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GENETIC MARKERS IN PLANT: CONCEPTIONS, TYPES AND ITS MEDICINAL AND BREEDING APPLICATION

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Abstract: Genetic markers are the biological features that are determined by allelic forms of genes or genetic loci and can be transmitted from one generation to another, and thus they can be used as experimental probes or tags to keep track of an individual, a tissue, a cell, a nucleus, a chromosome or a gene In terms of scientific progress, the old disciplines of quantitative genetics and plant taxonomy have been revived by the molecular marker approach. The progress made in DNA marker technology has been tremendous and exciting. DNA markers have provided valuable tools in various analyses ranging from phylogenetic analysis to the positional cloning of genes. The development of high-density molecular maps which has been facilitated by PCR-based markers, have made the mapping and tagging of almost any trait possible. Ever since their invention, they are being, constantly modified for enhanced utility as a means to solve problems and to bring about automation in the genome analysis, gene tagging, phylogenic analysis and selection of desirable genotypes. It is also evidence that molecular markers (non morphological markers) offer several advantages over the morphological markers (conventional phenotypic markers), as they provide data that can be analyzed objectively; giving new dimension to breeding especially with respect to the time required to developing new improved crop varieties as well utmost importance in pharmaceuticals for increase in yield as well in vaccine production. This review presents a basic description of different molecular techniques that can be utilized for DNA fingerprinting and molecular diversity analysis of plant species as well plant breeding.

Keywords: **PCR**: Polymerase Chain Reaction, **RAPD**: Random Amplification of Polymorphic DNA, **AFLP**: Amplified Fragment Length Polymorphism. **RFLP**: Restriction Fragment Length Polymorphism, **MAS**: marker- assisted selection

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INTRODUCTION

DNA markers are defined as a fragment of DNA revealing mutations/variations, which can be used to detect polymorphism between different genotypes or alleles of a gene for a particular sequence of DNA in a population or gene pool. Such fragments are associated with a certain location within the genome and may be detected by means of certain molecular technology. Simply speaking, DNA marker is a small region of DNA sequence showing polymorphism (base deletion, insertion and substitution) between different individuals. There are two basic methods to detect the polymorphism: Southern blotting, a nuclear acid hybridization technique [1], and PCR, a polymerase chain reaction technique.[2] Using PCR and/or molecular hybridization followed by electrophoresis (e.g. PAGE – polyacrylamide gel electrophoresis, AGE – agarose gel electrophoresis, CE – capillary electrophoresis), the variation in DNA samples or polymorphism for a specific region of DNA sequence can be identified based on the product features, such as band size and mobility. In addition to Sothern blotting and PCR, more detection systems have been also developed. For instance, several new array chip techniques use DNA hybridization combined with labeled nucleotides, and new sequencing techniques detect polymorphism by sequencing.

The theoretical advantages of using genetic markers and the potential value of genetic marker linkage maps and direct selection in plant breeding were first reported about eighty years ago.[3] However, it was not until the advent of DNA marker technology in the 1980s, that a large enough number of environmentally insensitive genetic markers generated to adequately follow the inheritance of important agronomic traits and since then DNA marker technology has dramatically enhanced the efficiency of plant breeding. DNA-based molecular markers have acted as versatile tools and have found their own position in various fields like taxonomy, plant breeding, genetic engineering e.t.c .[4]

A number of breeding companies have in the past two decades to varying degrees started using markers to increase the effectiveness in breeding and to significantly shorten the development time of varieties and therefore plant geneticist consider molecular marker assisted selection a useful additional tool in plant breeding programs to make selection more efficient.[5] over the last few decades plant genomics has been studied extensively bring about a revolution in this area, making molecular markers useful for plant genomic analysis, therefore becoming an important tool in this revolution.[4]

Molecular markers include biochemical constituents (e.g. secondary metabolites in plants) and macro-molecules, viz proteins and deoxyribonucleic acid (DNA). Analysis of secondary metabolites is, however restricted to those plants that produce a suitable range of metabolites which can be easily analyzed and which can be distinguished by varieties.[4] These metabolites

which are being use as markers should be ideally neutral to environmental effects or management practices. Hence, amongst the marker molecular markers used, DNA markers are more suitable and ubiquitous to most of the living organisms.

Diversity based on phenotypic and morphological characters, usually varies with environments and evaluation of traits requires growing the plants to full maturity prior to identification, but now the rapid development of biotechnology allows easy analysis of large number of loci distributed throughout the genome of the plants. Molecular makers have proven to be powerful tools in the assessment of genetic variation and in elucidation of genetic relationships within and among species .[6]

Molecular markers for classification of genotype are abundant, but unlike morphological traits, markers are not affected by environment. Collecting DNA marker data to determine whether phenotypically similar cultivars are genetically similar would therefore be of great interest in crop breeding programme.[7]

DNA-based molecular markers have proved their utility in fields like taxonomy, physiology, embryology, genetics, etc. As the science of plant genetics progressed, researchers have tried to explore these molecular marker techniques for their applications in commercially important plants such as food crops, horticultural plants, etc. and recently in pharmacognostic characterization of herbal medicine. It has been well documented that geographical conditions affect the active constituents of the medicinal plant and hence their activity profiles[8].

Molecular genetics or the use of molecular technique for detecting differences in the DNA of individual plants has many applications of value to crop improvement. The differences are called molecular markers because they are often associated with specific gene and acts as a ‘sign posts’ to those genes and such markers when very tightly linked to genes of interest, can be use to select indirectly for the desirable allele and this represents the simplest form of marker- assisted selection (MAS).[9]

Markers can also be used for dissecting polygenic traits into their Mendelian components or Quantitative Trait Loci (QTL) and this increasing understanding of the inheritance and gene action for such traits allows the use of markers – selection procedures.[10]

The molecular markers are no longer looked upon as simple DNA fingerprinting markers in variability studies or as mere forensic tools, but, they are constantly being modified to enhance their utility and to bring about automation in the process of genome analysis (Joshi et al, 2011). The discovery of polymerase chain reaction (PCR) was a land mark in this effort and proved to be a unique process that brought about a new class of DNA profiling marker, which has facilitated the development of marker-based gene tags, map-based cloning of agronomic

important genes, variability studies, phylogenetic analysis, synteny mapping, market assisted selection of desirable genotypes e.t.c. DNA markers offer several advantages over traditional phenotypic markers, as they provide data that can be analyzed objective. Therefore, several molecular marker types are available and they each have their advantages and disadvantages.[11]

MOLECULAR MARKERS

Molecular markers are specific fragments of DNA that can be identified within the whole genome. Molecular markers are found at specific locations of the genome. They are used to 'flag' the position of a particular gene or the inheritance of a particular character. Molecular markers are phenotypically neutral.

Molecular markers are identifiable DNA sequence, found at specific locations of the genome and associated with the inheritance of a trait or linked gene.[12] Molecular markers as naturally occurring polymorphism which include proteins and nucleic acids that are detectably different. Rapid advances are genome research and molecular biology has led to the use of DNA markers in plant breeding. Target genes in a segregating population can be identified with the assistance of DNA makers so as to accelerate traditional breeding programs.[13] Markers must be polymorphic (i.e. they must exist in different forms so that the chromosome carrying the mutant gene can be distinguished from the chromosome with normal gene by form of the marker it carries. Polymorhism can be detected at three levels: morphological, biochemical or molecular. Recently, the term DNA fingerprinting /profiling is used to describe the combined use of several single locus detection systems and are being used as versatile tools for investigating various aspects of plant genomes. These include characterization of genetic variability, genome fingerprinting, genome mapping, gene localization, analysis of genome evolution, population genetics, taxonomy, plant breeding and diagnostics.[4] The development of DNA (or molecular markers) has irreversibly change the disciplines of plant genetics and breeding.[14]An ideal DNA makers should however poses the following properties.(i) Highly polymorphism, which is the simultaneous occurrence of a trait at the same population of two or more discontinues variants or genotypes. [4]

(i) Co dominant inheritance- different form of marker should be detected in a diploid organism to allow discrimination of homozygote and heterozygote.

(ii) Frequent occurrence in genome

(iii) Selective neutral behaviour (the DNA sequences of any organism are neutral to environmental conditions or management practices)

(iv) Easy access (availability)

(v) Easy and fast assay

(vi) Reproducible – highly reproducibility and

(vii) Easy exchange of data between laboratories.

It is extremely difficult for a single genetic marker to possess all properties above. Depending on the type of study to be undertaken a marker system can be identified that would fulfil at least a few of the above characteristics.

I. Non-PCR based genetic markers

1. Restriction fragment length polymorphism

The first and forest molecular markers system called the Restriction Fragment length Polymorphism (RFLP), was developed in early 1980 .[15] The RFLPs are simply inherited naturally occurring Mendelian characters. Genetic information is stored in the DNA sequence on a chromosome and variation in this sequence is the basis for the genetic diversity within species. Plants are able to replicate their DNA with high accuracy and rapidity, but many mechanisms causing changes (mutation) in the DNA are operative.[4]

RFLP markers are the first generation of DNA markers and one of the important tools for plant genome mapping. They are a type of Southern- blotting based markers. In living organisms, mutation events (deletion and insertion) may occur at restriction sites or between adjacent restriction sites in the genome. Gain or loss of restriction sites resulting from base pair changes and insertions or deletions at restriction sites within the restriction fragments may cause differences in size of restriction fragments. These variations may cause alternation or elimination of the recognition sites for restriction enzymes. As a consequence, when homologous chromosomes are subjected to restriction enzyme digestion, different restriction products are produced and can be detected by electrophoresis and DNA probing techniques.

RFLP markers are powerful tools for comparative and synteny mapping. Most RFLP markers are co-dominant and locus-specific. RFLP genotyping is highly reproducible, and the methodology is simple and no special equipment is required. This marker was first reported by Botstein[16] in the detection of DNA polymorphism.[17]

II. POLYMERASE CHAIN REACTION BASED MARKERS

A decade after the emergence of AFLP, there was another breakthrough which involves the use of PCR in 1990.[15]PCR is an in vitro method of nucleic acid synthesis by which a particular segment of DNA can be specifically replicated .[18] The process involves two oligonucleotide primers that flank the DNA fragment of interest and amplification is achieved by a series of

repeated cycles of heat denaturation of the DNA, annealing of the primer to their complementary sequences, and extension of the annealed primers with a thermophilic DNA polymerase. Since the extension products themselves are also complementary to primers, successive cycles of amplification essentially double the amount of the target DNA synthesized in the previous cycle and the result is an exponential accumulation of the specific target fragment.

Genomic DNA from two different individual often produces different amplification and a particular fragment generated from one individual but not for other represent DNA polymorphism and can be used as genetic markers. The pattern of amplified bands so obtained could be use for genomic fingerprint. [19]

1. RANDOMLY - AMPLIFIED POLYMORPHIC DNA MARKER (RAPD)

In 1991 Welsh and McClelland[19] developed a new PCR-based genetic assay namely randomly amplified polymorphic DNA (RAPD). This procedure detects nucleotide sequence polymorphisms in DNA by using a single primer of arbitrary nucleotide sequence. In this reaction, a single species of primer anneals to the genomic DNA at two different sites on complementary strands of DNA template. If these priming sites are within an amplifiable range of each other, a discrete DNA product is formed through thermocyclic amplification. On an average, each primer directs amplification of several discrete loci in the genome, making the assay useful for efficient screening of nucleotide sequence polymorphism between individuals[20]. However, due to the stoichastic nature of DNA amplification with random sequence primers, it is important to optimize and maintain consistent reaction conditions for reproducible DNA amplification. They are dominant markers and hence have limitations in their use as markers for mapping, which can be overcome to some extent by selecting those markers that are linked in coupling[21]. RAPD assay has been used by several groups as efficient tools for identification of markers linked to agronomically important traits, which are introgressed during the development of near isogenic lines. The application of RAPDs and their related modified markers in variability analysis and individual-specific genotyping has largely been carried out, but is less popular due to problems such as poor reproducibility faint or fuzzy products, and difficulty in scoring bands, which lead to inappropriate inferences.

RAPD-based molecular markers have been found to be useful in differentiatingdifferent accessions of Taxus wallichiana[22],Neem [23],Juniperus communisL.[24],Codonopsis pilosula [25], Allium schoenoprasum L.[26] ,Andrographis paniculata [27]collected from different geographical regions.

Some variations in the RAPD technique include

DNA amplification fingerprinting (DAF).

Caetano-Anolles et al.[28] employed single arbitrary primers as short as 5 bases to amplify DNA using polymerase chain reaction. In a spectrum of products obtained, simple patterns are useful as genetic markers for mapping, while more complex patterns are useful for DNA fingerprinting. Band patterns are reproducible and can be analysed using polyacrylamide gel electrophoresis and silver staining. DAF requires careful optimization of parameters; however, it is extremely amenable to automation and fluorescent tagging of primers for early and easy determination of amplified products. DAF profiles can be tailored by employing various modifications such as predigesting of template. This technique has been useful in genetic typing and mapping.

Arbitrary primed polymerase chain reaction (AP-PCR).

This is a special case of RAPD, wherein discrete amplification patterns are generated by employing single primers of 10–50 bases in length in PCR of genomic DNA[29]. In the first two cycles, annealing is under non-stringent conditions. The final products are structurally similar to RAPD products. Compared to DAF, this variant of RAPD is not very popular as it involves autoradiography. Recently, however, it has been simplified by separating the fragments on agarose gels and using ethidium bromide staining for visualization.

Advantages associated with RAPD analysis include:

(i) Use of small amount of DNA which makes it possible to work with population that is not accessible with RFLP. It is fast and efficient in analysis having high-density genetic mapping as in many plant species such as alfafa ,[30] fabean bean [31] and apple[32]

(ii) Non involvement with radioactive assays [30]

(iii) Non – requirement of species specific probe librararies

(iv) Non – involvement in blotting or hybridization

Limitations of RAPD markers are:

(i) Its polymorphisms are inherited as dominant or recessive characters causing a loss of information relative to markers which show co-dominance.

(ii) Primers are relatively short, a mismatch of even a single nucleotide can often prevent the primer from annealing, hence leads to a loss of band.

(iii) Suffers from problems of repeatability in many systems, especially when transferring between populations or laboratories as is frequently necessary with marker assisted selection programs .[33]

2. AMPLIFIED FRAGMENT LENGTH POLYMORPHISM (AFLP)

A recent approach known as AFLP [34] is a technique based on the detection of genomic restriction fragments by PCR amplification and can be used for DNAs of any origin or complexity. The fingerprints are produced, without any prior knowledge of sequence, using a limited set of generic primers. The number of fragments detected in a single reaction can be 'tuned' by selection of specific primer sets. AFLP technique is reliable since stringent reaction conditions are used for primer annealing. This technique thus shows an ingenious combination of RFLP and PCR techniques[35]and is extremely useful in detection of polymorphism between closely related genotypes.

AFLPS are fragments of DNA that have been amplified using directed primers from restriction of genomic DNA. In this approach the sample DNA is enzymatically cut up into small fragments (as with RFLP analysis), but only a fraction of fragments are studied following selective PCR amplification [33]. It is a combination of RFLP and RADP methods. AFLP technique shares some characteristic with both RFLP and RAPD analysis [15] and combines the specifically of restriction analyses with PCR amplification. AFLP is extremely sensitive technique and the added use of fluorescent primers for automated fragment analysis system and software packages to analyze the biallelic data makes it well suitable for high thorough put analysis.

AFLP procedure mainly involves 3 steps

(a) Restriction of DNA using a rare cutting and a commonly cutting restriction enzyme simultaneously (such as *Mse*I and *Eco*RI*)*followed by ligation of oligonucleotide adapters, of defined sequences including the respective restriction enzyme sites.

(b) Selective amplifications of sets of restriction fragments, using specifically designed primers. To achieve this, the 5' region of the primer is made such that it would contain both the restriction enzyme sites on either sides of the fragment complementary to the respective adapters, while the 3' ends extend for a few arbitrarily chosen nucleotides into the restriction fragments.

(c) Gel analysis of the amplified fragments.

The major advantages of AFLP techniques [15] are: (i) generation of a large number of polymorphism.

(i) No sequence information is required

(ii) The PCR technique is fast with high multiplex ratio which makes the AFLP very attractive choice.

The problems associated with AFLPs are of three types and all are related with practical handling, data generation and analysis. These problems are not unique to AFLP technology but also associated with other markers systems.

An ideal marker should have sufficient variation for the problem under study, be reliable and simple to generate and interpret. Unfortunately, neither AFLP nor other DNA markers exhibit these qualities. Thus a specific technique or techniques selected on the basis of objectives be utilized collectively to achieve the best results .[36-37]

3. SIMPLE SEQUENCE REPEAT OR SHORT TANDEM REPEATS (SSRS) OR MICRO SATELLITES

A site in the genome that contains many short tandem repeat sequences (microsatellites). These sites are usually in the size range of 100-500 base pairs composed of dinucleotide and trinucleotide repeats. They are very polymorphic, scattered throughout genomes. Genomes typically contain 1,000s of SSRs! They are detected by PCR using primers flanking the repeats and resolved on gels.

"Microsatellite or short tandem repeats or simple sequences repeats (SSR) are monotonous repetitions of very short (one to five) nucleotide motifs, which occur as interspersed repetitive elements in all eukaryotic genomes".[38] However, strand slippage during DNA replication, where the repeats allow matching through excision or addition of repeats, results in disparity in the number of tandemly repeated units.[39]As slippage in replication is more expected than point mutations, microsatellite loci tend to be hypervariable. Hon et al. reported the efficiency of SSR markers in genetic authentication of two *Panax* species[40]. SSR markers used to identify and differentiate American ginseng and Oriental ginseng, cultivated and wild American ginseng [41]. Eight polymorphic microsatellite loci developed for the Chinese medicinal plant *Artemisia annua* L. (Asteraceae), useful for investigating the genetic diversity, genetic structure and gene flow within populations. [42] Li et al. [43] Zhou et al. [44] used 10 microsatellite (SSR) loci to investigate genetic diversity and differentiation in 16 natural populations of *Saruma henryi*Oliv. Use of cross-species SSR markers was reported in genetic diversity analysis of synthetic interspecific hybrid of *Hibiscus* which revealed a closer association of diploid genomes and high variability of tetraploid genomes [45]. Most recently, Katoch et al. [46] used DNA-based molecular marker techniques, viz. simple sequence repeats (SSR) and cytochrome P-450 markers to estimate genetic diversity in *Picrorhiza kurrooa*.

Although SSRs are usually considered just as evolutionary neutral DNA markers, the functional significance of a substantial part of SSRs has been proven by critical tests in various biological phenomena, as shown in Fig.1

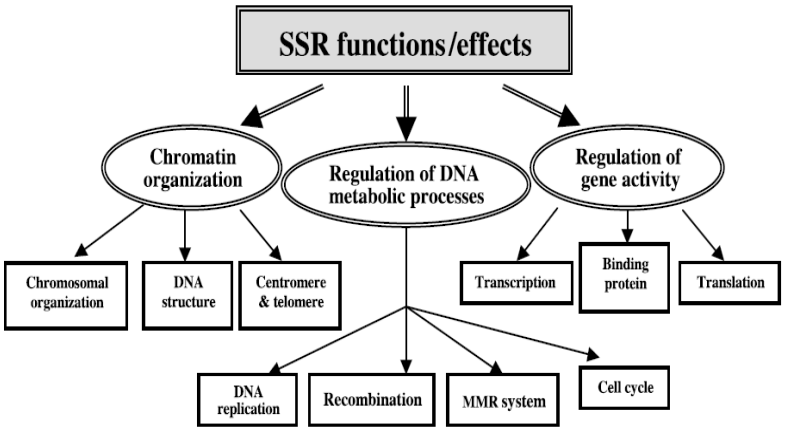

Fig.1 SSR putative functions/effects.[47]

III. MARKER ASSISTED SELECTION (MAS)

In plant breeding, MAS is a relatively new concept, nevertheless the original selection concept per se has not changed, that is, the purpose of the selection is to search and preserve the best genotypes, but using molecular markers. MAS can be used for manipulating both qualitative and quantitative traits. A highly saturated marker linkage map is necessary for effective marker based selection. Basically, MAS consists of identifying association between molecular markers and genes controlling agronomic traits, and using these to improve lines or populations [48]. Otherwise, based on the heritability of molecular markers, which is essentially 100 %, when the selection is performed for a low heritable trait, it will be more effective and potentially less expensive than phenotypic selection.[49] It is necessary to consider that effectiveness and cost of MAS is greatly influenced by the marker system used, therefore it must be chosen carefully [50]. Concerning MAS applied to qualitative traits, Huang et al. [51] using DNA marker-assisted selection developed rice lines containing two, three, and four resistance genes to the bacterial blight pathogen (Xanthomonas oryzae pv. orizae). MAS refer to the use of DNA markers that are tight-linked to target loci as a substitute for or to assist phenotype screening. By determining the allele of a DNA marker, plants that possess particular genes or quantitative trait loci (QTL) may be identified based on their genotype rather than their phenotype.

MARKER ASSISTED SELECTION OF DESIRABLE CHEMOTYPES

Along with authentication of species identity, prediction of the concentration of active phytochemicals may be required for quality control in the use of plant materials for

pharmaceutical purposes. Identification of DNA markers that can correlate DNA fingerprinting data with quantity of selected phytochemical markers associated with that particular plant, would have extensive applications in quality control of raw materials. AFLP analysis has been found to be useful in predicting phytochemical markers in cultivated Echinacea purpurea[52] germplasm and some related wild species. RAPD fingerprint has been developed to support the chemotypic differences in oil quality of three different genotypes of Pelargonium graveolens[53] and flavonoid composition of Aconitum[54] species. DNA profiling has been used to detect the phylogenetic relationship among Acorus calamus chemotypes differing in their essential-oil composition.[55] Artemisia annua, a source of antimalarial compound artemisinin, shows variation in artemisinin content all over India. These chemotype variants of A. annua L. have been characterized using RAPD markers. This study also revealed existence of high levels of genetic variation in the Indian population despite geographical isolation and opens out a possibility of further genetic improvement for superior artemisinin content. An attempt has also been made to study variation in essential-oil components and interspecific variations using RAPD technique.[56] Morphological, chemical and genetic differences in twelve basil (Ocimum gratissimum L.) accessions were studied to determine whether volatile oil and flavonoids can be used as taxonomical markers and to examine the relation between RAPDs and these chemical markers.[57]

IV. BREEDING OF POLYGENIC TRAITS

The utilization of markers can obviously prevent loss of quantitative trait loci (QTL) common with some crops DNA markers and this allow us to unravel the genetic basis of traits expressing continuous phenotypic variations as they are abundant and scattered throughout the genome. By using dense genetic marker maps, the contributions of separate regions of the genome on the trait values can be estimated once the mapping population is sufficiently large. In addition, agronomic important traits like nutritional quality, yield, flower time and durable resistance which appear to follow complex, polygenic inheritance patterns with multiple genes having small effects on the trait value can easily by analyzed using markers. Evidences obtained from various crops indicate that even such complex traits appear to be determined by only a few major factors/genes. [58-59]

DNA MARKERS AS NEW PHARMACOGNOSTIC TOOL

Traditionally, pharmacognosy mainly addressed quality related issues using routine botanical and organoleptic parameters of crude drugs. Pharmacognosy became more interdisciplinary because of subsequent advances in analytical chemistry. The new pharmacognosy includes all aspects of drug development and discovery, where biotechnology-driven applications will play an important role. Extensive research on DNA-based molecular markers is in progress in many

research institutes all over the world. This technique remains important in plant genome research with its applications in pharmacognostic identification and analysis. These markers have shown remarkable utility in quality control of commercially important botanicals like Ginseng, Echinacea, Atractylodes. In India several agricultural universities and research institutes are actively involved in exploring DNA-based techniques in genotyping of medicinal plants. Although considerable progress has been made in DNA marker technology, applications of these techniques for characterizing semi-processed and processed botanical formulations to ensure the desirable quality remain underutilized. Although DNA analysis is currently considered to be cutting-edge technology, it has certain limitations due to which its use has been limited to academia. In order to establish a marker for identification of a particular species, DNA analysis of closely related species and/or varieties and common botanical contaminants and adulterants is necessary, which is a costly and time-consuming process. Isolation of good-quality DNA suitable for analysis from semi-processed or processed botanicals is also a challenge. Another important issue is that DNA fingerprint will remain the same irrespective of the plant part used, while the phytochemical content will vary with the plant part used, physiology and environment. DNA fingerprinting ensures presence of the correct genotype but does not reveal the contents of the active principle or chemical constituents. Hence DNA analysis and pharmacognostic techniques for chemoprofiling such as TLC, HPTLC, etc. will have to be used hand in hand rather than in isolation. Identification of quantitative-trait loci[34] that are closely linked to a biologically active phytochemical will prove to be useful. Several attempts have been made in recent years, to correlate DNA markers with qualitative and quantitative variations in phytochemical composition among closely related species[60–64]. Proper integration of molecular techniques and analytical tools will lead to the development of a comprehensive system of botanical characterization that can be conveniently applied at the industry level for quality control of botanicals. Ayurvedic classification of medicinal plant is based on basic principles and therapeutic characters that may have a genetic basis. We have undertaken an exploratory study on the use of molecular markers for quick identification of botanical materials in crude, semi-processed and processed herbal formulations. Our strategy involves identification of species-specific marker after screening a number of species and/or varieties of the medicinal plant using random oligonucleotide primers, followed by cloning and subsequently converting it to SCAR markers for better specificity and reproducibility.

NEW ROLES OF PLANT BREEDING

The traditional roles of plant breeding will continue to be important. The technology for using plants as bioreactors to produce pharmaceuticals will advance; this technology has been around for over a decade. Strategies are being perfected for use of plants to generate pharmaceutical antibodies, engineering antibody-mediated pathogen resistance, and altering

plant phenotypes by immunomodulation. Successes that have been achieved include the incorporation of Streptococcus surface antigen in tobacco, and the herpes simplex virus in soybean and rice.

BREEDING PLANTS FOR NOVEL TRAITS

An application of genetic engineering to breed novel traits is the use of organisms as bioreactors to produce pharmaceuticals. One of the earliest applications of this technology was the commercial production of human insulin in microbial systems. Similarly, certain pharmaceuticals are commercially produced in mammalian milk of sheep, goats, and rabbits. The application is being applied to plants to produce selected chemical compounds. Plant-made vaccines are currently under development for protection against cholera, diarrhea (Norwalk virus), and hepatitis B. The most common plants that are being used in plant-made pharmaceuticals are corn, tobacco, and rice. Other crops being investigated include alfalfa, potato, safflower, soybean, sugarcane, and tomato. To be usable, the plant should be readily amenable to genetic engineering and capable of producing high levels of protein. Further, there should be an efficient method for extracting the protein products from the plant tissues. Another example of a plant-manufactured pharmaceutical is taxol, a secondary product derived from the Pacific yew tree. This product has been found to be effective against certain cancers.

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