilseed rape, rice, rye, sugar beet, soybean, sunflower (*Helianthus*), tomato, tobacco, walnut (*Juglans*), and wheat.

The first transgenic crops were ones changed in terms of modifying or enhancing traits that related directly to traditional farming activities. These included the control of insects, weeds, and plant diseases. More recently work has focused on more consumer-orientated goals such as altering end-use quality (including oil composition, starch, vitamin level, and even the inclusion of vaccines).

There has been considerable public debate concerning the application of plant transformation technology, particularly as new transgenic crops have been released into commercial cultivation and as the products have entered the human food chain. Plant breeders must be aware of the concerns as well as the regulations that apply to plants derived using recombinant DNA. This means that alongside the general social and environmental concerns, the breeder must be convinced that the techniques being used are the most effective for what is to be achieved and not simply assume that 'high tech' means most efficient! More recently a number of breeding groups have been utilizing plant transformation techniques to transfer desirable genes from wild weedy relatives to crop species. Plants developed using this system are not regarded as *transgenic* but rather *cisgenic*. The transformed genes (traits) in cisgenic plants could have been incorporated into the crop plants by traditional crossing and backcrossing and hence may not require the stringent regulatory testing or have some of the negative public conceptions that are associated with transgenic crop, where genes are transferred from completely unrelated species. The advantages of cisgenic over traditional breeding are (1) time to incorporate the gene of interest and (2) elimination of all the undesirable genes (even those with high linkage to the gene of interest) from the wild parent plant.

Molecular Markers in Plant Breeding

As noted initially, plant breeders have practiced their 'art' for many centuries, but genetics, as a subject, was only recognized in the twentieth century, with the rediscovery of Mendel's work. From that point, research in genetics has developed

rapidly but we still have little, or no, information in most crops about: (1) the locations of many of the loci of interest or even which chromosome they are on; (2) the number of loci affecting any trait we are interested in; and (3) the effect of an individual allele at a locus on the observed phenotype, except where there is an obvious major effect (e.g., height and dwarfing genes).

One idea for helping to increase our knowledge was to try to associate easily visualized markers with loci that were affecting variation in traits of interest, as proposed by Sax as early as 1923. The idea is that often characters of interest to plant breeders are difficult to evaluate, because they show continuous variation, or assessment is detailed and time-consuming, or the trait is only expressed after several years of growth. If the genes controlling these characters are closely linked to loci controlling a second, easily assessable trait, then it would be possible to select for the difficult characters by assessing the easily identifiable one.

The characteristics of a good marker system are

- the markers are easy, quick, and inexpensive to score,
- the markers have no deleterious effects on fitness,
- they have no effects on other traits,
- they show a high level of variation,
- they are stably expressed in different environments,
- they can be assessed in early growth phases of the plant (seedling level), and/or in tissue culture,
- the scoring of the markers can be achieved by nondestructive means,
- heterozygotes can be differentiated from either homozygous genotype (show codominance).

The types of markers that have been used in plant breeding include

1. Morphological markers, ones whose variation can be seen by simply looking at a plant's phenotype, such as pigmentation, dwarfism, leaf shape, absence of petals, etc.
2. Biochemical markers, such as variant forms of an enzyme, which are functionally identical but can be distinguished by electrophoresis (isozymes). Different forms of the enzyme will migrate to different points depending on their charge, size, and shape.
3. Molecular markers, which represent the variation present at the DNA level. So molecular markers are simply differences in the DNA itself between individuals, groups, species, taxa, etc. The main characteristics of molecular markers are that they are found in virtually all organisms; are not affected by the environment; show high levels of polymorphism; have no effects on the phenotype; and can be detected using small pieces of tissue.

Given the above characteristics and their relatively unlimited numbers, it is no surprise that the advent of molecular markers in the 1990s was seen as a major step forward.

Molecular markers can be used in plant breeding for:

1. Differentiating one cultivar from another (perhaps one already commercially available), or to prove proprietary ownership of specific cultivars (DNA fingerprinting). A further possibility of DNA fingerprinting is to assess how diverse genotypes are at the DNA level and hence assess their level of difference (genetic distance) for selecting parents when developing hybrid cultivars.
2. Marker-assisted selection (MAS) as a selection tool to identify genotypes with desirable genes, particularly when the gene (or genes) of interest are difficult to screen for phenotypically. MAS is especially effective when the molecular marker is indeed the gene of interest or is tightly linked to the gene of interest.
3. Marker-assisted backcrossing (MAB). When a gene of interest can be shown to be linked to a molecular marker, then assessment of the marker can help accelerate the backcrossing process. Mature plants would not need to be grown to identify which backcross individuals carry the allele of interest. Molecular markers can identify which of the backcross progeny have better restoration of the recurrent (adapted) parent and hence reduce the number of backcrossing generations necessary to eliminate the negative wild-type genes.
4. Quantitative trait loci (QTL) selection can be used if a genetic map based on a mapping population is available, as well as trait data collected from the same population. QTL markers can often indicate regions of chromosomes which have a high frequency of desirable genes for quantitatively important trails like yield. QTLs can be used to conduct selection based on the presence/absence of molecular markers genetically linked to a given QTL instead of running selection based on phenotypic assessments of those traits, reducing time and expenses.
5. Genomic-wide association studies (GWAS) have been suggested to overcome the lack of breeding relevance of QTL markers. GWAS approaches have been developed which enable genetic mapping to be carried out in set of genotypes rather than in mapping populations. For instance, an association mapping project might assemble several hundred individuals encompassing elite breeding lines, breeding germplasm, and other sources of genetic variation. The molecular markers most often used in association mapping approaches are SNPs. Using sophisticated analyses, statistically significant marker-trait associations are established which can be used to select genetically desirable breeding lines.

Future Potential

Plant breeding will continue to rely on classical techniques of crossing, trialing, and selection but will be helped by the application of the new approaches that are developing so quickly. These new techniques will be used, but in conjunction with the more established ones. Plant breeding will therefore continue to change to take into account new possibilities and new techniques but with a firm base in traditional approaches.

See also: **Plant Breeding and Genetics:** Marker-Assisted Selection; Molecular Markers; Mutation Techniques. **Postharvest Biology:** Genetic Engineering for Postharvest Quality. **Tissue Culture:** Somatic Hybridization.

Further Reading

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