Keynote Address

Progress in plant breeding techniques : scientific, social and global impact

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Today, it is generally recognized that the promotion of basic research and practical application of biotechnology are essential to maintain and even to enhance agricultural production, to conserve natural resources and to improve the environmental quality on a global scale for the present and future of mankind.

This concept has, in fact, been materialized in various areas of plant science, particularly in plant breeding. There are presently a large number of new varieties worldwide with outstanding characteristics that have been introduced by recombinant DNA techniques.

These achievements require us to consider genetically modified plants within the context of genetic changes occurring on the earth, either naturally -- which we cannot manage; or artificially -- which we can manipulate.

This further tells us that it is important to understand and utilize new biotechnological approaches within the comprehensive scope of plant breeding where imperfect materials are transformed into practically useful new varieties.

Genetic improvements

1. Sexual hybridization

The remarkable genetic improvement in crop plants achieved to date is largely based on sexual hybridization. As emerging cellular and molecular technologies develop further, sexual methods will continue to play a major role in crop improvement.

Although plant geneticists have frequently been able to manipulate and recombine the genome into agriculturally effective combinations using sexual breeding methods, they are limited by the lack of understanding of interactions among genes.

2. Tissue and cell culture / Totipotency

The ability of plant cells to regenerate a whole plant is called totipotency. Plant breeding by using tissue and cell culture is based on this unique characteristic of plant cells.

3. Somatic embryogenesis

Somatic embryogenesis is a typical example of totipotency of plant cells. Somatic embryos are formed directly or indirectly from various kinds of tissues. A large number of studies have been conducted to develop plant regeneration systems from plant cells.

4. Production of haploid plants through anther and pollen culture

For plant breeding, haploids are an attractive material. Since they do not have any alleles, a recessive character in a diploid can be detected in a haploid plant. In addition, we can easily obtain recombinant inbred lines through the doubling of chromosomes of haploid plants derived from hybrids. Anther culture and isolated microspore culture which are suitable techniques for obtaining a large number of haploid plants have been developed in more than 250 plant species since the first report in 1964.

For breeding, since a large number of doubled haploid plants are necessary, more efficient methods and transformation systems should be developed in haploid plants.

5. Protoplast isolation and fusion

The protoplasts can be fused to obtain somatic hybrid plants whose parents can not be cross-pollinated or into which genes from wild species can not be introduced.

Several cell fusion and screening methods have been developed; for example, fusion by treatment with polyethylene glycol (PEG) and electric pulse, asymmetrical fusion techniques and screening of fused cells by antibiotics, identification by restriction fragment length polymorphism (RFLP) and random amplified polymorphic DNA method (RAPD) analysis, etc.

We know that fusion remains a promising method for introducing genes into vegetatively propagated crops from their allies, re-synthesizing polyploid crops with various combinations of genome, and introducing cytoplasmic genes.

6. Identification and isolation of genes

To clarify the characteristics of genes controlling phenotypical traits and to improve plants by changing the genetic information, identification and isolation of genes corresponding to a trait are required. DNA fragment carrying a gene for target trait can be identified by utilizing a rice physical map produced by genome analysis.

The genes isolated and characterized as indicated above can be utilized for the improvement of not only rice but also other crops and the molecular information contained in these genes should contribute to the acquisition of fundamental knowledge in plant science.

7. Incorporation of foreign genes

(1) *Agrobacterium*-mediated gene transformation

Transformation enables to introduce new useful genes into plants without changing their original basic characters. Among the methods of introduction of foreign gene(s) into plants, *Agrobacterium*-mediated transformation is the most popular. Recently successful transformation of monocots, for example rice and maize, has also been achieved, thus widening the potential of this method.

For the utilization of transgenic plants in breeding, there are several barriers to overcome. First, efficient transformation systems must be developed in various plant species; second, more useful genes should be isolated; third, gene expression must be controlled. Finally we must determine the influence of transgenic plants on the environment, their suitability as foods, and win public acceptance.

(2) Electroporation

As a mechanical method for gene transfer, electroporation is used for species for which a protoplast culture system has been established.

(3) Microprojectile bombardment

Microprojectile bombardment employs high velocity metal particles to transfer DNA into plant cells or tissues. A wide range of plant species for which the *Agrobacterium*-mediated method or protoplast transformation is not effective have been transformed by microprojectile bombardment.

However, the transformation frequency by this method is still low. As a result, the experiments are labor-intensive and rather expensive. It is anticipated that improvements in hardware design and particle gun availability will enable to increase the efficiency and the number of crops that can be transformed using this technology.

Genetic resources

Genetic resources of crop species harbor the genes needed for future crop improvement. Germplasm collections provide food, feed and fiber security for mankind and materials to solve basic problems in the field of biology. Presently the utilization of genetic resources is hampered by the rapid decrease in diversity worldwide. The need to protect global genetic resources is more acute than ever before.

The Japanese government has ratified the "Convention of Biological Diversity", which is an international agreement to share the benefits and sustainable use of global genetic resources. Major focus in the research on genetic resources is placed on strengthening international networks for the conservation of plant diversity.

Prospects for improving food production and environment

Due to the rapid increase of the world population and the deterioration of the earth's environment, there is a great concern about global food supply. In many countries scientific research and innovative agricultural activities have been and are being conducted in order to improve the world food supply and global environment. In this regard, biotechnology, including the methods for the development and use of genetically modified organisms, is one of the key technologies. Many researchers are trying to develop novel plants and microorganisms, such as plants with low allergenicity and production of bio-degradative plastics, vaccines and other useful industrial materials.

Safety assessment of transgenic plants

On the other hand, the introduction of new molecular technologies in the early 1970s raised the issue about the safety of biotechnology procedures.

These discussions resulted in a number of national and international recommendations and the formulation of guidelines or regulations, and legislation. The OECD has set up general principles for the safe development of recombinant DNA organisms since 1983, such as "good industrial large-scale practice (GILSP)", "familiarity" and "substantial equivalence" pertaining to microorganism handling, agricultural applications and new foods, respectively. In many countries voluntary guidelines or laws are compatible with the national regulations currently in effect.

Many enterprises are moving rapidly towards the commercialization and marketing of agricultural and industrial products developed through modern biotechnology. Both public and private sectors have, therefore, emphasized the need for the harmonization of regulatory approaches to assess these products in order to reduce unnecessary duplication, facilitate administrative work, improve public acceptance, thus contributing to the safe application of biotechnology, promoting global exchange of biosafety information, and avoiding further trade barriers. Such an international framework should not give a negative impact on research and development in biotechnology and technology transfer. We consider that it is important to develop an internationally harmonized framework for the safe handling of recombinant DNA organisms within a few years.

Present state of agricultural use of transgenic plants

In Japan overall development, including handling and culture of recombinant DNA organisms is subject to guidelines. The guidelines formulated by the Ministry of Agriculture, Forestry and Fisheries (MAFF) cover all categories of organisms, including plants, animals and microorganisms, and their general release. The purpose of these guidelines is to define basic requirements concerning the appropriate application of recombinant organisms in agro-industries so as to ensure the safe use of recombinant organisms and to achieve sound overall development. In the case of plants, safety is assessed in four steps, two steps in green houses for research, the other two in field for practical use. So far, forty-two transgenic plants have complied with the MAFF guidelines formulated by the Ministry. The fifteen plant lines of virus-resistant tomato, petunia, rice, melon, tomato with delayed ripening and herbicide-tolerant canola and soybean are now being released in Japan.

The Ministry of Health and Welfare formulated guidelines relating to the food safety assessment of crop plants obtained through biotechnology in February 1996. Furthermore, MAFF also formulated guidelines for the safety assessment of animal feed and feed additives obtained through biotechnology in April 1996. Seven transgenic crops including herbicide-tolerant canola and soybean, and insect-resistant potato and corn are now under review in the Ministries.

Public acceptance

On the other hand, the general public is not familiar with recombinant DNA technology, and some people may feel uncomfortable with biotechnology. In order to promote agricultural biotechnology, it is essential to supply precise information about biotechnology, especially recombinant DNA technology, to the general public to join its acceptance of biotechnology-derived products.

Governments and developers should constantly promote activities to build and foster understanding on the part of the general public about recombinant DNA technology and safety to introduce the products while removing consumers' misgivings.

We wish to develop this technology even further so that it will contribute to the welfare of the people all over the world as we try to live in harmony with nature. Therefore, all scientists concerned, especially ourselves gathered here today, should continue the research work in order to secure a more stable supply of foods, better environment and necessary energy for the world.

Assistance extended to developing countries

There are various constraints on agricultural production: heat, drought, salinity, pests, diseases, etc. These biotic and abiotic constraints result in the instability of crop yield, which often leads to starvation and malnutrition of a large number of people in developing countries. In addition, the rapidly growing population accelerates the deterioration of natural resources, especially in marginal lands. We are now faced with the need to increase the sustainability of agricultural production, while the lack of materials and infrastructure necessary for agricultural production in most of the developing countries makes it difficult to increase and stabilize their agricultural productivity.

Moreover, recent progress in biotechnology is rapidly increasing the possibility of modifying crops genetically to make them highly tolerant to various kinds of adverse conditions. These new crops should contribute to the increase and stabilization of agricultural production with lower labor and financial input in developing countries.

On the other hand, lack of funding and human resources results in the application of limited aspects of biotechnology in most of these countries, where genetic engineering is too costly. It is hardly possible to carry out molecular breeding in the developing countries for the time being, although the farmers in these countries need the benefits of modern high technology.

Thus advanced research institutes in the public sector of industrialized countries may take the initiative to promote shuttle-type programs for the development of biotechnology techniques adapted to the specific conditions of individual developing countries in collaboration with international organizations.

How to assist developing countries

One of the major constraints on promoting biotechnology research and development in the developing countries is the shortage of appropriately trained researchers and technicians. Some advanced biotechnology research units of industrialized countries and international organizations now offer high-level training programs to researchers and technicians in developing countries. In addition to the training of researchers and technicians, training of research managers and leaders of national agricultural research systems in developing countries is urgently required.

Another issue is the enactment of regulations and guidelines necessary for the promotion of biotechnology research, development and use in developing countries. In order to stimulate the private sector to participate in this field in developing countries, intellectual property protection is a prerequisite. In addition, biosafety considerations are necessary to gain the public acceptance for genetically modified plants and microbes, a topic which we are going to discuss in detail in this symposium. The experiences of industrialized countries in this matter could be helpful to many developing countries.

Conclusion

In conclusion, I would like to say a few words which are related to the main theme of the present symposium. One of the most important goals to achieve is to create outstanding plant varieties which can really provide benefits to mankind. There will be two ways for further enhancing the effective use of outcome of biotechnology, especially biosafety field tests. One is to improve public understanding. Since public attitudes are shaped more by history, culture, and sociological factors than by scientific considerations, special efforts to improve public acceptance need to be strengthened by all sectors involved. Another is to promote international harmonization. These two mechanisms are actually interrelated as widely agreed harmonization will certainly result in increased public acceptance.

Above all, it is essential that we strictly maintain scientific quality and objectivity to evaluate biosafety issues.