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Spices research - A crystal gazing

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Abstract

The direction of research on spice crops especially black pepper (*Piper nigrum*) and cardamom (*Elettaria cardamomum*) during the present century has been conceptualized. The revolution that is likely to be created by genetic engineering and biotechnology in the areas of germplasm conservation, crop improvement, crop protection, crop management and post harvest technology, resulting in tremendous increase in productivity of spices has been highlighted.

Key words: biotechnology, black pepper, cardamom, research, spices.

Introduction

Aldous Huxley in his classical science fiction 'The Brave New World' vividly described cloning of humans tailored for different purposes and having different capabilities. That was in the 1930's, when and in the years that followed, it was considered by all as just a figment of imagination and as pure fiction. But as the century came to an end we realised how dangerously close we are to cloning human beings. Genetic engineering and biotechnology have virtually revolutionized the science of biology and the new brave biology is unlimited in potentialities. This is true of plant sciences as well. This new biology has its fall-out in the areas of agriculture, horticulture, forestry, fisheries and veterinary sciences. The theme of this article is how this new branch of biology is likely to affect the future of spices research. This is only a crystal gazing and the views expressed may appear imaginary.

The direction of spices research in the coming decades has been conceptualized and represented in Fig. 1. In the next two decades rapid progress can be anticipated in all areas of research, such as germplasm conservation, crop improvement, crop management, crop protection and post harvest technology. Not far away from now the boundary walls among these areas will blurr out and perhaps all the areas may merge into an unified one. As research in the new areas becomes more and more sophisticated, individual enterprises will give way to team work (close knit, coherent groups), as the best viable option to tackle problems successfully. Surely biotechnology will take the centre stage of spices research in the decades to come.

Spices in general are poor production systems, having low efficiency in converting solar energy into biomass. As such, spice plants cannot compete with many agri-horticultural crops. But the yield of high-value constituents in spices can be enhanced to very high levels through appropriate biotechnological interventions. Application of genetic engineering tools in plants necessarily requires good tissue culture protocols, and modern plant biotechnology has evolved out of an union of tissue culture technology and rDNA (recombinant DNA) technology during the last quarter of the 20th century.

Germplasm conservation

In germplasm conservation, molecular characterization may soon produce genetic finger prints of various accessions of different spices, and such finger prints will be useful in eliminating duplicates and in safeguarding unique genotypes. As this effort has already been initiated much progress will be achieved in the years to come and perhaps in 20 years time we may see the development of saturated molecular maps of all major spices such as black pepper, cardamom, ginger and capsicum. Genomic libraries and cDNA libraries will be developed for these spices,

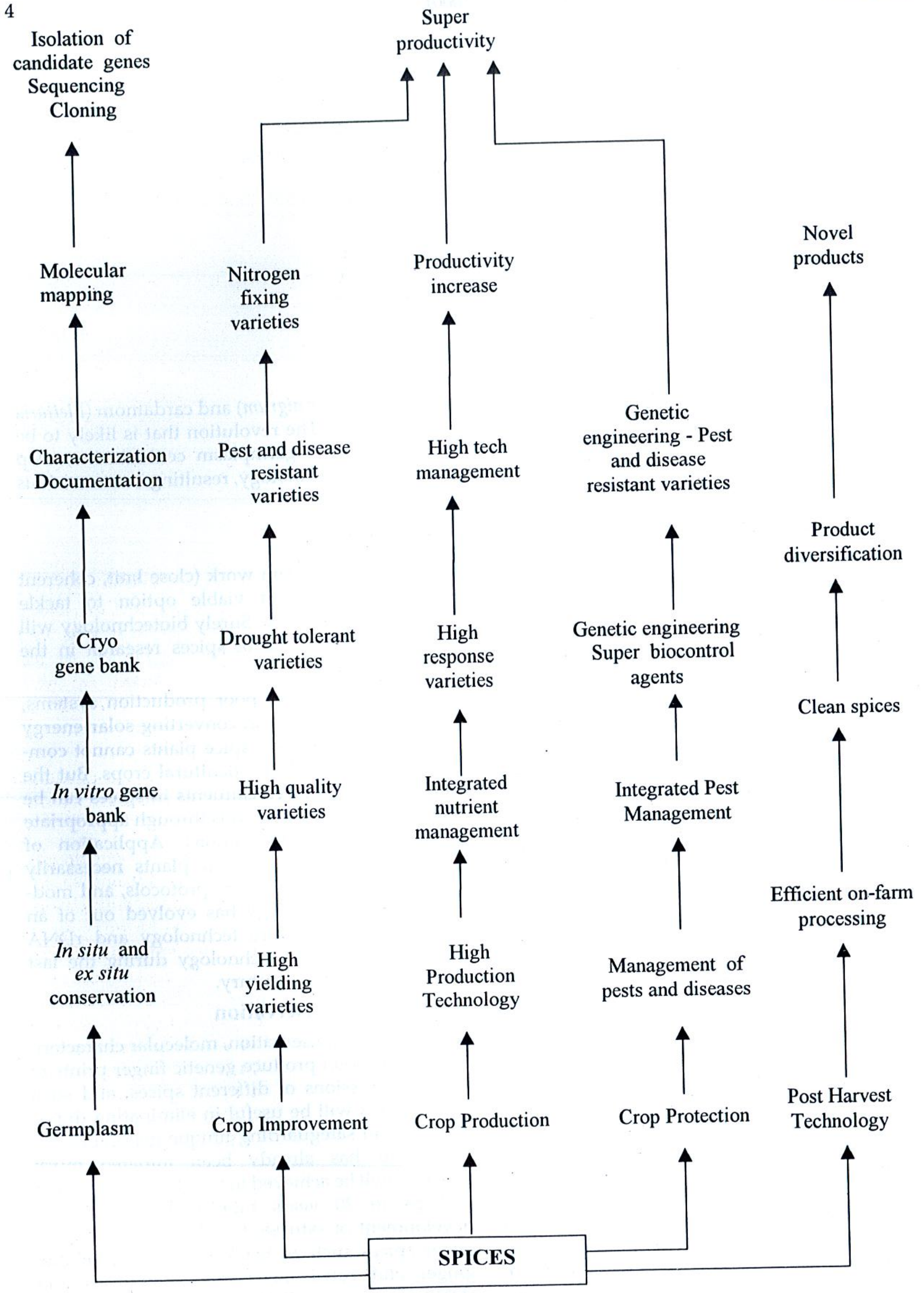

Fig 1. Spices research in 21st century

probably in the present decade itself. Total genome sequencing will take many more years to materialize, and capsicum will be the first spice to achieve this distinction possibly by the end of the first half of this century. Along with this, genes controlling important agronomic characters will be identified, isolated, cloned, sequenced and manipulated for upgrading agronomic and quality characters.

Along with *ex situ* conservation programmes, *in vitro* gene banks of spices will become fully operational. Efficient cryo-conservation methods for long term conservation will be evolved and cryogene banks will come into existence for conservation *ad infinitum* of genetic resources of spices for the benefit of posterity.

Crop improvement

In crop improvement, conventional breeding will give way to molecular breeding. Genetic modification of plants will become routine, and movement of genes across genera and families will help in pyramiding of genes from diverse sources for enhancing the expression of characters of importance. The present yield barriers will be broken by the use of super efficient promoters and modification of biosynthetic pathways. Possibly in another decade, scientists may achieve definite information on the nature of resistance/susceptibility to biotic and abiotic stresses, thereby helping researchers to design varieties resistant to these stresses. Perhaps by the end of the third decade of the present century, we may be lucky to see genetically modified varieties of black pepper, cardamom and ginger, combining high yield, high quality and resistance to diseases and pests.

Abiotic stress

Spice plants are sensitive to heat and cold, that restricts their cultivation to tropical or temperate lands. Extreme tolerance to heat/drought is found in plants growing in hot springs or in hot deserts. Similarly, plants that are highly tolerant to cold are found in the north polar region. Heat and cold resistance genes from plants growing in such extreme conditions can bestow extreme cold/heat tolerance to spices. Such spice plants, modified genetically for heat and cold tolerance, may be available by 2030. We may then see black pepper cultivation in Siberia or bumper harvests of saffron from Rajasthan deserts. Heat and cold resistance genes isolated from organisms living under extreme environments (extremophiles) are already finding way into plants, though the achievements at present are rudimentary. Many candidate genes that are implicated in stress tolerance have been isolated and are under active study. Advances in functional genomics will give a better insight into the genetics of cold, heat, stress and salinity tolerance, and genes contributing such characters will soon find their way into spice crops.

Building salt tolerance into plants is a formidable task. In the near future salt tolerant genes will be isolated from sea weeds or marshy plants, and these genes on incorporation in spices together with appropriate promoters may bestow salt resistance. Such salt resistant spice plants may become available by 2030. Salt resistance will be useful in cultivating vast stretches of sodic and marsh lands and will be a great boon. Such salt resistant transgenics can be grown near sea shores under sea water irrigation.

Crop management

By the end of the first half of the present century we may be lucky to see genetically engineered nitrogen fixing black pepper and ginger plants. The molecular biology of nitrogen fixation is well understood, and many genes involved in the process have been isolated and cloned. Yet there are many problems to be tackled, especially in creating an oxygen-free environment for nitrogenase action. A nitrogen fixing cereal will probably be developed by 2025, and a nitrogen fixing spice plant may become available by the end of 2040.

Chemical fertilizers may soon give way to efficient biofertilizers. Scientists will develop superefficient nitrogen fixers and phosphate solubilizers, so that chemical fertilizers can be brought down substantially. Such super efficient biofertilizers can be developed in a decade's time. High tech production systems will be developed for maximizing production and productivity. In such systems highly efficient biofertilizers can produce pure or 'organic spices', uncontaminated by any agrochemical. In such systems highly potent biocontrol agents will completely replace fungicides and insecticides.

The new god!

Biotechnology is the new god. The application of biotechnology is manifold and such applications will surely revolutionize production technologies in spices (Fig. 2). In addition to the achievements

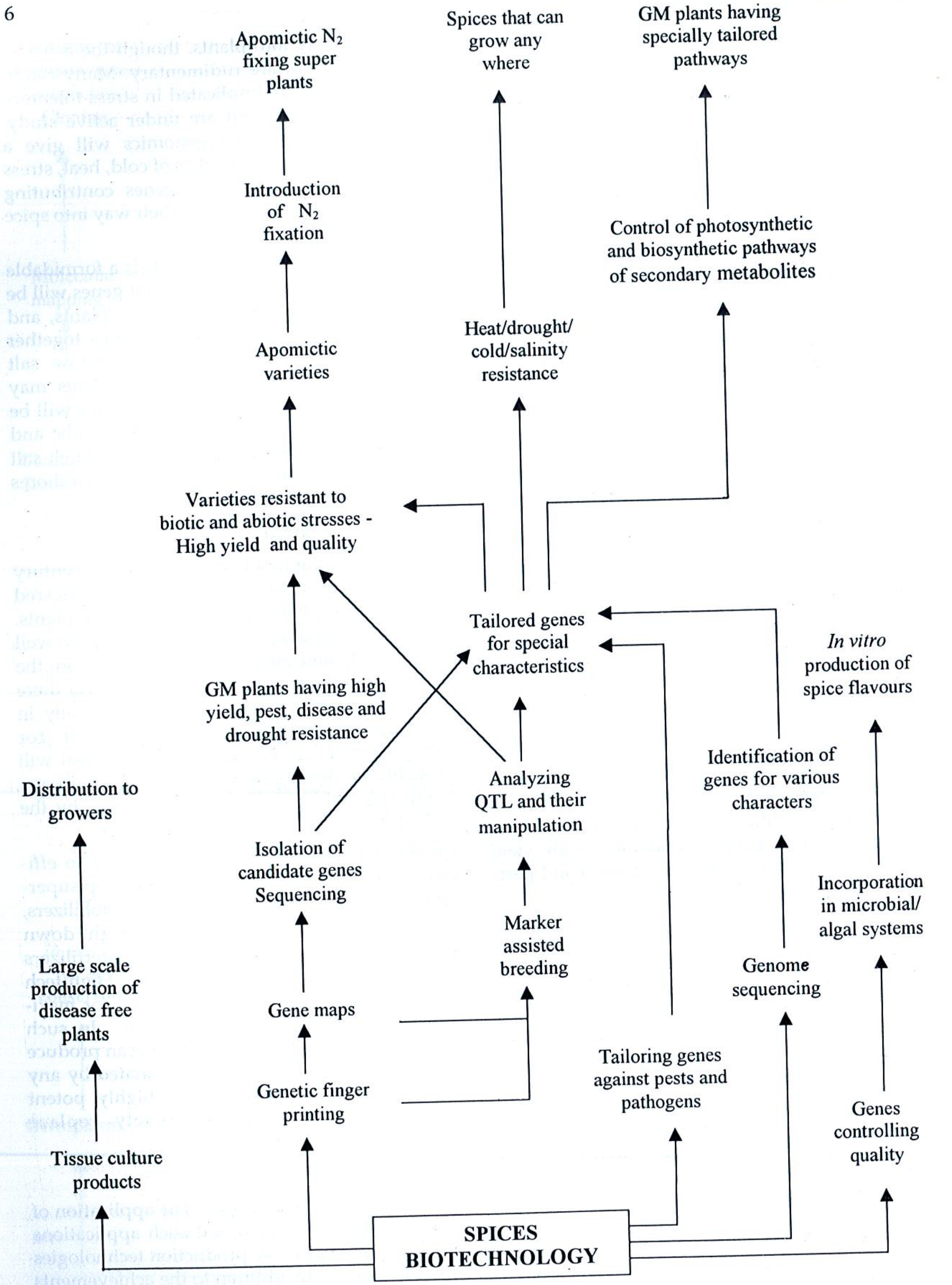

Fig. 2. Spices research in 21st century - Biotechnological approaches

mentioned above, biotechnological interventions will lead to the development of apomictic varieties, nitrogen fixing super varieties combining high yield, high quality and resistance to biotic and abiotic stresses, varieties with specialized characters, etc.

Human labour will become very costly and even may not be available for many agricultural operations, resulting in the need for mechanization of many labour intensive operations. This may necessitate modification of plant geometry so as to suit mechanical handling. Genetic engineering will come to our help to develop such crops. We may then have ginger plants with square or rectangular shaped rhizomes to suit mechanical peeling, and black pepper plants that can be harvested using centrifugal harvesters by which the berries alone are sucked in, so that harvesting and thrushing are combined into one operation.

Advancements in protein and gene engineering will lead to designing proteins that are a thousand times more active and efficient and tailored genes for such proteins will be synthesized and used in modifying plants in the near future. Possibly the 2020's will witness such advancements and use of such tailored genes will then push up productivity and quality, perhaps a hundred fold or more. The total sequencing of the first plant genome (*Arabiodopsi thaliana)* will bestow on scientists great insights into plant metabolic processes and their control points. Exploitation of this new knowledge will give spice scientists tremendous power to manipulate biosynthetic pathways, thereby pushing up the productivity levels beyond anything so far acheived or believed possible. Such knowledge will act as a catalyst for total gene sequencing of spices. With such advances spice farming will become super-tech and perhaps over production may even lead to a serious glut in the market.

However, everything would not be rosy in spices research, and spices producing and exporting countries may have to face serious blows with the advancements in biotechnology. Insights into the genetic machinery of spice plants will empower scientists in importing countries to produce spice flavours cheaply through microbial fermentation systems. Introduction of spice genes into alga like *Chlorella* or *Spirulina* can be highly efficient and can compete with conventional spice production systems as an efficient alternative to conventional spices farming, with accompanying disastrous consequences to spices growers and spices producing countries. Microbial production of vanillin has already been achieved and biovanillin has been produced by genetically engineering microbes with the vanillin genes from *Vanilla planifolia* Andrews. Similar advancements can be expected in the production of curcumin (biocurcumin), paprika pigments (capsorubin, capxanthin, etc.), colour and flavour compounds of saffron (crocin, picrocrocin and crocittin), etc.

Black pepper and cardamom - The future

As an extension of what has been visualized, specific examples of two spices namely, black pepper (*Piper nigrum* L.) and cardamom (*Elettaria cardamomum* Maton) have been chosen to do a crystal gazing of the shape of things to come in the present century in these spices.

Black pepper

The global scenario of black pepper production during the past indicated a growth rate of 2.7-3.6% on an average. This means that the demand for black pepper will go up to 360,000 t by 2050, this quantity being double the present figure.

The black pepper crop is facing serious biotic and abiotic stresses and diseases and nematodes are causing severe crop losses. Because of these factors research in the future will be tuned for productivity increase and disease and nematode resistance. We can expect much progress in these areas in the next two decades, and by the end of which we may get black pepper varieties having high resistance to *Phytophthora capsici*, *Radopholus similis* and little leaf virus.

In the area of crop breeding, homozygous lines through androgenic haploids will soon be produced, which will then help in evolving highly heterotic, complex hybrids, having high yield and quality combing multiple resistance to various diseases and pests.

Crop management will become more high-tech. Effective integrated pest management strategies, in combination with highly potential biofertilizers will reduce the use of fertilizers and agrochemicals and high production technologies for organic spices will soon take the place of the present high input production system. Efficient pest and disease management using genetically modified super efficient biocontrol agents will come into existence, and the use of phytochemicals will then recede to the back stage (Fig. 3).

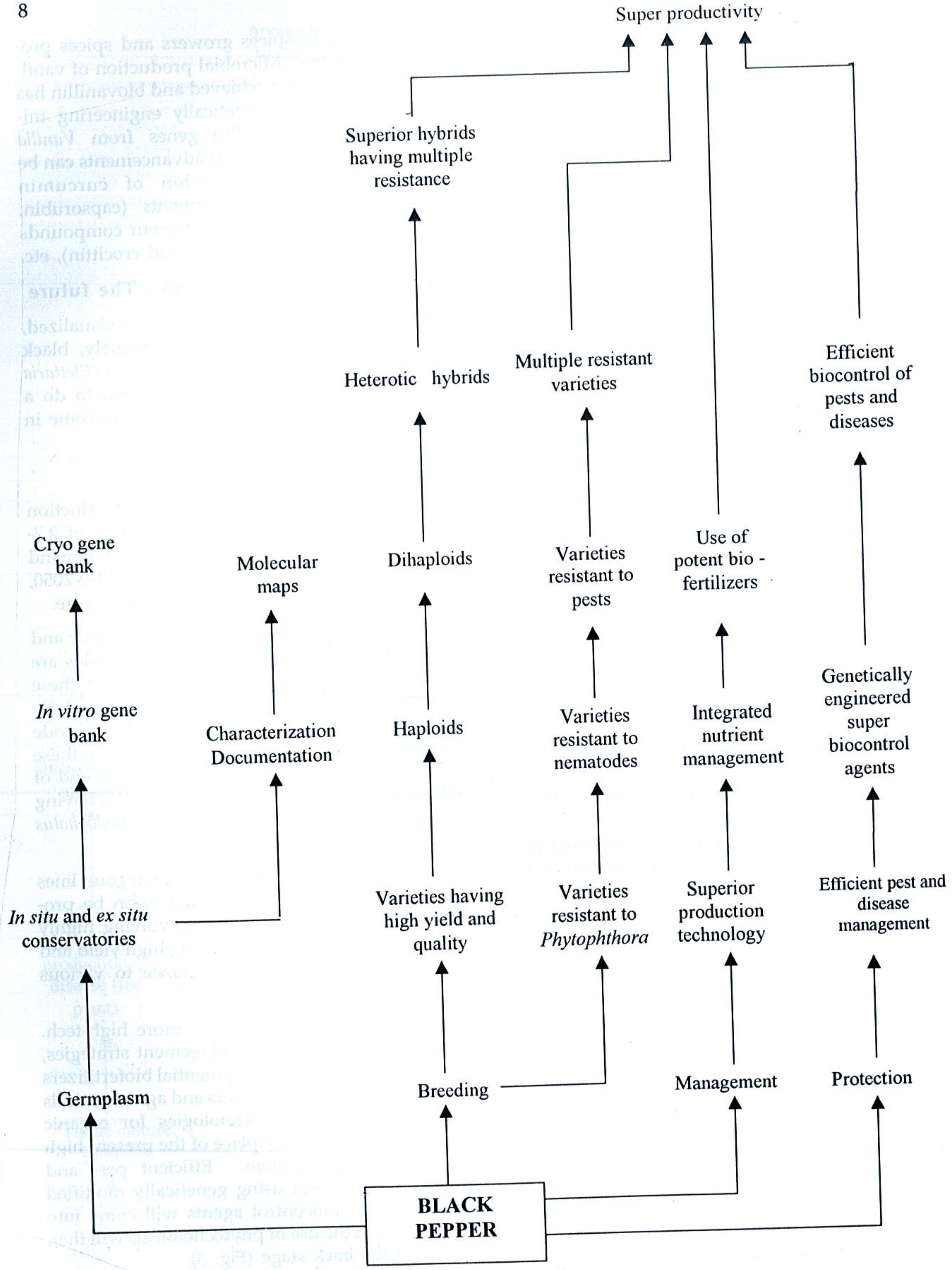

Fig. 3. Black pepper research - Crystal gazing

As in the case of most other crops, biotechnology will take the centre stage of black pepper research by the second decade of the present century. Tremendous progress can be anticipated by 2020's and many genetically modified lines having various desired characters will be evolved. Apart from black pepper genotypes that are immune to pests and diseases, we will also see complex black pepper transgenics having resistance to heat and drought, salinity and cold. Naturally, black pepper cultivation will spread to temperate regions or even to the Siberian tundras and perhaps even to the sodic marsh lands. The ultimate dream will be the apomictic, nitrogen fixing black pepper, but such advancements are expected only by the close of the present century. By that time we may also witness the total sequencing of the black pepper genome (Fig. 4).

Cardamom

In cardamom, the present level of biotechnological achievements are minimal, limiting to various tissue culture protocols. Revolutionary advancements in this crop are not anticipated, because the economic impact factor is not very encouraging. Still in the decades to come many developments may take place.

Success in anther culture, production of haploids and dihaploids will lead to many homozygous lines. Exploitation of heterosis will then become possible and hybrid seed production gardens will be established. High yielding hybrid cardamom may become popular by the end of the first decade of this century. Simultaneously large scale cloning of 'super clumps' of cardamom plants already located, and their popularization will push up the yield level considerably (5000-6000 kg dry cardamom/ha). Biotechnological interventions will help in the introduction of genes resistant to virus diseases and pests into the super lines thereby producing super varieties. These super varieties can then be modified for thriving well in plains and under fully open conditions through the incorporation of heat and drought resistant genes. Such advancements can be expected during the second and third decades of the present century. Introduction of apomictic genes may be achieved later in the century, which will help in avoiding multiplication through tissue culture.

Further ahead, we may get a nitrogen fixing, apomictic, super yielding varieties, resistant to various diseases and pests. Perhaps the second half of the present century may witness such developments. The total sequencing of cardamom genome can also be expected during that period. Simultaneously the dissecting out of the metabolic pathways and the control mechanisms involved will give us the opporutnity to manipulate metabolism for increasing yield and quality to higher levels. On the other hand, advancements in bioprocess engineering can help us produce important flavour components through microbial fermentation systems. These flavour components upon proper mixing can give near cardamom flavour, and such processes if happens to be cheaper, can upset cardamom production (Fig. 5).

These developments can also happen in other important spices too, as already mentioned. The unravelling of biochemical pathways and their control systems are essential for such advancements. This will be made relatively easy with the *Arabiodopsi* genome sequencing, which will provide many clues to the basic metabolic pathways in plants and their control systems. However, extensive investigations will be needed for every individual spice crop. Once such information is available, then genetic manipulaton becomes easier and the genes involved in any synthetic pathway can be incorporated into other plants producing higher physiological efficiency or into microbial systems. Use of powerful promoters and blocking of allosteric inhibition can lead to unlimited accumulation of target compounds.

Dreams and realities

The foregoing discussion may appear as a wild dream. Yes they are, and maybe the realities could be something different. We may not see revolutionary or path breaking discoveries or events in spices research at least during the present decade. Advanced research is yet to be initiated in most spice crops and the expertise available in spices research groups in the world is not of a high level. Perhaps what we expect for the next decade are:

1. Characterization of germplasm based on some of the molecular markers and preparation of molecular maps.

2. Perfecting protocols for protoplast culture, anther culture, etc. in important spices. Production of haploids and dihaploids and use of these homozygous plants in hybridization, production

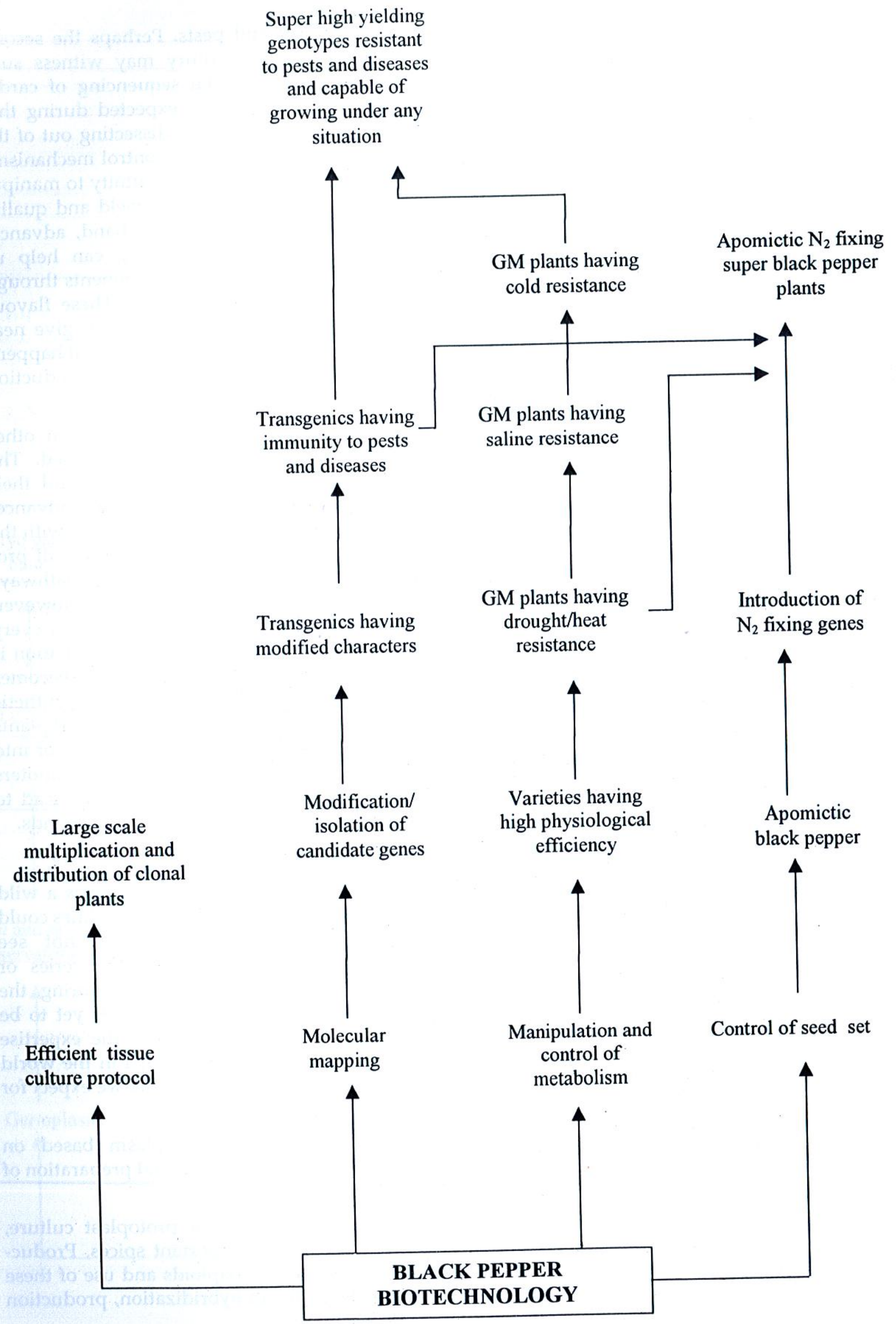

Fig. 4. Biotechnological approaches in black pepper research - Crystal gazing

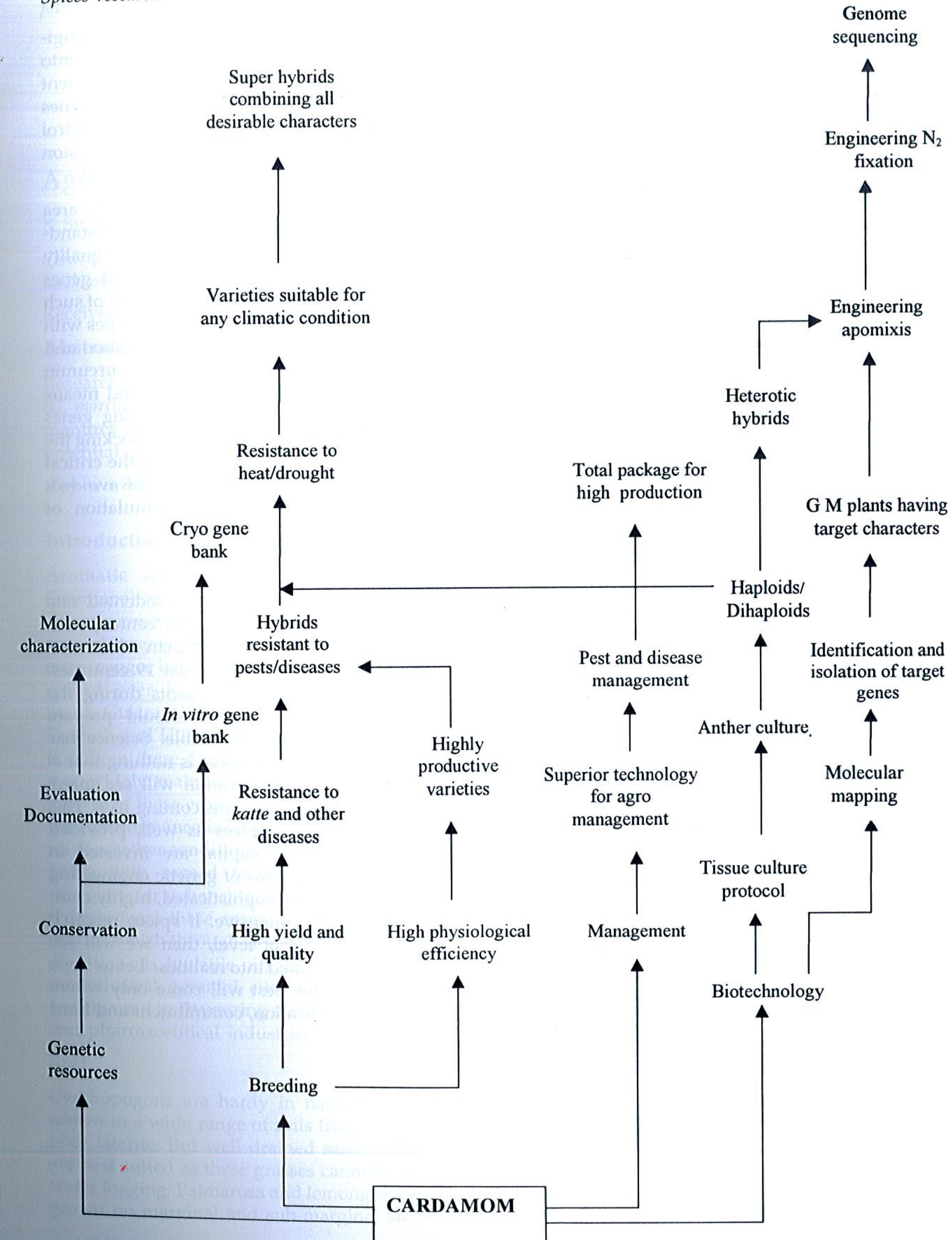

Fig. 5. Cardamom research - Crystal gazing

of high yielding hybrids, seed production gardens, etc.

3. Production of transgenics mainly with a view to incorporate disease resistance (antifungal and antibacterial genes) using the candidate genes currently available (Table 1). Gene pyramiding through incorporation of different genes in the same plant can lead to high resistance to fungal and bacterial pathogens. Such efforts may lead to resistant genotypes of spices maybe by 2020's.

4. Advancement in production technologies may push up productivity to much higher levels. Apart from development of resistant genotypes, highly potent biocontrol agents might lead to effective control of pests and diseases. Gene pyramiding can be effectively done in biocontrol agents, once the genes governing their mechanism of action is unravelled. Production of antifungal enzymes by biocontrol agents (eg. *Trichoderma*) can be increased many fold by genetically engineering more than one gene for such enzymes into the same organism, by using highly efficient promoter genes, and by using synthetic genes having increased activity. Such super biocontrol organisms will then provide efficient suppression of pathogenic fungi and bacteria in the soil.

Table 1. Candidate genes for producing transgenics against bacterial and fungal diseases

Gene	Gene source
Antibacterial	
Attacin	*Hyalophora aeropia*
Lysozyme	Chicken
Cecropin B	*H. cecropia*
Shiva 1, 2, etc.	Synthetic cecropins
Antifungal	
Glucanase	Tobacco, Alfalfa
Osmotin	Tobacco
Chitinase	Tobacco, *Serritia maseescens*, Rice
Disease Resistance	
Response Gene-49	
(DRRG-49)	Pea
Stilbene synthase	Grape
Hevein	Rubber

5. Advancements are also expected in the area of production physiology. A clearer understanding of biosynthetic pathways of various quality components can lead to identification of genes involved in these processes. Manipulation of such biosynthetic pathways can lead to genotypes with greater efficiency. For example, the isolation and cloning of genes involved in the curcumin biosynthetic pathway can give ways and means to enhance efficiency either by inserting genes with more powerful promoters, or by blocking the allosteric sites of enzymes controlling the critical steps, so that allosteric inhibition can be avoided, thereby leading to increased accumulation of curcumin.

Conclusion

Science has advanced at an unprecedented rate during the last century. The 20th century witnessed far greater achievements than what mankind has achieved during the past 19 centuries. We anticipate greater achievements during the coming decades and perhaps should prepare ourselves to see the unbelievable. Science has become so powerful, that there is nothing that is impossible. The next generation will see many wild imaginations and dreams coming true. This is true in the area of spices as well, provided enough expertise and capital are invested in spices research. The areas of genetic engineering and biotechnology are sophisticated, highly competitive and capital intensive. If spices research can be elevated to that level, then we will see dreams being translated into realities. Let us hope for the best, but the best will come only where there is talent, dedication, commitment and hard work.