STUDY ON MOLECULAR PLANT IN BIOTECHONOLOGY USING DNA TECHINIQUES- A RESERCH

M.Roopa Devi
M.Sc. Biotechnology
JNTU Hyderabad

Abstract

The life sciences offer open doors for changing human welfare exercises. Advanced by contributions from genomic explore, biotechnology is a noteworthy power for improvement in all nations. Weaved with culture and socio-moral esteems, biotechnology add to tackling issues like sustenance and water frailty that obstruct national advancement and debilitate peace in the creating scene. The absence of offices and expert abilities in biotechnology limits R and D activities in the creating and the slightest created nations (LDCs); and, confines their full interest in take-off exercises in national and independent local ventures in practical improvement. The act of biotechnology diverse in many creating nations is all things considered noteworthy. The foundation of biotechnology parks and therapeutic plant cultivates in a few creating nations is characteristic of biotechnology being concurred high strategy status in national improvement; of its importance in the annihilation of destitution; and of its utilization in the strengthening of ladies in applying the innovation for human and social welfare. This survey gives a few cases of various sorts of biotech exercises that are being utilized for advancement in the creating scene.

Keywords: *Biotechnology and development, Biotechnology parks, Capacity-building.*

1.0 Introduction:

Before farming, people lived as traveling seekers and could survive exclusively on wild plant and creature assets. Seeing the huge abundance of the plant and creature kingdoms, their effective endeavors to tame the wild species propelled farming. Until as of late, plant rearing still depended exclusively on the aggregated understanding of ages of agriculturists and raisers, that is, on sexual exchange of qualities between plant species. Nonetheless, late improvements in plant sub-atomic science and genomics now give us access to the information and comprehension of plant genomes and even the likelihood of changing them. Plant geneticists have received solely on the accumulated experience of generations of farmers and breeders, that is, on sexual transfer of genes between plant species. However, recent developments in plant molecular biology and genomics now give us access to the knowledge and understanding of plant genomes and even the possibility of modifying them. Plant geneticists have adopted Arabidopsis thaliana as a model organism some years ago because of its small diploid genome (the Arabidopsis genome, at about 120 Mb, is amongst the smallest known plant genomes), low repetitive DNA content, and rapid reproductive rate. Now, the complete sequence of the genome of this plant is known, which will lead to the identification of all its 26,000 Based on this success, the sequencing of various cultivated plant genomes is now well underway (e.g. rice). The rice genome sequence provides a foundation for the improvement of cereals, our most important crops Most importantly, our present knowledge of synteny indicates that, despite plasticity contributing to the diversity of the plant genomes, the organization of genes is conserved within large sections of chromosomes This validates a posteriori the considerable efforts made on model species. Such progress has encouraged a massive surge in plant biotechnology, which is currently changing our vision of crop production and protection. Indeed, this technological progress enables us to insert useful genes into cultivated plants at an incomparably fast rate and, doubtless, in a much more precise manner than with conventional genetic methods.

Figure: Biotechnology in industry

Plant transformation:

Genetic engineering techniques now allow us to transfer the genes of one species over to another species. Indeed, the intended uses aim to introduce new characters into an organism that otherwise would not have acquired them. These techniques can be applied generally to all living species: bacteria, fungi, viruses, animals, and plants. After undergoing genetic engineering, the organisms are referred to as genetically modified organisms (GMOs) to indicate that they are organisms that have had their genetic profile altered in a laboratory. In the case of plants, one of the elements of the current social debate is based on the fact that these new transplanted characters can originate from a species totally unrelated to the receiving plant.

Additional tools for plant improvement:

The plant breeding methods described above have enabled remarkable improvements to be made in the yield and quality of our modern crops. Additional tools are also used for genetic enhancement and propagation of crop plants. As with plant breeding, these methods have been used for many years to develop crop plants that better suit our needs.

Grafting and tissue culture techniques:

Grafting of tissues from two different varieties of a plant species has been used since ancient times in woody tree and vine crops such as citrus, peaches, walnuts, grapes, and ornamental trees. Surgically cutting a scion or bud from one variety and grafting it onto a rootstock from a different genetic variety is commonly used to enhance the disease resistance, productivity, and growth habit of these perennial crops. Superior varieties often developed through chance or induced mutations as well as through sexual crosses, can be rapidly and uniformly propagated by grafting buds onto the rootstocks of other varieties. Tissue culture has been used in crop improvement since the 1940s. In the simplest cases, this refers to culturing embryos or small plants in the laboratory on specific nutrient media until they can be moved into soil. The tiny growing tips of plants (meristems) can also be grown in culture to produce entire plants. In addition, plants have the unique property of being able to regenerate an entire plant from a single cell Under carefully controlled conditions, tissues can be taken from a plant, separated into individual cells that are grown in the laboratory as callus, then induced to develop back into whole plants These techniques have been used in a number of ways in crop propagation and improvement.

2.0 Literature review:

Edwards, J.D. and McCouch, S.R. (2007) our present knowledge of syntonic also indicates that, despite plasticity contributing to the diversity of the plant genomes, the organization of genes is conserved within large sections of chromosomes. In parallel, novel plant transformation systems have been proposed, notably with regard to plastid transformation and the removal of selectable marker genes in transgenic plants. Furthermore, a number of recent works considerably widen the potential of plant biotechnology.

H. Daniell, M.S. Khan, L. Allison (2002) Molecular markers are widely employed in plant breeding. DNA markers are being used for the acceleration of plant selection through marker-assisted selection (MAS). Genes of agronomic and scientific importance can be isolated especially on the basis of their position on the genetic map by using molecular markers technologies. In this review, the current status of marker development technologies for crop improvements will be discussed. It will also provide an outlook into the future approaches and most widely used applications in plant breeding in crop plants on the basis of present development.

Dasilva, edgar . j.(2010) Plant organelles have intrigued biologists since their defiance of Mend Elian inheritance and their endow symbiotic origins became apparent. The first application of organelle biotechnology was the role played by cytoplasmic male sterility in hybrid seed production, a contribution towards the "Green Revolution". In modern times, plant organelles are again leading the way for the creation of genetically modified crops. On a global scale, 75% of GM crops are engineered for herbicide resistance and most of these herbicides target pathways that reside within plastids.

T. Bouquin, M. Thomsen, (2002) for devising educational and capacity-building schemes that enable developing countries embark on sustainable development, possibly in network cluster groups once account has been taken of their level of research in biotechnology; of their capacities to produce and commercialize biotech products; of their degree of participation in developing national, regional and international biotech governance dealing with biosafety, conservation and trade of genetic diversity; of their capability and capacity for national education and training; and of their ability to engage in regional research.

T. Kunkel, Q.W. Niu, Y.S. Chan, N.H. Chua, (2004) Plastidial DNA transformation in certain species (tobacco in particular) is now under control. There are several advantages to developing this new technique. These include (a) the transfer of the desired genes solely via the female line (in most plant species, the pollen does not contain any plastids), thereby limiting the contamination of wild plants with transgenes carried on by pollen flow; (b) very high levels of transgene expression in genetically engineered plants (indeed, each plant cell contains many hundreds of plastids; furthermore, gene silencing, frequently observed in nuclear transgenic plants, has not been observed in genetically engineered chloroplasts); (c) targeted homologous recombination into the plastid genome As for the nuclear transgenic plants, several strategies have been developed for engineering chloroplasts that are free of antibiotic resistance markers

3.0 Random Amplified Polymorphic DNA's (RAPDs)

RAPDs are based on the PCR amplification of random DNA segments with primers of random nucleotide sequencesthat were inexpensive and easy to use. The primers bind to complementary DNA sequences and wheretwo primers bind to the DNA sample in close enough for successful PCR reaction. The amplified of DNA productscan then be visualized by gel electrophoresis RAPD markers have been widely used in diverse plant species for assessment of genetic variation in populationsand species, fingerprinting and study of phylogenetic relationships among species and subspecies Nevertheless, disadvantages of RAPD markers are the fact that it predominantly provides dominant markers,and incapability to detect allelic differences in heterozygotes. Polymorphisms are detected only as the presenceor absence of a band of a certain molecular weight, with no information on hetero additionally,because of their random nature of amplification and short primer length, they are not ideal for genome mapping.Moreover, these markers do not exhibit dependable amplification patterns and differ with the experimental conditions

Simple Sequence Repeats (SSRs):

During Simple sequence repeats (SSRs) which is also known as microsatellites were established and provided a choice for many genetic researches since they are amenable to low, medium and high-throughput approaches. They are randomly tandem repeats of short nucleotide motifs SSRs are frequently highly polymorphic sequences normally present in animal and plant species and can be used to study the relationship between inherited traits within a species Microsatellite markers are often derived from noncoding/ anonymous genomic regions, such as bacterial artificial chromosomes (BACs) and genomic survey sequences (GSSs). Therefore, development of SSR markers used to be expensive and laborious this assay is easily detectable by gel electrophoresis for few to hundreds of samples, which could be inexpensive by researchers with limited resources. Polymorphism is based on the variation in the number of repeats in different genotypes Since polymorphisms in longer pentad-nucleotide and tetra repeats are easier to make a distinction in a variety of detection systems and longer repeats may be more robustIn recent years, SSR markers can easily be developed in silicondue to the availability of large-scale gene (expressed sequence tag) EST sequence information for many plant species. Since EST sequencing projects haveprovided sequence data that is available in online databases and can be scanned for identification of SSRs The high degree of polymorphism as compared to RFLPs and RAPDs, their co-dominant nature and locus specific make them the markers of choice for a diversity of purposes including practical plant breeding. Therefore, (SSRs) have become a marker of choice for an array of applications in plants due to extensive genome coverage and hyper variable nature.

Research for Sustainable Agriculture and the Environment:

Molecular bases of plant-pathogen interactions

- Molecular bases of plant-symbiotic bacteria interactions
- Plant Breeding for resistance
- Stimulate defence mechanisms

Molecular bases of plant- symbiotic bacteria interactions: Plant-interacting bacteria can establish either mutualistic or pathogenic interactions that cause beneficial or detrimental effects respectively, to their hosts. In spite of the completely different outcomes, accumulating evidence indicates that similar molecular bases underlie the establishment of these two contrasting plant-bacteria associations. Recent findings observed in the mutualistic nitrogen-fixing Rhizobium-legume symbiosis add new elements to the increasing list of similarities. Amongst these, in this review we describe the role of plant resistance proteins in determining host specificity in the Rhizobium-legume symbiosis that resemble the gene-for-gene resistance of plant-pathogen interactions, and the production of antimicrobial peptides by certain legumes to control rhizobia proliferation within nodules.

Plant disease resistance:

Protects plants from pathogens in two ways: by pre-formed structures and chemicals, and by infection-induced responses of the immune system. Relative to a susceptible plant, disease resistance is the reduction of pathogen growth on or in the plant (and hence a reduction of disease), while the term disease tolerance describes plants that exhibit little disease damage despite substantial pathogen levels. Disease outcome is determined by the three-way interaction of the pathogen, the plant and the environmental conditions (an interaction known as the disease triangle Defense-activating compounds can move cell-to-cell and systemically through the plant vascular system. However, plants do not have circulating immune cells, so most cell types exhibit a broad suite of antimicrobial defenses.

Stimulate defense mechanisms: Defense mechanisms may result in healthy or unhealthy consequences depending on the circumstances and frequency with which the mechanism is used. In psychoanalytic theory, defense mechanisms German: Abwehrmechanismen are psychological strategies brought into play by the unconscious mind to manipulate, deny, or distort reality in order to defend against feelings of anxiety and unacceptable impulses and to maintain one's self-schema. These processes that manipulate, deny, or distort reality may include the following: repression, or the burying of a painful feeling or thought from one's awareness even though it may resurface in a symbolic form identification, incorporating an object or thought into oneself and rationalization, the justification of one's behavior and motivations by substituting "good" acceptable reasons for the actual motivations. In psychoanalytic theory, repression is considered as the basis for other defense mechanism

The plant as a factory to produce useful molecules:

A particularly fruitful area of research for the current interests of plant biotechnology concerns the improvement of the quality of plant products. It is clear that one can now alter the principal biosynthetic routes of the higher plants almost “at will” in order to make them synthesize new types of fatty acids, starch, and proteins Their metabolite content, which is indispensable to animal and human nutrition may also be modified. This is also the case for metabolites posing a problem to developing industrial applications lignin, the principal constituent in wood, which poses problems in the paper industry In the short and medium term, the potential for this area appears clearly to be much larger than those of resistance to herbicides or to insects that have been implemented until now. Although this is not an exhaustive list, this field of application is concerned with remodeling the constituents of oil and seed starch for not only nutritional and pharmaceutical but also industrial means (such as in detergents, lubricants, inks, polymers, cosmetics, plasticizers, biofuels, depolluting agents, etc.). It is also concerned with remodeling components essential to animal and human health (essential amino acids, vitaminsmanufacture, or therapeutic proteins and enzymes involved in lipid metabolism, vicinal plants, monoclonal antibodies, etc. This can be exemplified with some studies. The first exploits the specificity of plant metabolism and relates to the synthesis of essential amino acids and of principal vitamins. It would appear that the levels of biosynthesis in these components are well adapted to plant needs.

4.0 Applications and Strategies of DNA Markers in Breeding Programs

Gene pyramiding is defined as an assembly of multiple desirable genes which can be combined into a singlegenotype from multiple parents. This is referred as one of the major applications of marker assistant selection, as gene pyramiding via conventional plant breeding is difficult, if not impossible the methods for pyramiding favorable alleles can be used in the same way to accumulate QTL controlling different traits. A main difference in the model is that alleles at different trait *loci* to be accumulated may have different favorable directions, for instance negative alleles are preferable for some traits but positive alleles are Favorable for others. As a result, to meet breeding objectives one may need to combine the positive QTL alleles of some traits with the negative alleles of others Selection for multiple traits may be completed in one cycle if the population size is large enough to allow desirable individuals to combine different traitsNevertheless, the number of trait *loci* that can be manipulated in one cycle is restricted because the population size required covering the recombinants increases exponentially with the increase.

Improving the tolerance of plants to biotic stresses:

Environmental factors are essential components in crop yields. The introduction of resistance to heavy metals, salt, cold, and drought into crop plants has become a topic of major economic interest for agriculture. Genetically engineered drought- and salt-tolerant plants could provide an avenue to the reclamation of farmlands lost to agriculture because of salinity and a lack of rainfall. In the case of drought, we are beginning to understand certain extremely complex mechanisms through which seeds from orthodox plants acquire tolerance to desiccation during their final maturation period, when the seed becomes quiescent and its metabolism stops. Reminiscent plants, capable of supporting extreme hydrous stress, provide another model. Some of the genes associated with the acquisition of this tolerance to drought have been isolated and characterized. Based on this knowledge, there are several examples showing the feasibility of improving tolerance of plants to biotic stress by genetic engineering finally, it is interesting to note that the genetic engineering of the signaling pathway implicating the gibberellin hormones allows the creation of plants that are more resistant to bad weather.

Genotyping-by-Sequencing (GBS)

With the increased throughput of NGS platforms, re-sequencing for genome-wide surveys of genetic diversity became reasonableHowever, this assay is biotin dogmatically challenging, impartial estimation of genetic diversity across the genome in both coding and non-coding regions could be determined. Additionally, it allows for the detection of various types of genetic variation; this detection capability contains not only SNPs and small idles, but also large mega-base scale idles This assay involves the use of restriction enzymes for reducing the complexity of genomes followed by targeted sequencing of reduced proportions, in that way each marker can be sequenced at high coverage across many individuals at low cost and high accuracy A workflow of GBS has been presented in The main target for constructing GBS libraries was based on reducing genome complexity with restriction enzymes, which may reach important regions of the genome that are unreachable to sequence capture approaches The procedure has been demonstrated with barley (Oregon Wolfe Barley) at the recombinant inbred lines populations and maize (IBM) where about (25,000 to 200,000) sequence tags were mapped, respectively. With this method, species that lack a complete genome sequence can have a reference map settled around the restriction sites, which can be done in the process of sample genotyping. This system has been adjusted for reducing missing data points and improved SNP calls.

Capacity-building in biotechnology fordevelopment

Biotechnology is a cross-cutting technology encountered in wide application across several sectors of development. An amalgam of a variety of disciplines -biochemistry, the engineering sciences, genetics, informatics, molecular biology and microbiology, the neurosciences and nanotechnology amongst others, biotechnology makes important contributions to the new knowledge-based economy and markets. Developing countries, and especially the LDCs, face challenges in setting up the agendas of international cooperation in deriving benefits from biotech markets. The lack of professionals, sophisticated equipment, relevant infrastructure, deficiency of national legal instruments concerning patents and intellectual property rights, and of financial support widen rather than bridge the gap of R & D in biotechnology between the industrialized and developing countries. Hence there is a distinct need for education and capacity-building important elements in the use of biotechnology for development. The style, substance and scale of biotechnology in the developing world varies within a region, and from region to region. Hence the need for devising educational and capacity-building schemes that enable developing countries embark on sustainable development, possibly in network cluster groups once account has been taken of their level of research in biotechnology of their capacities to produce and commercialize biotech products; of their degree of participation in developing national, regional and international biotech governance dealing with biosafety, conservation and trade of genetic diversity; of their capability and capacity for national education and training; and of their ability to engage in regional research since the scope and scale of biotech literacy varies amongst countries in a region. Many of the advanced developing countries, unlike several LDCs, have well established centres and institutions with the capability to educate and provide training on general and specific issues.

Conclusions:

The development of these plant biotechnologies even now provokes many reactions. Among these are reactions to fields of application, modes of use, increased competence in agricultural procedures by agrochemical companies (appropriation of living organisms, modification of agricultural practices), and the reality and eventual consequences of transferring these techniques to developing countries Despite this, undeniable progress has been recorded on a scientific level, allowing, thanks to these technologies, useful modification of plants. This allows us to envisage the feasibility of improving the nutritional conditions for animals and humans (such as yield increase, correction of nutritional deficiency, elimination of anti-nutritional components, vitamin intake and health (protein and enzymes manufacture, vicinal plants), to encourage the arrival of multiple new industry-oriented products and to produce crops that work in greater harmony with the environment in terms of their water requirements, pesticides, and fertilizers. These techniques, although still not quite perfected, are now in place However, the parameters and conditions included in simulations may not fully reflect the complex situations of diverse plant breeding programs, the genetic gain per unit time and cost that has been achieved in simulations needs to be supported by the long-term selection response by comparing with other breeding approaches. In addition, this approach can technically lead to the discovery of thousands of SNPs in one single experiment. Moreover; it can be used of those plants that do not have the reference genome available.

References:

[1] Edwards, J.D. and McCouch, S.R. (2007) Molecular Markers for Use in Plant Molecular Breeding and Germplasm Evaluation. Marker-Assisted Selection-Current Status and Future Perspectives in Crops, Livestock, Forestry and Fish, Food and Agriculture Organization of the United Nations (FAO), Rome, 29-49.

[2] H. Daniell, M.S. Khan, L. Allison, (2002) Milestones in chloroplast genetic engineering: an environmentally friendly era in biotechnology, Trends Plant Sci. 7 84–91.

[3] Dasilva, edgar .j.(2010biotechnology,developing countries and globalization. world journal of microbiology and biotechnology, , vol.14, p. 463-486.

[4] T. Bouquin, M. Thomsen, L.K. Nielsen, T.H. Green, J. Mundy, M.H. Dziegie,(2002) Human anti-rhesus D IgG1 antibody produced in transgenic plants, Transgenic Res. 11 115–122.

[5] T. Kunkel, Q.W. Niu, Y.S. Chan, N.H. Chua,(2004) Inducible isopentenyl transferees as a high-efficiency marker for plant transformation, Nat. Bio-technol. 17 916–919