



**Joint FAO/IAEA Programme**  
Nuclear Techniques in Food and Agriculture

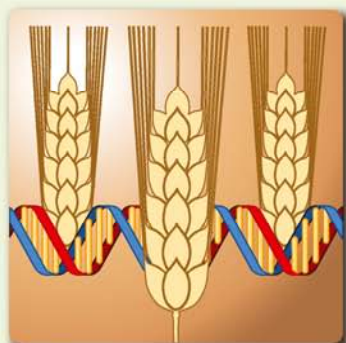
# Plant Breeding & Genetics Newsletter

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*Coffea* spp. (Photo ©FAO/Mario Marzot)

To learn more about naturally decaffeinated coffee, see News on page 25

## To Our Readers

Collegial greetings from Vienna!

This is the time of the year to look at things accomplished and to face challenges that lie ahead of us.

The Plant Breeding and Genetics Section (PBGS) in IAEA Headquarters, Vienna and the Plant Breeding and Genetics Laboratory (PBGL) in Seibersdorf are very grateful for the input and support of experts, consultants and lecturers from all the Member States that helped us implement our programmatic activities. We had the honour and the privilege to host 46 trainees, fellows, interns and scientific visitors for a total of 51 training months. Every single one has enriched this programme and we fondly remember our fruitful discussions. The Plant Breeding and Genetics Subprogramme provides technical support to Member States through the development and the promotion of technology packages based on mutation induction and efficiency enhancing molecular and biotechnologies applied to crop improvement through adaptive R&D in our Laboratory and four CRPs. This year, we worked on transferring these technology packages to 96 Member States by providing technical and scientific support through 63 national, regional and interregional Technical Cooperation Projects (TCPs) that are managed by IAEA's Department of Technical Cooperation. This transfer of technologies assists Member States in the implementation of national crop improvement programmes with specific breeding objectives or regional programmes addressing abiotic and/or biotic stresses, which represent serious threats impeding crop productivity in wide areas mostly in the developing world



**IAEA**  
International Atomic Energy Agency

under the growing driver of climate change and variability. Through the TCPs, we also provided policy advice, e.g. drafting of individual country programme framework (CPF), and technology support in the implementation of breeding programmes based on priorities expressed by Member States.

For example, banana and cassava are staple crops for up to 1 human out of 6 in developing countries particularly in tropical and subtropical regions. As these crops are vegetatively propagated, they face challenges not found in seed propagated crops. Improvement of vegetatively propagated crops is deemed especially pressing in the context of global population growth and climatic variations that continue to threaten food security and prices in the decades to come. Consequently, a CRP was started in 2004 on 'Molecular Tools for Quality Improvement in Vegetatively Propagated Crops Including Banana and Cassava' with the understanding that there were as yet uncharted opportunities for technology development, breeding and the use of mutations to advance progress in the crops, and to develop guidelines and strategies relevant to all vegetatively propagated species. As highlighted in this newsletter, the objectives of this CRP were largely met.

Another example comes from maize. Here, mutation induction applications in crop improvement are very efficient and sometimes appear in fields where they are least expected. Thus, application of mutation induction (and documented huge success) in hybrid maize breeding may surprise most readers since it is widely believed that outcrossing crops like maize have sufficient genetic variability. Therefore, mutation induction should play a limited role. Such a perception should be re-assessed against the great success of maize mutation breeding in Bulgaria as revealed through a regional European TC project scientifically and technically backstopped by PBGS and PBGL: about 80 mutant varieties of 14 different plant species include leading mutant maize and wheat varieties that are covering about 50% of maize growing area and almost 100% of durum wheat area, respectively. Novel mutations have not only played a role in improving resistance/tolerance to biotic/abiotic stresses, quality and nutrition traits, but also in facilitating hybrid seed production and enabling adaption to mechanization of crop production. Thousands of mutant lines are generated and preserved as germplasm collections and are being used in breeding programmes. In Eastern Europe, through the regional European TC project, over 200 well characterised advanced mutant lines of barley, wheat and maize have been developed for breeding and genetics studies with improved yield components and tolerance to drought. Several advanced mutants of tomato, pepper, potato and soybean with improved carotene, lycopene and tocopherol content have been characterised for breeding and genetics studies. A TILLING platform for barley has

been recently established at the Department of Genetics, University of Silesia in Katowice (Poland).

High demand for facilitation in training and network building also drives the activities of PBGS and PBGL.

Indeed, mutation induction is living a renaissance: introduction of new courses on Induced Mutagenesis in Plant Genetics at the University of Tirana, Albania. Over the years, scientists in South East Asian countries have been engaged in mutation breeding programmes. Since the wealth of these experiences was clustered in isolated institutions, we sponsored and supported the creation of a forum for exchange of scientific knowledge through the establishment of an Asia and Oceania Association of Plant Mutagenesis (AOAPM). The new association will be launched in the first quarter of 2011. Our activities assisted the government of Ecuador in organising a national training course on mutation induction. 70 scientists attended the lectures and experimental demonstrations and several Ecuadorian newspapers covered the event, which can really be considered a successful cooperation on the application of nuclear techniques to crop improvement for ensuring food security in the country: INIAP (Instituto Nacional Autónomo de Investigaciones Agropecuarias), which has a strong history in agriculture related research previously was not much involved in the use of nuclear techniques for crop improvement. The establishment of a TC project on Inducing Mutations in Agriculture with the Aid of Radiation has brought the technology under a new light. Scientists from INIAP, and other research centres are now increasingly using our technology packages developed through adaptive R&D at PBG.

In this issue, you will find more about services in support of activities in Member States, training and human capacity development and technology development/adaptation in the PBGL, including work on impact of NaCl stress on the biomass and mineral nutrient assimilation in different rice varieties, comparative studies of different mutagens on seed propagated crops, protocols and guidelines for mutation breeding lower cost molecular assays, and targeting deleterious mutations (including positive control kits).

We continue to coordinate closely with our FAO sister divisions and departments, as well as with the field offices of FAO to achieve the strategic objectives of our Member States. Application forms for the formulation of IAEA and FAO projects can be obtained respectively at <http://www-tc.iaea.org/tcweb/tcprogramme/default.asp> and [http://www.fao.org/tc/tcp/index\\_en.asp](http://www.fao.org/tc/tcp/index_en.asp). Highlights of our work in the TCPs and CRPs you will find inside this newsletter.

An important opportunity in terms of technology advancement is offered by the genetic improvement of crops that can adapt to the future climate conditions; i.e.

‘climate proofing’ crops. The main effects of climate change on agriculture will most probably be experienced through temperature fluctuations (increase in minima and maxima), altered changes in rainfall patterns (in amount, spatial and temporal distributions), elevated rates of evaporation, increased intensity and frequency of extreme events (floods and droughts), and raise of sea level affecting coastal areas where large quota of cultivated land are located (intrusion of salty water). Nuclear techniques for mutation induction can increase the adaptability of crops to climate change and variability, allowing for the exploitation of genetic diversity generated in mutated populations by assessing tolerance to stresses associated with climate change (i.e. increased temperature, more severe drought spells, salinity increase, resurgence or extension of diseases) in terms of yield and yield components. In addition, the application of existing tools to characterize physiological and biochemical responses to these stresses by application of stable isotope techniques will complete the tool box of the breeder to produce hardy crops in harsh environments. Consequently, we will strengthen

on-going efforts to integrate our activities with those of the Soil & Water Management and Crop Nutrition Section and Laboratory to foster the development of technology packages integrating best fit soil and water management practices with mutation induction and efficiency enhancing molecular and biotechnologies. The responses that crop production systems world-wide can put in place to cope with the expected impact of climate change and to reduce food insecurity range from institutional and policy levels to the best management practices and technology advancement.

Finally, I take this opportunity to wish you, your families and associates peace, health and prosperity for 2011, and success in all your endeavours.

*Pierre J.L. Lagoda*  
**Head,**  
*Plant Breeding and Genetics Section*



# Staff

**Joint FAO/IAEA Programme of Nuclear Techniques in Food and Agriculture, Wagramer Strasse 5, P.O. Box 100, A-1400 Vienna, Austria (Phone: +431 2600 + extension; Fax: +431 2600 7; Email: [Official.Mail@iaea.org](mailto:Official.Mail@iaea.org))**

Name	Title	Email	Extension
Qu Liang	Director	<a href="mailto:q.liang@iaea.org">q.liang@iaea.org</a>	21610

## Plant Breeding and Genetics Section

Name	Title	Email	Extension
Pierre J.L. Lagoda	Head of Section	<a href="mailto:p.lagoda@iaea.org">p.lagoda@iaea.org</a>	21626
Vacant	Plant Breeder/Geneticist		
Marie Madeleine Spencer	Plant Biotechnologist	<a href="mailto:m.m.spencer@iaea.org">m.m.spencer@iaea.org</a>	21623
Yvonne Rosaline Lokko	Plant Breeder/Geneticist	<a href="mailto:y.lokko@iaea.org">y.lokko@iaea.org</a>	21619
Sridhar Dharmapuri <sup>1</sup>	Consultant	<a href="mailto:s.dharmapuri@iaea.org">s.dharmapuri@iaea.org</a>	26843
Akbar Ali Cheema <sup>2</sup>	Consultant	<a href="mailto:a.cheema@iaea.org">a.cheema@iaea.org</a>	21617
Miriam Gaceri Kinyua <sup>3</sup>	Consultant	<a href="mailto:m.kinyua@iaea.org">m.kinyua@iaea.org</a>	21617
Katayoun Allaf	Senior Office Clerk	<a href="mailto:k.allaf@iaea.org">k.allaf@iaea.org</a>	21621
Mario Dani	Secretary	<a href="mailto:m.dani@iaea.org">m.dani@iaea.org</a>	21620

**FAO/IAEA Agriculture and Biotechnology Laboratory, Plant Breeding and Genetics Laboratory, A-2444 Seibersdorf, Austria (Phone: +431 2600 + extension)**

Name	Title	Email	Extension
Qu Liang	Acting Head, Agriculture and Biotechnology Laboratory	<a href="mailto:q.liang@iaea.org">q.liang@iaea.org</a>	21610/28274
Vacant	Head, Plant Breeding Unit		
Vacant	Mutation Breeder		
Bradley Till	Plant Molecular Biologist/Acting Lab Head	<a href="mailto:b.till@iaea.org">b.till@iaea.org</a>	28260
Mirta Matijevic	Technician	<a href="mailto:m.matijevic@iaea.org">m.matijevic@iaea.org</a>	28317
Joanna Beata Jankowicz-Cieslak	Technician	<a href="mailto:j.jankowicz@iaea.org">j.jankowicz@iaea.org</a>	28275
Guenther Berthold	Technician	<a href="mailto:g.berthold@iaea.org">g.berthold@iaea.org</a>	28418
Andreas Draganitsch	Technician	<a href="mailto:a.draganitsch@iaea.org">a.draganitsch@iaea.org</a>	28418
Souleymane Bado	Technician	<a href="mailto:s.bado@iaea.org">s.bado@iaea.org</a>	28208
Owen Anthony Huynh	Technician	<a href="mailto:o.huynh@iaea.org">o.huynh@iaea.org</a>	28279
Anne Lorenz	Secretary	<a href="mailto:a.lorenz@iaea.org">a.lorenz@iaea.org</a>	28274
Elisabeth Swoboda	Secretary	<a href="mailto:e.swoboda@iaea.org">e.swoboda@iaea.org</a>	28362

<sup>1</sup> Joined the IAEA in July 2010

<sup>2</sup> Joined the IAEA in August 2010

<sup>3</sup> Separated from the IAEA in September 2010

# Forthcoming Events

## **Second Research Coordination Meeting (RCM) on Improving Nutritional Quality by Altering Concentrations of Enhancing Factors Using Induced Mutation and Biotechnology in Crops, D2.30.28, Pretoria, South Africa, 11–15 April 2011**

Technical Officer: Y. Lokko

In addition to providing the major calories in human diets required to sustain life, plants are also the major source of health-beneficial agents, such as vitamins and minerals required for vital physiological processes. The major staple crops however, do not provide the essential vitamins and minerals in adequate quantities and quality. A sustainable way to ensure that adequate amounts of vitamins and minerals can be obtained from diets is to develop crops that provide the required amounts in their edible tissues. Induced mutations are proven tools in creating desirable genetic variability in plants that translate to enhanced accumulation of essential minerals; synthesis of precursors of vitamins; modified quantities and qualities of starch, proteins and oils as well as secondary plant metabolites that play critical roles in improving human health and nutrition.

This Coordinated Research Project (CRP) aims at utilizing the wealth of mutant germplasm in model crops, such as rice, tomato and barley to understand and identify genes involved in the biosynthesis of nutritional quality enhancing factors and to develop efficient screening methods to facilitate the genetic improvement of nutritional quality. The goal is to transfer knowledge and technologies of beneficial mutants associated with nutritional factors from model crops to improve nutritional quality into other crops.

Eleven Scientists from Botswana, Bulgaria, China, Denmark, Ghana, Germany, India, Kenya, South Africa, Ukraine, the United Kingdom and the United States of America working together on this CRP will be meeting in Pretoria, for the second Research Coordination Meeting (RCM).

## **IAEA/RAS Regional Coordination Meeting on Improvement of Crop Quality and Stress Tolerance for Sustainable Crop Production Using Mutation Techniques and Biotechnology, RAS/5/045, Bangkok, Thailand, 7–11 March 2011**

Technical Officer: Y. Lokko

Under the framework of the RCA project, the meeting will be held in collaboration with Chai Nat Field Crops Research Center of the Ministry of Agriculture and Cooperatives, Thailand.

The objectives of the meeting are:

- To assess the current status and progress achieved under the project since the last meeting;
- To discuss modalities for continuation in the next TC cycle and sustainable continuation of work in participating countries after the duration of IAEA assistance;
- To review recommendations for participation in Asia and Oceania Association of Plant Mutagenesis.

The Meeting is expected to have participation from plant geneticists and breeders from the IAEA Member States in the region, who are part of the Regional Technical Cooperation (TC) Project RAS/5/045. They should be engaged in research using induced mutation and molecular techniques in breeding and genetics of crops aimed at addressing the problems associated with the improvement of quality and stress tolerance in selected crops.

## **First Research Coordination Meeting on Climate Proofing of Food Crops: Genetic Improvement for Adaptation to High Temperatures in Drought Prone Areas and Beyond, D2.30.29, Vienna, Austria, 2–6 May 2011**

Technical Officer: M. Spencer

This CRP was initiated in 2010. The first RCM is planned to be held in Vienna, Austria, 2-6 May 2011. Approximately 11 contract holders (China (2), Colombia (2), Cuba, Mexico, Pakistan, the Philippines, Senegal, United Republic of Tanzania, Zimbabwe) and seven agreement holders (Australia, China, International Centre for Tropical Agriculture (CIAT), Japan, Spain, the United Kingdom (UK) and the United States of America (USA)) will meet in Vienna to discuss the effect of climate change and variability on the sustainability of crop production system. For detailed information, see Coordinated Research Projects (CRPs) and Research Coordination Meeting (RCMs).

## **IAEA/AFRA Workshop on Seeds and Planting Material Exchange in Africa and the World, RAF/5/056, Buea, Cameroon, 28 March–1 April 2011**

Technical Officer: M. Spencer

Course Director: X. Ndzana

This workshop will be organized under the regional TC project RAF/5/056 on Increasing Production of Nutritious Food through Mutation Breeding and Biotechnology (AFRA III-3).

The need for such a workshop was expressed by all participants during the last coordination meeting. In fact, one of the reasons mostly cited as a cause for the poor yields in African agriculture is the poor quality of the seeds and planting material and also the enormous difficulties encountered by breeders and farmers to assess the quality of their seeds, acquire or export good quality seeds and planting material. In recent years African leaders and officials in the most agricultural systems from the main regions, Northern, Western, Eastern and Southern Africa, have been working on the harmonization of the seed legislations and the agricultural trading regulations between neighbouring countries. All breeders and scientist involved in crop improvement, production and commercialization should be aware of the legislation, regulations, standards and policies governing the quality, sanitary status and legal rights (Intellectual Property Rights and Farmers' Rights) in force in their country and in the region.

The purposes of this workshop are to provide participants with (1) the opportunity to learn about the legislation, regulations, standards and policies governing the quality, sanitary status and legal rights (Intellectual Property Rights and Farmers' Rights) governing seeds production and exchanges; (2) to get a better understanding of their own rights as breeders and/or scientists over the innovations developed; (3) to get acquainted with the Standard Material Transfer Agreement (SMTA) for transfer of plant genetic resources for food and agriculture, including mutated lines and/or varieties and to provide information on the legal and political underpinnings for the relevant global convention and treaty.

The workshop will include lectures, roundtable discussion and consultations on the methodology and their application in various field situations.

The outputs to be expected are as follows:

- Increased understanding of participatory plant breeding and their applications under various situations.
- Improved participatory plant breeding workplan for each participating institute.

The scientists should be from participating Members States actively working in crop breeding including mutation breeding and hold at least a M.Sc. degree in plant breeding/genetics, plant biotechnology or related subjects.

As the workshop will be conducted in English only, participants should be proficient enough to have no difficulties in following lectures, reading and understanding written texts provided during the course. They should also be able to actively participate in the discussions.

## **Second Research Coordination Meeting (RCM) on Isolation and Characterization of Genes Involved in Mutagenesis of Crop Plants, D2.40.13, Vienna, Austria, 30 May–3 June 2011**

Technical Officer: P.J.L. Lagoda

Understanding the biological control of the process of DNA damage, repair, and mutagenesis, is not only scientifically important, but also vital for manipulation of mutation induction using recently emerging molecular tools. Knowledge of molecular genetic features of induced mutation is necessary for the selection of proper mutagens suitable for specific purposes in mutation induction.

The second RCM will provide a platform for the CRP participants to expose and discuss their results and assess progress in the CRP's endeavours: significant progress has recently been made in studies on the mechanisms underpinning the response of plant genomes to physical and chemical mutagens in the model plant *Arabidopsis*. Various genes have been identified to be involved in the biological process of response to external and internal DNA damages, although studies on genes responsive to physical mutagens are generally limited. Although induced mutations has been widely used in crop breeding and basic research, the fundamental processes that lead to mutations and the molecular nature of mutations induced by physical mutagens remain largely unknown in plants. The rapid progress in functional genomics has provided unprecedented opportunities to study the mechanisms underpinning the response of plant genomes to physical and chemical mutagens. Recent advances in genomics have created an opportunity to apply our current knowledge of DNA repair and mutagenesis in model systems to crop plants. Furthermore, emerging reverse genetics tools enable direct analysis of consequences of various mutagenic treatments at molecular genetic levels.

## **IAEA/AFRA Training Course on Advanced Molecular Marker Technologies and Data Analysis Applied to Crop Improvement, including Mutation Breeding, RAF/5/056, Pretoria, South Africa, tentatively planned for 27 June–1 July 2011**

Technical Officer: M. Spencer

Course Director: W.J. van Rensburg

This training course will be organized under the Regional TC Project RAF/5/056 on Increasing Production of Nutritious Food through Mutation Breeding and Biotechnology (AFRA III-3).

Following the steady and thorough implementation of the regional TC project, the training course is scheduled in order to enhance the results already obtained through mutation breeding in the specific projects of all participating countries. Classical breeding approaches associated with agronomy and biotechnologies has allowed less than 2% of the population of industrialized countries to produce ample food to satisfy their national needs. The tremendous advances in cell, molecular, nuclear and even nanotechnology afford scientists with powerful tools for exploring the living kingdom to the benefit of human kind. When considering the transfer of the enormous volume of genetic information available and the major technological developments together with their potential in crop improvement for developing countries, there is a great need of clear and wise assessment of their usefulness and applicability. Various approaches such as DNA sequencing, genomics, molecular markers, high throughput reverse genetics, including TILLING and other deep sequencing technologies, are totally applicable to breeding programmes in developing countries. Under national breeding programmes, where most of the basic classical breeding techniques and basic biotechnology are well-mastered, a range of techniques of molecular biology, developed since the late 1980s may be considered for application. The choice of molecular techniques requires a thorough consideration of:

- The relative costs (e.g. financial, social and political) of molecular techniques vs. the relative benefits (e.g. productivity and food security);
- The appropriateness of the various molecular techniques regarding their environmental impact, their impact on human health, the status with respect to intellectual property rights, the status with respect to bio-safety regulations and controls, the existing level of access to these technologies, the level of ca-

capacity-building or resources required to use them, their impact on food production and food security and the desire to definitely bridge the scientific gap.

The purposes of this training course are to provide participants with (1) the opportunity to enhance their scientific and practical knowledge on the most advanced molecular technologies available for breeding purposes (2) to get a better understanding of the pre-requisites for the application of these technologies in crop breeding (types of populations and screening protocols to be used) and (3) to get acquainted with the informatics tools for data analysis.

The training course will include lectures, hands-on experiments based on controlled populations of cowpea and *Phaseolus* (fields phenotyping, laboratory DNA experiments and computer based data analysis), roundtable discussion, consultations on the methodologies and their application in their own mutation breeding programmes.

The outputs to be expected are:

- Increased understanding of the use of advanced molecular marker technologies in crop breeding;
- Enhanced skills in the use of computer and web-based software for analysing field and molecular data collected.

The scientists should be from all participating Members States actively working in crop breeding including mutation breeding and hold at least a M.Sc. degree in plant breeding/genetics, plant biotechnology or related subjects.

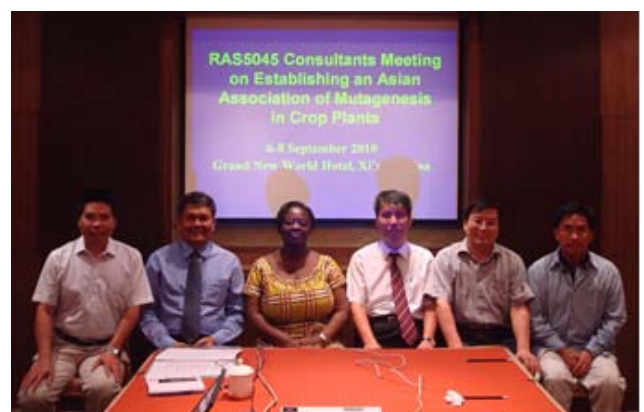
As the workshop will be conducted in English only, participants should be proficient enough to have no difficulties in following lectures; reading and understanding written texts provided during the course. They should also be able to actively participate in the discussions.

## Past Events

### Consultants Meeting on Establishing an Asian Association of Mutagenesis in Crop Plants (AAMCP), Xi'an, China, 6–8 September 2010

Technical Officer: Y. Lokko

The application of mutation techniques in plant improvement has resulted in the development of new germplasm and varieties of a number of plant species including food and non food crops and ornamentals with improved traits such as high yields, tolerance to drought and salinity, resistance to diseases and improved quality for consumers.





Over the past 50 years, scientists in Asia have actively used mutation techniques to improve the living standards and income of farmers and end-users. To date, more than half of the released mutant varieties in the world (over 3000, <http://mvgs.iaea.org>) are from Asia. In addition, thousands of desirable mutant germplasm of different crop species have been obtained and made available for breeding programmes and for functional genomics studies.

As of 2007, there was no regional professional association on plant mutagenesis. As such, the establishment of an Asian Association on Mutagenesis of Crop Plants (AAMCP) was discussed during the Project Planning Meeting of the IAEA/RCA TC project on Improvement of Crop Quality and Stress Tolerance for Sustainable Crop Production Using Mutation Techniques and Biotechnology (RAS/5/045), from 25 to 29 June 2007, in Kuala Lumpur, Malaysia. In collaboration with the Chinese Academy of Agricultural Sciences (CAAS), a consultants meeting was held in Xi'an, China, from 6 to 8 September, to formulate such an association, which recommended the name to be Asia and Oceania Association of Plant Mutagenesis (AOAPM).

Following extensive discussions and brainstorming, participants drafted the aims of the association, the name, its relevance in relation to existing national/regional/international associations, membership type and their responsibilities, the main activities of the association and modalities to ensure sustainability. A number of well-known experts in the region were recommended to serve as its founding board and to finalise the association's draft bylaws. It is expected that the association will be launched in the first quarter of 2011.

List of Founding Members, and Oceania Association of Plant Mutagenesis (AOAPM):

**Australia:** Chengdao Li, Dept of Agri. & Food Western Australia; Slade Lee, Centre for Tropical Crops and Bio-commodities, Queensland University of Technology

**China:** Luxiang Liu, Chinese Academy of Agricultural Sciences (CAAS); Qingyao Shu, Zhejiang University, Hangzhou china.

**India:** Prasanna Suprasanna, Nuclear Agriculture & Biotechnology Division, Bhabha Atomic Research Centre.

**Japan:** Hitoshi Nakagawa, National Agriculture and Food Research Organization (NARO); Atsushi Tanaka, Radiation-Applied Biology Division JAEA.

**Korea, Republic of:** Si-Yong Kang, Korea Atomic Energy Research, Jeongseup Republic of Korea.

**Malaysia:** Ibrahim Rusli, Malaysian Nuclear Agency, Kajang Malaysia.

**Vietnam:** Le Quang Luan, Center for Nuclear Techniques, VAEC.

**Yemen:** Abdulwahid Saif, National Atomic Energy Commission.

**IAEA:** Yvonne Lokko, Joint FAO/IAEA Division of Nuclear Techniques in Food & Agriculture.

## **Regional Training Course on the Use of Induced Mutations (TILLING) and DNA Markers in Cereal Genetics and Breeding, RER/5/013, Katowice, Poland, 13–24 September 2010**

Technical Officer: Y. Lokko

During the period of 13 to 24 September 2010, the Department of Genetics, University of Silesia in Katowice, Poland was the host of the Regional Training Course on Use of Induced Mutations (TILLING) and DNA Markers in Cereal Genetics and Breeding. The training course was organized under the IAEA regional TC project on Evaluation of Natural and Mutant Genetic Diversity in Cereals Using Nuclear and Molecular Techniques (RER/5/013). The purpose of the training was to provide participants with theoretical and practical aspects of the use of mutant populations and the applications of efficient techniques in screening for and utilization of useful mutants in breeding and genetics of cereals.



*Preparation of polyacrylamide gels for Li-Cor sequencers*

All together 13 scientists from Albania, Bulgaria, Croatia, Georgia, Kazakhstan, Serbia, the former Yugoslav Republic of Macedonia, Turkey, Uzbekistan and Poland participated in the training course. Two internationally recognized lecturers were invited to conduct the lectures and part of laboratory practicals. In the first week of the training, Dr. Brad Till from Plant Breeding and Genetics Laboratory, Joint FAO/IAEA Division, an expert in



TILLING method, shared his extensive knowledge on designing TILLING populations in various crops, dealing with TILLING optimisation and troubleshooting and on the applications of this technology in molecular biology and breeding. In the second week, Dr. Roberto Tuberosa from the Department of Agroenvironmental Science and Technology, University of Bologna, gave profound lectures on SNP markers development and their applications, QTL mapping and cloning, marker assisted selection, association mapping principles and application of molecular markers for linkage disequilibrium studies. Invited lecturers were accompanied with the local staff of the Department of Genetics, who introduced the participants to the methods of plant DNA extraction, PCR reactions and gel electrophoresis, the analysis of AFLP and SSR markers, DNA sequencing and the methodology of mutation detection by TILLING strategy. The wet lab exercises were complemented by the practicals of data analysis aimed on molecular maps construction and basics of bioinformatics analysis of DNA and protein sequences.

During the two-week stay in Poland, the participants had a chance to not only take part in scientific activities, but also to become familiar with some historical and cultural sites. Participants visited Krakow, the former capital city of Poland and Ogdzieniec, the little village with one of the best preserved ruins of a medieval castle in Cracow-Czestochowa Jurassic highland chain. Small group of participants chose to see the Nazis Concentration Camp in Auschwitz and learn more about this tragic though important part of European history.



The overall organization of the training course was evaluated as very high, due to the special emphasis on meeting the stated course objectives, good technical content of the course and quality of practicals, as well as the guidance by training personnel. Most participants declared that they will apply the knowledge they gained immediately, or not later than within one year after returning to their home institutions. The training course was for all a great opportunity to exchange ideas and experiences of their scientific work and expand their contacts in the scientific community for future collaborations.

## **Training Course on Biotechnology – Modern Tools and Methods for Breeding and Plant Genetic Resources, FAO-KAZ/3202, Astana, Kazakhstan, 20–24 September 2010**

Technical Officer: B. Till

The training course was organized under the auspices of the FAO-supported TC Project, KAZ/3202 to strengthen plant biotechnology capacity for sustainable utilization of plant genetic resources for food and agriculture. Twelve participants from across the country were invited and joined by the local team for seven days of lectures and practical exercises and computer laboratory work. Efforts were structured around the following tasks:

- Good laboratory practices, safety and security and standard molecular laboratory methodologies;
- DNA extraction, quantification and quality checks; Polymerase Chain Reaction.

There were demonstrations on the management of genetic and molecular data including the use of specialised statistical software for the analysis of molecular genetic data.



## **National Training Course on cDNA-Representational Difference Analysis (cDNA-RDA) as a Tool to Identify Genes Expressed in Mutated Plant Material, SAF/5/012, Pretoria, South Africa, 27 September–8 October 2010**

Technical Officer: Y. Lokko

Course Director: L. van Emmenes

This training course was organized by the Vegetable and Ornamental Plant Institute, Agricultural Research Council (ARC) in cooperation with the International Atomic Energy Agency (IAEA). The purpose of the training course was to provide knowledge and practical training in the use of cDNA-Representational Difference Analysis

(cDNA-RDA) as a molecular technique for screening and analyses of drought related genes in crops to facilitate crop breeding programmes aimed at developing new crop varieties with improved drought tolerance. The course was presented by Dr. Miroslaw Kwasnieski from the Department of Genetics, University of Silesia, Katowice, Poland. Nine local participants including researchers from the ARC-VOPI, the ARC-Institute for Tropical and Subtropical Crops (Nelspruit), the ARC-Infruitec Nietvoorbij (Cape Town) and the University of Stellenbosch attended the course. Dr. Kwasnieski was assisted by Dr. Christell van der Vyver from the University of Stellenbosch and Dr. Juan Vorster from the University of Pretoria. The course started on a high note with RNA extractions from mutated cowpea (*Vigna unguiculata*) and amaranth (*Amaranthus tricolor*) lines that had been subjected to drought stress for two weeks. High concentrations of RNA were obtained, which set the baseline for the following procedures and ensured high-spirited participation. Thereafter, the first amplicons were generated. It was a great relief when different products were obtained after the first round of hybridization! One additional round of hybridization was performed for amaranth and two extra rounds of hybridization were performed for cowpea. A further purification step for amaranth resulted in a long night, when the crew worked a late shift until 23:00 h! But once all the hybridizations were complete, two different products for Amaranth and eight difference products for cowpea were isolated and cloned. These products will be sequenced in order to determine their relevance with regard to tolerance to drought stress.



The course participants at the ARC-VOPI campus



Drs. Kwasnieski and Vorster in discussion with Ms. Gazendam with regard to PCR results

### Third IAEA/RER Coordination Meeting on Evaluation and Utilization of Natural and Mutant Cereals Germplasm, RER/5/013, Ohrid, the Former Yugoslav Republic of Macedonia, 19–21 October 2010

Technical Officer: Y. Lokko

This project was initiated in 2007 stemming from the fact that cereals make up the most important component of the daily human diet and have therefore been a subject of extensive studies aimed at improving their agronomical characteristics. It is well-known that diverse gene pools are the foundation of effective crop improvement programmes. In the 1970s experimental mutagenesis methods were widely explored in the countries of South Eastern Europe and with the IAEA's support, a wide diversity of mutant lines were developed. These mutant lines have been used extensively by genetic research institutes and breeding centres in combination with local and foreign accessions of crops and their wild relatives to create a broad diversity of lines. As a result of their incorporation in breeding programmes, a number of new common wheat, durum wheat and barley cultivars and maize hybrids have been released and registered. Evaluation of genetic variability within and among established populations is one of the cornerstones of modern plant breeding programmes directed towards more rational use of available genetic resources. A proper understanding of the fine structure of genetic diversity in cereals is a necessary prerequisite for revealing their genetic potential and is also essential for the implementation of new breeding strategies. Fluctuations and global climate changes, pathogen mutability and their influence on agricultural production sustainability present a new challenge to breeders. With many countries in South Eastern Europe, plagued by increased biotic and abiotic stresses, it was necessary to initiate strategies to improve the tolerance of cereal cultivars so as to achieve a further increase in their yield potential and quality. Information on the extent of biodiversity and biodiversity trends over time needs to be consolidated to permit an adequate response to the dynamics of climate and pathogen populations and to effectively utilize the potential of commercial and wild resources. It was envisaged that this will also permit the identification of accessions in germplasm collections with variant genomic regions or alleles of candidate genes having a favourable impact on the priority traits, making it possible to select superior genotypes having hitherto non-identified allele variants to broaden genetic diversity.

This meeting was held in collaboration with the Government of the Former Yugoslav Republic of Macedonia through the Faculty of Agricultural Sciences and Food, Ss. Cyril and Methodius University. The main purposes of the meeting were:



- To review the progress made in the project since the last meeting;
- To review workplan and needs for successful completion of project;
- To identify and discuss measures to ensure sustainable continuation of work after the completion of the project, including exchange of material and information.



A total of ten countries in the region from Albania, Bulgaria, Croatia, the Former Yugoslav Republic of Macedonia (2), Moldova, Poland, Serbia, Turkey and Ms. Yvonne Lokko (Technical Officer, from the Joint FAO/IAEA Division) attended the meeting.

Mr. Milan Drakalski, Head of the International Cooperation in the Ministry of Science and Education and Ms. Ivanka Solomonova of the Ss. Cyril and Methodius University formally opened the meetings and welcomed participants to the Former Yugoslav Republic of Macedonia.

Following participants' presentations on the status of work done in their respective countries since the last meeting, particularly in developing new mutant germplasm of the target cereals - wheat, maize and barley - the group evaluated the achievements from each country in contributing to the expected outputs of the project to date.

So far, significant progress has been made in most of the actively participating countries, following their involvement in the group activities and diligence in the work done in their respective institutions. The main improved characteristics are drought tolerance, improved yield and resistance to diseases. In Romania and Turkey some are already being utilised in breeding programmes. In Poland, a TILLING population of 9 600 individuals and their DNA is available. The counterpart also has 39 000 phenotypically fully characterized families available at the university.

Following participation in the project, most of the counterparts have enhanced their local and international collaborations. This includes collaboration between the counterparts from the Former Yugoslav Republic of

Macedonia and Romania to capitalise on their individual institutional capacities to enhance wheat genetic improvement in their respective countries. In addition, a total of 43 peer reviewed publications have been obtained from this project, from 2008 to date. Through participating in and providing services at training courses, the counterparts have also disseminated the skills and knowledge within their countries through regular undergraduate and postgraduate courses, workshops and training courses. From the 2009 academic year, Induced Mutagenesis in Plant Genetics has been introduced in the BSc and MSc curriculum in the University of Tirana, Albania.

We, in the Plant Breeding and Genetic Section, would like to say kudos to all our counterparts of the cereals component of the TC project RER/5/013.

### **Fourth and Final Research Coordination Meeting on Molecular Tools for Quality Improvement in Vegetatively Propagated Crops including Banana and Cassava, D2.30.27, Brasília, Brazil, 8–12 November 2010**

Technical Officer: B. Till



Fifteen scientists from thirteen countries (Bangladesh, Brazil, China, Cuba, Czech Republic, Ghana, India, Indonesia, Kenya, Mexico, Nigeria, Philippines and the United Kingdom) as well as the Technical Officer, and Dr. Chikelu Mba of FAO participated in the meeting. The meeting was organized by Dr. Luiz Carbalho of Empresa Brasileira de Pesquisa Agropecuária (EMBRAPA), with assistance by Dr. Robert Miller of the Universidade Católica de Brasília. The meeting was hosted by EMBRAPA.

Together, banana and cassava are staple crops for over one billion people in developing countries particularly in tropical and subtropical regions. Largely vegetatively propagated, these crops face challenges not found in seed propagated crops. The CRP was started in 2004 with the understanding that there were exciting opportunities for technology development, breeding and the use of muta-



tions to advance progress in the crops, and to develop guidelines and strategies relevant to all vegetatively propagated species. At the onset of the CRP, specific aims included:

- Stimulate the development of mutant populations for analyzing key physiological pathways for quality, post-harvest duration, abiotic and biotic stresses.
- Generate doubled-haploids for genetic mapping, heterosis application and breeding.
- Develop and adapt genomic resources and tools such as Conserved Orthologous Sequences, EST libraries and expression analysis datasets.
- Develop a knowledge base, core of trained staff and foster collaboration between advanced labs and national programs.
- Produce and publish results, including guidelines for the use of nuclear techniques and molecular tools for improvement of vegetatively propagated crops.

The objectives of the CRP were largely met. Participants at the meeting presented their results covering the entire five year span of the CRP with specific emphasis on new results obtained since the 2008 meeting held in Vienna. Presentations emphasized achievements, publications, weaknesses or pitfalls of approaches and also a look towards the future and continued work beyond the scope of the CRP. The group welcomed the presentation by Dr. Chikelu Mba on on-going efforts at the FAO for strategies for national plant breeding capacity building. A great deal of progress has been made in banana and cassava, over the five year duration of the CRP, with a clear roadmap emerging for future endeavours for improvement of these two very important food security crops. While detailed results and achievements are too numerous to list here, some specific achievements include:

- Publication of over 25 scientific manuscripts from work related to the CRP.
- Development of strategies for mutation induction including tissue culture and cell-suspension strategies.
- Development of mutant populations in cassava and banana and the discovery of novel phenotypic traits including enhanced quality, lengthened storage stability, and disease resistance.
- Large collections of genomic resources including novel molecular markers, digital differential display (DDD) resources, and allelic diversity studies at the candidate gene and whole genome level.
- Collection and characterization of natural and mutant genetic resources.

All participants recognized the importance of dissemination of their research to the broader research community

to assist Member States in their work for the improvement of vegetatively propagated crops. This was deemed especially pressing in the context of global population growth and climatic variations that continue to threaten food security and prices in the decades to come. As such, major efforts at the meeting were given to develop a strategy for publication that maximizes impact of the results. While the CRP is coming to a close, the importance of the work continues and the strong collaborative networks developed through this CRP between scientists in developing and developed Member States will no doubt continue to serve the laudable goals of sustainable food security for vegetatively propagated crops.

### **IAEA/RER Coordination Meeting on Evaluation of Natural and Mutant Resources for Increased Phytonutrient Levels in Solanaceous Food Crops, RER/5/013, Vienna, Austria, 17–19 November 2010**

Technical Officer: Y. Lokko

The improvement of quality traits in food crops is one of the most important goals in plant breeding and is gaining more and more attention for nutritional, health and marketing purposes. In 2006, the IAEA approved the regional technical cooperation project RER/5/013 on Evaluation of Natural and Mutant Genetic Diversity in Cereals Using Nuclear and Molecular Techniques, which aims at evaluating and increasing genetic diversity in major cereals using nuclear techniques, molecular genetics and biotechnological tools. For the 2009-2011 TC cycles, a second project on the evaluation of natural and mutant resources for increased phytonutrient levels in solanaceous food crops was incorporated into this project. During the project design stage between staff of the IAEA and counterparts, it was decided that this project be included in the existing TC project RER/5/013 as a new component with separate funding and participating counterparts for its activities.

The second coordination meeting of the project was held at IAEA headquarters in Vienna, Austria, from 17 to 19 November 2010. The purposes of the meeting were:

- To review the progress made in the project since the last meeting;
- To review work plan and needs for successful completion of project;
- To identify and discuss measures to ensure sustainable continuation of work after the completion of the project, including exchange of material and information.

Project counterparts from Albania, Bulgaria, Hungary, the Former Yugoslav Republic of Macedonia, Moldova, Turkey, Serbia and Ukraine attended the meeting.

Participants gave technical presentations on the use of mutant genetic resources in breeding and the genetics of tomato; methods in improving levels of and screening for anti-oxidants in breeding; the importance of standardisation of anti-oxidant levels in food crops; and country reports on the status of breeding for improved levels of anti-oxidants in target solanaceous food crops including the use of mutants. Each presentation was discussed and the key points were incorporated in the later discussions on the main activities to be carried out with the project. At the end of the meeting key group activities identified as important for the successful implementation of the project were agreed upon.

### **Third Coordination Meeting on Responding to the Transboundary Threat to Wheat Black Stem Rust (Ug99), INT/5/150, Ankara, Turkey, 6–10 December 2010**

Technical Officer: P.J.L. Lagoda

The third Coordination and Steering Meeting of INT/5/150 was hosted by Turkey and congregated 26 participants and experts from 19 countries (Algeria, Australia, China, Egypt, India, Islamic Republic of Iran, Iraq, Jordan, Kenya, Lebanon, Oman, Pakistan, South Africa, Saudi Arabia, Sudan, Syrian Arab Republic, Tunisia,

Uganda, Yemen), FAO, IAEA and ICARDA. In a one week workshop, the recent positive developments were discussed and the roadmap for the coming year plotted. More about the recent outputs will be presented in the following newsletter under Ug99 'Highlights', when some of the positive developments can be consolidated.

### **Second Research Coordination Meeting on Enhancing the Efficiency of Induced Mutagenesis through an Integrated Biotechnology Pipeline, D2.40.12, Vienna, Austria, 13–17 December 2010**

Technical Officer: B. Till

The CRP was initiated in 2008, with the first RCM being held in Vienna, Austria from 25 to 29 May 2009. The CRP aims at evaluating and adapting methods and strategies to increase the efficiency of using induced mutations for crop improvement. For detailed information, see Coordinated Research Projects (CRPs) and Research Coordination Meetings (RCMs).

# Coordinated Research Projects (CRPs) and Research Coordination Meetings (RCMs)

Project Number	Ongoing CRPs	Scientific Secretary
D2.40.12	Enhancing the Efficiency of Induced Mutagenesis through an Integrated Biotechnology Pipeline	B. Till
D2.30.28	Improving Nutritional Quality by Altering Concentrations of Enhancing Factors Using Induced Mutation and Biotechnology in Crops	Y. Lokko
D2.40.13	Isolation and Characterization of Genes Involved in Mutagenesis of Crop Plants	P.J.L. Lagoda
D2.30.27	Molecular Tools for Quality Improvement in Vegetatively Propagated Crops Including Banana and Cassava	B. Till
<b>New CRP starting in 2011</b>		
D2.30.29	Climate Proofing of Food Crops: Genetic Improvement for Adaptation to High Temperatures in Drought Prone Areas and Beyond	M. Spencer

## Climate Proofing of Food Crops: Genetic Improvement for Adaptation to High Temperatures in Drought Prone Areas and Beyond, D2.30.29 (New)

Technical Officer: M. Spencer

This CRP was initiated in 2010. The first RCM is planned to be held in Vienna, Austria, 2-6 May 2011.

Climate change is now largely accepted as a real and pressing global problem. It has recently been estimated that developing countries will bear 70-80% of the costs of climate change damage with agriculture being the most impacted sector. The main impacts of climate change on agriculture will most probably be experienced through higher temperatures (increase in minima and maxima), altered changes in rainfall patterns (in amount, spatial and temporal distributions), increased rates of evaporation, increased intensity and frequency of extreme events (floods and droughts), and raise of sea level affecting coastal areas where large quota of cultivated land are located (intrusion of salty water). The responses that agriculture systems world wide can put in place to cope with the expected impact of climate change and to reduce the food insecurity range from institutional and policy levels to the best management practices and technology advancement. An important opportunity in terms of technology advancement is offered by the genetic improvement of crops that can adapt to the future climate conditions; i.e. 'climate proofing' crops. Additionally, data

collected from the selected crops: rice and common bean during the course of the CRP may be useful for a study of mathematical models for responses to high temperature in association with AquaCrop – FAO.

This CRP will focus on improving the grain yields of rice (cereals) and common bean (legumes), two essential staples in the diets of millions of impoverish and vulnerable populations, to high temperature stress in the face of climate change. The approach is to use (1) the whole plant (2) gene expression markers and (3) physiological/biochemical responses to high temperature to identify valuable germplasm.

- 1) Exploit genetic diversity (existing mutated populations,  $M_2$  and up) to assess tolerance to high temperature in terms of yield and yield components.
- 2) Analyse and exploit mutations in functional genomics using molecular tools such as positional cloning of critical genes, whole genome sequencing projects, SNP diversity analysis, global genomes expression analysis, and associated bioinformatics tools to evaluate large datasets and visualize metabolic pathways affected by stresses and/or genotypes.
- 3) Apply existing tools to characterize physiological and biochemical responses to high temperature on nodulation/nitrogen fixation and/or water use efficiency by application of stable isotope techniques.



## The AquaCrop Conceptual Framework



*Through the contribution of the AquaCrop modeling system, which integrates historical and current data from the atmosphere, the crop and its specific management system, and from the soil in the targeted region, one will be able to predict the trends and establish mathematical models for a better handling of crop productivity under the foreseen variations caused by the climate change and variability - Photos courtesy of Dr. Pasquale Steduto, Land and Water Division, FAO, Rome, Italy*

### Overall objective

To identify high yielding food crop germplasm contributing to sustainable food security (with a focus on a major cereal - rice and a grain legume - common bean) with improved resource use efficiency (water and nitrogen) and adaptation to high temperatures (increased minima and maxima) as anticipated by climate change and variability for the next 20 to 40 years.

### Specific objectives

1. To explore whole plant genetic variability and identify high yielding genotypes from existing natural and mutated germplasm of cereals (preferably rice) and nitrogen fixing grain legumes (preferably common bean) for adaptation to high temperature; establishing robust experimental protocols for physiological, genetic and molecular studies; use advanced biotechnology, bioinformatics and genomics tools for whole plant analysis and data visualization.
2. To facilitate technology transfer, sharing of genetic and knowledge resources (through electronic means, peer reviewed publications, workshops,

training courses, field days) and foster networks between participating research groups and potential end users for their mutual benefit

### Expected research outcomes

- Availability of advanced mutant crop resources for breeders, geneticists in National Agriculture Research Systems (NARS) in MS countries with adaptation to increased temperatures;
- Availability of phenotypic, physiologic and high throughput genetic tools for the characterization of adaptability to high temperature together with data sets for AquaCrop modelling;
- Successful implementation of the CRP and related publication reports, technical documents, and newsletters published.

### Expected research outputs (Results)

1. Data set on genetic variability for high temperature tolerance (HTT) in (a) a cereal and (b) a grain legume compiled;
2. High temperature tolerant germplasm by (a) whole plant performance, (b) molecular marker technolo-

gy, and (c) stable isotope techniques identified and tested for current and future climate change scenarios using crop simulation models;

3. A web-based platform to share knowledge, tools and technologies related to HTT in crops activated and maintained by participating members.

### Criteria for selecting participants

Projects must be built on the relevant past achievements of nuclear, genetic and genomic technologies. Based on these, the expected outputs must ensure that systems are sustainable in the face of climate change and do not adversely contribute to climate change. In addition, the following elements should be fulfilled:

#### Crop selection

- Existing genetic resources and mutants/mutant products for the particular crop (at least M<sub>2</sub> generation).
- Existing cereals and/or legumes related knowledge base and ongoing genetic, mutation or molecular work.

#### Target environment

- Agro-ecological zones most exposed to high and variable temperature as a result of climate change during crop growing season.

#### Capacity

- Must have the needed resources in terms of expertise (mutation techniques, physiology and/or molecular genetics), infrastructures, equipments including a sustainable national support funding for the project and an associated interdisciplinary team (e.g. an agronomist, statistician, etc.).
- Evidence of national/international collaborative research will be an asset.

#### Nuclear-Related Techniques Used Within the CRP

- Mutation: gamma irradiation, fast neutron, ion beam.
- Stable isotopes (<sup>15</sup>N, <sup>13</sup>C, <sup>18</sup>O).

#### Molecular techniques

- Forward and reverse genetics using mutant population screening.
- Extensive bioinformatics and data visualization technology.

#### Parameters and Methodologies to be used

- Cereal crops and grain legumes: yield and its components.
- Physiological responses to high temperature using isotopes (water use efficiency and nodulation).

- Basic molecular technologies (e.g. PCR, cloning, gel electrophoresis, etc.) and high throughput gene expression analysis of different plant tissues.
- Bioinformatics for large scale data handling, visualization and cross-species comparisons.

Up to 10 research contracts are expected to be awarded and four no cost agreement holders from advanced laboratories with experience in the selected technologies and fields of expertise to share their experience with the contract holders. In addition, it is foreseen that two technical contracts will be awarded for services in identification and isolation of mutant genes in model crops. Coordination and technical management will be handled by the scientific secretary in the Plant Breeding and Genetics Section.

### Enhancing the Efficiency of Induced Mutagenesis through an Integrated Biotechnology Pipeline, D2.40.12

Technical Officer: B. Till

The concept is to develop a series of different modules, from which combinations can be made to create a tailored pipeline specific for the crop and objectives of the researcher. Modules include methods to evaluate the density and spectrum of mutations using different mutagens, doses, and tissues and modes of induction. This allows guidelines to be produced on how to develop the most efficient mutagenized population. For example, the density of induced mutations can be used to determine the minimal population size required to obtain a mutation in a target gene. Knowledge of the spectrum of mutations (i.e. % of knocked-out alleles versus missense changes) allows for calculations on the probability of obtaining functionally deleterious alleles given a set population size. Different materials are being subjected to treatment with mutagens such as cell suspension cultures, microspores and seeds. Other modules include methods for selection of plants with desirable traits, the dissolution of chimeric sectors after mutagenesis, and reverse-genetic strategies to target mutations in candidate genes and obviate the need for large field trials. The target crops for this CRP are banana, barley, cassava and rice so that major biological systems and production constraints can be covered, including key differences between seed and vegetatively propagated species. The next RCM, scheduled for 13-17 December 2010, will focus on achievements since the 2009 meeting and plans for future activities. Specific attention will be given to planning for a new module with a recently added agreement holder to test rapid evaluation of mutation induction in banana cell cultures.

## Improving Nutritional Quality by Altering Concentrations of Enhancing Factors Using Induced Mutation and Biotechnology in Crops, D2.30.28

Technical Officer: Y. Lokko

This CRP started with a Consultants Meeting in 2008 in Vienna, Austria. The staff of the Joint FAO/IAEA sub-programme and four experts formulated a proposal for a new Coordinated Research Project (CRP) on 'Enhancing nutritional, nutraceutical and pharmaceutical value of crops using mutation induction techniques'. The experts consulted were Dr. S.K. Rasmussen (Denmark) and Dr. D.X. Wu (China), plant breeders with expertise in breeding for improved micronutrient content; Prof. P. White (UK), a Plant Nutritionist with expertise on plant nutritional genomics (including biofortification); and Dr. N. Tomlekova (Bulgaria), a geneticist with expertise in improving beta carotene content in vegetables.

The first RCM was held in Vienna, Austria, from 29 June to 3 July 2008. Eleven Scientists from Botswana, Bulgaria, China, Denmark, Ghana, Germany, India, Kenya, South Africa, Ukraine, the United Kingdom (UK) and the United States of America (USA) attended the meeting in Vienna to discuss the individual project activities, adapt the workplans, and facilitate possible collaboration between the research teams. The second RCM is planned to be held in Pretoria, South Africa, 11–15 April 2011.

Food security, nutrition and health are key issues in the national agenda of government planning in many countries in the world. Humans require more than 25 mineral elements. Recommended daily intakes and safe upper levels of many of these elements have been defined. The availability and intake of nutritious foods provides the required amount of calories, vitamins and minerals and ensures good health. In addition to forming the major components of human diets, providing the required calories and nutrients to sustain life, crop plants also contain most of the essential vitamins and, either directly or indirectly, deliver many of the essential mineral elements to the human diet. These vitamins and minerals are required to prevent common micronutrient disorders. However, the major staple crops are often deficient in some of these vitamins and minerals and, in many areas of the world the basic diet does not provide sufficient quantities. Thus, malnutrition, with respect to micronutrients like vitamin A, iron and zinc, affects >40% of the world's population. It is estimated that, of the 6 billion people in the world, 60-80% are Fe deficient, over 30% are Zn deficient and certain social groups do not receive sufficient Ca and Mg in their diets. Thus, the Copenhagen Consensus 2004 concluded that providing sufficient dietary micronutrients was among the most important research priorities for advancing global welfare.

Methods used to enable humans to get sufficient vitamins and minerals in their diets include supplementation (food

and tablets) or fortification of food or crops by fertilisation. However, this has not been very successful, particularly in developing countries due to inadequate resource. A more sustainable approach is to cultivate genotypes that accumulate greater concentrations of vitamins and minerals in their edible tissues and/or have increased bio-availability of minerals, particularly when low in soils and fertilizers are expensive, by increasing the concentrations of promoter substances that enhance the uptake minerals by humans (e.g. carotenoids and tocopherols) and decreasing the concentrations of anti-nutrient compounds that inhibit the absorption of minerals (e.g. phytate and oxalates). Both promoter and anti-nutritional compounds are synthesized by the plant and their levels can be changed genetically.

The overall objectives of this Coordinated Research Project (CRP) aims at utilizing the wealth of mutant germplasm in model crops, such as rice, tomato and barley, to understand and identify genes involved in the biosynthesis of nutritional quality enhancing factors, and develop efficient screening methods to facilitate the genetic improvement of nutritional quality. The goal is to transfer knowledge and technologies of beneficial mutants associated with nutritional factors from model crops to improve nutritional quality in other crops.

The results of the activities will be widely disseminated to provide a proof of concept on the use of ionomics, genomics and induced mutation in model crops to study genes affecting nutritional quality traits and their application in crop improvement. In the long term, induced mutants, genomic tools, and knowledge developed under this project will increase the efficiency and breeding for enhanced nutritional quality in the crops for the sustainable delivery of adequate levels of essential vitamins and minerals to reduce the negative health effects of vitamins and minerals deficiencies in Member States. This will ultimately lead to improved livelihoods and food security of the human populations that rely on these selected crops for sustenance.

Specifically the CRP will employ induced mutants, bioinformatics, functional genomics and ionomics, as well as appropriate genetic mapping populations, to identify molecular markers for marker-assisted selection (MAS) of nutritional quality traits. These traits are resistant starch (HR starch), increased carotenoid and tocopherol concentrations, and decreased phytate and oxalate concentrations. Mutants will be verified and assayed to confirm no detrimental effects on their mineral composition.

The specific objectives are to:

- 1) Use new and existing mutants in rice, tomato and soybean with the target nutritional quality traits, to define target genes for manipulating these traits in other crops.



- 2) Identify beneficial alleles for nutritional quality in the target crops.
- 3) Identify the consequences of reduced levels of anti-nutrient factors on mineral content (Fe, Zn, Ca, and Mg).
- 4) To generate mutant germplasm with reduced levels of oxalate in spinach, to understand and transfer knowledge of its biosynthesis to other leaf vegetables.
- 5) Facilitate technology and germplasm transfer between member states, and establish a catalogue of beneficial mutants in biochemical pathways improving nutritional quality.

The expected research outputs (results) include the generation of the following resources by the end of the project:

- 1) New mutant germplasm collections from elite varieties of spinach and target crops.
- 2) Developed efficient phenotypic screening methods for resistant starch, increased carotenoids, decreased oxalate and increased tocopherols.
- 3) Developed efficient genotypic screening methods for genes affecting synthesis of resistant starch, increased carotenoids, decreased oxalate, decreased phytate and increased tocopherols.
- 4) Increased knowledge of the metabolic pathways and genes affecting phytate, carotenoids and tocopherol biosynthesis.
- 5) Identification of genes affecting the production of resistant starch and oxalate crystals.
- 6) Mutant lines with beneficial traits affecting resistant starch, increased carotenoids, decreased oxalate, decreased phytate and increased tocopherols and increased mineral concentration (Fe, Zn, Ca, Mg).
- 7) Increased capacities in NARS to utilise induced mutations and genomic tool in breeding programmes.
- 8) Published and disseminated research results.

The expected outcomes are threefold:

- 1) Availability of advanced mutant resources for nutritionists, breeders, geneticists.
- 2) Availability of efficient phenotypic and genotypic screening technologies and protocols for target nutritional quality enhancing factors.
- 3) Established research linkages between scientists in Member States to address genetic improvement of nutritional quality enhancing factors in crops.

## Isolation and Characterization of Genes Involved in Mutagenesis of Crop Plants, D2.40.13

Technical Officer: P.J.L. Lagoda

This CRP started with a Consultants Meeting in 2008 in Vienna, Austria, gathering five experts, Drs. K. Riha (Austria), H. Puchta (Germany), A. Levy (Israel), B. Hohn (Switzerland), and A. Britt (USA), who were invited to present their work in the Concurrent Session number two of the International Symposium on Induced Mutations in Plants (ISIMP). They worked out the proposal for this CRP on Plant DNA Damage, Repair and Mutagenesis.

The first RCM was held in St. Louis, Missouri, USA in conjunction with the ninth International Plant Molecular Biology Congress (IPMB), from 26 to 31 October 2009. Eight research contract holders (Argentina, Bulgaria, China, India, Republic of Korea and Poland) and five agreement holders and consultants (Germany, Switzerland and USA), participated in this CRP. The second RCM is planned to be held from 30 May to 3 June 2011 in Vienna (see forthcoming events).

The overall objective of this CRP is to understand the mechanisms of mutagenesis and molecular genetic features of induced mutation in plants, to provide the scientific basis for developing more efficient mutation techniques, and their proper use for crop improvement. This is translated into the specific research objective of isolating and characterizing genes involved in mutagenesis. Further finding out types of mutations induced by various physical mutagens in studied crop plants; and generating mutants deficient in DNA repair pathways in crops is one of the mandatory steps in achieving this objective. The results expected will facilitate assessment of their usefulness for efficiency enhancement of mutation induction.

The expected research outputs include:

- 1) A Plant Mutagenesis Database, publicly accessible for all Member States. It will include plant homologs of DNA damage response and repair genes from a variety of species, and a list of existing mutants defective in these genes.
- 2) Characterized homologs of genes involved in DNA repair and mutagenesis in crop plants.
- 3) A better understanding of the spectrum (the molecular genetic feature) of mutations induced by various mutagens.
- 4) Improved protocols for mutation induction and screening, i.e. proper mutation screening method for particular mutagens.

- 5) Genetic resources that could be used for efficient mutation induction. This refers to mutant lines deficient in genes involved in mutagenesis.

The yearly reports are being evaluated and the CRP is on track.

### **Molecular Tools for Quality Improvement in Vegetatively Propagated Crops Including Banana and Cassava, D2.30.27**

Technical Officer: B. Till

This CRP was initiated in 2004. The first RCM was held in Vienna, Austria from 18 to 22 July 2005. The second

RCM took place in Thiruvananthapuram, Kerala, India, from 5 to 9 February 2007, and the third RC in Vienna, Austria from 11 to 16 August 2008 in conjunction with the International Symposium on Induced Mutations in Plants. The fourth and final RCM was just completed in Brasilia, Brazil from 8 to 12 November 2010. Please see the past events section to read more on the results of this RCM.

**IAEA Coordinated Research Activities Web Site:**

<http://www-crp.iaea.org/html/forms.html>

# Technical Cooperation Field Projects

The Plant Breeding and Genetics subprogramme currently has technical responsibilities for the following technical cooperation projects that are managed by the IAEA's Department of Technical Cooperation.

## Continuing Projects

Project Number	Country	Title and Objective(s)	Technical Officer
AFG/5/003	Afghanistan	Sustainable Increase in Crop Production in Afghanistan	Y. Lokko in collaboration with Soil and Water Management Section
ALG/5/023	Algeria	Protection of Date Palm Trees Against Bayoud Disease	M. Spencer
ALG/5/024	Algeria	Improvement of Cereals for Tolerance to Drought and Resistance to Disease	M. Spencer
ANG/5/006	Angola	Improvement of Food Crops Through Mutation Breeding and Biotechnology	M. Spencer
BGD/5/026	Bangladesh	Increasing Agricultural Production in the Coastal Area through Improved Crop, Water and Soil Management	S. Dharmapuri/Y. Lokko in collaboration with Soil and Water Management Section
BOT/5/003	Botswana	Mutational Improvement of Groundnut Varieties	S. Dharmapuri/M. Spencer
CAF/5/003	Central African Republic	Development of New Varieties of Cassava Through Mutation Breeding and Biotechnology Techniques	M. Spencer
COS/5/027	Costa Rica	Generation of Promising Strains of Beans through Induced Mutations in Calluses and Seeds to Increase Competitiveness	M. Spencer
CPR/5/017	China	Construction of Radiation-Induced Mutant Libraries and Function Analysis of Mutated Genes in Crop Plants	M. Spencer/Y. Lokko
ECU/5/023	Ecuador	Inducing Mutations in Agriculture with the Aid of Radiation	M. Spencer
ERI/5/004	Eritrea	Improving Crop Productivity and Combating Desertification	Y. Lokko/A.A. Cheema in collaboration with Soil and Water Management Section
INS/5/035	Indonesia	Application of Nuclear Techniques for Screening and Improving Cash Crop Plants in Coastal Saline Lands	S. Dharmapuri/M. Spencer
INS/5/036	Indonesia	Genetic Improvement of Artemisia Cina Using Irradiation Technique	M. Spencer
<b>Awaiting Financing</b>			
IRQ/5/017	Iraq	Optimization of Land Productivity Through the Application of Nuclear Techniques and Combined Technologies	S. Dharmapuri/P.J.L. Lagoda in collaboration with Soil and Water Management Section
JAM/5/010	Jamaica	Plant Breeding and Diagnostics Technologies	Y. Lokko



<b>Project Number</b>	<b>Country</b>	<b>Title and Objective(s)</b>	<b>Technical Officer</b>
MAR/5/018	Mauritius	Improvement of Banana and Tomato Varieties Through the Use of Nuclear Techniques for Mutation Induction and Biotechnology	M. Spencer/Y. Lokko
MYA/5/016	Myanmar	Development of Rice Varieties with Improved Iron Content/Bioavailability Through Nuclear Techniques	S. Dharmapuri/Y. Lokko
NIR/5/035	Nigeria	Adding Value to Root and Tuber Crops Through the Use of Mutation Induction and Biotechnologies	Y. Lokko
PAK/5/044	Pakistan	Improvement of Drought Tolerance in Chickpea Through Induced Mutations	M. Spencer
PER/5/030	Peru	Genetic Improvement of Quinoa and Kiwicha Using Mutation Induction and Biotechnology	Y. Lokko
QAT/5/002	Qatar	Developing Biosaline Agriculture in Salt-Affected Areas in Qatar	A.A. Cheema/P.J.L. Lagoda in collaboration with Soil and Water Management Section
RAF/5/056	Regional Africa	Field Evaluation and Dissemination of Improved Crop Varieties Using Mutation Breeding and Biotechnology Techniques	M. Spencer/S. Dharmapuri
RAS/5/045	Regional Asia	Improvement of Crop Quality and Stress Tolerance for Sustainable Crop Production Using Mutation Techniques and Biotechnology (RCA)	Y. Lokko/A.A. Cheema
RAS/5/048	Regional Asia	Mutation Induction and Supportive Breeding and Biotechnologies for Improving Crop Productivity (ARASIA)	P.J.L. Lagoda
RER/5/013	Regional Europe	Evaluation of Natural and Mutant Genetic Diversity in Cereals Using Nuclear and Molecular Techniques	Y. Lokko/S. Dharmapuri
SAF/5/010	South Africa	Development of New Maize and Sorghum Germplasm with Enhanced Nutritional Content	Y. Lokko
SAU/5/003	Saudi Arabia	Improving Fertilization under Saline Conditions for Sustainable Crop Production	A.A. Cheema/P.J.L. Lagoda in collaboration with Soil and Water Management and Crop Nutrition Section
SEN/5/030	Senegal	Integrated Approach to Develop Sustainable Agriculture in Senegal	M. Spencer in collaboration with Soil and Water Management and Crop Nutrition Section
SIL/5/009	Sierra Leone	Improving Sorghum Productivity Through Nuclear and Biotechnology	S. Dharmapuri/Y. Lokko
SUD/5/030	Sudan	Increasing productivity of Selected Crops Using Nuclear Related Techniques	A.A. Cheema/M. Spencer in collaboration with Soil and Water Management Section
TUN/5/023	Tunisia	Radiation-Induced Mutations for Improvement of Cactus	Y. Lokko
TUN/5/024	Tunisia	Development of Improved Strains of Olive Tree Through Mutation Breeding and Biotechnology	Y. Lokko

Project Number	Country	Title and Objective(s)	Technical Officer
UZB/5/004	Uzbekistan	Development of Mutant Cotton Breeding Lines Tolerant to Diseases, Drought and Salinity	Y. Lokko
YEM/5/008	Yemen	Introduction of Gamma Ray Irradiation Techniques for Agriculture Purposes	Y. Lokko
ZAI/5/016	Democratic Republic of the Congo	Mutation Techniques for Improving Nutritional and Medicinal Plants with a Curative Effect on Human Diseases and Alimentary Plants	M. Spencer
ZIM/5/013	Zimbabwe	Development of Drought Tolerant and Disease Resistant Grain Legumes, Phase I	Y. Lokko

## Projects Started in 2009

Project Number	Country	Title and Objective(s)	Technical Officer
AFG/5/004	Afghanistan	Enhancing Crop Productivity through Mutation Breeding and Pest Control	Y. Lokko in collaboration with Insect Pest Control Section
BKF/5/007	Burkina Faso	Improving Voandzou and Sesame Based Cropping Systems through the Use of Integrated Isotopic and Nuclear Techniques	M. Spencer
BOL/5/018	Bolivia	Enhancing Food Security Using Conventional and Nuclear Techniques for the Acquisition of Climate-Change Tolerant Commercial Potato Seed	M. Spencer/S. Dharmapuri
COL/5/023	Colombia	Enhancing Mutagenesis and Biotechnology Used in the Improvement of Rice	Y. Lokko/S. Dharmapuri
COS/5/028	Costa Rica	Generating Promising Strains of Beans through Induced Mutations in Calluses and Seeds to Increase Competitiveness (Phase II)	M. Spencer
ECU/5/025 Awaiting Financing	Ecuador	Inducing Genetic Variability in Soya, Banana and Rice	M. Spencer
INS/5/037	Indonesia	Applying Nuclear Techniques for Screening and Improving Cash Crop Plants in Coastal Saline Lands	S. Dharmapuri/M. Spencer in collaboration with Soil and Water Management Section
INS/5/038	Indonesia	Using Induced Mutations to Improve Rice Productivity through a Hybrid Rice Breeding Programme	S. Dharmapuri/M. Spencer
INT/5/150	Interregional	Responding to the Transboundary Threat of Wheat Black Stem Rust (Ug99)	P.J.L. Lagoda/S. Dharmapuri
IVC/5/031	Cote d'Ivoire	Improving Plantain and Cassava Yields through the Use of Legume Cover Crops	M. Spencer
KAZ/5/002	Kazakhstan	Improving Wheat and Maize Using Nuclear and Molecular Techniques	Y. Lokko/A.A. Cheema
KEN/5/029	Kenya	Developing Appropriate Artemisia Varieties for Management of Malaria	Y. Lokko/M. Spencer
MAG/5/018	Madagascar	Improving Cereal Production (Rice and Maize) through Mutation Breeding for Tolerance/Resistance to Striga ( <i>Striga asiatica</i> )	M. Spencer

<b>Project Number</b>	<b>Country</b>	<b>Title and Objective(s)</b>	<b>Technical Officer</b>
MAK/5/006	Macedonia, the Former Yugoslav Republic of	Improving Wheat, Barley and Triticale for Food and Feed in Drought-Prone Areas, Using Nuclear Techniques	Y. Lokko
MAL/5/028 <b>Awaiting Financing</b>	Malaysia	Enhancing the Production of Bioactive Compounds in a Local Herbal Plant by a Soilless Planting System and <i>In Vitro</i> Mutagenesis	Y. Lokko/M. Spencer
MYA/5/017	Myanmar	Studying Yield Improvement of Local Rice Varieties though Induced Mutation	S. Dharmapuri/Y. Lokko
MYA/5/019 <b>Awaiting Financing</b>	Myanmar	Developing Thermo-Insensitive (Cold-Tolerant) Green Gram Genotypes, Using Mutation Techniques	Y. Lokko
NAM/5/009	Namibia	Using Mutation Breeding and Integrated Soil Plant Management Techniques to Develop Sustainable, High Yielding and Drought Resistant Crops	Y. Lokko in collaboration with Soil and Water Management Section
NER/5/014	Niger	Improving the Productivity of Cowpea/Finger Millet Based Cropping Systems	M. Spencer in collaboration with Soil and Water Management Section
RLA/5/056	Regional Latin America	Improving Food Crops in Latin America though Induced Mutation (ARCAL CV)	M. Spencer/Y. Lokko
SAF/5/012	South Africa	Analysing the Level of Drought Tolerance in Mutant Gerplasms of Cowpea and Amaranthus Using Molecular Biotechnology	Y. Lokko
SEN/5/032	Senegal	Improving the Productivity of Jatropha Curcas Plantations in Semi-Arid Areas	M. Spencer
THA/5/049	Thailand	Increasing Productivity of Selected Crops Using Nuclear Related Techniques	Y. Lokko/A.A. Cheema
TUR/5/025	Turkey	Using Molecular Techniques for Enhancing the Efficiency of Mutation Induction and Utilization of Mutants in Agriculture	A.A. Cheema/Y. Lokko
URT/5/026	United Republic of Tanzania	Improving Rice Varieties through Mutation Breeding and Biotechnology in Zanzibar	S. Dharmapuri/Y. Lokko
UZB/5/005	Uzbekistan	Developing Mutant Cotton Breeding Lines Tolerant to Diseases, Drought and Salinity (Phase II)	Y. Lokko/P.J.L. Lagoda
YEM/5/010	Yemen	Using Induced Mutations and Efficiency Enhancing Bio-molecular Techniques for Sustainable Crop Production	Y. Lokko
ZAM/5/026	Zambia	Improving Crop Varieties through Use of Nuclear Techniques	Y. Lokko in collaboration with Soil and Water Management Section



## TC Projects Closed in 2010

Project Number	Country	Title and Objective(s)	Technical Officer
ROK/5/035	Republic of Korea	Using a Gamma Phytotron for Mutant Induction to Improve Food and Ornamental Crops	P.J.L. Lagoda

For details, see the IAEA Technical Cooperation Programme's Website at:

<http://www-tc.iaea.org/tcweb/default.asp>

## TC Project Highlights

### Lanka Cherry – A new cherry type tomato variety through induced mutations, RAS/5/045



Dry seeds of Manik variety were treated with 320 Gy gamma rays from  $^{60}\text{Co}$  source. Generations from  $M_1$ - $M_4$  were established and evaluated. In  $M_2$  generation, an attractive cherry type mutant was identified, among the other mutants. The plant is an indeterminate type. The fruit is pear shaped small, weighing 8.0 g, dark red in colour and has a brix value of 6.0. It fetches a higher price than the normal tomato varieties. The shelf life is about 14 days. Bacterial wilt (BW) screening studies in the field and laboratory revealed that it is resistant to BW. The yield evaluation studies in  $M_4$  generation, using RCB design with the check varieties T245 and Thilina at research station trials in different agro-ecological zones revealed that it has a yield potential of 17 t/ha. The variety was also tested in farmer fields for its adaptability and acceptance. The farmers reported that the new variety is easy to cultivate as it is less prone to pest and diseases. Lanka cherry variety is an improvement over the original variety Manik which has irregular shape, non-attractive colour and large locular cavities. The Lanka variety cherry was officially released by the Department of Agriculture in Sri Lanka on 22 June 2010. This is the first cherry type tomato variety released by the Department of Agriculture in Sri Lanka. At present, there is a big demand for this variety from airlines, shipping lines and super markets.

*Ranjani Peiris  
Horticultural Crops Research and Development  
Institute, Gannoruwa, Peradeniya, Sri Lanka*

# News

## Using mutagenesis to develop naturally caffeine free coffee crops in Brazil

In 2004, a group of Brazilian scientists reported in *Nature* the discovery of a coffee genotype whose seeds after dehydration are naturally caffeine free. However, attempts for commercially exploiting this naturally decaffeinated coffee turned out to be quite frustrating due to the low productivity of the plants originating from Ethiopia.

Now, using mutation induction, a researcher from the State University of Campinas (UNICAMP) obtained, from irradiated commercial coffee seeds, seven mutant plants combining good productivity and the absence of caffeine in the seeds.

The study, completed with the assistance of FAPESP, was performed by Dr. Paulo Mazzafera, Professor at the Plant Biology Department and Director of the Biology Institute in UNICAMP. According to Mr. Mazzafera seeds from the commercial coffee: Red Catuai (arabica) were treated. About 28000 plants were analysed with a qualitative method to score the presence or absence of caffeine in the seeds.

‘With the treatment, we obtained seven coffee plants which almost totally lacked caffeine. These plants are quite vigorous and are currently flowering. We have already performed a large part of the biochemical and molecular analysis of these plants and we are looking for a Brazilian company interested in commercially developing these naturally decaffeinated varieties’, said the FAPESP agency.

The scientist explained that in the experimental field where this study was conducted, there are now 250 plants. With the seeds available, it is possible to plant five Ha for further testing.

‘If everything goes right, we will proceed with these seeds and take all the genotypes to the fields by 2011’,

said Mr. Mazzafera, who is one of the members of the coordination team in agronomy and veterinary in FAPESP.

According to Mr. Mazzafera, the commercial interest for decaffeinated coffee is quite small in Brazil, in striking contrast to other countries. Less than 1% of the commercial coffee in Brazil is decaffeinated. On the other hand, in Europe and in the USA, the broadcast of the negative effects of caffeine has driven a steady increase of the consumption of decaffeinated coffee.

‘Decaffeinated coffee corresponds to about 10% of the commercialized coffee in the world. It is certainly a very interesting market with high added value. The alternative of having a decaffeinated coffee without having to go through the industrial processing appears very promising in terms of marketing’, said the scientist.

The plants obtained through this process bear one problem: the structure of the coffee flower normally warrants that the plant has a high degree of self-pollination - about 95% - but the flowers of the mutant plants prematurely open and therefore the rate of self-fertilization is quite different.

‘As the flowers open prematurely, this could allow cross-pollination. In fact, the problem is not that bad, because we can plant blocks with solely the mutant plants, separated from segregations from the normal plants. Or we could also place bees on the plants with low content in caffeine and thus increase the rate of self-pollination’, said Mr. Mazzafera.

One of the main results of the mutation induction method was the time saving. ‘Conventional genetic improvement would require a long time to develop productive decaffeinated plants’ said the scientist.



# Success Story

## Radiation Breeding of Hybrid Rice II You 838 in China

In 1968, the Sichuan Institute of Atomic Energy (SIAE) initiated research to improve the quality of hybrid rice through radiation induced mutations. The first successful restorer line, Fuhui 06, was developed in 1983. This was followed by the development of more than 20 new fine quality rice varieties. Among them, II You 838 occupies pride of place.



*Field trial of II You 838*

II You 838 is a new middle-season hybrid rice and was developed by combining hybridization and radiation mutation technology. It is the first rice variety developed in the Sichuan Rice Breeding program with at least a 5% yield increase over Shanyou 63, the widest distributed check variety in China. It is regarded as the outstanding achievement of this program in the last 15 years. Under a comprehensive evaluation conducted by Chengdu Institute of Biology, the Chinese Academy of Sciences, and Chongqing Crops Research Institute, II You 838 tested superior in rice quality when compared to Shanyou63. It was approved for release by the Sichuan Provincial government and the National Approval Committee for Crop Variety in 1995 and 1999, respectively. In 2005, II You 838 was designated as the check variety for late maturing medium indica rice group in China.

II You 838 has proven to be of the most adaptable rice varieties in the world. It is planted in large areas of the Northern and Southern hemispheres, in China and abroad. According to our estimates, II You 838 has been planted in 10 000 000 ha since 1995 and is becoming the largest planted rice variety in China after Shanyou63.

II You 838 has already made a tremendous contribution to the increase of grain output in many countries. From 1997 to 2010, it was planted in Vietnam, Bangladesh, Pakistan, Nepal, Myanmar, Indonesia, Malaysia, Guinea, Brazil, Peru, and Namibia over an area of more than

3 000 000 ha, with the largest acreage in Vietnam. Between 1998 and 2006, more than two-thirds of the rice seeds imported by Vietnam from China, belonged to the II You 838 variety. It increased the grain production of Vietnam dramatically, transforming it from a grain-importing country to an exporter. It also became the national check variety of Vietnam



*Vietnam delegation visiting SIAE*

## Principle characteristic

The chief varietal characteristics of II You 838 are a high rate of ear head formation, a strong stalk leading to high lodging resistance, moderate resistance to blast disease and fine rice quality. It has a high and stable yield and is tolerant to high temperature, cold and drought. A series of field experiments and studies have been conducted and confirm these characters.

High yields of II You 838 was measured over wide planted areas in Sichuan, Henan, Hubei, Jiangxi, Shaanxi, Yunnan, Guizhou, Fujian, Guangxi and Guangdong. Hundreds of tests performed in the southern rice-growing areas in China showed that II You 838 had significant yield increases (8-20%) when compared with Shanyou63. In provincial and national trials monitored over a decade, the stable yield ability of II You 838 was higher than Shanyou 63.



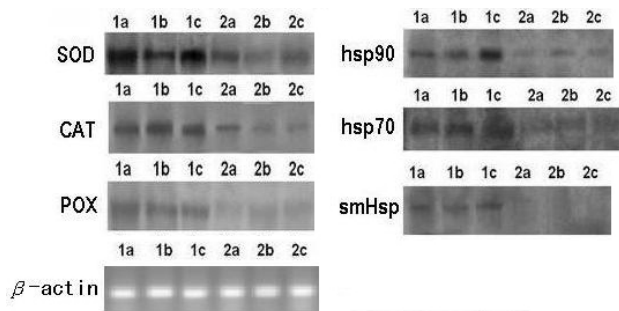
*Ear heads of II You 838 in the field*



II You 838 is tolerant to high temperature and drought. It was field tested in comparison to 30 varieties including Fuhui 838 and Shanyou 63. The tolerance of II You 838 to high temperatures (daily average of 36°C) during tassel period was significantly higher than other combinations including Shanyou 63. The seed setting rate also did not change after significantly high temperature treatment for three days, and it still remained 55% above control after 4 days of treatment. Genetic analysis indicated that heat tolerance was mainly inherited from the male parent and was higher than that of either parent.

Heat tolerant gene expression was investigated by molecular approaches. A number of genes were identified in the leaves. The relationship between heat tolerance genes, heat shock protein genes such as *Hsp* 90, *Hsp* 70, *Hsp* 18 and antioxidant enzyme genes including *SOD*, *CAT*, *POX* was also studied. The result showed that the expression of these six genes in II You 838 and its filial generations was significantly up-regulated after high temperature treatment. This suggested that these genes were associated with heat tolerance and could be inherited stably in II You 838.

During the domestic promotion process, II You 838 was also found to be more cold tolerant than other rice varieties. Further studies on the breeding and qualifying characteristics of II You 838 are currently being undertaken. II You 838 has already demonstrated strong resistibility, high and stable yield performance and wide adaptability in China and Southeast Asia area. Its plantation area will further be enlarged in the coming years. Its mechanism of heat, drought and cold tolerance is a topic of intense research interest. Given these useful characters, this variety can play a significant role in attaining food security for vulnerable areas.



Expression of genes related to heat tolerance in II You 838 and its filial generations. 1. Heat treated; 2. Control; a. Fuhui 838; b. II-32A; c. II You 838

# Developments at the Plant Breeding and Genetics Laboratory, Seibersdorf

## Introduction

Through the three pronged approach of technology development; adaptation and transfer, the provision of technical and analytical services, and human capacity building, the dedicated staff of the Plant Breeding and Genetics Laboratory support efforts in Member States for the development of superior crop varieties, with a special emphasis on the utility of using mutation based strategies. Most of this work takes place in the Seibersdorf laboratories, but staff also participates in off-site training and meetings, and support staff of the Plant Breeding and Genetics Section in the normative functions of the FAO/IAEA Joint Division. Below you will find an overview of some of our activities over the past six months.

## Technology Development and Adaptation

If you quiz scientists, a common theme that emerges is that most of us feel there is more work to do than hours in the day. If this describes you, please take a moment now and look out your window. If you are lucky, you have a view that includes plants and animals. As biologists, we are fascinated by the diversity of life on this planet. This fascination is not merely academic, nor for the sake of beauty; diversity is a cornerstone to continued and sustainable agricultural production. It is the tool the plant breeder uses to generate crops with improved traits. More fascinating still is the fact that much of the diversity we observe can be explained by changes in DNA sequence. Where useful diversity is not available in nature, the plant breeder can exploit induced mutations that mimic what occurs in nature, but on scale and time-frame that is orders of magnitude greater, and practical for the breeder. The PBGL continues to focus efforts on developing and adapting technologies suitable for addressing pressing needs in developing Member States, with the majority of its efforts going towards this cause.

## Impacts of NaCl stress on the biomass and mineral nutrient assimilation in different rice varieties

Salinity stress, which usually occurs in arid and semiarid regions, is a major environmental constraint to plant growth and the crop productivity. Excessive soil salinity can result from natural processes, or from crop irrigation such as using saline irrigation water and/or poor drainage conditions. Plant physiological responses to salinity and their relation to salinity resistance have been much researched and frequently reviewed in recent years. One assumption is that plants with increased salinity resistance are expected to maintain higher rates of growth than susceptible plants when experiencing equivalent levels of salinity stress. Salinity stress occurs in three

ways: low water potential of the root medium leads water deficit, toxic effects of excess accumulation of ions such as  $\text{Na}^+$  and  $\text{Cl}^-$ , and nutrient imbalance caused by depression in uptake and/or shoot transport in saline soils. Plant species and cultivars within a crop species can differ greatly in their response to salinity. Genetic variability within a species is therefore a valuable tool for screening and breeding for higher salt tolerance. Unfortunately, the production and introduction of salinity resistant crops has been a relatively slow process. Crucial to supporting traditional and mutation breeding approaches are early and rapid screening methods to identify plants with improved responses. In order to determine predictive screening parameters that can be applied at early development stages for crops such as rice, a study in the PBGL is ongoing to evaluate the impacts of the salinity stress on the biomass and inorganic components with a total six rice varieties (two tolerant wild types, two moderates and two susceptible varieties).

## Comparative studies of different mutagens on seeds propagated crops

We are continuing our comparative studies of the effect of gamma and X-ray irradiation on rice plants. The main objective is to produce optimized protocols for X-ray treatment and subsequent early phenotypic evaluation of mutagenized plants, especially the development of methods for accurate dose selection for bulk irradiation. For this purpose, five Malagasy rice accessions have been used and irradiated with gamma and X-ray with following doses: 75, 150, 300, 450 and 600 Gy. The effect of irradiation has been observed on the  $M_1$  generation and following phenotypic traits have been recorded for the dose effect evaluation: germination/survival rate, root and shoot length at the seedling stage, shoot length, number of tillers, number of leaves at the mature stage and shoot length, number of tillers, leaves, number and length of panicles, and number and weight of grains and flowering time. Statistical methods have been applied in order to measure the level of significant changes.

One of the five accessions has been selected for further analyses in the  $M_2$  stage to evaluate heritability of the induced mutation events. Four thousand and six hundred  $M_2$  rice plants have been germinated. Phenotypic characterisation of the  $M_2$  plants is ongoing and the data are being collected for the same characteristics as at the  $M_1$  stage. In addition to those measurements, growth rate and chlorophyll content data have been taken as well as the cytological studies of the microspores. Preliminary observations of the  $M_2$  plants show significant differences in the response to the various mutagens and sources of

irradiation. In addition to supporting research activities of counterparts from Madagascar, and the PBGLs efforts to update guidelines on the use of X-ray irradiation for mutation breeding, materials produced in this project will be evaluated and incorporated into the ongoing CRP 'Enhancing the Efficiency of Mutagenesis through an Integrated Biotechnology Pipeline'; one example of how the PBGL is working to synergize its research and capacity building capabilities.

### **Mutation discovery and reverse-genetic strategies: improving protocols and guidelines for mutation breeding and targeting deleterious mutations**

Readers of the newsletter are no strangers to the fact that the use of induced mutations for breeding improved crop varieties has been enormously successful resulting in over 3000 induced varieties and income gains estimated in the billions of US dollars. That said, modern biotechnologies are enabling strategies to greatly increase the efficiency of this process, and consequently the chances of success, for mutation breeding projects. Over the past several years, the PBGL has invested in developing mutation discovery platforms to evaluate the density and spectrum of induced mutation events in mutagenized populations. Much has been learned in the past decade of the consequence of chemical mutagenesis on the genome and the inheritance of mutations in seed-propagated species, including crops. Less is known for physical mutagens such as gamma and X-ray irradiation and the density, spectrum and inheritance of resulting mutations in vegetatively propagated species. With such knowledge, it becomes possible to develop statistically significant estimations for the minimal population size required for a high probability of recovering desired mutant alleles. More nuanced estimations such as the efficiency of using induced mutations for modifying quantitative traits also becomes possible.

The PBGL is continuing efforts to screen a population of mutagenized triploid bananas that were mutagenized and propagated in tissue culture. This is a common approach for vegetatively propagated species such as banana and plantain, and cassava; major staples and food security crops for nearly one billion people in the developing world. Markedly different than approaches for seed-propagated crops, extant protocols and guidelines were developed using visual phenotypic assays relying mostly on dominant alleles. However, many alleles are expected to be recessive and therefore genomic mutation screening should allow increased resolution for evaluating mutant populations. Mutation screening in banana supports this, although the data set is complex due to the method of clonal propagation that is commonly employed to dissolve genetically chimeric tissue types. As experiments continue, preliminary data suggests that the strategy employed is robust, with a spectrum and density of muta-

tions expected for triploids, and stable inheritance of mutations. The guidelines that emerge from this work will be applicable to a variety of species where seed-mutagenesis strategies are less desirable, not efficient, or impossible.

Efforts have been ongoing in cassava as well. Since the last newsletter, screening for mutations in a starch synthesis gene in a population of 80 irradiated samples previously characterized to have defects in starch biosynthesis has been completed. Quality control assays for primer design show that this step remains inefficient due to incomplete genomic sequence availability. No candidate mutations that could be causative for phenotypes were found, suggesting that such candidate gene approaches are likely less efficient than traditional TILLING reverse-genetic approaches that employ larger populations. Efficiency is expected to increase as novel sequencing technologies allow higher-throughput and lower cost screening of larger genomic regions.

Work also continues on seed-propagated rice, and like with cassava and banana, is tied to the ongoing Coordinated Research Project on 'Enhancing the Efficiency of Mutagenesis through an Integrated Biotechnology Pipeline', where the PBGL is supporting the project by adapting and providing mutation discovery and phenotypic screening assays. For rice, the focus has been on developing target-specific primers for genes involved in iron content in seeds and herbicide resistance. Planning the next phase of this research will be finalized at the Research Coordination Meeting for the CRP that will be held in Vienna in December 2010.

### **Lower cost molecular assays**

Staff of the PBGL are continually focused on the development and adaptation of lower cost molecular assays that are suitable for developing Member States where resources can be limited. While many of our efforts focus on mutation discovery (see below), even the most basic procedures such as extraction of genomic DNA from plant tissues can be made lower cost and higher-throughput by developing do-it-yourself protocols rather than relying on expensive kits and equipment. Towards this end, the PBGL has developed a working protocol for DNA extraction that is non-toxic and very low cost. The strategy, however, suffers from low throughput as samples are processed in single tubes. To remedy this, we are working on strategies to make the assay suitable for a 96-well plate format. Currently, a key limitation has been sufficient tissue maceration for the recovery of sufficient quality and quantity of DNA for downstream molecular applications. To remedy this, we have machined several grinding tools with different diameter pins to develop a method of sufficient tissue grinding (see below). Evaluation and optimization experiments are underway. If such a device is deemed suitable, it will represent a sufficient



cost-reduction over currently used equipment for grinding.



The work of the PBGL to develop and adapt low-cost mutation discovery assays is ongoing. The group has recently published a study in the journal *Theoretical and Applied Genetics* showing the development of a fluorescence based Ecotilling platform for polymorphism discovery in diploid and polyploid *Musa* (banana and plantain accessions). As described in the last newsletter and the subsequent peer-reviewed publication, this was deemed a rapid, highly accurate, and robust approach. With this serving as a baseline, progress has been made to adapt a low-cost approach using enzymatic mismatch cleavage to induce double strand DNA breaks followed by resolution of cleaved fragments using standard agarose gels. The protocol was adapted for banana for screening local germplasm and induced mutants as part of the research fellowship work of Ms. Babita Dussoruth of Mauritius (Fellowship Code No.:C/MAR/9005). The PBGL is also working on adapting the method for use in other species with research fellow Ms. Wahiba Tiliouine (Fellowship Code No.: C6 /ALG/10051) who is developing the method for characterization of Algerian barley accessions.

### Protocols and guidelines and positive control kits

We invite readers to visit the section of Mutant Variety and Genetic Stocks database (MVGS) developed specifically for protocols, guidelines and information on services and materials provided by the PBGL (<http://mvgs.iaea.org/LaboratoryProtocols.aspx>). In addition to protocols and advice on topics such as inducing mutations and mutation discovery, we also provide information and protocols for our positive control kits for TILLING and Ecotilling. Most recently we have sent the kits to counterparts in Pakistan and Mauritius. Also on this site are instructions and request forms for the two major services we provide: genotyping and mutagenesis. New to the site is our manual for the popular interregional training course series that assembles protocols covering topics from molecular markers, population genetics

and TILLING. Have suggestions for new protocols? Please let us know. We can also provide video protocols for specific topics on request. If you have developed a protocol that you think would be beneficial to researchers in developing member states, we will happily consider it for our website.

### Services in Support of Activities in Member States

The irradiation service requests from Member States executed from June to December 2010 are summarized below. In all eight Member States requested support with irradiating the seeds or propagules of nine different plant species with seven being food crops. A further two requests for radio-sensitivity tests were fulfilled and performed on four crop varieties.

#### Summary of irradiation services provided by PBGL between January and June

Member State	Crop species
United Kingdom	<ul style="list-style-type: none"> <li>• <i>Primula vulgaris</i></li> <li>• <i>Triticum aestivum</i></li> </ul>
Jordan	<ul style="list-style-type: none"> <li>• Durum wheat</li> </ul>
Madagascar	<ul style="list-style-type: none"> <li>• <i>Oryza sativa</i> (calli)</li> </ul>
Kenya	<ul style="list-style-type: none"> <li>• <i>Hordeum vulgare</i></li> </ul>
Spain	<ul style="list-style-type: none"> <li>• <i>Euphorbia lathyris</i></li> <li>• <i>Zantedeschia aethiopica</i></li> </ul>
Turkey	<ul style="list-style-type: none"> <li>• <i>Sesamum indicum</i></li> </ul>
Philippines (IRRI)	<ul style="list-style-type: none"> <li>• <i>Oryza sativa</i></li> </ul>
Poland	<ul style="list-style-type: none"> <li>• <i>Lupinus angeustifolius</i></li> </ul>
Bulgaria	<ul style="list-style-type: none"> <li>• <i>Triticum aestivum</i></li> </ul>

Genotyping services included AFLP, SSR and DNA sequence analysis of mutant and natural varieties of crop species in support of fellowship research for counterparts in Syrian Arab Republic, Algeria, Zambia, and Senegal, along with activities directly supporting colleagues in other laboratories in the FAO/IAEA Joint Programme, and the PBGL. A total of 2656 samples were analysed in the second half of 2010. Flow cytometry was also performed to characterize ploidy levels in local *Musa* accessions from Mauritius in support of fellowship activities.

### Training and Human Capacity Building

Each year the PBGL trains many scientists on mutation induction and efficiency enhancing biotechnologies for the development of superior crop varieties. While fellows visit the PBGL to learn new techniques, PBGL staff also learns from research fellows. This helps to broaden staff understanding of the needs and pressing issues facing agricultural production in developing countries, and enables a demand-driven aspect for adaptive research and development activities of the PBGL. As described above,

this is highlighted by the recent work on low-cost assays. Through research fellows, the PBGL staff are also exposed to a range of diverse crop species that provide a good base for continued efforts for protocol adaption. For example, current fellow Mr. Hussam Alfaoury (SYR/10017) is working with PBGL staff to adapt TILLING and Ecotilling protocols for mutants and accessions

of Syrian crops cotton, garlic, sesame, *Daucus* (including carrot) and date palm. Please see the table below for a description all interns, research fellows and scientific visitors of the last six months.

### Fellows

Name	Country	Areas of training	Period
Mr. Hussam ALFAOURY	Syrian Arab Republic	<ul style="list-style-type: none"> <li>Genetic diversity study of different accessions of <i>Daucus</i>, cotton and date palm varieties from Syria</li> <li>TILLING and Ecotilling for characterization of Syrian <i>Daucus</i> and cotton germplasm and putative sesame and garlic mutants.</li> </ul>	August-December 2010
Ms. Wahiba Amri Epse TILIOUINE	Algeria	<ul style="list-style-type: none"> <li>Molecular biology techniques for the genotyping of putative barley mutants.</li> </ul>	September-November 2010
Ms. Marilyn ALFORQUIE	Philippines	<ul style="list-style-type: none"> <li>Induced mutagenesis facilitated by <i>in vitro</i> techniques</li> <li><i>In vivo</i> and <i>in vitro</i> techniques for screening plants for virus resistance</li> </ul>	October-December 2010

### Interns

Name	Country	Areas of training	Period
Ms. Kamila KOZAK	Poland	<ul style="list-style-type: none"> <li>Cytological and phenotypic analyses of M<sub>2</sub> rice plant</li> <li>Technology transfer to the Lupines sps</li> </ul>	August 2010 – March 2011

### Scientific visitors

Name	Country	Areas of training	Period
Mr. Tsegay Berhane GHEBREMARIAM	Eritrea	<ul style="list-style-type: none"> <li>Induced mutations in crop improvement and related biotechnologies</li> </ul>	18-22 October 2010

### Laboratory Staff Publications

Tadele Z., Mba C., Till B.J. (2010) TILLING for mutations in model plants and crops. In *Molecular Techniques in Crop Improvement*, 2<sup>nd</sup> edition; Springer, Netherlands. pp 307-332.

Till B.J. (2010) Mutation detection in plants by enzymatic mismatch cleavage. In *The Handbook of Plant Mutagenesis and Mutant Screening (Mining of Natural and Induced Alleles)*; Wiley-Blackwell-VHC, Germany. pp 131-148.

Till B.J., Jankowicz-Cieslak, J., Sagi L., Huynh O.A., Utsushi H., Sweenen R., Terauchi R. and Mba C. (2010) Discovery of nucleotide polymorphisms in the *Musa* gene pool by Ecotilling. *Theoretical and Applied Genetics*, 121(7): 1381-1389.

Lamien C.E., Lelenta M., Goger W., Silber R., Tuppurainen E., Matijevic M., Luckins A.G. and Diallo A. (2010) Real time PCR method for simultaneous detection, quantitation and differentiation of capripoxviruses.

### **Plant Breeding and Genetic Laboratory Staff Travel to the Member States**

#### **Mr. B. Till**

*Katowice, Poland:*

- Served as an expert lecturer on TILLING for the Regional Training Course on the use of Induced Mutations (TILLING) and DNA Markers in Cereal Genetics and Breeding, Katowice, Poland (RER/5/013), 15-17 September 2010.



*Cluj-Napoca, Romania:*

- Presented the PBGL's research into efficiency enhancing strategies for incorporating induced mutations into technology packages for vegetatively propagated species at the BIOVEG symposium on Plant Biodiversity and Food Diversification, Cluj-Napoca, Romania, 27-29 September 2010.



*Brasilia, Brazil:*

- Served as Technical Officer at the Fourth and Final Research Coordination Meeting on Molecular Tools for Quality Improvement in Vegetatively Propagated Crops Including Banana and Cassava, Brasilia, Brazil, 8-12 November 2010.

#### **Mr. S. Bado**

*Astana, Republic of Kazakhstan:*

- Served as an expert for a training course on Biotechnology and Modern Tools and Methods for Breeding and Plant Genetic Resources, National Center for Biotechnology of the Republic of Kazakhstan, Astana, 20-24 September 2010



### **TILLING and Ecotilling positive control kit**

The Plant Breeding Unit in Seibersdorf has developed a positive control kit for TILLING and Ecotilling that it is now available for researchers in Member States upon request. The kit contains genomic DNA with known nucleotide polymorphisms, gene-specific oligonucleotide primers, buffers and enzymes for mutation and polymorphism discovery. Along with the protocols, examples of high quality data produced with these materials are provided to serve as a reference point for assay optimizations.



## Reports

### Visit of FAO Permanent Representatives – Asia Group, Vienna, Austria, 5–6 August 2010

DIR-NAFA (AGE): Q. Liang

SHs: P.J.L. Lagoda, G. Viljoen, M.-L. Nguyen, J. Hendrichs and D. Byron



We had the honour to host 13 Rome based FAO permanent mission representatives from 10 countries (Bangladesh (2), China (2), India, Islamic Republic of Iran, Japan, Malaysia, Myanmar, the Philippines (2) and Thailand(2)) at the VIC in Vienna and the FAO/IAEA Agricultural and Biotechnology Laboratories in Seibersdorf. This fruitful visit is one further step in fostering closer coordination and cooperation with the regions in Food and Agriculture, based on strengthened interactions between the IAEA and FAO. For the sake of efficiently and effectively implementing IAEA TC projects, the support of our FAO sister divisions and departments, as well as the regional, sub-regional and national FAO offices is paramount and we hereby acknowledge their invaluable input.

## Announcements

### Conference on Plant & Animal Genome XIX, San Diego, California, USA, 15–19 January 2011

For details, visit:

<http://www.intl-pag.org/>

### Plant Breeding Today: Genomics and Computing Advances Bring Speed and Precision, Washington, D.C., USA, 17–21 February 2011

A series of talks and a discussion meeting. Saturday, February 19, 2011, 10:00 - 11:30 am. Walter E. Washington Convention Center, 147A. AAAS meeting, Washington.

Speakers:

**Molecular approaches speed up plant breeding of medical and developing country crops**

Ian Graham, Centre of Novel Agricultural Products, University of York

### Dissecting the Genetics of Complex Agronomic Traits for Crop Improvement

Edward S. Buckler, Cornell University

Discovery of genes for crop improvement from wild ancestor plants

Susan Rotherford McCouch, Cornell University

For details, visit:

<http://aaas.confex.com/aaas/2011/webprogram/Session2825.html>

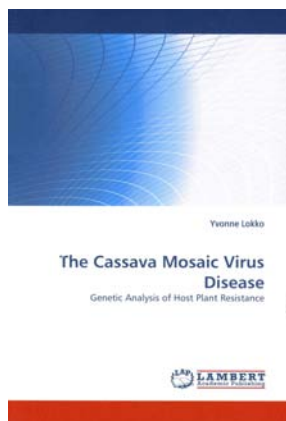
Queries to: Elspeth Bartlet [elspeth.bartlet@york.ac.uk](mailto:elspeth.bartlet@york.ac.uk)

## Publications

### Staff Publication in the Field of Plant Breeding and Genetics

#### The Cassava Mosaic Virus Disease – Genetic Analysis of Host Plant Resistance

Yvonne Lokko



#### Abstract

The cassava mosaic virus disease (CMD) is considered the most devastating disease of cassava in Africa. Annual yield loss on the continent amounts to over 2.2 billion

tonnes of storage root, at a cost of US\$ 440 million. The use of resistance cultivars is the most effective means of control, however, over the years a single source of resistance has been used in breeding and cultivar development. To ensure that durable resistance is maintained within the African cassava germplasm, additional sources of resistance with a wider genetic base are required to diversify resistance to the disease, and allow the accumulation of desirable gene combinations that would be difficult for the pathogens to circumvent in the long term. To efficiently utilise these new sources of resistance in breeding programmes, it is essential to compare the various resistant genotypes with each other to determine if the loci for resistance are similar, and how their effects complement each other to enhance resistance. The study aimed to evaluate the genetic relationships among the various sources of resistance to CMD and identify molecular markers linked to genes for resistance to CMD.

2010

LAP LAMBERT Academic Publishing

ISBN 978-3-8383-7909-8

# Publications within Coordinated Research Projects (CRPs) 2009-2010

## Molecular Tools for Quality Improvement in Vegetatively Propagated Crops including Banana and Cassava (D2.30.27)

Resmi L. and Nair A.S. Differential effect of cytokinins in the micropropagation of diploid (AA & AB) and triploid (AAA & AAB) Musa cultivars from South India submitted in *Biologia Plantarum*.

Smitha P.D. and Nair A.S. (2010) Somatic Embryogenesis and Plant Regeneration in Diploid Banana Cultivars from Kerala Indian J. Plant Genet. Resour. 23(1): 69-72.

Gayathri and Nair A.S. (2010) Isolation purification and characterization of polygalactouranase enzyme from *Musa acuminata* cv. Palayamcodan fruits, Indian Science Congress.

Mohandas H. and Nair A.S. (2010) HPLC analysis of Beta carotene in Banana cultivars from Kerala, Golden Jubilee National Symposium on Plant Diversity, Utilization & Management May 2010.

Bayoumi S.A.L., Rowan M.G., Beeching J.R. and Blagbrough I.S. (2010) *Phytochemistry* 71, 598.

Hřibová E., Neumann P., Matsumoto T., Roux N., Macas J., Doležel J. (2010) Repetitive part of the banana (*Musa acuminata*) genome investigated by low-depth 454 sequencing. – *BMC Plant Biol.* 10: 204.

Hřibová E., Čížková J., Christelová P., Taudien S., de Langhe E. and Doležel J. The ITS1-5.8S-ITS2 sequence region in the Musaceae: structure, diversity and use in molecular phylogeny. – **submitted**

Christelová P., Valárik M., Hřibová E. de Langhe E. and Doležel J. A multi gene sequence-based phylogeny of the Musaceae (banana) family. – **submitted**

James A., Ortiz R., Miller R., Baurens F. and d'Hont A. (2010) Map-Based or Positional Cloning. In: *Encyclopedia of Plant Genomics* (Ed. Prof. Chittaranjan Kole).

Togawa R.C., Mello Romero Santos C., Miller R.N.G., Teixeira Souza Júnior M., Martins N.F. (2010). DATAMusa - a Database for Ortholog Genes from Musa. *Tree and Forestry Science and Biotechnology*.

Miller R.N.G., Passos M.A.N., Emediato F.L., de Camargo Teixeira C., Pappas Júnior G.J. (2010) Candidate Resistance Gene Discovery: Resistance Gene Analog Characterization and Differential Gene Expression Analysis in *Musa-Mycosphaerella* Host-Pathogen Interactions. *Acta Hort.* (ISHS).

Baurens F.C., Bocs S., Rouard M., Matsumoto T., Miller R.N.G., Rodier-Goud M. Mbeguie-a-mbeguie D. and Yahiaoui N. (2010) Mechanisms of haplotype divergence at the RGA08 nucleotide-binding leucine-rich repeat gene locus in wild banana (*Musa balbisiana*). *BMC Plant Biology* 10: 149.

Hippolyte I., Bakry F., Seguin M., Gardes L., Rivallan R. Risterucci A.-M., Jenny C., Perrier X., Carreel F., Argout X., Piffanelli P., Khan I, Miller R.N.G., Pappas Georgios J., Mbeguie-a-mbeguie D., Matsumoto T., De Bernardinis V., Huttner E., Kilian A., Baurens F.C., D'Hont A. Cote F., Courtois B. and Glaszmann J.C. (2010) A saturated SSR/DArT linkage map of *Musa acuminata* addressing genome rearrangements among bananas. *BMC Plant Biology* 10: 65.

Miller R.N.G., Passos M.A.N., Menezes N.N.P., Souza M.T., do Carmo Costa M.M., Rennó Azevedo V.C., Amorim E.P., Pappas G.J. and Ciampi A.Y. (2010) Characterization of novel microsatellite markers in *Musa acuminata* subsp. *burmannicoides*, var. *Calcutta 4*. *BMC Research Notes* 3: 148.

Till B.J., Jankowicz-Cieslak J., Sagi L., Huynh O.A., Utsushi H., Sweenen R., Terauchi R. and Mba C. (2010) Discovery of nucleotide polymorphisms in the *Musa* gene pool by Ecotilling. *Theoretical and Applied Genetics* 121(7): 1381-1389.

## Effects of Mutagenic Agents on the DNA Sequence in Plants (D2.40.11)

Contreras Rojas M., Pérez J.C., Ceballos H., Baena D., Morante N. and Calle F. (2009) Introduction of inbreeding and analysis of inbreeding depression in eight *S<sub>1</sub>* cassava families. *Crop Sci.* 49:543-548.

Rice Starch Quality Research and Utilization. Edited by Wu D.X. and Shu X.L. China Agriculture Publishing House. May 2009.

Sánchez T., Salcedo E., Ceballos H., Dufour D., Mafla G.T., Morante N., Calle F., Pérez J.C., Debouck D., Jaramillo G. and Moreno I.X. (2009) Screening of starch quality traits in cassava (*Manihot esculenta* Crantz). *Starch/Stärke* 61:12-19.

van der Vyver C. (2009) Submitted paper: 'RDA, RAPD and SSR analysis of radiation-induced genome alterations in *Vigna unguiculata*', *Annals of Botany*.



Wang M., Caetano-Anollés G. (2009) The evolutionary mechanics of domain organization in proteomes and the rise of modularity in the protein world. *Structure* 17:66-78.

#### **In press:**

Dimitrova A., Todorovska E., Christov N., Stoilov L., Atanasov A. and Gecheff K.. Molecular characterization of ionizing radiation-induced mutational alterations in barley. *Genetics and Breeding*.

Gecheff K., Manova V., Bonchev G., Kitanova M., Vlahova M. and Stoilov L. Position-specific effects in the action of mutagenic agents on the chromosomes of barley (*Hordeum vulgare* L.) *Genetics and Breeding*.

Sanchez, T., A. Rosero, A.P. Tofiño, K. Denyer, A. Smith, H. Ceballos, N. Morante, and J.C. Pérez. (2009). Induction and identification of useful mutations for root quality traits in cassava. *FAO/IAEA International Symposium on Induced Mutations in Plants*. 12-15 August, Vienna, Austria.

#### **Pyramiding of Mutated Genes Contributing to Crop Quality and Resistance to Stress Affecting Quality (D2.30.25)**

Chen H., Qian N., Guo W.Z., Song Q.P., Li B.C., Deng F.J., Dong C.G. and Zhang T.Z. (2009) Using three overlapped RILs to dissect genetically clustered QTL for fiber strength on chro.24 in Upland cotton. *Theor Appl Genet*. 119: 605-612.

Song X.L. and Zhang T.Z. (2009) Identification of Quantitative Trait Loci Controlling Plant Architectural Traits in Cotton. *Plant Sci*. 177: 317-323.

Wu C.T., Zhou B.L. and Zhang T.Z. (2009) Isolation and characterization of a sterile dwarf mutant in Asian cotton

(*Gossypium arboreum* L.). *J Genetics & Genomics* 36: 343-353.

Ivanova B. and Bojinov B. (2009). Identification of QTLs for fiber quality in a Bulgarian cotton breeding collection. *Genetics and Breeding (Blg.)*, accepted.

Mondal S. and Badigannavar A.M. (2009). Molecular diversity and association of SSR markers to rust and late leaf spot resistance in cultivated groundnut (*Arachis hypogaea* L.). *Plant Breeding*. DOI:10.1111/j.1439-0523.2009.01635.x.

Badigannavar A.M., Mondal S. and D'Souza S.F. (2009) Success story of radiation based induced mutagenesis in groundnut (*Arachis hypogaea* L.). *International Conference on Peaceful Uses of Atomic Energy*, Sept 29 – Oct 1, 2009, New Delhi. Vol .II Pp: 544-545.

Do Khac Thinh, Nguyen Thi Cuc, Hung Phi Oanh & Dao Minh So et al, (2009) Mutation Breeding and Socio-economic Effectiveness in Southeastern & Highlands 2009, *Proceedings of National Conference of Vietnam on Application of Nuclear Techniques*, organized, 20-22<sup>th</sup> August, 2009, Nha Trang, Vietnam.

Guzy-Wrobelska J., Nawrot M., Janiak A., Adamska K., Matyszczyk I. and Szarejko I. (2009) Molecular mapping of genes responsible for root architecture in barley. The 4th Conference of Polish Society of Experimental Plant Biology. *Experimental Plant Biology. Why not?!* 21-25 September, 2009 Kraków.

Park H.H., *et al.*, (2009). Analyzing seed protein in a high lysine barley mutant using proteomics approach. *Spring Annual Meeting of The Korea Society of Crop Science*, pp. 133.

# List of Plant Breeding and Genetics Section Publications

## Plant Mutation Reports

Year	Edition	Contents (a sampling of the papers are listed below):	Reference No.
2010	Vol.2, No. 2	<ul style="list-style-type: none"> <li>• Mutation breeding in Bulgaria</li> <li>• Mutagenesis in clusterbean</li> <li>• Doubled haploid durum wheat</li> <li>• Improvement of indigenous rice</li> <li>• Mutant variety of groundnut</li> <li>• Gamma phytotron</li> <li>• Gamma greenhouse</li> <li>• Gamma field</li> </ul>	ISSN 1011-260X
2008	Vol. 2, No. 1	<ul style="list-style-type: none"> <li>• Induced genetic variability in kacholam</li> <li>• Mutagenesis of guar</li> <li>• Cocoyam radiation sensitivity</li> <li>• Virus resistant rice variety</li> <li>• Cold tolerant mutant rice</li> <li>• Proton radiation</li> <li>• Tomato adapted to low water supply</li> <li>• Increasing crossability of mungbean</li> </ul>	ISSN 1011-260X
2007	Vol. 1, No. 3	<ul style="list-style-type: none"> <li>• Mutation breeding and genetics in Korea</li> <li>• Genetic enhancement of groundnut</li> <li>• Virus resistant banana</li> <li>• Ion beams implantation on wheat</li> <li>• Trombay mutant groundnut varieties</li> <li>• Lodging tolerant rice variety</li> </ul>	ISSN 1011-260X
2006	Vol. 1, No. 2	<ul style="list-style-type: none"> <li>• 30 years rice mutation breeding and genetics</li> <li>• Mutant groundnut varieties in Bangladesh</li> <li>• Shortening durum wheat plants</li> <li>• Seedless mutant sweet orange</li> <li>• Colorful chrysanthemum mutations</li> <li>• Radiosensitivity of cassava <i>in vitro</i> culture</li> </ul>	ISSN 1011-260X
2006	Vol. 1, No. 1	<ul style="list-style-type: none"> <li>• Rice mutation breeding in China</li> <li>• Long grain aromatic rices and induced mutations</li> <li>• Significant contribution of mutation techniques to rice breeding in Indonesia</li> <li>• Use of induced mutants in rice breeding in Japan</li> <li>• Katy deletion mutant populations</li> <li>• Rice mutation breeding in Vietnam</li> </ul>	ISSN 1011-260X

## Mutation Breeding Newsletter and Reviews

Year	Edition	Contents (a sampling of the papers are listed below):	Reference No.
2005	No. 1	<ul style="list-style-type: none"> <li>• High yielding mutants in cotton</li> <li>• Drought resistant tomato</li> <li>• Groundnut resistant to foliar diseases</li> <li>• Lodging resistant glutinous rice</li> <li>• First ever oilseed mustard mutant</li> </ul>	ISSN 1011-260X

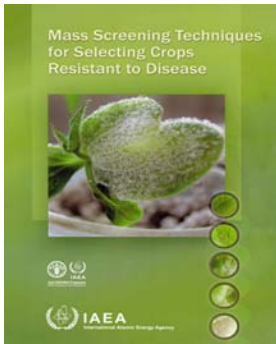
## Mutation Breeding Review (published until 2004)

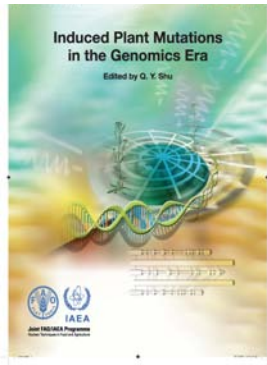
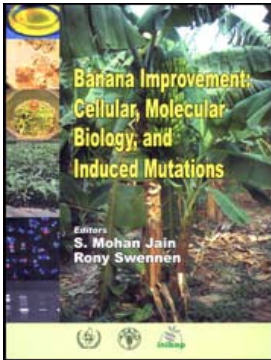
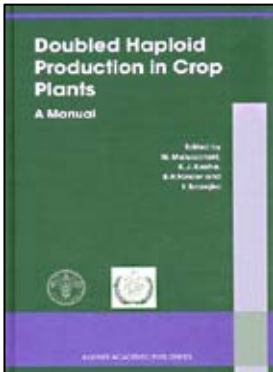
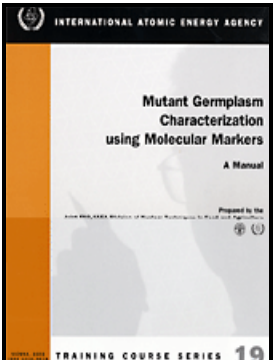
Year	Edition	Title	Reference No.
2004	No. 14	Officially released mutant varieties in China	ISSN 1011-2618
2001	No. 13	Grain legume cultivars derived from induced mutations, and mutations altering fatty acid composition	ISSN 1011-2618
2000	No. 12	Officially released mutant varieties – The FAO/IAEA database	ISSN 1011-2618
1999	No. 11	Oilseed cultivars developed from induced mutations and mutations altering fatty acid composition	ISSN 1011-2618

## Mutation Breeding Newsletter (published until 2003)

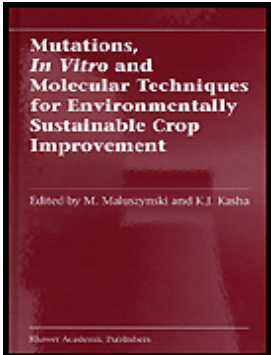
Year	Edition	Title	Reference No.
2003	No. 46	Index Issue No. 21-44	ISSN 1011-260X
2001	No. 45	Issue No. 45	ISSN 1011-260X
1999	No. 44	Issue No. 44	ISSN 1011-260X

## Books

Year	Edition	Title	Book Cover	Reference No.
2010		Mass Screening Techniques for Selecting Crops Resistant to Disease		ISBN 978-92-0-105110-3

Year	Edition	Title	Book Cover	Reference No.
2009		Induced Plant Mutations in the Genomics Era		ISBN 978-92-5-106324-9
2004		Banana Improvement: Cellular, Molecular Biology, and Induced Mutations		ISBN 1-57808-340-0
2003		Doubled Haploid Production in Crop Plants – A Manual		ISBN 1-4020-1544-5
2002	Training Course Series No. 19	Mutant Germplasm Characterization using Molecular Markers – A Manual		ISSN 1018-5518



Year	Edition	Title	Book Cover	Reference No.
2002		Mutations, <i>In Vitro</i> and Molecular Techniques for Environmentally Sustainable Crop Improvement		ISBN 1-4020-0602-0

## Technical Publications

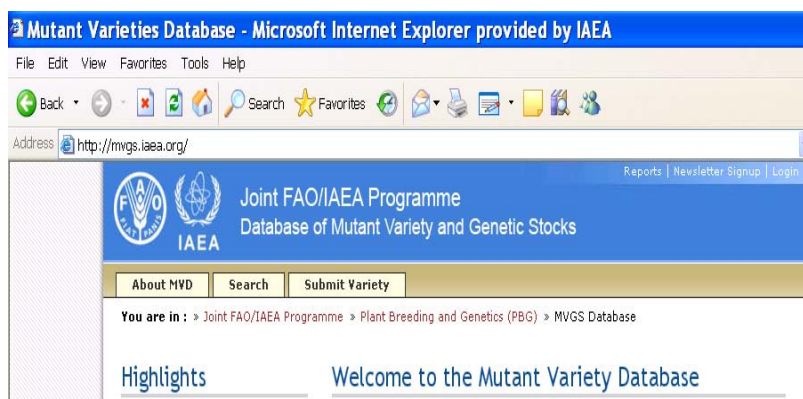
Year	Type of Publication	Title	Reference No.
2009	IAEA-TECDOC-1615	Induced mutation in tropical fruit trees	ISBN 978-92-0-1027-09-2
2006	IAEA-TECDOC-1493	Mutational analysis of root characters in food plants	ISBN 92-0-103106-8 ISSN 1011-4289
2004	IAEA-TECDOC-1384	Low cost options for tissue culture technology in developing countries	ISBN 92-0-115903-X ISSN 1011-4289
2004	IAEA-TECDOC-1426	Genetic improvement of under-utilized and neglected crops in low income food deficit countries through irradiation and related techniques	ISBN 92-0-113604-8 ISSN 1011-4289
2003	IAEA-TECDOC-1369	Improvement of new and traditional industrial crops by induced mutations and related biotechnology	ISBN 92-0-101603-4 ISSN 1011-4289
2001	IAEA-TECDOC-1195	Sesame improvement by induced mutations	ISSN 1011-4289
2001	IAEA-TECDOC-1216	Induced mutations in connection with biotechnology for crop improvement in Latin America	ISSN 1011-4289
2001	IAEA-TECDOC-1227	<i>In vitro</i> techniques for selection of radiation induced mutations adapted to adverse environmental conditions	ISSN 1011-4289
2001	IAEA-TECDOC-1253	Radioactively labeled DNA probes for crop improvement	ISSN 1011-4289
1998	IAEA-TECDOC-1010	Application of DNA based marker mutations for improvement of cereals and other sexually reproduced crop plants	ISSN 1011-4289
1998	IAEA-TECDOC-1047	Use of novel DNA fingerprinting techniques for the detection and characterization of genetic variation in vegetatively propagated crops	ISSN 1011-4289
1997	IAEA-TECDOC-951	Improvement of basic food crops in Africa through plant breeding, including the use of induced mutations	ISSN 1011-4289
1996	IAEA-TECDOC-859	Use of mutation techniques for improvement of cereals in Latin America	ISSN 1011-4289
1995	IAEA-TECDOC-800	<i>In vitro</i> mutation breeding of banana and plantains	ISSN 1011-4289
1995	IAEA-TECDOC-809	Improvement of root and tuber crops in tropical countries of Asia by induced mutations	ISSN 1011-4289
1994	IAEA-TECDOC-781	Mutation breeding of oil seed crops	ISSN 1011-4289

For details on IAEA Publications, visit: <http://www-pub.iaea.org/MTCD/publications/publications.asp>

# New FAO/IAEA Database of Mutant Varieties and Genetic Stocks

Welcome to our new FAO/IAEA Database of Mutant Varieties and Genetic Stocks! At the moment, we just completed construction of the part for Mutant Variety Database, which is still in the process of information updating. We will add the other part for Mutant Genetic Stocks in due time. The new database has improved over

the FAO/IAEA Mutant Variety Database in many ways. We are working to make the new database as the global information source of mutant varieties and mutant genetic stocks, as well as activities and events related to plant mutation breeding and research.

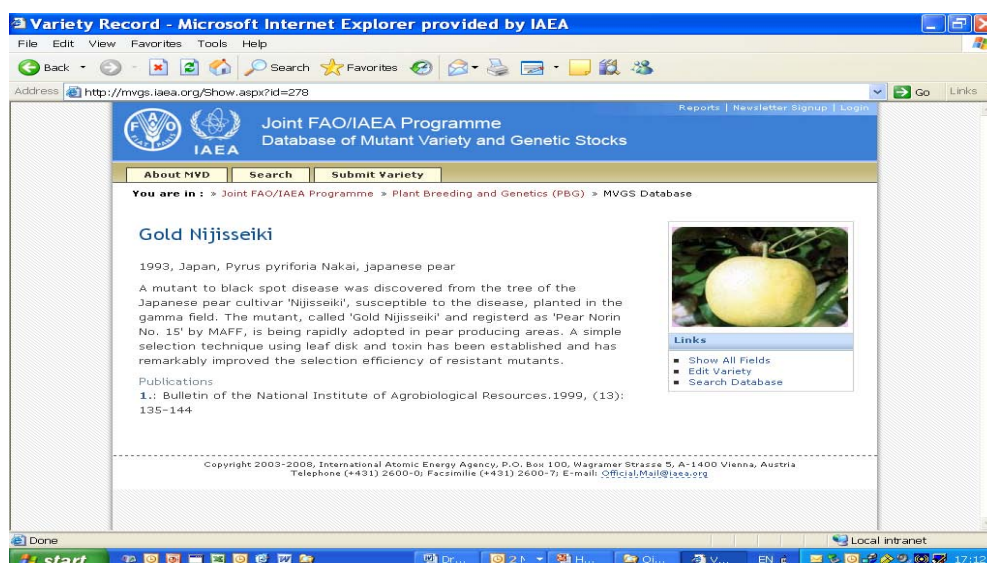


The key feature of the database is that you can register your mutant varieties from your desktop. For this purpose, you need first register an account; then you will be authorized to submit or edit a mutant variety.

We would greatly appreciate your support by registering your mutant variety in our database. Once the variety is registered, it will have its own 'homepage' (see below). Therefore, you can use it as an important platform to

showcase your new varieties (The introduction of this variety may be shown in local language).

Please visit the website <http://mvgs.iaea.org> and send us your valuable suggestions and comments regarding the structure and content of this database. Please also send us other information, related to plant mutation breeding and mutant varieties, genetic stocks; we may post them on the website.



**YOU MAY STILL SEND US INFORMATION ON YOUR MUTANT VARIETY AND WE WILL UPLOAD THEM INTO THE SYSTEM, IF IT IS DIFFICULT FOR YOU TO DO SO.**



# Plant Mutation Reports

## Author's Guidelines for Manuscript Submission

### Scope

Plant Mutation Reports (PMR) publishes (mini) reviews, short communications and complete research papers in all areas of plant mutation research which focuses on mutagenesis, phenotyping and genotyping characterization, of mutant populations and the application of mutation breeding and biotechnology in crop improvement. It also publishes description papers on mutant germplasm and mutant varieties. Papers on social-economic impact analysis of induced mutations and mutant varieties are also accepted.

### Style

The manuscript should be accurately and concisely written in English with the following sections:

#### Title page

- Title: The title should be brief and informative 10 to 12 words (excluding 'and,' 'of,' and similar conjunctions and prepositions). As much as possible use common names for crops and avoid abbreviations.
- Authors: The names of all authors should follow the title line initials of given names followed by full family name. Place an asterisk (\*) after the name of the corresponding author (i.e. the person from whom reprints are to be requested). If authors are from different institutions, indicate institutional affiliation with numbers in *superscript font* (<sup>1,2...</sup>).
- Affiliation(s)/Address(es):
- Email address: Provide the corresponding author's email address.

#### Abstract and keywords

Provide brief and informative paragraph summarizing the content of article on the second manuscript page. The abstract not exceed 150 words. Do not cite references. Each paper should have 3-5 keywords.

#### Main text

- The main text should follow the title page and abstract.
- Review articles may be organized according to their specific requirements.

- Research articles should be arranged as in the following order: Introduction (which includes the literature review), Materials and Methods, Results, Discussion, Conclusions (optional), Acknowledgments (optional), and References, followed by any Figure captions and then Tables. Use the 'Title Case' for each section.
- Results and Discussion may be combined and conclusions can be given at the close of the Discussion section.
- Start each section (including figure captions and tables) on a new page and number all pages.
- New mutant germplasm should include a short description of initial material used and the mutagen and doses applied; selection process; mutated characteristics and its genetic and agronomic analysis. Description of mutant variety should, in addition, include its performance in yield trials for varietal release and the releasing committee, when applicable and proof of entry in the MVGS (<http://mvgs.iaea.org/>).

#### Acknowledgements

- Acknowledgements of grants, support etc, should follow the text and precede the references.

#### References

The literature should be cited using the author-year notation system, either as John (1990) for single author paper, John and Johnson (2000) for papers with two authors, or John *et al.* (2000) for papers with more than two authors throughout the text. In the list of references, arrange alphabetically by author following the style shown below:

- Periodicals: Shamsuzzaman K.M. and Shaikh M.A.Q. (1991) Early maturing and high seed yielding chickpea mutant. *Mut Breed Newslett* 37: 4-5.
- Books (edited by someone other than author of article): Maluszynski M. (1990) Gene manipulation in plant improvement. In: Gustafsson J.P. (ed), *Induced Mutations in Plant Improvement*. Plenum press, New York. Pp239-250.
- Books (identical author and editor) van Harten A.M. (1998) *Mutation Breeding, Theory and Practice*. Cambridge University Press, Cambridge, U.K. pp. 237-240.



All single author entries should precede multiple-author entries for the same first author. Use chronological order only within entries with identical authorship (alphabetizing by title for same-author, same-year entries). Add a lowercase letter a, b, c, etc. to the year to identify same-year entries for text citation.

### Figures

- Figures, e.g. photographs, graphs and diagrams should be referred to as 'Fig.' numbered consecutively (1, 2, etc.)
- Submit figures in high-resolution, individual files (one figure per file) and identify each file accordingly.
- A figure caption should be brief, but informative. All figure caption should be placed at the end of the main text.
- Identify curves, symbols, or structures with a legend within the figure itself, not in the caption. Define abbreviations in the caption and define symbols used in the caption or in the legend.
- Indicate the scale for micrographs, either in the illustration or the caption.

### Tables

- All tables should be prepared with the 'Tables' feature in your word processor, (do not use tabs, spaces, or graphics boxes) and must be numbered consecutively with brief headers explaining the content of the table. Use footnotes for detailed explanation of the tables.

Each datum should be in an individual cell. Define all variables and spell out all abbreviations. Tables should be placed at the end of the main text document on with each table on a separate page.

- The \*, \*\*, and \*\*\* are always used in this order to show statistical significance at the 0.05, 0.01, and 0.001 probability levels, respectively, and cannot be used for other notes. Significance at other levels is designated by a supplemental note. Lack of significance is usually indicated by NS.
- Footnote should be placed immediately below the table. The footnotes should be identified by symbols in this order: †, ‡, §, ¶, #, ††,
- Do not use boxes; use horizontal lines only. Figures and tables should be placed on separate pages.

### Abbreviations

All abbreviations should be fully defined when first mentioned in abstract and also in the main text, and then the abbreviation may subsequently be used

### Nomenclature and identification of materials.

Give the complete binomial and authorities at first mention (in Abstract or text) of plants, pathogens, and insects

### Units and symbols

The standard SI units (Système International de Unités) and symbols should be used throughout ([www.scenta.co.uk/tcaep/science/siunit/index.htm](http://www.scenta.co.uk/tcaep/science/siunit/index.htm)).

## Impressum

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