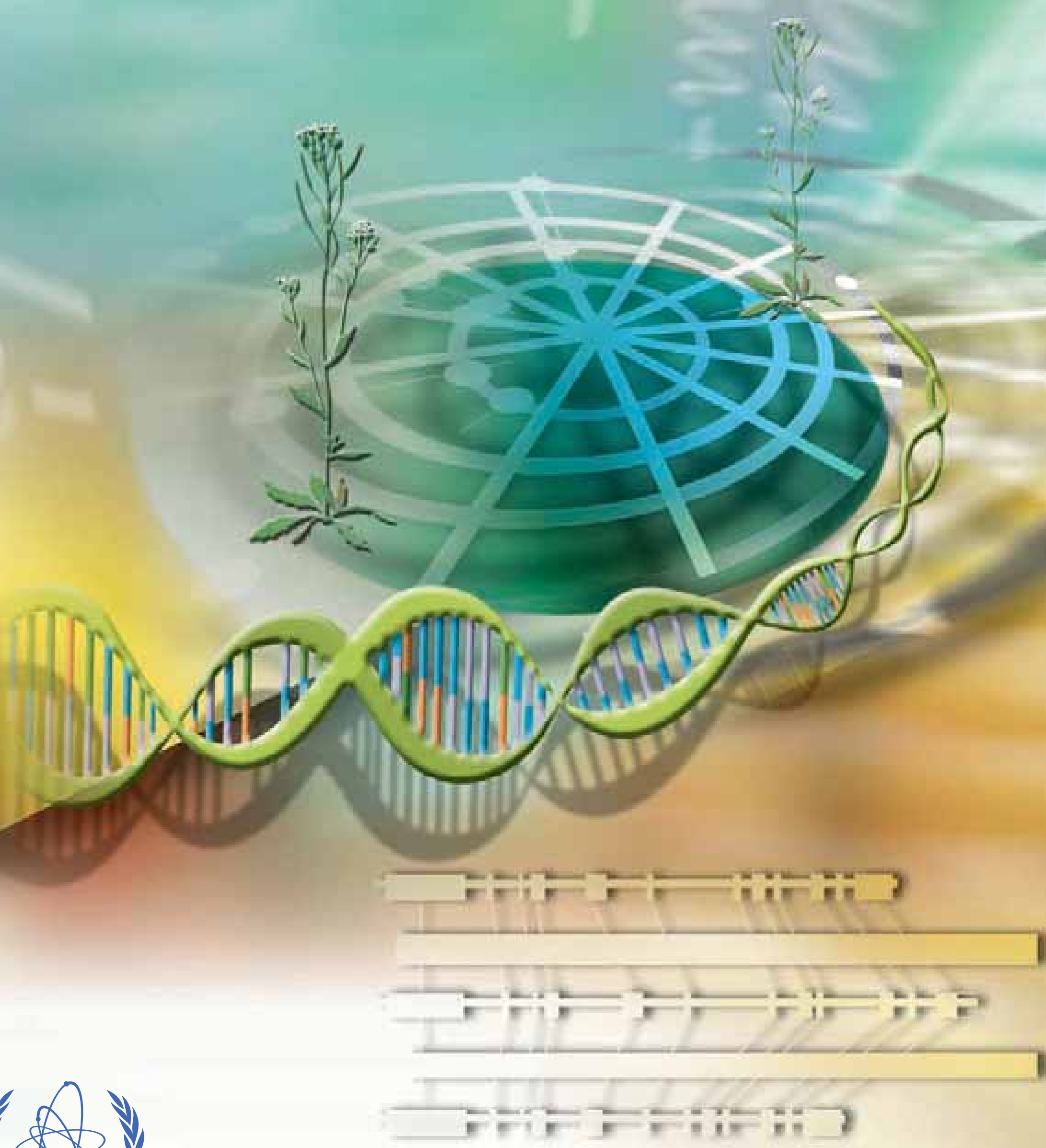
Induced Plant Mutations in the Genomics Era

Edited by Q. Y. Shu



Joint FAO/IAEA Programme
Nuclear Techniques in Food and Agriculture

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Preface

The year 2008 marks the 80th anniversary of mutation induction in plants. The application of mutation techniques, i.e. Gamma-rays and other physical and chemical mutagens, has generated a vast amount of genetic variability and has played a significant role in plant breeding and genetic studies. The widespread use of induced mutants in plant breeding programmes throughout the world has led to the official release of more than 2,700 plant mutant varieties. A large number of these varieties (including cereals, pulses, oil, root and tuber crops, and ornamentals) have been released in developing countries, resulting in enormous positive economic impacts.

During the last decade, with the unfolding of new biological fields such as genomics and functional genomics, bioinformatics, and the development of new technologies based on these sciences, there has been an increased interest in induced mutations within the scientific community. Induced mutations are now widely used for developing improved crop varieties and for the discovery of genes, controlling important traits and understanding the functions and mechanisms of actions of these genes. Progress is also being made in deciphering the biological nature of DNA damage, repair and mutagenesis. To this end, the International Symposium on Induced Mutations in Plants was organized by the International Atomic Energy Agency (IAEA) and the Food and Agriculture Organization (FAO) of the United Nations through the Joint FAO/IAEA Division of Nuclear Techniques in Food and Agriculture.

The Symposium comprised an open session, two plenary sessions and ten concurrent sessions, covering topics from induced mutations in food and agriculture, plant mutagenesis, genetic diversity, biofortification, abiotic stress tolerance and adaptation to climate changes, crop quality and nutrition, seed and vegetatively propagated plants, gene discovery and functional genomics. A workshop on low phytate rice breeding was also organized. About 500 participants from 82 Member States of the IAEA and FAO, and nine international organizations/institutions attended the Symposium, with a good balance between the private and public sector, as well as developing and developed Member States. The Symposium received valuable assistance from the cooperating organizations and generous support from the private sector, for which the sponsoring organizations are most grateful.

This publication is a compilation of peer-reviewed full papers contributed by participants. They were either oral or poster presentations given in different sessions except Concurrent Session 3 (which will be compiled by the Human Health Division in a separate publication). These papers not only provide valuable information on the recent development in various fields related to induced mutations, but also on the social and economic impact of mutant varieties worldwide. Therefore, these Proceedings should be an excellent reference book for researchers, students and policy makers for understanding applications of induced mutations in crop improvement and biological research.

Qu Liang
Director
Joint FAO/IAEA Division of Nuclear Techniques in Food and Agriculture
IAEA

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Opening Remarks by W Burkart

Deputy Director General of the IAEA
Department of Nuclear Sciences and Applications

Dear colleagues, ladies and gentlemen,

It is a pleasure to welcome you to the Vienna International Centre for the International Symposium on Induced Mutations in Plants.

This international Symposium, promoted by the Joint FAO/IAEA Division, is the eighth of its kind – the first was held in 1969 - dedicated to harnessing and disseminating information on current trends in induced mutagenesis in plants. These symposia have dealt with themes relating to the development of efficient protocols for induced mutagenesis and their role in the enhancement of quality traits, as well as resistance to biotic and abiotic stresses in crops and the integration of *in vitro* and molecular genetic techniques in mutation induction.

The Joint FAO/IAEA Division has been promoting the efficient use of mutation techniques since the late 1960's, in line with the Agency's "atoms for peace" agenda, very much related to agricultural policy and practices of some our main donor nations. In 1960, for example, in the United States, disease heavily damaged the bean crop in Michigan — except for a promising new variety that had been made by radiation breeding, which quickly replaced the old bean.

The Manual on Mutation Breeding, edited by the Agency and first published in 1970, updated in 1979 and reprinted several times afterwards, was the first book of its kind in the world. It has been widely used both as textbook in universities (translated into a couple of local languages) and reference book for breeders in their profession. Together with the training provided to scientists in developing countries and the support and coordination of research activities in this area, this manual has greatly promoted the correct and efficient use of mutation techniques in crop improvement.

At a time when the world is facing a food and energy crisis of unprecedented proportions, plant mutation breeding can be a catalyst in developing improved, higher-yield, saline-resistant sturdier crop varieties. More and more, the interest of the scientific community in this discipline has focused on the discovery of genes that control important traits, and on understanding the functions and mechanisms of actions of these genes.

The year 2008 will mark the 80th anniversary of mutation induction in crop plants. The widespread use of induced mutants in plant breeding programmes throughout the world has led to date to the official release of close to 3,000 mutant varieties from more than 170 different plant species. Many of these varieties (including cereals, pulses, oil, root and tuber crops, and ornamentals) have been released in developing countries, resulting in considerable positive economic impacts, which are measured in billions of US dollars and tens of millions of hectares of cropping area.

In effect, the application of mutation techniques, i.e. Gamma rays and other physical and chemical mutagens has generated a vast amount of genetic variability and plays a significant role in plant breeding and genetics and advanced genomics studies. There will be many recent mutation induction success stories presented here, in a wide range of disciplines. Please allow me to just cite two in the field of plant breeding and genetics, fostered by the Agency through the Joint FAO/IAEA Division in collaboration with the Technical Cooperation Department:

The first example is:

(i) Mutant barley varieties that thrive in an up to 5,000 meter altitude in the inclement highlands of Peru. The adoption and cultivation of these mutant varieties account for over 52% yield increase between 1978 and 2002 translating to significant increases in income generation for the Indian farmers. For the socio-economic impact of improved barley mutant varieties, the Peruvian government has awarded the 2006 Prize of "Good Governmental Practices" to the principal counterpart.

The second example relates to:

(ii) Mutant rice varieties that thrive in the high salinity region of the Mekong Delta in Vietnam. The breeder of one of these varieties, with export quality that made it up amongst the five top export rice varieties, got the 2005 National Science and Technology Prize of Vietnam for this variety because of socio-economic impact.

These are just two examples of many that showcase the ability of mutation induction to produce hardier cops adapted to harsh environments.

Ladies and gentlemen, this year, 2008, will be remembered as the year in which the global conscience understood the realities of climate change, the food crisis and the energy debate and its link to hunger. These big issues are intimately interlinked, and translate in the agronomy field into a competition between food, feed and fuel for soil, water, human and financial resources.

Mutation induction has proven flexible, workable, and ready to use on any crop. In addition, it is a non-hazardous and low-cost technology that has the ability to address current challenges in agriculture. The breeding of new mutant varieties - with a higher yield potential, more productive biomass for energy use, better nutrient composition for human health, better adaptation to climate change and variability, or a heightened potential to sequester carbon - will be the driving force to meet the challenges of the 21st century.

Combined technology packages based on mutation induction, the most advanced genomic screening techniques and nuclear techniques applied to good agricultural practices will foster powerful new tools to improve plant breeding. In this respect, this Symposium brings together key players in basic research, as well as in the development and application of technologies relating to the efficient use of induced mutations for crop improvement and empirical genetic studies.

Ladies and gentlemen, dear colleagues, before you begin your deliberations, I would like to remind you that this Symposium is representative of one of the best collaborations in the United Nations system – the Joint FAO/IAEA Division - two sister agencies working for the welfare of humanity- a partnership that is already 44 years old.

I wish you fruitful discussions and a successful participation at the Symposium.

Opening Remarks by S Pandey

Representative of the FAO
Director of Plant Production and Protection
Agriculture and Consumer Protection Department

Mr. Chairman, distinguished guests, ladies and gentlemen,

On behalf of the Director General of the Food and Agriculture Organization in Rome, I bring you greetings and welcome you to the Joint FAO/IAEA International Symposium on Induced Mutations in Plants.

It has been 12 years since the last Symposium and I take this opportunity to congratulate Mr. Liang and his staff for organizing this Symposium at this time.

Crop plants form the major components of human diets, providing the required calories and nutrients to sustain life. With recent soaring of food prices, which is one of the immediate causes of the current food security crisis, the need to efficiently increase food availability through the production of high yielding crop varieties under the contrary effect of climate change and variability, plays a key role in ensuring food security.

As such, holding this Symposium at this moment in time is a significant and blissful coincidence.

An essential aspect of crop improvement is the utilization of the available genetic variation to produce new crop varieties. Induced mutations are a proven tool in creating a wealth of desirable genetic variability in plants, and its success in crop improvement abound.

Currently the Joint Division's mutant varieties database registers over 2,700 mutant and mutant-derived varieties. Furthermore, some of these mutant varieties have contributed significantly to the livelihoods of farmers and their respective country's economy. For instance:

- *Diamant* and *Golden Promise* being the progenitors of most of the cultivated barley varieties used in brewery industry in Scotland and most of Europe, contributing over 20 million US dollars per annum in additional income to farmers annually;
- *Durum* wheat varieties used in the pasta industry in Italy arising from mutants account for tens of millions of US dollars in additional income to farmers per annum;
- Grapefruit in the US with *Rio Star* mutant accounting for 75% of the US grapefruit industry;
- The mutant pear variety *Gold Nijesseiki* in Japan contributes 30 million US dollars in additional income to farmers annually;
- The rice mutant variety *Zhefu 802* yield 10.6 million ha in China, giving a yield increase of 10.5% between 1980 and 1995. This translates into providing food to an extra two million people per year; and
- In Vietnam, where rice export is one of the main national revenue sources, eight high-yielding mutant rice varieties with other socio-economic value traits including high quality, tolerance to salinity and short duration allowing up to three harvests per year have been developed and adopted by farmers, providing them an extra income of 300 million US dollars this year.

These successes and many more were achieved by including induced mutations in the plant breeding scheme.

The efficiency of mutation induction is directly related to producing, handling and assaying the required large numbers of mutant stocks and could be expensive, laborious, time-consuming and often dependent upon the growing season of the crop. However, recent advances in genomics, especially the quantum leap in the volume of publicly available genomics resources, imply that a high throughput platform such as Targeted Induced Local Lesions in Genomes (TILLING) which utilize induced mutations, will make the rapid evaluation of mutant stocks for specific genomic sequence alteration more practicable.

The history of the Joint FAO/IAEA Division dates back to 1964, and since then its activities have been aimed at promoting the use of nuclear technologies in crop breeding and genetics in our Member States.

Let me express my deep satisfaction with the efficient synergistic link the Joint FAO/IAEA Division provides between FAO and IAEA. Indeed, their input to FAO's programmes in agriculture is highly valued by the sister divisions in FAO, and I congratulate the Joint FAO/IAEA Division for 44 years of flawless service to the Member States of both organizations. In return, FAO is pleased to note the added value it could offer in return to the IAEA programming.

More of the successful results of our continued support to Member States, I believe, will be detailed to you during the presentations over the next three days.

Still there are challenges ahead. The looming adverse effects of climate change and variability affecting crop productivity requires intervention to produce new varieties which can perform more efficiently under severe water and climatic conditions, and also ensure a continued maintenance of the existing biodiversity.

Malnutrition, with respect to micronutrients like vitamin A, iron and zinc, affects more than 40% of the world's population. While various interventions such as supplementation and fortification have been proposed, providing major staple crop varieties that accumulate greater concentrations of vitamins and minerals in their edible tissues, will be a sustainable intervention particularly for low-income populations.

The demands for bioenergy crops and/or efficient use of existing crops to provide both food and fuel without threatening the current food supplies, pose yet another challenge for our crop improvement programmes.

I encourage you to consider these challenges in your deliberations and the potential of induced mutations by addressing these efficiently.

Ladies and gentlemen, again I welcome you to this Symposium.

Summary of the FAO/IAEA International Symposium on Induced Mutations in Plants by T Ishige

President
National Institute of Agrobiological Sciences, Japan

Thank you Mr. Chairman.
Ladies and gentlemen and distinguished guests,

On behalf of the Steering Committee, I would like to express my sincere appreciation to all the speakers and participants for their informative presentations and discussions during the Symposium. I would like to acknowledge and thank the members of this meeting for supporting this conference here in Vienna.

In the flyer, it is written that 2008 is the 80th anniversary of induced mutation breeding. The commercial utilization of approximately 3,000 mutant-induced and mutant-derived varieties strongly shows that mutation breeding is a useful tool for generating new germplasm for crop improvement.

Mutation induction by radiation is the method of choice in China, India and Japan. In plant genetic and breeding research, induced mutations have contributed to the discovery and identification of gene functions following the completion of genome sequencing projects in *Arabidopsis* and rice.

In this Symposium, a wide range of topics related to mutation breeding was discussed. In the keynote presentation, Dr. Ron Phillips showed the importance of expanding gene variation for crop improvement. He presented modern mutation technologies which are useful for practical plant improvement and plant science. Dr. Lagoda showed the importance of international cooperation in plant mutation genetics and breeding. He introduced the role of the joint FAO/IAEA Programme.

In Plenary Session 1, history and topics of mutation breeding of rice, barley, legumes and other crops in the world were presented.

In Concurrent Session 1, "Mutational Enhancement of Genetic Diversity and Crop Domestication" was discussed. The key genes responsible for domestication in barley, such as the 6-rowed spike, were identified by the use of natural variation and mutants. Unique agronomically useful characteristics were isolated in wheat, sunflower, soybean, and lupine.

In Concurrent Session 2, the topic "Plant Mutagenesis - DNA Damage, Repair and Genome" was discussed. Following the completion of the genome sequencing in *Arabidopsis* and rice, we can identify a particular mutation, such as deletion size and the point of mutation at the molecular level. Furthermore, the mechanism of gene function and gene repair can be identified.

In Concurrent Session 3, "Biofortification of Staple Food Crops for Improved Micronutrient Status" was discussed. One of the most important activities of the IAEA that of human nutrition, was presented. DNA changes that provide variations useful for human nutrition are seen to become increasingly important, including the transgenic approach (for example, golden rice). Many of these variations useful in human nutrition will be introduced together with new traits desired by growers (for example, submergence tolerant rice). Complex interaction such as the role of phytic acid in micronutrient availability was discussed.

In Concurrent Session 4, "Induced Mutations for Traits That Affect Abiotic Stress Tolerance and Adaptation to Climate Change" was discussed. Various approaches to develop crops with tolerance to abiotic stresses, including drought, salinity, and tolerant root systems were presented.

In Concurrent Session 5, "Induced Mutations for Enhancing Crop Quality and Nutrition" was discussed. Mutation research for seed phosphorus and useful starch mutants were presented. We also learned how a mutant trait can be modified by marker-assisted selection as in the example of Quality Protein Maize (QPM).

In Concurrent Session 6, "New Techniques and Systems for Mutation Induction" were discussed. New mutation technologies, such as transposon Tos17, restriction endonuclease, space irradiation, and ion beams, were presented.

In Concurrent Session 7, "High Throughput Techniques for Mutation Screening" was discussed. Screening for mutants is an important step in mutation breeding, as well as mutation induction. Recently, very useful molecular screening tools, such as TILLING, using the genome sequences of agronomically useful traits in some crops, have become available.

In Concurrent Session 8, "Mutation Induction and Breeding of Ornamental and Vegetatively Propagated Plants" was discussed. Mutation breeding is very useful for improvement in asexual crop species, where hybridization is not possible. Typically, in this area, breeding objectives are focused primarily on flower colour and shape, which can be easily screened by observation.

In Concurrent Sessions 9 and 10, "Induced Mutations in Seed Crop Breeding" was discussed. There are many useful traits induced by mutations, such as semi-dwarfness, resistance to diseases, and quality components of grains and beans.

In Plenary Session 2, "Induced Mutations in the Genomics Era: New Opportunities and Challenges" was discussed. Genomic analysis and metabolite profiling of induced mutants has become an excellent approach for the analysis of gene function.

In this Symposium, we discussed practical mutation breeding and the analysis of gene function originating from many mutants. As you know, now, genome science and molecular biology are very powerful tools to analyse gene function. Genome sequencing of rice was completed in 2005 by the International Rice Genome Sequence Project (IRGSP), with NIAS (National Institute of Agrobiological Sciences), in Japan playing the central role. This effort has resulted in dramatic changes in the mutation breeding of rice, as well as other monocot crops which show similar genomic synteny with rice. Molecular genetics and information technology based on genome sequencing will be presumably powerful tools for the selection of mutants exhibiting specific characteristics. NIAS is now extensively applying genome science to organisms such as rice, wheat, barley, soybean, silkworm and pig. The Radiation Breeding Institute of NIAS, directed by Dr. Nakagawa, is studying practical radiation breeding and creating of new genetic resources by mutation.

Furthermore, mutation technologies can provide many new genetic resources induced by radiation, chemicals, and several kinds of genetic engineering. Those new genetic resources are very useful not only for practical breeding, but also for plant science. We can utilize and combine the multitude of useful genetic resources and modern molecular technologies.

Finally, I would like to draw some conclusions from this wonderful conference.

(1) Direct mutation in specific genes for specific traits

Scientists have utilized natural variation from spontaneous mutations, as well as induced mutations for many years. But now there are new technologies to direct mutation in specific genes for specific traits. With more and more genome and gene sequence information and more knowledge about gene regulation and gene networks, we will see the development of more "direct mutation" techniques. I predict that the next IAEA conference will feature direct mutagenesis methods as a major topic.

(2) Variation inherent in various species

We will probably also see more powerful methods for recognizing and utilizing the variation inherent in various species.

(3) Advancement of many new alleles at loci of interest.

Another advancement that became clear at this conference is our ability to recognize many new alleles at loci of interest.

(4) Base change by various mutagens

The new technologies presented here demonstrate the ability to produce many base changes by various mutagens.

(5) We also saw how even base changes not leading to a mutant phenotype are useful, for purposes such as tracing variation and farm products.

I believe this Symposium and the proceedings of the presentation, which will be published following this Symposium, will help establish the valuable role that mutation breeding has played in the disciplines of plant science and world agriculture. I hope that we will meet again in the near future and discuss the progress of the mutation research after this meeting.

Finally, we applaud the IAEA and FAO for organizing this excellent conference – as well as all their other activities of coordinating research, conducting research, development of database, and so forth.

Thank you so much.

Closing Statement by A M Cetto

Deputy Director General of the IAEA
Department of Technical Cooperation

Distinguished delegates, ladies and gentlemen

I am very pleased to speak to you today, as the International Symposium on Induced Mutations in Plants draws to a close.

This Symposium was the eighth in the Joint FAO/IAEA Programme's Symposium series, dedicated to harnessing and disseminating information on current trends in induced mutagenesis in plants. The first meeting was held in 1969, and the most recent in 1995. Thirteen years on, in a climate of increasing food shortages, it was high time that this Symposium was held. Although mutation breeding is 80 years old, as we just heard, new applications continue to be found and will continue to be developed. The application of mutation techniques, such as Gamma-rays and other physical and chemical mutagens, has generated a vast amount of genetic variability and has played a significant role in plant breeding and genetic studies in countries throughout the world. The importance of these techniques is reflected in the large number of participants gathered here: a total of 500 delegates from 81 countries that are IAEA and FAO Member States and nine organizations. I believe that the extensive scientific programme, which included 126 oral and 252 poster presentations, is an indicator of the range, depth and relevance of the topic.

Being aware of the scientific nature of this Symposium, let me however, say a few words on behalf of the IAEA's TC Programme. The programme provided financial support to several participants to attend this Symposium, but more importantly, many of the participants have been trained through the Technical Cooperation Programme, supported by the FAO/IAEA Joint Division. Technical training of this sort is a core component in the implementation of the IAEA mandate "Atoms for Peace, Health and Prosperity."

Over the past five years, food and agriculture has accounted for one of the largest areas of the IAEA Technical Cooperation Programme around the world, and projects in this area are on the increase in the 2009–2011 programme cycle. The Agency's combination of technical and managerial expertise offers Member States significant benefits in the formulation and development of projects, transfer of technology, infrastructure development and capacity building. Support is delivered through training courses, fellowships, expert and scientific visits, and through setting up of laboratories.

Ladies and gentlemen, Pandit Jawarlal Nehru, the great former prime minister of India, remarked once that "Everything else can wait but not agriculture," and this remark remains relevant today. There is no short-term magic formula to solve the world's food problems. We must take advantage of all possible modes of intervention and action. Nuclear technology will continue to play an essential role in strengthening conventional breeding through induced mutations and efficiency-enhancing biomolecular technologies. As we have just heard, there is now a trend towards directed mutagenesis. I am sure that our technical colleagues have taken note of the suggestion to dedicate the next Symposium to this topic. These will hopefully continue to represent safe techniques, respectful of nature and the environment, supporting and speeding up natural processes to develop food and agricultural products with improved characteristics and increased value.

I sincerely hope that you have enjoyed your stay in Vienna, and that the Symposium has stimulated you to find innovative ways to face the challenges that face us all. Your work is key to ensuring the sustainability of agriculture and to meeting the ever-growing global demand for food resources.

I wish you all the best in your continuing endeavours and a safe journey back to your home countries.

Thank you.

A Summary of the International Symposium on Induced Mutations in Plants

Q Y Shu

1. Organization of the Symposium

The International Symposium on Induced Mutations in Plants was held from 12-15 August, 2008 at the Vienna International Centre. Almost 500 participants, more than half of them from developing countries, attended the Symposium. Nine international organizations and 82 member countries, among these 60 developing countries, were represented at the Symposium.

The Symposium was opened by IAEA's Dr. W. Burkart, Deputy Director General, Department of Nuclear Sciences and Applications. Dr. S. Pandey, Director, delivered opening remarks on behalf of the Food and Agriculture Organization of the United Nations. Also in the opening session, Professor R. Phillips (USA), Vice President of the International Crop Science Society, delivered a keynote address on "Expanding the Boundaries of Gene Variation for Crop Improvement," and Dr. P.J.L. Lagoda, Head of the Plant Breeding and Genetics Section in the IAEA, introduced the role of the Joint FAO/IAEA Programme on Nuclear Techniques in Food and Agriculture in networking and fostering of cooperation in plant mutation genetics and breeding. Dr. T. Ishige, President of the Japanese National Institute of Agrobiological Sciences summarized the opportunities and the way forward for plant mutagenesis in the genomics era, and Dr. A.M. Cetto, Deputy Director General of the Department of Technical Cooperation in the IAEA, officially closed the Symposium after highlighting the role of technical cooperation in promoting the use of mutation techniques in Member States.

Apart from opening and closing session, the Symposium was organized into two Plenary Sessions, 10 Concurrent Sessions, and one Workshop. Four hundred twenty-four abstracts were submitted for the Symposium - out of these, 129 papers were selected for oral presentation (including invited talks) and 184 for poster presentation.

The Symposium was organized by the International Atomic Energy Agency (IAEA) and the Food and Agriculture Organization (FAO) of the United Nations through the Joint FAO/IAEA Division of Nuclear Techniques in Food and Agriculture. The great cooperation and support of the following organizations contributed to the success of the Symposium and is highly commended: Bhabha Atomic Research Centre (BARC, India); Chinese Society of Agricultural Biotechnology, European Association for Research and Plant Breeding, Indian Society of Genetics and Plant Breeding, and National Institute of Agrobiological Sciences (Japan).

2. Overview of topics of the Symposium

The year 2008 marked the 80th anniversary of mutation induction in crop plants. Although induction of mutations had often been considered as accidental for a long time since it was discovered, it has been globally explored and has significantly contributed to increased agricultural production over the past half century. During the past 10 years, more and more molecular biological studies have proven that mutations are not mere "accidents," but that they could be deliberately induced, using various methods. Meanwhile, induced mutations have become more and more useful and important in modern genetic studies, such as gene discovery and function elucidation. By integrating molecular techniques, such as high throughput mutation screening techniques, induced mutations are now widely expected to play an even greater role in plant improvement than ever before. Progress in all those fields was repeatedly reported by various groups at the Symposium, which not only demonstrated the recent renaissance of mutation techniques but also outlined a bright future for these classic techniques.

2.1 Induced mutations for world food security

The widespread use of induced mutants in plant breeding programmes throughout the world has led to the official release of more than 2,700 mutant plant varieties. A dozen presentations convincingly documented the contribution of induced mutations to the increase of agricultural production that is valued in the billions of US dollars and millions of hectares of cultivated area. It is noteworthy that a large number of mutant varieties had been developed and widely cultivated in developing countries, hence greatly improving food security in those countries.

With population growth, the demand for food and feed is growing as well, while natural resources are limited. Erratic rain falls, sudden and severe drought conditions, excessive floods, etc., often related to climate change, even deteriorate crop production conditions. The yield potential of crop plants has to be significantly increased to combat the worsening food security situation. Traditionally, induced mutants with favourable traits have been directly or indirectly used in breeding new varieties. During the last decade, induced mutations have also been gaining increasing importance in plant molecular biology as a tool to identify and isolate genes and to study their structure and function. Knowledge of genes controlling important agronomic and quality traits are critical for plant breeders to develop proper strategies and efficiently implement breeding programmes. Therefore, induced mutations can contribute further to increasing global food production both directly and indirectly by increasing yield potential and stability.

2.2 Genetic diversity, crop domestication and improvement

Genetic variability is a very basic asset for crop domestication and improvement, as well as genetic research. While some plant species of cultivated crops have rich genetic diversity, others have very limited genetic variation. In Concurrent Session 1 alone, mutational enhancement of genetic diversity was reported in 17 plant species; those mutated populations will become important genetic resources for breeding, gene discovery, and functional analysis of various genes.

The work on lupine provided an excellent example of how spontaneous and induced mutations were utilized in the domestication and improvement of a modern crop. The introduction of the narrow leaf mutation resulted in the domestication of lupine as a dominant legume crop in Australia. The herbicide tolerance mutation showed the potential to double the yield by growing lupine under irrigation when herbicide is used for weed control.

Radiation was also proven to be effective for the production of wheat-alien translocation lines, which thus sets up a unique method for tapping genetic variability of wild species into cultivated crop plants. A research group in China established a sophisticated protocol and has used the introduced genetic variation for the development of several elite wheat varieties.

The potential of induced mutations for soil decontamination was demonstrated in sunflower. During the past two decades, the use of plants has been proposed as an alternative technique to remove toxic metals from contaminated soils. The efficiency of this technique largely depends on the biomass production and toxic metal uptake of plants grown in contaminated soils. In this direction, sunflower mutants generated through chemical mutagenesis were reported to have the capacity for the extraction of cadmium, zinc, and lead, three to five times higher than their wild type parent.

2.3 Induced mutations for quality and nutrition improvement

Induced mutation has long been used for the enhancement of crop quality and nutrition - two sessions (Concurrent 3 and 5) and one workshop were devoted to discourse on these topics. Many attributes of these two characteristics were covered. The research covered the enhancing of mineral elements (biofortification) and amino acids essential for humans and animals, alteration of protein and fatty acid profiles for nutritional and health purposes, as well as change of physicochemical properties of starches for different end-uses, to the enhancement of phytonutrients in fruits and reduction of anti-nutrients in staple foods. Several mutant genes have been successfully introduced into commercial crop varieties and significantly enhance the nutritional value of those crops. A few examples are given below:

1. **Quality Protein Maize (QPM).** Maize endosperm protein is deficient in two essential amino acids, lysine and tryptophan. The *opaque 2* mutant gene, together with endosperm and amino acid modifier genes, was used for the development of QPM varieties. QPM has almost twice as much lysine and tryptophan, and 30% less leucine, as normal maize, and has shown to have dramatic effects on human and animal nutrition, growth and performance. QPM varieties are now grown on hundreds of thousands of hectares.

2. **Low phytic acid (LPA) crops.** Much of the phosphorus is deposited as phytic acid and its salt form (phytate) in seeds. Since phosphorus and mineral elements such as iron and zinc in the form of phytate cannot be digested by humans and monogastric animals, reduction of phytic acid would increase the bioavailability of phosphorus and micronutrient mineral elements. Reports at the meeting showed a number of LPA mutations have been induced in barley, rice, wheat, and soybean, and new LPA barley varieties were released for commercial use. It is anticipated that LPA varieties will be eventually developed in various crops, which might ultimately reduce both phosphorus-pollution and increase bioavailability of phosphorus and micronutrient minerals in cereal grains and legume seeds.

3. **Oilseeds with optimised fatty acid compositions.** The optimal composition of plant oils depends on their end uses, for example, unsaturated fatty acids (oleic, linolenic) are desirable for salad and cooking oils, but increased concentration of saturated fatty acids (stearic, palmitic) is preferred for oils used in the food industry, since high temperature processes (frying) require oils resistant to thermo-oxidation. A number of papers described the alteration of fatty acids composition by mutation induction in soybean, sunflower, and other oil crops. By using the mutated genes, new varieties have been developed for numerous purposes.

2.4 New techniques for mutation induction, screening and utilization

In the past decade, there have been substantial technological developments in the induction, screening, and utilization of mutated genes. Traditionally, plant materials are treated with physical or chemical mutagens, such as Gamma-rays, ethyl methane sulphonate (EMS), and mutants are screened from the progenies in the field for morphological traits, or in the laboratory for chemical components. Due to the usually low mutation frequency, large populations need to be screened, which makes it rather expensive and sometimes impossible to identify a target mutation. The new developments reported at the Symposium include the use of space flight, ion irradiation, and transposable elements for mutation induction, the use of restriction endonuclease for site-directed, homologous recombination; DNA markers linked to mutated genes for marker-assisted selection and tracing of the gene, and target-induced local lesions in genomes (TILLING), as well as different variant versions for high throughput screening of mutated alleles.

New techniques for mutation induction. In China, spacecraft, recoverable satellites, and high altitude balloons have been used to bring seeds into space for mutation induction. During the past 10 years, more than 60 new mutant varieties were developed through this special programme. Ion beam irradiation has also been explored for plant mutation induction, mostly in China and Japan. Low energy ion beams are commonly used in China (mostly for agriculturally important crops), while high-energy ion beams are adopted in Japan (mostly for ornamental plants of high market value). The reports raised much interest among the participants. The transcriptional activation of transposable elements and their incorporation into various regions of the genome was reported in rice and maize. Since the position of transposable elements could be identified using PCR techniques, the mutation caused by transposition could be precisely positioned, and mutated genes could be easily identified. Another development of mutation induction is the use of restriction endonucleases (REs) for inducing double-strand breaks in plant genomes and consequently resulting mutations. This technology has been further integrated with zinc finger DNA binding proteins –zinc finger nucleases, and is now used for site-specific mutagenesis.

High throughput techniques for mutation discovery. One concurrent session was devoted specifically to presentations on this fast developing topic. Reports demonstrated that the TILLING technology is high throughput, cost-effective, and applicable to most organisms, hence its application is rapidly expanding. While TILLING was originally designed for screening mutations in mutant populations generated with chemical mutagens, such as EMS, it was also shown that it could be adopted to use mutant populations developed through physical mutagenesis, such as gamma and fast neutron irradiation. For example, the De-TILLING technique could be effectively used to detect a specific mutant in a pool of 6,000 plants. The other trend of mutation discovery technique is the use of high-throughput DNA sequencing techniques; several next-generation sequencing instruments are already available and it is foreseen that the mutation discovery will become more and more efficient and cost-effective.

A number of papers presented results on identification of DNA markers linked to mutated traits. These findings will be useful for marker-assisted selection and eventually for the cloning of mutant genes. This type of work has become an integrated part of mutation programmes in developed countries, and more recently in developing countries as well.

2.5 Integration of induced mutations with new "omics" techniques

While many papers across the different sessions discussed the use of molecular techniques or investigation at molecular level, the second plenary session highlighted the new trend of the integration of induced mutations with newly emerged "omics" techniques. These include genomics and functional genomics, microarray technology and transcriptomics, metabolite profiling, and spectral models of phenomes.

Induced mutants are investigated in a systematic way by the use of "omics" techniques, which coincides with the newly emerged subject "systems biology." Since the genetic background of mutants and their parent varieties only slightly differs, genes and pathways of mutated traits could be identified through comparative studies using various "omics" techniques. For example, results of metabolite profiling of low phytic acid mutants and their parents were indicative of the genes mutated in rice and soybean, and the deleted genes were identified through comparative genomics analysis in Citrus. These reports should be read not only for their importance of the particular subject, but also for the new direction and for the possible fields in which induced mutation can play a role in plant science.

2.6 Understanding the molecular basis of plant mutagenesis

Understanding the genetic control of plant mutagenesis is vital for the proper application and manipulation of mutation induction for enhancing genetic variation and plant mutation breeding. A session was assigned for some deliberations of progress in this field. Presentations covered topics such as recombination and extrachromosomal DNA on genome stability and evolution, and the role of human disease gene homologues for the maintenance of plant genomes, DNA repair mechanisms in the extremely radio resistant bacterium, the influences of environmental stresses (radiation, toxic metals, etc.) on plant genome stability, and DNA damage caused by various mutagenic agents. While most studies are not directly related to experimental mutagenesis, particularly for mutation induction, they could nevertheless provide useful information for studies on the molecular mechanisms of induced mutagenesis in plants.

3. The role of the Joint FAO/IAEA Programme on the Nuclear Techniques in Food and Agriculture

For more than 40 years, the Joint FAO/IAEA Programme has been promoting the research, development, and application of nuclear techniques in food and agriculture in the Member States. The use of nuclear techniques for plant genetics and breeding is a major field. The great achievements of the Joint Programme and the vital assistance given to Member States were widely and gratefully acknowledged by the participants during and after the Symposium.

3.1 Symposia on plant mutation breeding and genetics

With the successful completion of the International Symposium on Induced Mutations in Plants, the Joint FAO/IAEA Programme has already organized nine international symposia and numerous meetings in the field of Plant Breeding and Genetics. These events are the major international scientific forums for researchers working in this field. New ideas are sparked at the meetings and research topics conceptualized through discussion have become a source of force driving the progress of this scientific discipline. More than 80 letters of appraisal from participants demonstrate the high relevance and quality of the recent Symposium.

3.2 IAEA Technical Cooperation projects

The IAEA Technical Cooperation projects (TC projects), technically backstopped by the Joint FAO/IAEA Programme, play a great role in capacity building and efficient use of nuclear techniques for plant breeding in the Member States. Many of the success stories presented at the Symposium are from project counterparts, such as barley mutants in Peru, legume variants in India and Pakistan, as well as mutant rice in China, Tanzania, and Vietnam.

3.3 IAEA coordinated research projects

The IAEA's coordinated research projects (CRPs) are playing a unique role in promoting research and networking among scientists of both developing and developed countries. Three research coordination meetings of ongoing CRPs were organized in conjunction with the Symposium, and 17 CRP participants made oral presentations at the Symposium.

3.4 Future directions

As reflected by the presentations at the Symposium, induced mutations are playing an important role in modern plant improvement; their efficiency and application as a technology for crop improvement and plant research is foreseen to grow in the years to come. Mutation breeding is approaching a molecular era, which requires the extensive use of molecular techniques in mutation induction, screening, and utilization. Therefore, Member States will need to substantially improve their capacities to make full use of the opportunities of scientific advance. In this regard, the Joint Programme should continue to assist Member States through CRPs, TC projects and other efficient instruments.

The Symposium also highlighted some fields that the Joint Programme should pay special attention to. For example, the understanding of genetic control of the process of DNA damage, repair, and mutagenesis has been limited. This is vital for the proper application and manipulation of mutation induction for enhancing genetic variation and plant mutation breeding. High throughput techniques, such as TILLING, provide great promise, but their applicability and effectiveness in practical crop improvement programmes, particularly in developing nations, needs to be critically evaluated.