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Nuclear Techniques in Food and Agriculture

Plant Breeding & Genetics Newsletter

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To Our Readers

Pierre Jean Laurent Lagoda (15 June 1961 – 16 June 2016).

Dear Colleagues,

It is with great sadness that we announce the death of Pierre Jean Laurent Lagoda. Pierre began work in the Plant Breeding Section in 2002 originally as a Technical Officer, but rose to become Head of the Section in 2004. From the onset he was a fierce and tireless champion for all FAO/IAEA programmes in plant mutation breeding and passed on his boundless energy in motivating his staff and colleagues. Pierre died in hospital after a long battle with illness. From his hospital bed he continued to support and drive his team forward in their various plant breeding activities with Member States and aimed to be out of hospital as soon as possible, sadly this was not to be. His funeral (1st of July Feuerhalle, Vienna) captured what was typical and well-loved of Pierre, there was a boisterous, rousing march, and poignantly his daughter (11 years old) played Beethoven's 'Ode to Joy' on her trumpet. And it is with joy that we will remember Pierre the most. We shall miss him greatly, as our section head and as a friend. Our condolences and thoughts go out to his family, especially his wife, Helena and daughter, Vivienne.

REST IN PEACE

Public Event in the Vienna International Centre (VIC): Long Night of Research, 22 April 2016.

On 22 April 2016 the Long Night of Research took place at the Vienna International Centre (VIC) for the seventh time across Austria in order to promote science and research in particular amongst the younger generation. More than 1300 external visitors were welcomed by nuclear scientists and had the chance to learn about the variety of nuclear applications in various fields and how nuclear science and technology are used to improve lives around the world. At this event, many activities of the Joint FAO/IAEA Division were highlighted, with emphasis on Laboratories. Plant Breeding and Genetics Laboratory (PBGL) booth was visited by many people. The visitors were particularly interested in learning about the mutation breeding for crop improvement, the broad usage of mutant crops in our daily food and the high socioeconomic impacts of many mutant varieties. They had a unique chance for hands-on experience and discussion with the scientists.

The mission of the Joint FAO/IAEA Division is continually evolving to address novel challenges in MSs and nuclear applications continue to provide added value to conventional approaches in addressing a range of agricultural problems and issues. Member States are increasingly concerned with climate change and have expressed their support in responding to emerging challenges related to transboundary plant diseases.

The PBG subprogramme is pleased to announce that this year a new project on Capacity Building for Latin American Countries against Coffee Leaf Rust was initiated sponsored by a grant of the OPIC Fund for International Development (OFID). In partnership with OFID, the project will be implemented from Plant Breeding and Genetics Subprogramme of the Joint FAO/IAEA Division. A technical workshop was held from 4 to 8 April 2016 with participation of coffee scientists from Costa Rica, El Salvador, Guatemala, Honduras, Nicaragua, Peru and experts from World Coffee Research, Portugal and Austria. The one-week technical workshop resulted in the establishment of network for collaboration on using mutation breeding in coffee to fight against coffee leaf rust and defined technical and scientific approaches among participating countries from Latin America. (please see page 14).

I would like to inform you that a new Coordinated Research Project (CRP D2.50.05) on Mutation Breeding for Resistance to *Striga* Parasitic Weed in Cereals for Food Security will hold its first Research Coordination Meeting (RCM) in October 2016, and for which we are encouraging submission of relevant research contract proposals. Information on how to apply for research contracts and research agreements can be found at http://www-naweb.iaea.org/nafa/pbg/crp/coordinated-research-pbg.html. The aim of this CRP will be to support the generation of novel sources of variation, using mutation breeding, by developing efficient screening protocols for *Striga* resistance in cereals for research capacity building and improvement of food security in Member States.

In response to Member States' requests to consider the possibilities of developing and applying mutation breeding techniques on tolerance to abiotic stress in crops, we will be holding a Consultants Meeting to develop a new Coordinated Research Project on Improving Crop Resistance to Abiotic Stresses through Mutation Breeding for Sustainable Agriculture. The project objectives will be to increase the efficiency of mutation screening techniques for crop breeding and genetic research through the generation of resources and development of technology packages which can be transferred to Member States.

I am very glad to report that we concluded the 2014–2015 cycle of the IAEA Technical Cooperation Programme with a number of technical cooperation projects supporting our Member States that reached their expected outcomes. In 2016, we are starting the new 2016–2017 cycle with new projects that have been developed together with counterparts in our Member States and approved by the IAEA Board of Governors (please see list of TC project on page 25).

About the R&D at the Plant Breeding and Genetics Laboratory (PBGL) in Seibersdorf, we can report new Protocol for Pre-Field Screening of Mutants for Salt Tolerance in Rice, Wheat and Barley (available on http://www.springer.com/us/book/9783319265889). In-house research at the FAO/IAEA Plant Breeding and Genetics Laboratory is focused on molecular discovery of induced mutations and optimizing mutation induction protocols using x-ray and ion beam as alternative irradiation techniques to gamma rays (see Developments at the Plant Breeding and Genetics Laboratory on page 29).

I would like to draw your attention that 2016 is the International Year of Pulses (IYP 2016). The IYP 2016 aims to heighten public awareness of the nutritional benefits of pulses as part of sustainable food production aimed towards food security and nutrition. Plant Breeding and Genetics subprogramme jointly with Soil and Water Management and Crop Nutrition subprogramme will organize Side Event on Enhancing Pulses for Food Security by Nuclear Applications during 60th Regular Session of the IAEA General Conference in Vienna, Austria from 26 to 30 September 2016.

Finally, I would like to thank all our collaborators and counterparts for their support and significant inputs to our joint activities. Also, I would like to express my appreciation to the staff of the Plant Breeding and Genetics Section at the IAEA's Headquarters in Vienna as well as the staff of the Plant Breeding and Genetics Laboratory in Seibersdorf, Austria for their dedication and competence in support to the Member States (MSs) by developing and transferring sustainable plant mutation breeding techniques to them.

Ljupcho Jankuloski
Acting Head
Plant Breeding and Genetics Section

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[2] Separated in April 2016
[3] Separated in May 2016
[4] Separated in March 2016

In Memoriam

Pierre J.L. Lagoda (15 June 1961 – 16 June 2016).

On 16 June 2016 Pierre Jean Laurent Lagoda, Head of the Plant Breeding and Genetics Section, passed away at the age of 55. Pierre was ill for a long time but his death was unexpected and deeply shocked us. This is a very big loss for his family and for all of us.

Pierre was an excellent scientist and a very fine human being.

Pierre, originally from Luxembourg, started his scientific career with research in molecular oncology, a field in which he received his doctor's degree in 1990 at Saarland University, Germany. After his PhD he went to the University of Michigan for a Postdoc fellowship. Open minded as he was, Pierre shifted to banana research, motivated by reading an article on properties of the banana peel to fight against cancer. In 1992 he found out that the French Agricultural Research and International Cooperation Organization (CIRAD) was looking for a molecular biologist to work on banana research, which led to the beginning of his career in banana genetics research at CIRAD, Montpellier. For the next 10 years, he led the team that developed the first Musa genetic map and discovered the wild ancestors of the most popular bananas. He also introduced the use of microsatellite markers in banana, of which he was one of the first specialists in the world. Out of the 22 papers with high Impact Factor, published throughout this collaboration, 'Microsatellites, from molecules to populations and back' has been cited more than 800 times. This gave Pierre lasting international stature in the field of plant genetics. When he left CIRAD, he left his mark on an area of major technological and scientific developments. Pierre was instrumental in the establishment of the Global Musa Genomics Consortium (GMGC), now the Genomics Thematic Group of MusaNet.

In 2002, Pierre moved to the International Atomic Energy Agency (IAEA) in Vienna. After two years he was appointed the head of the Plant Breeding and Genetics Section of the Joint FAO/IAEA Division. Here he was a tireless supporter of the banana work in counterpart institutes and at the PBG's laboratory in Seibersdorf. He also worked as a Technical Officer providing strong support to mutation breeding projects in many different countries on a variety of crops. One of his major recent achievements was the initiation and technical backstopping of an Interregional Technical Cooperation project on Responding to the Transboundary Threat of Wheat Black Stem Rust (Ug99), which resulted in release of UG99 resistant wheat mutant varieties in Kenya within five years after seeds have been irradiated in Seibersdorf. In recognition of this success, Pierre and his team in November 2014 received the IAEA Superior Achievement Award, the highest Agency award given by the Director General.

Pierre was famous for his way of explaining the concept of mutation breeding to a broad audience. On stage at the 2012 IAEA scientific forum on 'Food for the Future: Meeting the Challenges with Nuclear Applications' he took out from his pocket four dice, rolled them onto the table and by this got the idea of the random nature of mutations creating genetic diversity across to everybody. The dice were a kind of trademark of Pierre. He also used them earlier in an interview with the New York Times, which became widely noted and often cited. Up to now journalists are contacting us referring to this article.

Pierre never shied away from leaving the ivory tower of science in order to make knowledge accessible to others and to share his enthusiasm. He enjoyed and was very good

in explaining seemingly complicated facts in a simple, straightforward manner. Unforgotten is his self-made thermocycler he used in our interregional training course on molecular markers to visualize the principle of PCR. By means of three heatable water baths and a crane made from Fischertechnik, being controlled automatically by an old Commodore computer he impressively demonstrated the core of the matter. A little later his machine was incorporated in an exhibition at the German Museum in Munich.

His vision and outstanding contribution to the development of plant biotechnology methods to understanding of crop diversity and improvement will long be remembered.

Pierre was not only widely recognized for his scientific work, but also for his passion and idealism, always striving for the benefit of humanity. He never hesitated to help scientists around the world to fight against hunger and for the improved livelihoods of those in need.

We always will bear in remembrance Pierre as an extremely competent, dedicated and warm-hearted boss, colleague and friend.

The Plant Breeding and Genetics team
and Nicolas Roux (Bioversity International)

Staff News

Laboratory Head

I have the pleasure to introduce our new Laboratory Head, Mr Ivan Ingelbrecht.

Mr Ingelbrecht was born in Belgium and holds a BSc in biotechnology and obtained a PhD in plant molecular biology, both from the Faculty of Sciences at Ghent University, Belgium. His PhD research contributed to uncovering transgene silencing phenomena in plants, nowadays recognized as an epigenetic mechanism involved in gene regulation and pathogen defence responses.

Mr Ingelbrecht brings with him extensive R&D experience in applied agricultural sciences having worked at the interface of plant biotechnology and plant breeding for over 20 years, the majority in an international agricultural research setting, the CGIAR (Consultative Group of International Agricultural Research). Mr Ingelbrecht is former Head of the Central Biotechnology Laboratory at the International Institute of Tropical Agriculture (IITA) in Ibadan, Nigeria where he lived and worked for over 10 years. At IITA, Mr Ingelbrecht also served as Coordinator of the Program on Agricultural Biotechnology, Plant Breeding and Plant Genetic Resources and was an active member of IITA's Research-for-Development Council mandated to support R&D strategic development for the Institute. In these capacities, Mr Ingelbrecht acquired extensive experience in R&D project, personnel and also laboratory management. As a Postdoctoral Researcher and later Senior Biotechnologist at IITA, he has worked on a range of tropical crops including cassava, banana/plantain, maize, and cowpea. As a Research Associate at Texas A&M (USA) he was involved in the genetic improvement of sugarcane and grapefruit using *in vitro* tissue culture and transgenesis approaches and molecular diagnostics. At IITA and Texas A&M, Mr Ingelbrecht has supervised many degree and non-degree students at the graduate and post-graduate level in *in vitro* tissue culture, molecular biology and molecular genetics tools for crop genetic diversity assessment, molecular marker development and also diagnostics. He has worked in the private sector as Intellectual Property Rights Liaison and has a sound understanding of Intellectual Property Rights as they relate to plant genetic resources.

If you want to contact Ivan, please feel free to use the following electronic address: I.Ingelbrecht@iaea.org.

Consultant

I have the pleasure to introduce Mr Brian Peter Forster who will accompany the Plant Breeding and Genetics subprogramme as a consultant for two months.

Mr Foster served three years at the IAEA as the Head of Plant Breeding and Genetics Laboratory.

Mr Foster obtained his BSc in Agricultural Botany from the University of Leeds, England, UK and his PhD in Agricultural Botany from the University of Edinburgh, Scotland, UK. He was employed for more than 20 years by the Scottish Crop Research Institute (SCRI) in various positions with increasing responsibilities as plant breeder/geneticist, interrupted for two times one year as sabbatical leave at the Svaloev Weibull AB (International Seed and Breeding Company), Svaloev, Sweden and as a consultant at the FAO/IAEA Plant Breeding and Genetics Laboratory, Seibersdorf, Austria. Mr Foster currently works for PT Timbang Deli Indonesia Medan, South Sumatra. Brian works closely with plant breeders and has expertise in high throughput trait selection methods, plant reproductive biology, tissue culture and DNA marker diagnostics. He is knowledgeable about temperate and tropical cereals (wheat, barley, rye and rice, including landraces and wild relatives), rapeseed (Europe and North America) and tropical plantation crops (oil palm, rubber, cocoa and banana). He has published extensively in high standing peer reviewed journals.

Farewell

It is time to say goodbye to our dear friend and colleague Andreas Draganitsch, who served Plant Breeding & Genetics Laboratory for 32 years. It is impossible to summarize how important he has been for our team. We thank him for making sure the lab ran smoothly and for sharing his knowledge and experiences in plant tissue culture with countless trainees. Every single banana plant which was distributed at the General Conference or at a special

event such as Long Night of Research went through Andi's magical hands. You can feel his presence as you visit the many offices in Seibersdorf and Vienna where there is a happy banana plant growing in a pot. Andi served with dedication and hard work. We will surely miss him! It is not all sad news because the reason Andi is leaving is that he is retiring. A loss for us is therefore a gain for his friends and family and also the forests and mountains that he now has more time to visit. We wish him all the best.

Au revoir!

Our colleague and dear friend Mr Souleymane Bado left the Agency in April 2016 after eight years of hard work

and dedicated service. With a passion for *in vitro* plant tissue culture, Bado together with Andreas formed the backbone of the *in vitro* tissue culture activities at PBGL during many years. In November 2014 he was a recipient of the 'Superior Achievement One-House Award – Rapid Response in Protecting Wheat from Ug99', together with other PBGL team members, a testimony to his many contributions to the PBGL. We will miss his fine work ethic and good nature and wish him all the best!

Forthcoming Events

Third Research Coordination Meeting (RCM) on Integrated Utilization of Cereal Mutant Varieties in Crop/Livestock Production Systems for Climate Smart Agriculture, Darkhan, Mongolia, 1–5 August 2016

Technical Officer: L. Jankuloski

Chief Scientific Investigators (CSIs) from participating countries (Austria, People's Republic of China, Indonesia, Kuwait, the Former Yugoslav Republic of Macedonia, Malaysia, Mongolia, Peru and UK) will present progress made during the last two years, since the second RCM held in Bogor, Indonesia in 2014.

The major objectives of this CRP are to maximize the yields of mutant varieties. The crops chosen are those that can be used for both human food and animal fodder. The project involves eight participating countries and four major crops, namely wheat (the Former Yugoslav Republic of Macedonia, Mongolia), rice (People's Republic of China, Malaysia), barley (Austria, People's Republic of China, Kuwait and Peru) and sorghum (Indonesia). The project aims to improve the agronomy of the crop especially in respect to soil and water management, improve nutritional value and improve the knowledge and skills base of participating Member States.

This CRP began in the third quarter of 2012 and will be concluded in the third quarter of 2017. The first RCM took place in Vienna in December 2012 from which short term objectives were established. These included the identification of mutant varieties or advanced mutant lines for food and feed. The Second RCM was held in Bogor, Indonesia in August 2014. The Second RCM provided an opportunity to assess progress across all participating countries and to move the programme forward. Indonesia has made significant progress in this CRP as the counterpart has identified advanced mutant lines and mutant varieties of sorghum that performed well in various locations.

Regional ARCAL Training Course on Basic Mutation Breeding, RLA/5/068, Havana, Cuba, 12–16 September 2016

Technical Officer: S. Nielen

In second half of 2016 two Regional training courses related to TC project RLA/5/068 on Improving Yield and Commercial Potential of Crops of Economic Importance (ARCAL CL) will be organized.

The first course in on Basic Mutation Breeding and will take place at the Instituto Nacional de Ciencias Agrícolas (INCA), Havanna, Cuba. The purpose of the training is to provide to new participants of the project basic knowledge and skills in the use of mutation induction in crop improvement programs. The course will cover mutation induction and *in vitro* and *in vivo* selection methods in both sexual and vegetatively propagates crops. Target traits to be discussed in the course include agronomic performance, quality characters and tolerance to biotic and abiotic stress. Also statistical methods applied in mutation breeding will be part of the course.

Regional Training Course on Handling of Mutation Rice Production in Selection through Advanced Marker Added Technique on Biotic and Abiotic Traits, RAS/5/073, Mymensingh, Bangladesh, 2–11 October 2016

Technical Officer: L. Jankuloski

The regional training course will be held under the TC project RAS/5/073 on Supporting Climate-Proofing Rice Production Systems (CRiPS) Based on Nuclear Applications-Phase II. This training course will be organized by the International Atomic Energy Agency in cooperation with the Government of Bangladesh through Bangladesh Institute of Nuclear Agriculture.

Participants will be plant breeders and geneticists from participating Member States involved in project RAS/5/073 (Bangladesh, Cambodia, People's Republic of China, Indonesia, People's Democratic Republic of Lao, Malaysia, Mongolia, Myanmar, Nepal, the Philippines, Thailand and Viet Nam).

The negative effects of climate variability and change such as flood, drought, heat and salinity are major constraints affecting sustainable agricultural productivity globally Rice farming in Asian countries suffers from low and fluctuating productivity. With increasing climate change and variability, it is important to select and evaluate improved rice varieties with enhanced adaptability to harsh conditions of high temperatures, drought, low and erratic rainfall, soil salinity, acidity or low nutrient availability, as well as resistant to diseases and pests.

In this context, mutation induction technique has shown potential as a valuable tool in developing improved crop cultivars tolerant to abiotic and biotic stresses. The use of mutation induction for creating useful new germplasm and developing new cultivars is a profitable approach to crop improvement.

The training course will include lectures, demonstrations (laboratory, greenhouse and field), and practical exercises on plant mutation breeding and related biotechnologies applied for abiotic and biotic stress tolerance breeding, particularly salinity, submergence and drought tolerance.

First RCM on Mutation Breeding for Resistance to the Parasitic Weed (*Striga* spp) in Cereal Crops, D2.50.02, Vienna, Austria, 10–14 October 2016

Technical Officer: A.M.A. Ghanim

Striga parasitic weeds are major constraints to cereal and leguminous crop production on a global scale, especially in Africa, Asia and Australia. Losses in crop yield are reported to be over 40% and could reach 100% during heavy infestation, especially in low-input farming in nutrient poor and drought stricken areas. Mutation breeding addresses this new threat by facilitating the development of new varieties resistant to *Striga*. The major objective of the CRP is to develop technology packages and protocols for the efficient screening of mutant populations of major cereals for resistance to *Striga*.

The meeting objectives are to review and consolidate the work plan, build the networking of the team and coordinate the work to maximize the use of resources to achieve the targeted objectives.

It is expected to have 15 participants (agreement, technical and research contract holders) from Africa, Asia, Europe and North America attending the RCM.

Regional AFRA Training Course on Basic Mutation Breeding Techniques for Crop Improvement, RAF/5/076, Morogoro, United Republic of Tanzania, 24–28 October 2016

Technical Officer: F. Sarsu
Course Director: P. Kusolwa

This training course will be organized by the International Atomic Energy Agency in cooperation with the the Government of United Republic of Tanzania through Sokoine University of Agriculture. It is open to project partners/candidates from the project RAF/5/076 (AFRA) on Improving Crops Using Mutation Induction and Biotechnology through a Farmer Participation Approach.

The purpose of this course is to provide participants with theoretical as well as practical information on 1) Mutation induction and application in seed/vegetative propagated crops; 2) Accelerated breeding techniques (such as doubled haploid, anther/embryo culture etc.); and 3) Mutant line screening in phenotyping and genotyping for biotic and abiotic stress tolerance in crop breeding.

The course will include lectures and practical sessions on:

- Mutation breeding basic concepts knowledge on mutagenic agents, the establishment of dosimetry assays, determination of LD 50 doses for various crops;
- Induced mutation in seed/vegetatively propagated crops;
- Establishment of the proper starting material for specific project objectives;
- Handling of mutated populations to develop new varieties;
- Supportive techniques to mutation breeding such as Doubled haploid, anther/embryo culture techniques;
- Phenotyping and Genotyping Screening for Mutants in laboratory and in the field.

The participants should be from all participating Member States involved in the TC project RAF/5/076. Additionally they should be currently and actively working in mutation breeding and have basic knowledge in crop breeding. The course will surely enrich scientists with at least a M.Sc. degree involved in plant breeding/genetics. Participants should have a strong affinity and interest in modern plant breeding methods involving induced mutation, mutation screening (high-throughput phenotyping and genotyping) and techniques that can facilitate the breeding process.

Regional Training Course on Mutation Breeding and other Related Techniques for the Development of Heat Tolerant Cotton Mutants, RAS/5/075, Faisalabad, Pakistan, 7–18 November 2016

Technical Officer: L. Jankuloski

Cotton has a special significance and plays an important role in the economies of Australia, People's Republic of China, India, Islamic Republic of Iran, Myanmar, Pakistan, Viet Nam and Bangladesh. This leading fibre crop is grown on 20.5 million hectares in the three main cotton producing countries of the Asia and Pacific region i.e. People's Republic of China, India and Pakistan, with their annual contribution of about 60–65% in total world cotton production. Emerging demands from Viet Nam and Bangladesh for their cotton mill use signifies the increased role of cotton production in the economy of regional countries.

Use of induced mutations in recent years has become an important approach to plant breeding for improvement of crops and a large number of early, high yielding, disease and insect resistant varieties of various crops have been released in different countries by use of induced mutations. Development of high yielding, heat/drought tolerant and nutrient use efficient germplasm is necessary for sustainable cotton production in the region.

This training course will be organized by the International Atomic Energy Agency in cooperation with the Government of Pakistan through Nuclear Institute of Agriculture and Biotechnology, Faisalabad, Pakistan

The purpose of this training course is to introduce participants of TC project RAS/5/075 to mutation breeding in crops for abiotic stress tolerance, particularly for cotton improvement for heat tolerance.

Theoretical and practical lectures will be provided to participants to improve their knowledge in cotton breeding and genetics.

Participants should be plant breeders and geneticists from participating Member States involved in the TC project RAS/5/075 on Improving Sustainable Cotton Production through Enhanced Resilience to Climate Change. Participants should have a strong affinity and interest in modern plant breeding methods involving induced mutation, mutation screening (high-throughput phenotyping and genotyping) and techniques that can facilitate the breeding process.

Regional ARCAL Training Course on Mutation Breeding for Resistance to Biotic Stresses, RLA/5/068, Santo Domingo, Dominican Republic, 5–9 December 2016

Technical Officer: S. Nielen

The second training course related to TC project RLA/5/068 on Improving Yield and Commercial Potential of Crops of Economic Importance (ARCAL CL) will be organized and held at the Centro de Tecnologias Agricolas (CENTA), Santo Domingo, Dominican Republic, on mutation breeding for resistance to biotic stresses. The course should provide knowledge on screening techniques to identify plants with qualitative and quantitative resistance to fungi, bacteria, viruses and other diseases. Additionally the mechanisms of plant defence will be part of the course programme.

Past Events

First Coordination Meeting on Supporting Mutation Induction and Supportive Breeding and Biotechnologies for Improved Wheat and Barley – Phase II, RAS/5/076, Muscat, Sultanate of Oman, 14–16 December 2015

Technical Officer: F. Sarsu

The new TC regional project RAS/5/074 is a follow-up project of RAS/5/058 thereby it was the last coordination meeting of the previous project and the first of the current project. Representatives of seven countries (Iraq, Jordan, Lebanon, Oman, Saudi Arabia, Syrian Arab Republic and Yemen) attended the meeting. The participants presented their country reports which included the information on implementation of SMTA, national report and work plans.

Participants discussed national achievements and progress made in their last projects. The major achievements include: Jordan (developed 11 barley), and Yemen (developed four bread wheat, two durum wheat and two barley pre-released mutant varieties, of which at least half are expected to be released by 2017. Also, all participating countries have developed advance mutant lines in barley and wheat with outstanding traits such as drought tolerance, rust resistance, earliness, tolerance to salinity, lodging, and higher yield. These mutant lines need to be further evaluated under multi locational trials before starting the release process.

Meeting participants together with the National Liaison Officer (NLO) of Oman at the Sultanate of Oman Ministry of Agriculture and Fisheries facilities.

First RCM on Efficient Screening Techniques to Identify Mutants with Disease Resistance for Coffee and Banana, D2.20.05, Vienna, Austria, 7–11 December 2015

Technical Officers: B. Till and S. Nielen

This CRP was approved in October 2015 with the first RCM being held in Vienna on December 2015. Ten research contract and agreement holders plus three official observers attended the meeting. In addition, one participant joined the meeting by Skype. In total, 11 countries were represented. Project participants and PBGL staff gave presentations on their work in coffee and banana and work plans for the CRP. The group also visited the Plant Breeding and Genetics Laboratory in Seibersdorf to see the capacity of the lab and discuss how the lab can support the CRP. This meeting focused on a thorough review of the CRP objectives, activities and outputs and the workplans of the individual participants. Coordination of activities, sharing of materials and short and long-term objectives were also discussed. The consensus of the meeting is that efficient techniques are needed to screen the requisite thousands of mutant plants in order to recover the rare variant with improved resistance to disease. The primary focus will be on Fusarium wilt tropical race four (TR4) in banana and Coffee Leaf Rust (CLR) for coffee. An additional challenge is the efficient induction of mutations in coffee where it can take years to produce seed. Also, both edible banana and *Coffea arabica* are polyploids which represent additional bottlenecks to mutation induction and the expression of novel traits. The CRP will address these and other identified bottlenecks. By the end of the meeting the participants were confident that these issues can be evaluated and improved methods developed within the time-frame of the project.

National Training Course on Basic Mutation Breeding Techniques for Crop Improvement, IVC/5/039, Daloa, Cote D'Ivoire, 8–12 February 2016

Technical Officer: F. Sarsu
Course Director: K.Y. Justin

The national training course was part of TC project on Improving Maize Production in Savannah Areas with Severe Pedoclimatic Degradation in the North of Côte

d'Ivoire through the Cultivation of Induced Mutants Adapted to these Areas. It was implemented in the Université Jean Lorougnon Guédé. Ms Nasia Tomlekova, Mr Nikolai Christov and Ms Fatma Sarsu were participated as the lecturers for the course. Twenty five trainees from four institutions, Université Nangui Abrogoua, Abidjan; Université Félix Houphouet-Boigny, Abidjan; Université Jean Lorougnon Guédé, Daloa and CNRA Ferké, Abidjan participated in the event.

The training course included lectures and practical sessions on:

- Mutation induction for crop breeding;
- Establishment of the proper starting material for specific project objectives for maize breeding;
- Application of mutation breeding in self-pollinated and cross pollinated crops – Handling of mutated population;
- Phenotyping and genotyping screening of mutant in lab and greenhouse;
- Practical of statistical analysis and evaluation of various radio sensitivity tests;
- Basics of plant bio-techniques, preparation of mutant plant material for molecular characterization;
- Plant tissue culture techniques for the use of mutation induction in plant breeding;
- Doubled haploid techniques to support maize mutation breeding;
- Phenotyping and genotyping screening for biotic/abiotic stress. Root phenotyping and genotyping of aluminium tolerance genes in maize;
- Protocols for mutagenesis (physical (60Co), chemical (EMS, DMS, NMU, NEU, etc.) and combined (^{60}Co and EMS) treatment; protocol for inducing mutations via Accelerating aging treatment of maize seeds.

At the end of the course, round table discussions were conducted and groups were organized to discuss and prepare the projects on mutation breeding from irradiation to release of mutant varieties. Also the course participants prepared different mutation breeding programmes on different crops starting from irradiation to the release of new variety. Each of the groups presented a short PowerPoint presentation on the project. The discussions were made through cross-questioning which helped them to fine-tune the projects.

Participants discussing on radio sensitivity test.

Participants discussing on optimum dose calculation.

Regional (AFRA) Training Course on Farmer Participatory Plant Breeding Approach in Dissemination of Mutant Varieties, RAF/5/076, Harare, Zimbabwe, 29 February–4 March 2016

Technical Officer: F. Sarsu
Course Director: P. Matova

This training course was organized by the International Atomic Energy Agency in cooperation with the Government of Zimbabwe through Crop Breeding Institute (CBI), Department of Research and Specialist Services (DR&SS), Ministry of Agriculture, Mechanisation and Irrigation Development (MAMID). It was open to project partners/candidates from TC project RAF/5/066 (AFRA) on Improving Crops Using Mutation Induction and Biotechnology through a Farmer Participation Approach (AFRA) project partners.

Group photo of the participants and the course facilitators.

The course was attended by 17 international participants from 17 African countries (Algeria, Cameroon, Democratic Republic of Congo, Ghana, Gabon, Kenya, Lesotho, Malawi, Madagascar, Mauritius, Mozambique, Namibia, Senegal, Sudan, United Republic of Tanzania, Uganda and Zambia). Two experts, Mr Hussein Shimelas and Mr Yahya Shakhatreh were facilitating the training course. There were 14 national participants coming from local institutions which included Crop Breeding Institute, Agronomy Research Institute, Cotton Research Institute, Horticulture Research Institute, Arda Seeds and University of Zimbabwe. In total there were 12 female participants and 23 male participants.

The training was composed of PowerPoint presentations and lectures from the facilitators, group discussions and presentations and also a field tour of on-farm trials in Beatrice/Chiota and on-station trials and nurseries at Harare Research Centre. The purpose of the training was to learn from the facilitators' and also participants' experiences on the following aspects:

- Handling of mutated populations;
- Evaluation and selection of mutant varieties together with farmers in farmers' fields;
- Participatory plant breeding and participatory variety selection with farmers;
- How to increase the rate of dissemination and adoption of mutant/released varieties;
- Strategies to quicken variety release.

In Beatrice/Chiota the tour was on the advanced cowpea mutants that are being evaluated on-farm. Two sites were visited and the group discussed on how mutation breeding programmes can make selections in farmers' fields together with the farmers themselves. Farmers were also given a chance to give an account of how they planted and how they were managing their crops. They were also asked to give their preferred traits in cowpea varieties. Some farmers reported that they wanted varieties that give good leafs and grain yield. They also highlighted that they noted drought tolerance and aphid resistance cowpea lines. After selection with farmers participatory, the project counterpart decided to start release process of these farmers' preferred lines.

Participants and farmers viewing a field with cowpea mutants on-farm in Beatrice/Chiota.

National Training Workshop on Mutation Breeding of Brachiaria Grass and Dolichos Lablab Species, Nakuru, Kenya, 14–18 March 2016

Technical Officer: S. Nielen

From 14 to 18 March 2016 a National Training Workshop on Mutation Breeding of Brachiaria Grass and Dolichos Lablab Species was organized in Nakuru, Kenya. This workshop was an activity of the National TC project KEN/5/034 on Using Irradiated Improved Brachiaria Grass and Dolichos Lablab Species for Increasing Quantity and Quality of Milk Production and Reproduction for Smallholder Dairy Farms in Drought Prone Areas. The main counterpart institute and workshop organizer was the Beef Research Institute Kenya, which is part of the Kenya Agricultural & Livestock Research Organization (KALRO). Apart from the project team, 11 scientists from other Kenyan institutes were invited to the training. Most of the participants had a background in agronomy/animal husbandry, however, with broad knowledge on forage crops. The course was opened by Dr Eliud K. Kireger, Director KALRO. Lectures related to genetics and breeding of the forage crop Brachiara were given by the invited expert Dr Cacilda Borges do Valle of Embrapa Beef & Cattle, Brazil. Dr Borges do Valle is a leading expert in biology, cytogenetics, and breeding of Brachiaria contributed substantially to the enormous success of this forage crop in her country. Now coming for the first time to the country of origin of Brachiaria, she shared her knowledge with the participants. The Technical Officer covered in his lectures various aspects on mutation breeding, mutation induction techniques and handling of mutant populations. Prof Miriam Kinyua from Moi University, Eldoret, joined the team of lecturers and reported on her success stories in mutation breeding, such as development of drought tolerant wheat and recently UG99 resistant wheat varieties. Members of the project team presented the progress in their respective field of activity.

The majority of Brachiaria species is apomictic, meaning that they reproduce through a mechanism of asexual formation of seeds, retaining the maternal genotype in the progeny. Apomixis makes crop improvement through conventional breeding very intricate. One of the aims of mutation induction therefore could be to break apomixes. However, it is very important to determine if the genotype of choice reproduces sexual or asexual. During the training course Dr do Valle has demonstrated how this question can be answered using cytology. The technique included collection of flowers at the proper stage for determining mode of reproduction, extracting ovaries under the stereoscope, fixation and clarifying of the ovaries and finally microscopic observation. The practical demonstrations and hands-on experience were of particular value for the success of this training workshop, which was very positively acknowledged by all participants.

Collection of flowers from Brachiara for determining mode of reproduction.

Extracting ovaries under the stereoscope.

Visit of Brachiaria experimental plot.

First Technical Workshop on Capacity Building Programme for Latin American Countries against Coffee Leaf Rust, Vienna, Austria, 4–8 April 2016

Technical Officer: S. Nielen

Coffee Leaf Rust has been a latent threat in Central America since its first outbreak in 1976 in Nicaragua and is widespread within the region. Recently, in 2012, a second large outbreak of Coffee Leaf Rust created national warnings in the coffee sector for some countries. The impact of Coffee Leaf Rust was huge in the countries of Central America, not only in terms of production decrease but also in the social and economic component that induced poverty on producing families. There is a strong worry that resistance will further break down and new crises of leaf rust will emerge, necessitating the need for improving resistance in cultivars.

From 4 to 8 April 2016 a workshop was organized at the Agency's Headquarters in Vienna as part of the OFID (Opec Fund for International Development) funded projected 'Capacity Building Programme for Latin American Countries against Coffee Leaf Rust'. Representatives of six institutes in coffee growing countries in Latin America (Costa Rica, El Salvador, Guatemala, Honduras, Nicaragua, Peru) and FAO/IAEA with participation of Promecafe/IICA (Programa Cooperativo Regional para el Desarrollo Tecnológico y Modernización de la Caficultura/Inter-American Institute for Cooperation on Agriculture), CIFC (Centro de Investigação das Ferrugens do Cafeeiro), University of Natural Resources and Life Sciences, Vienna, and World Coffee Research (WCR) came together and have established the Coffee Mutation Network (CMN) for collaboration on using mutation breeding in coffee. While networks of coffee research already exist, the consensus at the meeting was that there is a strong need for a network specifically dedicated for the use of induced mutations to overcome the narrow genetic base of coffee and provide new avenues for enhanced disease resistance. The use of induced mutations represents a new technology for coffee and a network is deemed important because each country has varying capacities and strengths which can be expanded through collaboration and sharing of materials and methods. Further, different countries plan to take different approaches for using induced mutations (choice of tissue type, genotype mutagenized, type of mutagen used) and important results from this work and lessons learned can be easily shared within the framework of the network. The meeting deemed relevant collaboration by:

- Common use of existing facilities (e.g. for tissue culture) and capacities (including existing capacity in mutation breeding, e.g. in Peru and Costa Rica);

- Coordinated approach for disease screening (in collaboration with CIFC, Portugal);
- Exchange of information, protocols, germplasm;
- Direct bi- or multi-lateral collaboration on specific R&D objectives;
- Joint efforts for raising grants to fund the R&D activities;
- Identification of possible collaborators/counterparts (national, regional, interregional);
- Joint efforts to raise awareness for the technology in the region.

The network will also closely collaborate with CRP D2.20.05 on Efficient Screening Techniques to Identify Mutants with Disease Resistance for Coffee and Banana.

It was agreed that there is a major priority to establish efficient mutation induction techniques in coffee. There is a large constraint on time owing to issues of chimerism and long seed to seed time. One of the options discussed was the use of cell cultures for mutagenesis. The advantage would be that regenerated plants from single cells would be non-chimeric in contrast to plants from irradiated seeds. A pilot project for cell culture mutagenesis, plant regeneration and mutation discovery was planned during the workshop.

A training meeting on Mutation Induction in Coffee will take place 3–14 October 2016 at the FAO/IAEA Plant Breeding and Genetics Laboratory in Seibersdorf. Scientists affiliated with the institutes participating in the network will be invited to be trained on lectures, demonstrations (laboratory and greenhouse), and laboratory exercises on mutation breeding procedures/methodologies and handling of mutated populations and related techniques, on tissue culture techniques relevant for mutation induction in coffee, and on appropriate technologies for mutant phenotyping and genotyping.

Visit of PBGL during the first Workshop on Coffee Mutation.

Regional Training Course on Mutation Detection Methods Applied to Floods, RAS/5/069, Putrajaya, Malaysia, 11–15 April 2016

Technical Officer: L. Jankuloski
Course Director: A.R. Harun

The regional training course was organized in Putrajaya, Malaysia under the TC project RAS/5/069 on Complementing Conventional Approaches with Nuclear Techniques towards Flood Risk Mitigation and Post-Flood Rehabilitation Efforts in Asia. This five-day training course was officiated by Dr Dahlan Bin Hj. Mohd, Deputy Director General of Malaysian Nuclear Agency.

Training course participants.

A total of 40 participants took part in the course, in which 15 participants were international participants cutting across various countries such as Bangladesh, People's Republic of China, Indonesia, Myanmar, Pakistan, the Philippines, Thailand and Sri Lanka. Local participants consist of professionals and students from Malaysian Nuclear Agency as well as research institutes and universities.

The purpose of this training course was to provide participants with theoretical as well as practical information on modern techniques in mutation breeding combined with efficiency enhancing biotechnologies to improve the capacity to generate and develop flood/submergence tolerant crops using nuclear techniques.

The programme of the training course consisted on the following topics:

- Concepts of plant mutation breeding;
- Identification, evaluation and handling of target traits for submergence tolerant breeding;
- Physiological and phenotypic analyses of traits associated with tolerance to submergence;

- Mapping of rice submergence tolerance trait: principles of bulk segregation analysis, QTL mapping of sub1 gene;
- Mechanism associated with tolerance to submergence during germination and seedling growth;
- Utilization of appropriate technologies for screening of target traits toward flood risk mitigation and post-flood rehabilitation;
- Molecular genetic analysis and cloning of rice submergence tolerance gene: fine mapping and cloning of the sub1 gene and mutant analysis with next generation sequencing technology;
- Marker assisted breeding of rice submergence tolerance: principle of marker assisted selection via backcrossing breeding of Swarna Sub1.

This course was conducted through series of lectures during the first two days, followed by series of practical sessions for the next two days and a technical visit during the last day. Two experts appointed by the IAEA, Dr Yuwei Shen from China and Dr Ramani Kumar Sarkar from India took part in the course. All lectures were given by Dr Abdul Rahim Harun, Dr Yuwei Shen and Dr Ramani Kumar Sarkar. On the other hand, the practical session was focused on induced mutation using gamma ray, radio-sensitivity test, screening of submergence tolerance trait, DNA extraction as well as preparation of PCR and genotyping. The group activities during practical sessions were facilitated by a group of trained facilitators from Malaysian Nuclear Agency.

The participants showed great interest in the training course and they actively participated in the theoretical and practical lectures. Discussions were made with the participants and experts on the applications on mutation breeding for crop improvement particularly for flood/submergence tolerance in rice.

The course ended with a closing speech by Dr Khairuddin Abdul Rahim, Course Advisor and Director of Agrotechnology and Biosciences Division from Malaysian Nuclear Agency and certificates were presented to all lecturers, participants and facilitators.

Mutation induction and radiosensitivity test at Gamma Greenhouse, Malaysian Nuclear Agency.

First Regional (AFRA) Coordination Meeting on Improving Crops Using Mutation Induction and Biotechnology through a Farmer Participation Approach, RAF/5/076, Dakar, Senegal, 18–22 April 2016

Technical Officer: F. Sarsu

The new regional TC project RAF/5/076 is a follow up of the project RAF/5/066 thereby the meeting was the third coordination meeting of the previous project also the first coordination meeting of the current project. Within previous project, well-characterized stable mutants of a wide range of crops such as high yield, resistance to some biotic stress (such as aphid, mosaic virus etc.), abiotic stresses (such as drought, acidic soil etc.) and also quality traits were produced. Also three pre-released mutant varieties were identified, further processed in multi locational experiments and expected to be released in 2017/18.

Representatives from 20 countries (Burundi, Benin, Burkina Faso, Central African Republic, Cameroon, Democratic Republic of Congo, Egypt, Ethiopia, Ghana, Kenya, Libya, Madagascar, Mauritius, Morocco, Namibia, Senegal, Sierra Leone, United Republic of Tanzania, Zambia and Zimbabwe) attended the meeting. The participants presented their progress made till date and the achievements reached during the implementation of the project RAF/5/066. Each national coordinator prepared a PowerPoint presentation and a report (Word document) on the activities carried out, achievements, challenges they faced and their national work plans for 2016–2019, including support needed from the IAEA through the regional project RAF/5/076.

During the meeting, the project achievements within project RAF/5/066 were discussed and follow up activities were defined within the national work plans for 2016–2019, and within the frame of the new TC project RAF/5/076 ‘Improving Crops by Using Mutation Induction and Biotechnology through a Farmer Participatory Approach’.

Review and analysis of project RAF/5/066, revealed the need to apply mutation breeding combined with related bio techniques to develop new crop varieties in order to improve food security, especially in the region. The crops of interest within the project are potato, maize, amaranth, Bambara nut, sesame, cassava, wheat, rice, safflower, sesame, Chinese yam (esculenta), barley, peanut, colocasia, onion, cowpea, pearl millet, sorghum and velvet bean.

A separate session was held to identify the challenges that Member States confronted during the implementation of their national activities and the lessons learned. To

successfully complete the ongoing regional project, national and regional project activities were agreed based on the lessons learned.

Meeting participants visiting cowpea drought tolerance screening experiments in Senegal.

First Regional Coordination Meeting on Improving Sustainable Cotton Production through Enhanced Resilience to Climate Change, RAS/5/075, Goiania, Brazil, 2–6 May 2016

Technical Officer: L. Jankuloski

Cotton has a special significance and plays an important role in the economies of Australia, People's Republic of China, India, Islamic Republic of Iran, Myanmar, Pakistan, Viet Nam and Bangladesh. This leading fibre crop is grown on 20.5 million hectares in the three main cotton producing countries of Asia and Pacific region i.e. People's Republic of China, India and Pakistan, with their annual contribution of about 60–65% in total world cotton production. Emerging demands from Viet Nam and Bangladesh for their cotton mill use signifies the increased role of cotton production in the economy of regional countries.

The First Regional Coordination Meeting was held under the TC project RAS/5/075 on Improving Sustainable Cotton Production through Enhanced Resilience to Climate Change. Project coordinators from six countries (Bangladesh, Islamic Republic of Iran, Myanmar, Pakistan, Syrian Arab Republic and Viet Nam) attended this meeting. The participants presented their current status of cotton breeding and cotton mutation breeding, expected roles of nuclear and enhancing biotechnologies in the project, as well as gaps and needs for the application of mutation breeding techniques.

After reviewing national cotton breeding programmes, it was concluded that:

Existing cotton varieties grown in the region are highly sensitive to extreme periods of heat stress resulting in decreasing yield and deteriorated fibre quality.

Development of high yielding, heat/drought tolerant and nutrient use efficient germplasm is necessary for sustainable cotton production in the region.

Use of induced mutations in recent years has become an important approach to plant breeding for improvement of crops and a large number of early, high yielding, disease and insect resistant varieties of various crops have been released in different countries by use of induced mutations.

It is important to use nuclear techniques and related approaches for the development of cotton germplasm tolerant to heat and drought, and other important traits in cotton.

During the meeting network among participants was established. Seed material of two promising cotton mutant lines developed in Pakistan will be disseminated to the participating countries and field trials in different countries will be carried out.

Mutation breeding in cotton will be initiated in Bangladesh, Myanmar, Islamic Republic of Iran, Syrian Arab Republic and Viet Nam. Pakistan as a leader in cotton mutation breeding in the region will provide services for mutation induction and expertise in cotton mutation breeding.

During the meeting the RAS/5/075 work plan was fine-tuned and further project activities were finalized. Based on needs of participating countries, it was proposed to organize training course on cotton mutation breeding for abiotic stress tolerance in Pakistan.

The First Regional Coordination Meeting was held in conjugation with the World Cotton Research Conference (WCRC–6) and 2016 Biennial Conference of the International Cotton Genome Initiative (ICGI) — two of the largest events in cotton research. The WCRC–6 was attended by more than 500 participants from the world, presenting the most recent developments in cotton breeding and genetics. This event provided unique opportunity to the counterparts of RAS/5/075 to contact many cotton researchers from around the world to share knowledge and to establish collaboration.

RAS/5/075 participant from Pakistan, Mr Manzoor Hussain Manj, gave two presentations during the WCRC–6 parallel sessions on 'Improving for Sustainable Cotton Production through Enhanced Resilience to Climate Change with Reference to Pakistan' and on 'Evolution of NAIB–KIRAN, an Early Maturing, High Yielding, Fine Quality Fiber Cotton Variety having Enhanced Resilience against CLCuD and Heat under the Changed Climatic Scenario' presenting success of mutation breeding programme for cotton improvement in Pakistan.

Meeting participants.

IAEA/RAS Regional Training Course on Application of Mutation Breeding and Screening for Biotic and Abiotic Stresses in Cereals Supporting Mutation Induction and Supportive Breeding and Biotechnologies for Improved Wheat and Barley – Phase III, RAS/5/074, Amman, Jordan, 2–12 May 2016

Technical Officer: F. Sarsu
Course Director: A. Al-Yassin

Training course participants.

This training course was organized by the International Atomic Energy Agency in cooperation with the Government of Jordan through National Center for Agricultural Research and Extension (NCARE). Mr Yahya Khalil Shakhatreh organized the training course which was facilitated by Mr Adnan Al-Yassin (course director). Dr Michael Ghanem from ICARDA and Mr Mehboob Rahman from Pakistan attended as lecturers. The course was attended by 18 participants from Iraq (3), Jordan (6), Kuwait (1), Lebanon (2), Oman (1), Saudi Arabia (1), Syrian Arab Republic (2) and Yemen (2).

The purpose of this course was to provide participants with advanced theoretical as well as practical information on mutation breeding and screening for biotic and abiotic stresses and their application in cereal (mainly wheat and barley) breeding programmes.

The two-week training course consisted of lectures, demonstrations (laboratory and greenhouse), and laboratory exercises on:

1) Mutation breeding procedures/methodologies and handling of mutated populations (from mutation induction to release variety);
2) Methodologies for screening of mutant population, identification and detection of mutants for biotic/abiotic stress resistance/tolerance;
3) Genetics and physiology of abiotic stresses (e.g. drought and salinity) including lectures and practical sessions on application of physiology in cereal breeding; phenotyping techniques: what to measure and when; Use of modeling in physiological breeding; practical work and data analyses exercise;
4) Breeding cultivars with resistance to disease (e.g. wheat stem, yellow and leaf rust);
5) Screening of mutant lines for desired traits of cereals in the field/greenhouse/laboratories including lectures and practical sessions on mutation detection methods, screening of mutant lines and utilisation of molecular techniques in mutation breeding;
6) Utilization of appropriate technologies for mutation breeding and screening for biotic/abiotic stresses including lectures and practical sessions on genotyping by sequencing, etc. were redelivered. In the second session, participants were exposed to PCR technique, electrophoresis, and primer designing technique.

At the end of the training course participants were divided in to three groups. Based on obtained knowledge and experience from the training course, participants prepared workplan and gave presentation on how the mutation breeding can be applied in their national breeding programmes.

Participant's discussion on plant mutation breeding.

National Training Course on Methodologies and Mechanisms for Screening against Abiotic (Salinity and Drought) and Biotic Stresses, OMA/5/004, Muscat, Oman, 8–19 May 2016

Technical Officer: A.M.A. Ghanim

A two-week national training course was organized in the context of the implementation of TC project OMA/5/004 on Building Capacity for the Improvement of Major Crops through Induced Mutation Using Nuclear and Related Techniques. The course was hosted by the Date Palm Research Center, Directorate General of Agricultural and Livestock Research, Ministry of Agriculture, Muscat, Oman. Dr Mehmet Ihsan Tutluer from Turkey and Dr Qahir Sohail from Pakistan were invited as lecturers to the training course. The training covered the basic principle of mutation induction in seed and vegtatively propagated crop plants, development and handling of mutant populations and screening for biotic and abiotic tresses with emphasis on diseases, drought and salt tolerance. Main lectures and practical sessions included:

- Introduction to mutation breeding including mutation induction and development of mutant population;
- Screening for abiotic stresses with emphasis on drought and salinity;
- Screening for biotic stress mainly diseases with emphasis on yellow leaf rust (wheat and barley);
- Efficiency enhancing techniques (introduction to bases of doubled haploid technologies and molecular markers).

Training course participants and the lecturer, Dr M.I. Tutluer, in the center.

The PBGL supplied the irradiated seeds and *in vitro* propagated M_1V_1 banana for the practical sessions of radio-sensitivity assessment. Eighteen researchers from the national programmes participated in the training. The training was successfully implemented and the trainees expressed satisfaction. The acquired skills are expected to contribute to the successful implementation of the national TC project.

Participants working in the lab for measurements of M_1 plants for radiosensitivity test.

Regional (AFRA) Training Course on Field Experimental Design and Data Analysis in Mutation Breeding, RAF/5/066, Cotonou, Benin, 9–14 May 2016

Technical Officer: F. Sarsu
Course Director: C.B. Gandonou

This training course was organized by the International Atomic Energy Agency in cooperation with the Government of Benin through the University of Abomey-Calavi under the TC project RAF/5/076 on Improving Crops Using Mutation Induction and Biotechnology through a Farmer Participation Approach (AFRA). It was attended by 22 young breeders from Algeria, Burundi, Burkina Faso, Benin (five local participants) Central African Republic, Cameroon, Cote D'Ivoire, Egypt, Ghana, Gabon, Liberia, Madagascar, Namibia, Niger, Senegal, Seychelles, Sierra Leone and Zambia.

Group photo - participants and lecturers.

Mr Ali Ustun from Turkey and Mr Samuyel Amuteye were invited as lecturers.

The training course included lectures and practical sessions on:

- Application of mutation breeding for crop improvement;
- Establishment of the proper starting material for specific project objectives;
- Breeding procedures, methodologies and handling of mutated population;
- Experimental designs for accurate data collection, analysis and interpretations;
- Design and analysis of experiments;
- Randomization/replication/control of experimental error;
- Computer applications to agricultural experiments using statistical packages;
- Statistical analysis for breeding;
- Analysis of variance for plant breeding;
- Analysis of multiple locations and years;
- Interpretations of statistical data for crop improvement;
- Stability and its importance in plant breeding;
- Using molecular marker data to detect mutations and use of markers in breeding.

IAEA/RCA Regional Training Course on the Applications of *In Vitro* Techniques in Mutation Breeding, RAS/5/070, Jakarta, Indonesia, 23–27 May 2016

Technical Officer: F. Sarsu
Course Director: S. Human

Training course participants.

This training course was organized by the IAEA in cooperation with the Government of Indonesia, the National Nuclear Energy Agency, Center for Isotopes and Radiation Application (CIRA) and National Nuclear Energy Agency of the Republic of Indonesia (BATAN). It was open to project partners/candidates from TC project RAS/5/070 (RCA) on Developing Bioenergy Crops to Optimize Marginal Land Productivity through Mutation Breeding and Related Techniques. The training course was attended by 28 participants from 14 countries, including Bangladesh, Peoples Republic of China, India, Indonesia, Cambodia, People's Democratic Republic of Laos, Republic of Korea, Malaysia, Mongolia, Myanmar, Pakistan, the Philippines, Sri Lanka and Vietnam. The purpose of the training course was to provide participants with theoretical and practical information on mutation induction, mutation breeding and mutation related biotechnologies, as well as screening of target traits for crops. The training course included lectures, roundtable discussions and practical sessions (laboratory, greenhouse and field) on:

- Introduction to mutation induction for crop improvement;
- Mutation breeding for improvement of bioenergy crops in Indonesia;
- Basics of plant biotechnology and mutation induction;
- Using *in vitro* techniques in mutation breeding;
- Bioenergy crop production technology in marginal land areas;
- Handling of subsequent *in vitro* derived mutated populations;
- Utilization of appropriate technologies for mutation screening for target traits in bioenergy crops;
- Establishment of adequate screening protocols *in vitro* and/or *in vivo*;
- Practical sessions comprised radio sensitivity tests for Sorghum seeds including the establishment of LD50;
- Divers *in vitro* techniques: media preparation, plantlet preparation, culturing plantlets on media, doubled haploid, anther/embryo culture techniques to support mutation breeding.
- Roundtable discussions: how the participants will use this training experience — mutation breeding and appropriate *in vitro* and *in vivo* technologies — in their national breeding programmes to release improved varieties.

Dr Taryono, Faculty of Agriculture, Gajah Mada University, Yogyakarta, Dr Human, CIRA-BATAN, Jakarta, and Dr Sangwan, Université de Picardie Jules Verne, France were lecturers at the training course. Participants showed great interest in the training course, participated actively in the roundtable discussions and asked many questions about the applications of *in vitro* techniques in mutation breeding. They also gained experience in the establishment of the proper screening

protocols (*in vitro* and/or *in vivo*) to be used in the field conditions. The training course was well appreciated and highly evaluated by the participants. They acknowledged that the knowledge and skills, gained during this training course, are greatly relevant for their future work in their home countries.

Participants visiting BATAN's greenhouse: Handling of in vitro — derived mutation populations.

Laboratory experiments by the participants: In vitro anther culture of sweet sorghum.

Coordinated Research Projects

Project Number	Ongoing CRPs	Scientific Secretaries
D2.30.29	Climate Proofing of Food Crops: Genetic Improvement for Adaptation to High Temperatures in Drought Prone Areas and Beyond	F. Sarsu
D2.30.30	Integrated Utilization of Cereal Mutant Varieties in Crop/Livestock Production System	L. Jankuloski
D1.50.13	Approaches to Improvement of Crop Genotypes to High Water and Nutrient Use Efficiency for Water Scarce Environment	Joseph / L. Jankuloski
D2.20.05	Efficient Screening Techniques to Identify Mutants with Disease Resistance for Coffee and Banana	B. Till and S. Nielen

	New CRPs	Scientific Secretaries
D2.50.05	Mutation Breeding for Resistance to *Striga* Parasitic Weeds in Cereals for Food Security	A.M.A. Ghanim and L. Jankuloski

Mutation Breeding for Resistance to *Striga* Parasitic Weeds in Cereals for Food Security, D2.50.05, New

Scientific Secretaries: A.M.A. Ghanim and L. Jankuloski

Severly damaged sorghum field by heavy Striga infestation.

A new CRP (D2.50.05) on Mutation Breeding for Resistance to *Striga* Parasitic Weeds in Cereals for Food Security was open for proposals from Member States during the reporting period. The parasitic weeds *Striga asiatica* and *S. hermonthica* are major biological constraints to cereal production in most of sub-Saharan Africa and semi-arid tropical regions of Asia. The CRP proposes the use of physical mutagenesis and associated screening technologies to broaden the genetic base of resistance. The main objective is to develop screening protocols for mutant population to identify and advance resistant mutants. Screening packages will be optimized for laboratory, screen house and field to *Striga asiatica* and *S. hermonthica*. Allelism and mechanism of resistance will be analyses to classify different source of resistance which can be combined to produce durable resistance. Accelerating techniques such as rapid cycling of crop generation and efficiency enhancing technologies of doubled haploid, genomics and molecular markers will be adapted as appropriate. So far, 14 contacts were approved for participation in the CRP and additional 2–3 are expected. The initial notification was send to contract holders inviting them to the first RCM meeting. (For more information, see Forthcoming Events on page 9).

Efficient Screening Techniques to Identify Mutants with Disease Resistance for Coffee and Banana, D2.20.05

Scientific Secretaries: B. Till and S. Nielen

The constant evolution of plant pathogens combined with global climate change and variation contributing to new outbreaks means that plant breeding for disease resistance requires continuous efforts. Induced mutations provide novel genetic diversity for the plant breeder. This is especially important in crops where there is a limited genetic base, or there are bottlenecks to sexual propagation. Genetic improvement of banana is hindered by the fact that edible sweet bananas are triploid, sterile and parthenocarpic. Breeding of *Coffea arabica* is slowed down by the years it takes to go from seed to seed. The productivity of both crops is threatened by fungal pathogens whose spread is exacerbated by global climate change and variation. Exported Cavendish bananas are clones and susceptible to diseases including Fusarium wilt caused by tropical race four (TR4). In recent years TR4 has been identified in nine countries suggesting that it is spreading geographically and threatening global banana production. Coffee is the second most traded commodity behind petroleum. Coffee leaf rust is devastating to

plantations. This CRP has a main focus on developing efficient screening methods for large mutant populations. In addition, low-cost tissue culture and efficient methods to dissolve chimeric sectors that result from mutagenesis of multicellular tissues are needed to make mutation breeding in coffee and banana efficient. This CRP began in the fourth quarter of 2015, with the first RCM being held in December of 2015. The efforts to improve coffee in this CRP are further supported with capacity building funds from the Opec Fund for International Development (OFID).

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

Scientific Secretary: F. Sarsu

This CRP has 11 research contract holders from Colombia, China, Cuba, India, Mexico, Pakistan, the Philippines, Senegal, the United Republic of Tanzania and Zimbabwe and three agreement holders from Spain, the United Kingdom and International Center for Tropical Agriculture (CIAT).

The overall objective of this CRP is 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 temperature extremes (increased minima and maxima) as anticipated by climate change and variability for the next 20 to 40 years. The aim is to develop tools that allow plant breeders to use mutation programs together with efficiency enhancing plant biotechnologies to develop improved crop varieties with higher and wider adaptability to temperature variations.

The project focused on improving the grain yields of rice grown in harsh conditions such as high temperature stress, as foreseen under the climate change. Our goal is to improve rice yields by screening and selecting induced mutants for high temperature stress tolerance.

In terms of technology advancement, the genetic improvement of crops that can adapt to future climate conditions, i.e. 'climate proofing' crops, is an important opportunity. The seedling and flowering stages in rice were identified as key growth stages that are sensitive to heat tolerance. Thereby, a protocol for screening methodologies for high temperature in the greenhouse, growth chamber and under field conditions was developed in order to identify valuable mutant populations which are tolerant to heat stress. Simple and quick methods are also provided to screen seedlings for heat tolerance in hydroponics and pots in growth chamber/greenhouse conditions. The seedling test takes 3–4 weeks and allows the screening of several hundred seedlings. The test can be used to screen mutant lines and cultivars, as well as M_3 generations to advanced mutant generations.

Each participating country has been establishing at least one or two protocols for whole plant, physiological, genetic and molecular studies. Mutant line screening protocols for rice and common beans have been developed under growth chamber and field conditions for confirmation of heat tolerance. In the particular case of beans, responses to nodulation and nitrogen fixation are closely monitored in order to identify the best symbiosis: plant/rhizobium is performing satisfactorily under heat conditions. Additionally, gene expression was investigated under increased temperature conditions. Some genes involved in high temperature response, both in rice and beans, showed a significant change in expression patterns, which may play a role in stress tolerance. A detailed characterization of those genes under heat stress has yet to be carried out.

All participating countries generated new mutant populations in rice and beans. Also, some countries used characterized mutant lines from previous projects. All of them have mutant lines at least at the M_4 generation, which tested tolerant for their responses to increased temperatures. Cuba released a new variety 'LP 20' which has good yield under heat stress conditions and low water supplies. Some participants have advanced/pre released mutant lines in rice and beans which is planned to be released to farmers by 2018.

Significant progress has been achieved so far in major areas of research on rice and common beans to accomplish the objectives of the CRP. It is expected that the identification of high yielding rice and bean germplasm, and the establishment of experimental protocols for physiological, genetic and molecular characterization, will be completed and prepared for dissemination by the end of this year.

Integrated Utilization of Cereal Mutant Varieties in Crop/Livestock Production Systems, D2.30.30

Scientific Secretary: L. Jankuloski

This CRP began in the third quarter of 2012 and will be concluded in the third quarter of 2017. In December 2012, we held our first RCM in Vienna, Austria and the second RCM was held in August 2014 in Bogor, Indonesia.

The objectives of this CRP are:

1. To identify mutant varieties or advanced mutant lines for food and feed;
2. To evaluate mutant cereal varieties/lines for agronomic performance and feed quality;
3. To develop crop management systems for cereal mutant varieties with respect to improved yield and quality;

4. To determine biomass, harvest index and nitrogen use efficiency of mutant varieties and advanced lines;
5. To validate and publish protocols and guidelines for speeding up the establishment of useful mutants in desirable genetic backgrounds;
6. To perform pilot tests of superior mutant varieties/lines on-farm through participatory farmer approaches.

The crops chosen are those that can be used for both human food and animal fodder. The project involves nine participating countries and four major crops, namely wheat (the Former Yugoslav Republic of Macedonia, Mongolia), rice (People's Republic of China, Malaysia), barley (Austria, People's Republic of China, Kuwait and Peru) and sorghum (Indonesia). The project aims to improve the agronomy of the crop especially in respect to soil and water management, improve nutritional value and improve the knowledge and skills base of participating MSs.

All participants have identified promising mutant lines that are now subject to farming management practices to maximize yields in challenging conditions. Success in tailoring agronomy for mutant varieties will be judged by take up by farmers but there are already impressive outcomes, particularly in Indonesia where mutant varieties are now grown in several regions on an increasing area.

All project participants have submitted project progress report and all contracts have been renewed for 2015–2016. The mid-term review was positively evaluated by the Committees for Coordinated Research Activities (CCRA), thus the project will continue as planned.

The Third RCM will be held 1–5 August 2016 in Darkhan, Mongolia.

Approaches to Improvement of Crop Genotypes with High Water and Nutrient Use Efficiency for Water Scarce Environments, D1.50.13

Scientific Secretaries: J. Adu-Gyamfi and L. Jankuloski

This CRP is in its final year. Ten research contract holders (Bangladesh, People's Republic of China, Kenya, Malaysia (two participants), Mexico, Pakistan, Peru, South Africa, Uganda and Vietnam), one technical contract holder (Peru) and one agreement holder (South Africa) are participating in the CRP. The research project was started in December 2011 and three RCMs have been carried out so far to review project progress and present preliminary results. The overall objective of this CRP is to increase crop productivity and food security by developing improved crop varieties and soil, water, nutrient and crop management technologies and making them available to farmers, and ensure their cropping systems are resilient to biotic and abiotic stresses in water scarce environment.

Key outputs of the CRP to date include:

1. The total area covered by ratooning rice cultivars (one planting and two harvests) from 2012–2015 is 42,000 ha in China with yield up to 14,500 kg/ha over two harvests.
2. 20–30% yield increase of two elite potatoes with high fertilizer use efficiency at four locations (Njoro, Kabiana, Marigat and Molo) using a combination of animal manure and nitrogen fertilizer have been recorded by farmers.
3. Three genotypes of wheat with high water and nutrient use efficiencies are being tested in 25 farmers field (0.5 ha per farmer) in six districts (Peshawar, Nowshera, Charsadda, Lakki- Marwat, Swabi and Dir) in Pakistan.
4. Three varieties and one advanced mutant line of barley, and five improved genotypes of quinoa suitable for high altitude have been identified and are being tested in the high altitude mountains and coastal areas of Peru.

The final RCM will be held in Vienna, Austria from 7 to 11 November 2016.

General information applicable to all coordinated research projects

Submission of Proposals
Research contract proposal forms can be obtained from the IAEA, the National Atomic Energy Commissions, UNDP offices or by contacting the Technical Officer. The form can also be downloaded from the URL:
http://www-crp.iaea.org/html/forms.html.

Complementary FAO/IAEA Support
IAEA has a programme of support through national Technical Cooperation (TC) projects. Such support is available to IAEA Member States and can include additional support such as equipment, specialized training through IAEA training fellowships and the provision of technical assistance through visits by IAEA experts for periods of up to one month. Full details of the TC Programme and information on how to prepare a project proposal are available at the URL: http://pcmf.iaea.org/.

Technical Cooperation Field Projects

Project Number	Country/Region	Title	Technical Officer(s)
ALB/5/007	Albania	Supporting the Improvement of Plant Productivity Using Radiation Techniques	L. Jankuloski/F. Sarsu
ALG/5/026	Algeria	Increasing the Genetic Variability for the Improvement of Strategic Crops (Wheat, Barley, Chickpeas and Dates) for Enhanced Tolerance to Biotic and Abiotic Stresses and the Development of Biotechnology Capacities	A.M.A. Ghanim
BGD/5/029	Bangladesh	Evaluating Promising Abiotic Stress Tolerant Crop Mutants/Varieties and Measuring the Suitable Management Practices for the Promotion of Sustainable Production at Saline, Submergence and Drought Prone Areas	L. Jankuloski in collaboration with Soil and Water Management and Crop Nutrition Section
BUL/5/014	Bulgaria	Screening of Cereal Germplasm Stress Response and Adaptation Potential by Advanced Nuclear, Omics and Physiological Approaches	L. Jankuloski/S. Nielen
BKF/5/016	Burkina Faso	Using Nuclear Techniques for Improving Rice Yield and Quality	L. Jankuloski/I. Ingelbrecht
BDI/5/001	Burundi	Improving Cassava Productivity through Mutation Breeding and Better Water and Nutrient Management Practices Using Nuclear Techniques	S. Nielen/I. Ingelbrecht in collaboration with Soil and Water Management and Crop Nutrition Section
CAF/5/008	Central African Republic	Improving Cassava Yield through Improved Crop Variety and Best Soil Management Practices Using Nuclear Techniques	I. Ingelbrecht/A.M.A. Ghanim in collaboration with Soil and Water Management and Crop Nutrition Section
COL/5/024	Colombia	Supporting Mutagenesis and Functional Genomics Applied to the Improvement of Rice	S. Nielen/B. Till
ZAI/5/025	Congo, Democratic Rep. of the	Increasing Genetic Variability in Cassava and Maize for Enhanced Tolerance to Biotic and Nitrogen Stresses	L. Jankuloski/F. Sarsu
IVC/5/039	Cote d'Ivoire	Improving Maize Production in Savannah Areas with Severe Pedoclimatic Degradation in the North of Cote d'Ivoire through the Cultivation of Induced Mutants Adapted to these Areas	F. Sarsu/I. Ingelbrecht
ERI/5/008	Eritrea	Supporting the Livelihood of Barley Farmers through Mutation Techniques and N15 Technology to Improve Malting, Food and Feed Barley Production	A.M.A. Ghanim
GHA/5/036	Ghana	Screening Oil Palm M2 Population for Useful Mutants	L. Jankuloski/F. Sarsu
INS/5/039	Indonesia	Enhancing Food Crop Production Using Induced Mutation, Improved Soil and Water Management and Climate Change Adaptation	L. Jankuloski/B. Till
IRA/5/014	Iran, Islamic Republic of	Improving Wheat Yield and Stress Tolerance for Sustainable Production	B. Till/L. Jankuloski
KEN/5/037	Kenya	Using Climate Smart Bracharia Mutants to Develop Integrated Farm Model Technologies for Improved Livelihood Among Smallholder Farmers	S. Nielen/F. Sarsu

Project Number	Country/Region	Title	Technical Officer(s)
KUW/5/002	Kuwait	Implementing Mutation Induction to Improve Barley Production under Harsh Environmental Conditions	L. Jankuloski/A.M.A. Ghanim
LAO/5/001	Lao, P.D.R.	Enhancing Food Security through Best Fit Soil-Water Nutrient Management Practices with Mutation Induction for Drought Resistant Rice	L. Jankuloski
LES/5/004	Lesotho	Using Nuclear Techniques for Improvement of Crop Yield, Quality and Stress Tolerance for Sustainable Crop Production (Continuation of the on-going project)	S. Nielen/A.M.A. Ghanim
LES/5/005	Lesotho	Improving Crop Yield, Quality and Stress Tolerance for Sustainable Crop Production, Phase II	S. Nielen/A.M.A. Ghanim
MAG/5/023	Madagascar	Promoting Climate Smart Agriculture to Face Food Insecurity and Climate Change with Regard to Basic National Foods (Rice and Maize)	L. Jankuloski/F. Sarsu
MAU/5/006	Mauritania	Contributing to the Improvement of Rice Crop Yields through the Application of Nuclear Techniques to Water Management and Soil Fertility	L. Jankuloski/F. Sarsu in collaboration with Soil and Water Management and Crop Nutrition Section
MAR/5/020	Mauritius	Developing Stress Tolerant Banana and Tomato Varieties by Enhancing the National Capacity in Mutation Induction and Biotechnology	B.Till/S. Nielen
MAR/5/023	Mauritius	Improving Landraces of Crucifers (Cauliflower and Cabbage) and Carrot through the Use of Nuclear Techniques for Mutation Breeding and Biotechnology	F. Sarsu/L. Jankuloski
MON/5/021	Mongolia	Improving the Productivity and Sustainability of Farms Using Nuclear Techniques in Combination with Molecular Marker Technology	L. Jankuloski/S. Nielen in collaboration with Animal Production and Health Section
MOR/5/033	Morocco	Using Nuclear Techniques to Support the National Programme for the Genetic Improvement of Annual and Perennial Plants and to Develop Agricultural Production	A.M.A. Ghanim
MOZ/5/007	Mozambique	Enhancing Mutation Breeding of Sorghum and Pearl Millet to Develop High Yield, Disease Resistance and Drought Tolerance	S. Nielen/A.M.A. Ghanim
NAM/5/012	Namibia	Developing High Yielding and Drought Tolerant Crops through Mutation Breeding	F. Sarsu/S. Nielen
NAM/5/014	Namibia	Evaluating Efficient Water and Nutrient Use, Molecular Characterization and Nutritional Composition of Mutant Germplasm Populations	F. Sarsu/S. Nielen in collaboration with Soil and Water Management and Crop Nutrition Section
NEP/5/003	Nepal	Improving Crop Yield for Food Security and Economic Growth by Using Nuclear and Molecular Techniques	S. Nielen/L. Jankuloski
NER/5/019	Niger	Improving Sesame Plant Productivity by Obtaining High-Yielding Induced Mutants Adapted to Semi-Arid Conditions	I. Ingelbrecht/A.M.A. Ghanim
OMA/5/004	Oman	Building Capacity for the Improvement of Major Crops through Induced Mutation Using Nuclear and Related Techniques	A.M.A. Ghanim/I. Ingelbrecht

Project Number	Country/Region	Title	Technical Officer(s)
PAK/5/047	Pakistan	Developing Germplasm through TILLING in Crop Plants Using Mutation and Genomic Approaches	B. Till/S. Nielen
PAL/5/009	Palestine	Enhancing the Performance of Durum Wheat Landraces by Induced Mutation (Phase II)	L. Jankuloski/A.M.A. Ghanim
QAT/5/006	Qatar	Enriching Genetic Diversity and Conserving Plant Genetic Resources Using Nuclear Techniques and Related Technologies	A.M.A. Ghanim/L. Jankuloski
RAF/5/076	Regional Africa	Improving Crops by Using Mutation Induction and Biotechnology through a Farmer Participatory Approach	F. Sarsu/S. Nielen
RAS/5/069	Regional Asia	Complementing Conventional Approaches with Nuclear Techniques towards Food Risk Mitigation and Post-Flood Rehabilitation Efforts in Asia	L. Jankuloski/S. Nielen
RAS/5/070	Regional Asia	Developing Bioenergy Crops to Optimize Marginal Land Productivity through Mutation Breeding and Related Techniques (RCA)	F. Sarsu/S. Nielen
RAS/5/073	Regional Asia	Supporting Climate-proofing Rice Production Systems (CRiPS) Based on Nuclear Applications-Phase II	L. Jankuloski/S. Nielen
RAS/5/074	Regional Asia	Enhancing Wheat and Barley Productivity through Induced Mutation with Supportive Breeding and Related Biotechnology Techniques (Phase III) (ARASIA)	F. Sarsu/L. Jankuloski
RAS/5/075	Regional Asia	Improving Sustainable Cotton Production through Enhanced Resilience to Climate Change	L. Jankuloski/F. Sarsu in collaboration with Soil and Water Management and Crop Nutrition Section
RAS/5/077	Regional Asia	Promoting the Application of Mutation Techniques and Related Biotechnologies for the Development of Green Crop Varieties (RCA)	S. Nielen/F. Sarsu
RLA/5/068	Regional Latin America	Improving Yield and Commercial Potential of Crops of Economic Importance (ARCAL CL)	S. Nielen/L. Jankuloski
SEN/5/034	Senegal	Using an Integrated Approach to Develop Sustainable Agriculture in a Context of Degrading Soil Fertility, Climate Change and Crop Diversification	F. Sarsu
SIL/5/017	Sierra Leone	Selecting and Analyzing Bio-Enriched and Bio-Fortified Rice and Cassava Lines and their Efficient Postharvest Transformation to Popular Food Products	S. Nielen/I. Ingelbrecht
SRL/5/045	Sri Lanka	Establishing a National Centre for Nuclear Agriculture	F. Sarsu
SUD/5/037	Sudan	Applying Nuclear Techniques to Improve Crop Productivity and Livelihood of Small-scale Farmers in Drought Prone Areas	F. Sarsu/S. Nielen in collaboration with Soil and Water Management and Crop Nutrition Section
URT/5/030	Tanzania, United Rep. of	Improving Rice and Barley Production through Application of Mutation Breeding with Marker Assisted Selection	L. Jankuloski/F. Sarsu
URT/5/032	Tanzania, United Rep. of	Developing Maize Cultivars for Improved Yield and Resistance to Viral Disease	F. Sarsu/L. Jankuloski

Project Number	Country/Region	Title	Technical Officer(s)
THA/5/054	Thailand	Increasing Adaptability for Adverse Environment Tolerance in Rice Germplasm Using Nuclear Techniques	F. Sarsu/S. Nielen
UZB/5/005	Uzbekistan	Developing Mutant Cotton Breeding Lines Tolerant to Diseases, Drought and Salinity (Phase II)	F. Sarsu/S. Nielen
VIE/5/018	Viet Nam	Adapting Rice-Based Cropping Systems to the Impact of Climate Change by Nuclear Mutation Breeding and Improving Nitrogen Use Efficiency Using Nitrogen-15 for Vegetables in Main Growing Areas	L. Jankuloski
ZIM/5/021	Zimbabwe	Assessing and Promoting Sustainable Agricultural Production in Communal and Newly Resettled Farms	F. Sarsu/A.M.A. Ghanim in collaboration with Soil and Water Management and Crop Nutrition Section

Developments at the Plant Breeding and Genetics Laboratory (PBGL)

The first half of 2016 has been a period of intense activity at the PBGL as well as reflection as we said goodbye to two long-serving colleagues and friends, Mr Souleymane Bado and Mr Andreas Draganitsch.

In terms of research, transboundary pests and diseases in crops were affirmed as a major topic for adaptive R&D at PBGL, in line with priorities set at the programme level and recommendations by the IAEA's Standing Advisory Group for Nuclear Applications (SAGNA).

Two recently initiated CRP's focus on three crop epidemics: Panama disease in banana caused by Fusarium oxysporium sp. cubense TR4; Coffee Leaf Rust, a disease that is devastating susceptible coffee plantations in Central America; and the parasitic weed *Striga* causing annual losses in food crops such as sorghum and maize estimated at over 10 billion US$, mainly in Sub Saharan Africa.

We further considered that developing markers for agronomically important traits created through mutation breeding offers exciting opportunities to link our molecular work with field applications that are of value to the Member States. A multidisciplinary approach integrating plant breeding and genetics with biotechnology and phenotyping will be followed to link causative mutations with traits and develop molecular markers for accelerated introgression of these traits in adapted lines.

Accordingly, under the CRP on 'Integrated Utilization of Cereal Mutant Varieties in Crop/Livestock Production Systems', D2.30.30, a first concept note has been drafted focusing on a barley orange lemma mutant called 'rob 1'. This mutation results in reduced lignin content and mutant barley with low lignin has potential as animal feed due to its higher digestibility. Using molecular markers, this recessive trait could be introgressed more rapidly into adapted lines. This work will be carried ou in cooperation with Prof H. Grausgruber, University of Natural Resources and Life Sciences, Tulln, Austria.

As for the CRP on 'Efficient Screening Techniques to Identify Mutants with Disease Resistance for Coffee and Banana', D2.20.05, we have opted to use regenerable *in vitro* cell cultures as source materials for gamma irradiation in case of coffee. This approach should enhance the recovery of homohistonts and allow generating a virtually limitless number of mutant *in vitro* plantlets. A first and time consuming step is to develop the protocols for producing the cell cultures, in first instance from selected genotypes of Coffea arabica, the tetraploid, self-pollinating species that is susceptible to Leaf Rust. In vitro tissue culture of coffee initiated at PBGL is currently further pursued in cooperation with Prof M. Laimer, BOKU University, Vienna, Austria. So far, primary cell cultures are being established from different tissues, including from anthers to establish haploid cell cultures. There are over 50 races of the fungus Hemileia vastatrix causing Coffee Leaf Rust with different host specificity, virulence and spread across the affected countries. A rapid, low cost molecular diagnostics tool is required to facilitate resistance breeding. The PBGL is well positioned to take on this task in partnership with coffee institutions in South America and Europe.

Over the years, PBGL has established a solid foundation for molecular discovery of mutations and further progress in this area is covered in this Newsletter. Proof-of-concept has been demonstrated in case of rice for mutations induced by gamma irradiation. There is a clear opportunity to build on these successes and move towards mutation discovery and marker development targeting agronomically important traits, thus linking molecular work with applications in the field. A collection of candidate crop/trait is being compiled at the time of writing. Appropriate linkages with National Programmes would ensure that this work meets the needs of our Member States.

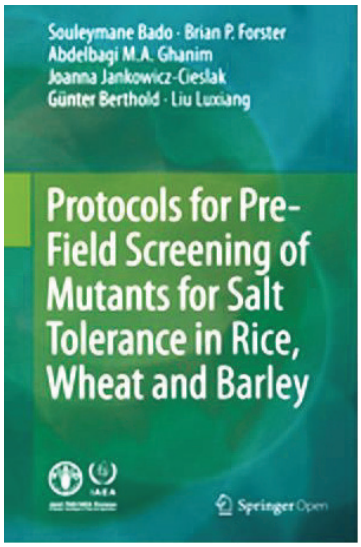
The PBGL continues to host an impressive number of trainees with 11 fellows from 10 different Member States already in the first half of 2016. A similar number is expected for the second half of 2016. Later this year, PBGL will further host a practical workshop on *in vitro* tissue culture of coffee in the context of the 'Capacity Building Programme for Latin American Countries against Coffee Leaf Rust', funded by OFID.

The PBGL has continued its irradiation services for plant mutation induction to MS. At the time of writing, 25 irradiation requests have been processed from 21 MS covering over 32 different plant species. To better serve our clients as well as improve our to internal knowledge management, the 'Irradiation Service Request Form' and the 'Guidelines for Sample Preparation' have been updated. These forms will be made available online through the PBGL website later this year.

Finally, with regards to outreach and communication, new information sheets to promote our R&D flagship projects as well as other draft promotional materials are under preparation. These are intended to on the one hand illustrate and promote the power and global impact of plant mutation breeding as an effective tool for crop

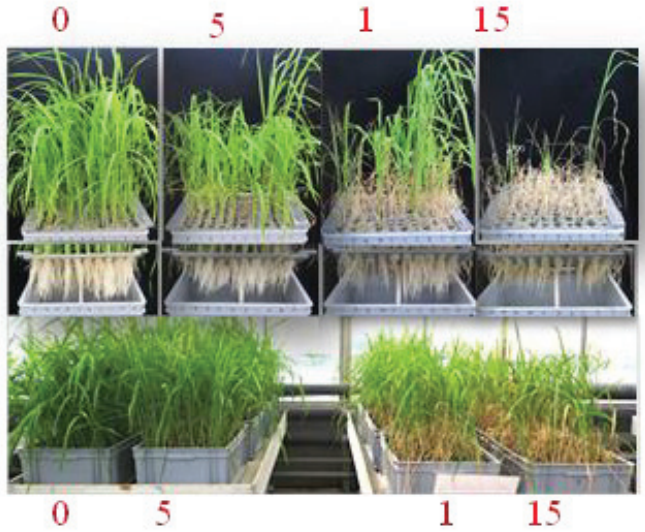
improvement across a wide range of crops and traits, and on the other showcase our own activities and R&D projects. Thanks to the inputs and efforts from the entire PBS team, a colourful display illustrating the approaches and successes of plant mutation breeding was already presented at the '2016 Long Night of Research' which turned out to be a tremendous success!

Protocols for Pre-Field Screening of Mutants for Salt Tolerance in Rice, Wheat and Barley



Mutation induction in plants aims to generate novel genetic diversity for plant breeders targeting improvement in yield, quality, resistance to pests and diseases, and tolerance to abiotic stresses such as salinity, drought and heat. Salinity is a major abiotic stress limiting crop yields in many parts of the world. Soil salinity affects more than 800 million hectares worldwide, equivalent to over 6% of all land on Earth There is continual pressure on plant breeders to develop higher yielding crop cultivars and adapted varieties to stress environment to sustain food security under climate change ad limited arable land. Plant mutation breeding has great potential to meet these demands. However, screening method is often a bottleneck for the ability to select mutants carrying desired traits. PBGL has published a book on pre-field screening of mutants for salt tolerance in rice, wheat and barley. The book of protocols has three main sections: (1) a brief introduction to the problem of soil salinity, (2) a protocol for measuring soil salinity, and (3) a protocol for screening for salt-tolerant cereal genotypes. The protocols are aimed to assist plant breeders and especially breeders who need to screen cereal populations, such as mutant populations of rice, wheat and barley, for salt tolerance. The protocols are designed to be effective, low cost and user friendly. The booklet provides simple and quick methods for soil sampling and analysis for water-soluble salt content, both of which are critical for the downstream screening. With these easy-to-follow protocols, users can conduct analyses in a quick and effective manner. Simple and quick methods are also provided to screen seedlings for salt tolerance in hydroponics. The seedling test takes 4–6 weeks and allows the screening of several hundred seedlings. The test can be used to screen segregating populations, standard lines and cultivars, as well as M_2 populations and advanced mutant generations. The book is open access and can be downloaded free at http://www.springer.com/us/book/9783319265889

The PBGL furthering expanding its R&D of the screening protocols by adapting the protocols to other crops and expanding the scale of the protocol by expanding the range of the stress and the time of exposure and by validate the screening methods under soil condition and for extended time until seed setting.

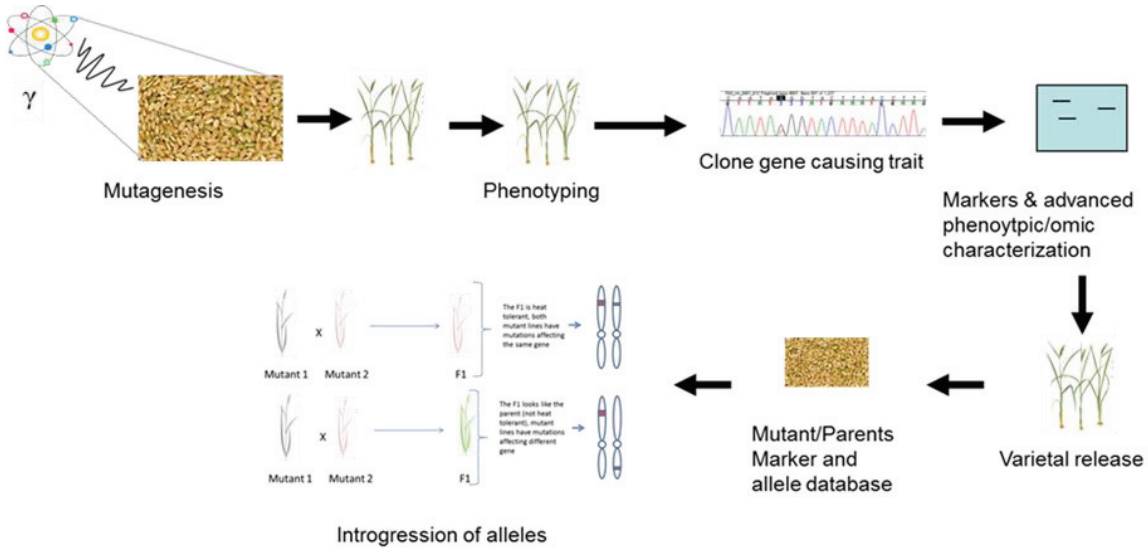


Comparisons of screening under hydroponic and soil condition for salt tolerance under continuous salt stress at concentrations (0, 5, 10 and 15 dS/m).

Screening for Induced Mutations using Molecular Methods

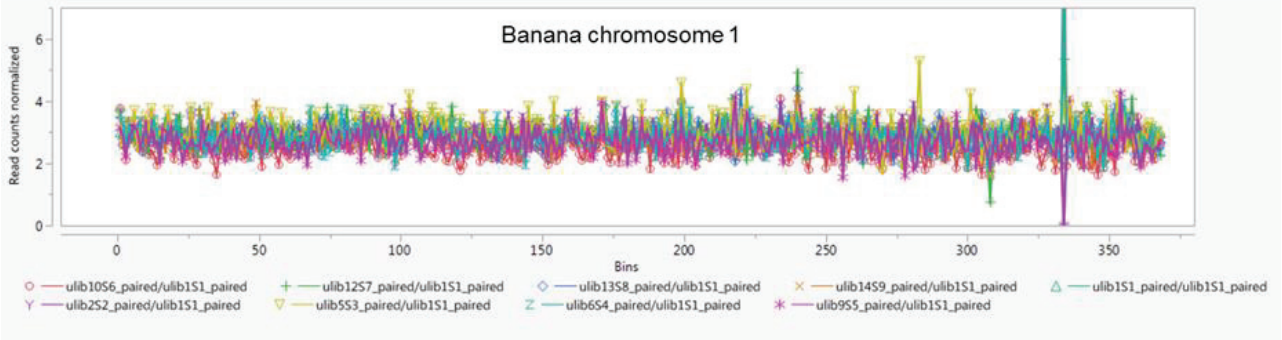
Readers of the newsletter will know that the successes of mutation breeding as exemplified by the over 3200 officially released mutant varieties are oft-lauded, and rightly so. The genetic improvement of crops has led, amongst others, to improved yield and quality, better resilience to climate change, improved disease resistance. Yet, in defence of scepticism, how induced mutations are identified and used deserves a critical evaluation. On one hand, we know that released varieties have made a tremendous impact. On the other hand, we don't know why. More specifically, we don't know how the mutations have affected the plant genome to cause the improved

trait. Knowing the mutation (or causative allele) is important for several reasons. First, adding to the knowledge of gene and gene function will advance the field of plant science. Second, identification of the causative alleles can lead to direct practical improvement of mutation breeding. So-called perfect molecular markers can be created to speed-up breeding and also allow rapid introgression of mutant alleles into other germplasm. This latter step would reduce the need for making new mutant populations in different countries where the same trait improvement is sought, thus saving many years and much money. To promote this approach the Plant Breeding and Genetics Laboratory a concept for allele discovery and introgression (see figure).



A pipeline for allele discovery and introgression. Mutation breeding follows standard steps of irradiation followed by phenotypic selection. Once traits are stable (M4 or higher), next generation sequencing is performed to identify genes causing the improved traits. Markers are developed and used to speed-up the release of a new variety. A database is created containing information on mutant alleles, traits and location of seed. When the same trait is desired in a different genetic background, a crossing plan is generated for the introgression of the allele. A new mutant population is not required, saving both time and money.

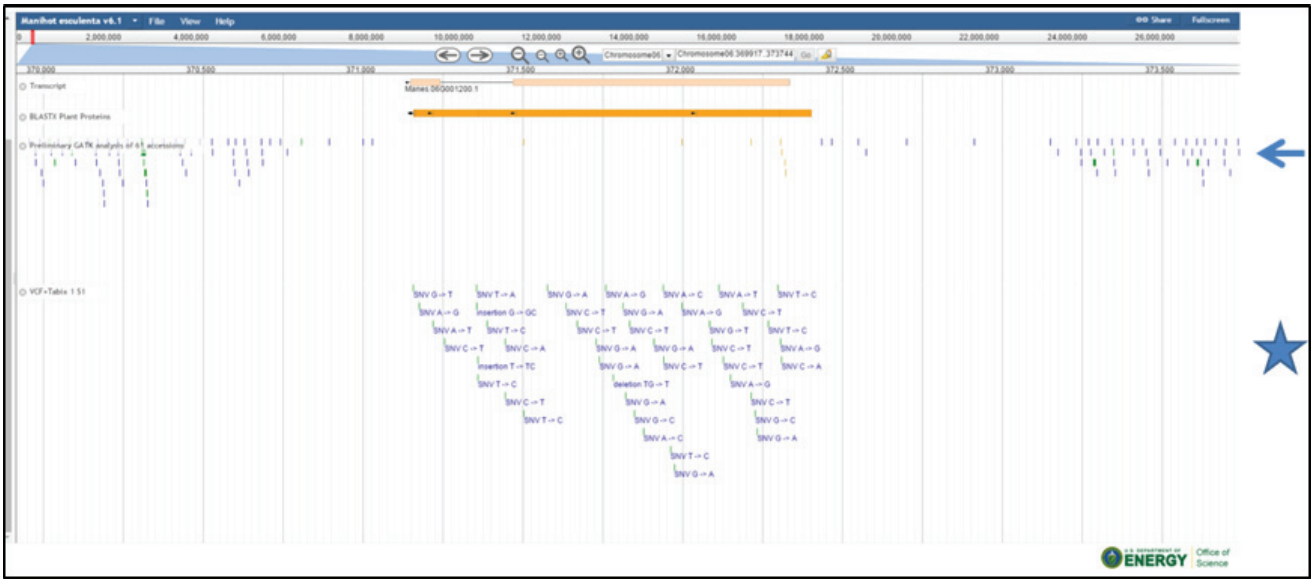
An important step is the cloning of alleles and development of perfect markers. This involves the acquisition of genomic sequence from mutant and non-mutated plants, bioinformatic comparison of the two data sets, development of molecular markers, and validation of molecular markers. The PBGL has been developing different strategies for the discovery of induced mutations in crops. In previous newsletters we described the use of exome capture sequencing for recovery of gamma induced mutations in sorghum, and whole genome sequencing methods for recovery of irradiation induced mutations in rice. Sequencing and analysis methods have been established in the PBGL. With this in hand, we are now adapting the methods for polyploid species. We are using banana as a model triploid. This work further supports the CRP "Efficient Screening Techniques to Identify Mutants with Disease Resistance for Coffee and Banana", where we seek to measure the rate of induced genomic lesions in isolated meristems so that we can better optimize the mutagenesis procedures. We have now collected preliminary data on gamma irradiated bananas in our NGS facility and have begun in-house analysis of the data (see figure).



A graphical representation of MiSeq reads from banana chromosome X. Colors represent different control and mutated samples.

Extra Budgetary Funding to Support Improvement of Cassava

In the last newsletter we described the beginning of a collaborative research project between PBGL and CIAT (International Center for Tropical Agriculture) for the genotyping of cassava accessions held at CIAT. Financial support for this work is provided by the Colombian granting agency COLCIENCIAS. Since the last report PBGL has collected data from 1728 cassava accessions using the MiSeq. To facilitate this we developed a pipeline for genomic DNA production, gene selection and sequencing. Sequencing was carried out in the PBGL NGS facility using the Illumina MiSeq. Preliminary data analysis suggests the strategy we developed was successful and we recovered over 7000 SNP and Indel variants, including unique variation not previously discovered (see figure).



Visual evaluation of variants and comparison with resequencing data on Phytozome. Region marked with an arrow represents publically available data from resequencing 61 African accessions. Star indicates data collected in PBGL. The orange bar at the top represents exonic sequence.

Kits

The Plant Breeding and Genetics Laboratory has developed positive control kits to assist Member States in optimizing PBGL protocols in their own laboratories for their own species. Each kit contains a detailed protocol along with the material needed to successfully complete the protocol. Kits are available upon request.

Kits distributed since the last newsletter:

- Low cost DNA extraction kits distributed to Iran, Islamic Republic of.
- Low cost enzyme extraction for mutation discovery: Indonesia, Pakistan, Thailand.

Professional Networking

The PBGL thanks all of you who have connected with us on LinkedIN If you haven't already linked with us, please feel free to connect (http://at.linkedin.com/pub/iaea-plant-breeding-and-genetics/31/4b6/aa3). We are now linked with354 researchers and are happy to continue to expand our connections to the broader plant sciences and agricultural community.

Human Capacity Development

Group Training on Next Generation Sequencing for the Discovery of Gamma Induced Mutations in Rice

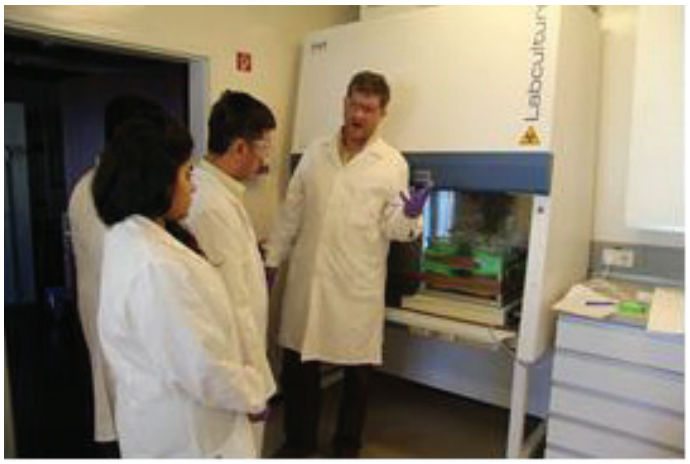
An *ad hoc* training course on whole genome sequencing of rice was held at the PBGL from 8 to 16 February 2016.

The course was organized by Mr B. Till and Ms S. Datta. Course topics included genomic DNA fragmentation, library preparation and next generation sequencing. Visitors from Madagascar, Sierra Leone, and Thailand attended the course.

Top: Whole genome library preparation using the Illumina TrueSeq Nano DNA library preparation kit is being demonstrated. From left to right: Sneha Datta, Reunreudee Kaewcheenchai, J.V. Sesay and Harimialimalala Jhonny Rabefiraisana. Bottom: Quantification of NGS libraries using the Fragment Analyzer is explained. From left to right: Reunreudee Kaewcheenchai, J.V. Sesay, Harimialimalala Jhonny Rabefiraisana, Bradley Till, and Sneha Datta.

Group Training on EMS Mutagenesis of Barley

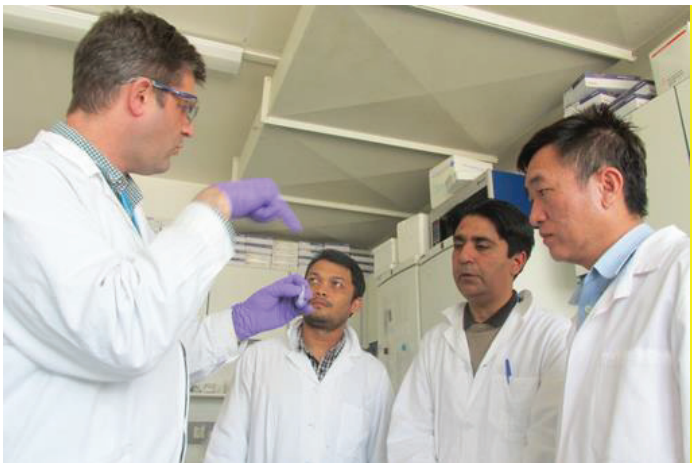
An *ad hoc* training course on chemical mutagenesis of barley seed was organized by Mr B. Till from 12 to 14 April 2016. The training covered a standard protocol for treatment of seed with EMS and post-treatment handling. Visitors from Pakistan and Thailand attended the course.



The principles of chemical mutagenesis are described in a dynamic fashion.

Group Training on Low-Cost Purification of Single-Strand-Specific Nucleases for Mutation Discovery

An *ad hoc* training course was organized by Mr B. Till from 25to 26 April 2016 covering low-cost protocols developed at the PBGL for, bench-top enzyme purification and mutation discovery. Training focused on extraction of enzyme from mung bean sprouts as this is readily available in the trainees' home countries. Visitors from Indonesia, Pakistan, and Thailand attended this course.



Group training on methods for purifying enzyme from mung bean sprouts for plant mutation discovery. From left to right: Bradley Till, Yassier Anwar, Farooq Azam, and Vichai Puripunyavanich.

Irradiation Services

Requests for Crop Irradiation Services by the Member States that is provided by the PBGL continue to be in high demand. At the time of writing 25 requests have been processed covering a minimum of 32 different crop species (see table) with the total number of requests now reaching 1465. For each new species and also upon request from a Member State, we carry out a radiosensitivity testing to determine the optimal dose for mutation induction. After irradiation, the M_1 seed or propagules in case of vegetatively propagated crops or trees, are returned to the Member State. To better serve our clients and improve our internal knowledge management, the Irradiation Service Request Form as well as the Guidelines for Sample Preparation have been updated during this period.

Request number	Country	Species
1441	Cambodia	Cassava
1442	Tanzania, United Rep. of	Maize, Barley
1443	Uzbekistan	Paulownia
1444	Germany	Various ornamental plants
1445	Cote D'Ivoire	Maize
1446	Sri Lanka	Onion
1447	Nepal	Rice
1448	Germany	*Salvia hispanica* (chia)
1449	Burkina Faso	Rice
1450	Oman	Date palm, Banana
1451	Cambodia	Cassava
1452	Sudan	Pearl Millet, Sorghum, Groundnut
1453	Sierra Leone	Cassava, Cowpea, Maize, Soybean
1454	Czech Republic	Barley
1455	Mongolia	Wheat, Oat, Rye, Barley, Soybean, Pea, Flax
1456	UK/India	Watermelon
1457	Germany	Various ornamental plants
1458	Niger	Sesame
1459	Spain	Citrus clementine
1460	Burkina Faso	Rice, Cowpea
1461	Sri Lanka	Mungbean, Soybean, Millet, Cowpea, Chilli, Onion, Sorghum, Horse gram
1462	Bulgaria	Wheat
1463	Eritrea	Barley
1464	Mauritania	Rice
1465	Germany	Various ornamental plants

Individual Training Activities

Name	Country	Status	Topic / Areas of Training	Period
Mr Yassier Anwar	Indonesia	Scientific Visitor	Mutation induction in barley, screening and accelerated breeding	April 2016
Mr Mohammed Jouhar	Syrian Arab Republic	Scientific Visitor	Development of low cost method for disease diagnostic	May 2016
Ms Sneha Datta	India	Intern	Plant mutation detection	October 2015–October 2016
*Ms Lina Kafuri	Colombia	Intern	Discovery of natural mutations in cassava	August 2015–February 2016
*Mr Daniel Tello	Colombia	Intern	Discovery of natural mutations in cassava	August 2015–February 2016
**Ms Prateek Gupta	India	Intern	Plant mutation detection	June–July 2016
Mr Harimialimalala Jhonny Rabefiraisana	Madagascar	Individual Fellow	Mutation detection in maize and rice	October 2015–February 2016

Name	Country	Status	Topic / Areas of Training	Period
Ms Reunreudee Kaewcheenchai	Thailand	Individual Fellow	Mutation detection in rice	October 2015–February 2016
Mr Romaric Nzoumbou-Boko	Central African Republic	Individual Fellow	Mutation detection in cassava	September–February 2015
Ms Geralde Gado Yamba Kassa	Central African Republic	Individual Fellow	Mutation detection in cassava	September 2015–February 2016
Ms Junatsu V. Sesay	Sierra Leone	Individual Fellow	Mutation induction	January–March 2016
Mr Farooq Azam	Pakistan	Individual Fellow	Mutation detection in wheat	March–May 2016
Mr Hamid Bachiri	Algeria	Individual Fellow	Mutation detection/phenotyping	March–May 2016
Mr Vichai Purupunyavanich	Thailand	Individual Fellow	Mutation detection/genotyping in rice	March–June 2016
Mr Zeremariam G. Mosazghi	Eritrea	Individual Fellow	Mutation detection/phenotyping	March–May 2016
Mr Alfred Ubalus	Nigeria	Individual Fellow	Mutation induction	April–May 2016

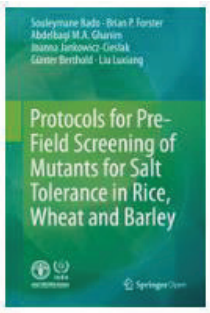
*Funded by CIAT (Centro Internacional de Agricultura Tropical) Cali, Colombia; **Funded by the University of Hyderabad, India.

Visitors to the PBGL

The PBGL continues to welcome visitors almost on a weekly basis. During 2016 until the time of writing, we have received about 25 visitor groups representing some 27 different Member States. These included high level visits from the EU, Japan, USA, Nepal, Bahrain and Bulgaria as well as a visit from the DG IAEA at the occasion of the inauguration of the Friedenstrasse, the new access road to the Seibersdorf campus. Selected members of the Standing Advisory Group on Nuclear Applications SAGNA visited PBGL on the occasion of their Eighteenth Meeting held at IAEA headquarters, Vienna. Several representatives of our host country, Austria, visited us on the occasion of the outreach event 'The Long Night of Research', amongst others. Visitors also included R&D organizations such as INRA, France, numerous representatives of the EU, a working group on Coffee Leaf Rust from 6 different Central American countries, as well as students and fellows from IAEA or universities from various Member States. These visitors showed great interest to learn more about plant mutation breeding, its global impact on food security and job creation, and how their countries could further benefit from PGBL activities and services, in the face of global challenges such as climate change.

Publications

Books



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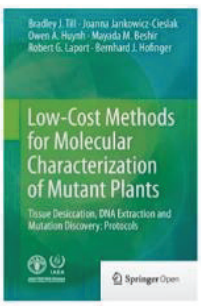
http://rd.www.springer.com/book/10.1007%2F978-3-319-26590-2.



TILL, B.J., J. JANKOWICZ-CIESLAK, O.A. HUYNH, M.M. BESHIR, R.G. LAPORT, B.J. HOFINGER (2015) Low-Cost Methods for Molecular Characterization of Mutant Plants: Tissue Desiccation, DNA Extraction and Mutation Discovery, A Protocol. Springer ISBN: 978-3-319-16258-4 (Print) 978-3-319-16259-1 (Online), ISBN: 978-3-319-16258-4 (Print) 978-3-319-16259-1 (Online).

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TILL, B.J., S. DATTA, J. JANKOWICZ-CIESLAK. TILLING: The Next Generation. In: Advances in Biochemical Engineering/Biotechnology. Rajeev K. Varshney et al. (eds.), Springer *(in press)*.

JANKOWICZ-CIESLAK, J., B.J. TILL (2016). Forward and Reverse Genetics in Crop Breeding. Chapter 8 in: Advances in Plant Breeding Strategies Volume In: Breeding, Biotechnology and Molecular Tools. J.M. Al-Khayri et al. (eds.), Springer.

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JANKOWICZ-CIESLAK J., B.J. TILL. Chemical Mutagenesis of Seed and Vegetatively Propagated Plants using EMS. Current Protocols in Plant Biology (*in press*).

JHURREE-DUSSORUTH, B., J. JANKOWICZ-CIESLAK, B.J. TILL (2016). Genetic Diversity Study of Dessert-type Banana Accessions in Mauritius using Low-Cost SNPs Detection Technology Acta Horticultura 1114. ISHS 2016. DOI 10.17660/ActaHortic.2016.1114.6 XXIX IHC – Proc. Int. Symp. Banana: ISHS-ProMusa Symposium on Unravelling the Banana's Genomic Potential Eds.: I. Van den Bergh et al.

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TILL, J.B. (2016). Mutation Discovery Technologies for Forward and Reverse-Genetics. International Conference on Plant Genetics & Breeding Technologies II, Vienna, Austria, 1–2 February 2016, Abstract Book pp.14.

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HOFINGER, B.J., R. ELIAS, M. JAWHAR, A. ALBATERNI, A. SKIHEITA, Y. BAKRI, M.I.E. ARABI, N.M. ALI, B.J. TILL (2016). Ecotilling as A Low-Cost Screening Method for Gene Variations in the Plant Pathogenic Fungus Cochliobolus sativus. International Conference on Plant Genetics & Breeding Technologies II, Vienna, Austria, 1–2 February 2016, Abstract Book pp.27.

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Websites and Links

- Plant Breeding and Genetics Section:

  http://www-naweb.iaea.org/nafa/pbg/index.html

- InfoGraphic for Mutation Breeding:

  http://www-naweb.iaea.org/nafa/resources-nafa/Plant-Mutation-breeding.mp4

- Mutant Variety Database:

  http://mvd.iaea.org

- IAEA Plant Breeding and Genetics LinkedIn:

  http://at.linkedin.com/pub/iaea-plant-breeding-and-genetics/31/4b6/aa3

- Joint FAO/IAEA Division of Nuclear Techniques in Food and Agriculture:

  http://www-naweb.iaea.org/nafa/index.html

  http://www-naweb.iaea.org/nafa/news/index.html

- Joint FAO/IAEA Publications

  http://www-naweb.iaea.org/nafa/resources-nafa/publications.html

- Food and Agriculture Organization of the United Nations (FAO):

  http://www.fao.org/about/en/

- FAO Agriculture and Consumer Protection Department:

  http://www.fao.org/ag/portal/index_en/en/

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