SOUTH ASIA
BIOSAFETY PROGRAM

October 2011
Vol.7 No.10

NEWSLETTER

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www.cera-gmc.org

SABP

The South Asia Biosafety Program (SABP) is an international developmental program initiated with support from the United States Agency for International Development (USAID). The program is implemented in India and Bangladesh and aims to work with national governmental agencies to facilitate the implementation of transparent, efficient and responsive regulatory frameworks for products of modern biotechnology that meet national goals as regards the safety of novel foods and feeds and environmental protection.

SABP is working with its in-country partners to:

- Identify and respond to technical training needs for food, feed and environmental safety assessment.
- Develop a sustainable network of trained, authoritative local experts to communicate both the benefits and the concerns associated with new agricultural biotechnologies to farmers and other stakeholder groups.
- Raise the profile of biotechnology and biosafety on the policy agenda within India and Bangladesh and address policy issues within the overall context of economic development, international trade, environmental safety and sustainability.

BIOSAFETY CLEARING HOUSE — A MAJOR LMO INFORMATION SHARING TOOL

Dr. Ranjini Warrier, Director, Ministry of Environment & Forests and BCH Focal Point for India - Email: warrier@nic.in

Biosafety Clearing House (BCH) is an information exchange mechanism established under Article 20 of the Cartagena Protocol on Biosafety (CPB). It comprises a central portal and a distributed network of external components/portals. The Secretariat of the Convention on Biological Diversity (SCBD) manages and hosts the central portal of the BCH, which is critical to the implementation of the Protocol and is intended to be a repository of up-to-date information on LMOs and biosafety. Information available on BCH is extremely useful for decision-makers in countries around the world as well as other stakeholders, specifically, scientists, research institutions, universities, biotechnology industries and enforcement agencies.

All signatories or ratifiers of the Protocol are obliged to make information available through the BCH. The Protocol requires that countries enter and manage their own data. It is therefore imperative that countries have the basic infrastructure as well as technical capacity including equipment, tools and practical know-how to fulfill their obligations and to take advantage of the benefits of the BCH.

The BCH provides a dynamic platform where interested users can retrieve and submit information to the BCH databases through a secure facility known as the "Management Centre". Access to the Management Centre is restricted to BCH national focal points (BCH-NFPs) and other national authorized users (NAUs) who are issued with login accounts and passwords. Registered information is validated (i.e., verified for accuracy and authenticity) by the BCH-NFPs before it is made publicly available, which helps to ensure the highest level of security and reliability of the information. Aside from the NFPs and NAUs, other stakeholders can also register reference records such as projects, publications, websites and training, which are validated by SCBD before being placed in the BCH.

Recently the Ministry of Environment and Forests organised a series of three BCH capacity building workshops customized for target stakeholders, mainly decision makers, enforcement agencies, scientists from the public and private sectors and industry. The first workshop, targeted at key government agencies, provided an understanding of BCH record formats, registration and publication procedures for biosafety-related decisions and discussed the issues/challenges in strengthening compliance to BCH requirements. The second workshop, for agencies involved in regulatory enforcement, provided an understanding of BCH and familiarized stakeholders with the transboundary movement of LMOs/GMOs with the modalities of using BCH as an information tool. The third workshop was to publicize the BCH and its utility to concerned central and state government representatives, research institutions, industries and other key stakeholders. The workshop's 135 participants were introduced to online e-learning tools for training and self-directed learning. UNEP deputized BCH regional advisors to assist in conducting the workshops. Relevant case studies were discussed to give participants greater understanding when using the BCH Central Portal to find and share information on LMO status; regulations, laws and guidelines; decisions and declarations. Overall, the workshops contributed toward improving the institutional expertise in BCH at a national level and better understanding and integration of requirements for registering national information on BCH.

BCH CAPACITY BUILDING WORKSHOP

OPPORTUNITIES AND CHALLENGES OF FISHERIES BIOTECHNOLOGY RESEARCH IN BANGLADESH

Prof. Dr. Md. Samsul Alam, Department of Fisheries Genetics and Biology, Bangladesh Agricultural University (BAU), Mymensingh, Email: samsul_bau@yahoo.com

The fisheries sector plays a significant role in the agro-based economy of Bangladesh. The role of this sector in the national economy increases daily. Fish contribute 3.74 per cent to GDP, 22.23 per cent to agriculture and 2.70 per cent to export earning as well as providing 58 per cent of national animal protein intake. The livelihood of about 10 per cent of the country is directly or indirectly dependent on this sector. The government has planned to increase fish production by 25 per cent by 2021. Since capture fisheries are static, aquaculture production must increase considerably to maintain the availability of fish and shellfish. Increased production is likely to come from more intensive culture, marine farming, sea ranching and an increased number of species. Genetic engineering of farm animals offers great potential for improvement of selected traits of economic significance. Several species of fish have been used to exploit this technology for commercial purposes. Examples include the attempted induction of freeze resistance in transgenic salmon using an antifreeze protein gene and production of growth enhanced fish such as salmon, tilapia, common carp and loach using novel growth hormone genes.

PROSPECT OF GENETIC ENGINEERING IN FISH

Fish have many advantages in genetic engineering and biotechnological research, namely,

i. large numbers of eggs laid, in some cases year-round;

ii. development is external;

iii. short generation times (for example only six months in tilapia);

iv. gene transfer through egg injection by glass needles is relatively easy in many species; and

v. availability of possible genome manipulation techniques such as triploidy and homozygous gynogenetic lines.

Most established gene transfer methods such as microinjection, electroporation, particle bombardment, liposome-mediation have been successfully used for gene transfer in fish.

Several traits have been introduced into fish. These include genes for growth enhancement, cold tolerance, disease resistance and traits to increase the ability to metabolise plants in the diet.

GROWTH ENHANCEMENT

Gene transfer in fish was initially attempted for two major reasons: growth enhancement and cold tolerance. Initially, growth hormone (GH) genes from mammals including humans were transferred however, with a view to increasing consumer acceptability, "all-fish" gene constructs (all components of the gene construct taken from fish) were designed and used to produce transgenic fish. Growth enhancement obtained thus far has varied from 2 times to an extraordinary 30 times that of the non-transgenic control fish. For example, 2.5 times growth and 20 per cent greater feed conversion efficiency was obtained in tilapia; 2-13 times increased growth in Atlantic salmon, 10-30 times in Coho salmon and 20-150 per cent increased growth in common carp.

COLD TOLERANCE

Cold-water fish have body fluids with the same freezing point as seawater (-1.7 to -2°C) and contain antifreeze proteins (AFP) that inhibit ice crystal growth and lower the freezing point of bodily fluids. Fish rich in AFPs include winter flounder, sea raven and ocean pout. Ocean pout type III AFP was successfully expressed and inherited in lines of goldfish and Atlantic salmon showing cold tolerance.

DISEASE RRESISTANCE

Disease is a major limitation in the aquaculture industry. The immune system is weakened by high-density culturing and stress. Use of antibiotics and vaccines are limited and labour intensive. Several antibacterial peptides have been isolated for genetic research. For example, Cecropins, small, antibacterial peptides first identified in the silk moth, are already being used in transgenic plants like tobacco and potato. Channel catfish and Japanese medaka genetically engineered with the Cecropin gene have shown increased disease resistance to several pathogens. Transgenic zebrafish with the chicken lysozyme (nonspecific antibacterial enzymes) gene also showed increased disease resistance.

METABOLISM OF LAND-BASED PLANTS

Plant ingredients are generally favoured over marine-based fish feed due to lower costs. However, use of plant ingredients introduces nutritional and metabolic problems. These diets are low in omega-3 fatty acids EPA (eicosapentaenoic acid) and DHA (Docosahexaenoic acid) and contain indigestible compounds such as phytate. Long-chain omega-3 fatty acids DHA and EPA are valued for their numerous health benefits. Plant oils contain the precursor ALA (α-linolenic acid). In fish, the enzyme n-6-desaturase is lacking, which is considered as the rate-limiting step in ALA conversion. So, fish cannot digest phytate and eventually excrete it as waste. Zebrafish with an n-6-desaturase transgene showed increased EPA (1.4 times) and increased DHA (2.1 times). The enzyme phytase breaks down phytate, the major form of phosphorus (P) in plants. The phytase gene from *Aspergillus niger* was recently expressed in Japanese medaka, which showed increased survival on diets rich in phytate.

DESIRED OUTCOME FOR TRANSGENIC RESEARCH

A stable integration of a single copy of the transgene into a single location in the genome, and not in a functional gene or regulatory element and germline transmission is desirable. However, integration of multiple copies at one locus or insertion of transgene at multiple locations in the genome is very common. Insertion of the transgene into a host gene may turn the host gene off, sometimes affecting the viability or health of the host. Expression of transgenes should have no undesired effects on the expression of other host genes or health of the host. Moreover, expression of a transgene can have pleiotropic effects, that is, effects upon multiple traits of the host resulting in morphological and metabolic abnormalities. Hazards stemming from insertional events can be identified by screening and managed by culling individuals that have desired events

Although Bangladesh is rich in fish biodiversity (260 freshwater fish species and 475 marine fish species), 12 exotic species have been introduced to boost aquaculture production. As there is a fear of escape of transgenic fish in flood-prone Bangladesh, it is possible to transfer economically important genes into exotic fish that have already adapted to the environment. For example, tilapia and common carp,

(continued on page 3 - see Bangladesh)

Bangladesh- *continued from page 2*

two important species with existing gene transfer methods can be used.

The exotic Nile tilapia (*Oreochromis niloticus*) was first introduced to Bangladesh in 1974. The popularity of tilapia as an aquaculture species is growing rapidly in Bangladesh as it is in many other countries. Because there is no indigenous counterpart to this species, transgenic tilapia cultured in Bangladesh brings no threat of gene transfer to other indigenous species. The gene transfer technique of this valuable fish species has already been developed. In Cuba, to assay the food safety of transgenic tilapia that contained and expressed tilapia GH transgene and grew twice as quickly as non-transgenic controls, 22 human volunteers were fed the transgenic tilapia twice daily for 5 consecutive days. No abnormalities in clinical and biochemical parameters were found in hemoglobin, total serum proteins, glucose, creatinine, cholesterol, leukocytes and erythrocytes content.

CHALLENGES OF FISH GENETIC ENGINEERING AND BIOTECHNOLOGICAL RESEARCH IN BANGLADESH

Genetic engineering research requires a good laboratory set up with high end equipment to conduct gene cloning, DNA sequencing, gene transfer and analysis of foreign gene integration and expression in the host organism and subsequent generations. Bangladesh research institutes and universities currently lack such facilities. Most efforts in biotechnological research have been devoted to various crop plant in Bangladesh. There is presently only one scientist who has earned a Ph.D. in gene transfer in fish working in Bangladesh (at BAU, Mymensingh). Therefore, human resource development with genetic engineering and biotechnological research capabilities is another big challenge facing fish genetic engineering research in Bangladesh. International collaboration and technical cooperation to develop capacity and capability of biotechnological research and human resources are highly desirable.

The Reading List

. . . new and notable articles

APPLICATION OF A HIGH-SPEED BREEDING TECHNOLOGY TO APPLE (*MALUS* X *DOMESTICA*) BASED ON TRANSGENIC EARLY FLOWERING PLANTS AND MARKER-ASSISTED SELECTION

Flachowsky H, LeRoux PM, Peil A, Patocchi A, Richter K, Hanke MV

Breeding of apple (*Malus* × *domestica*) remains a slow process because of protracted generation cycles. Shortening the juvenile phase to achieve the introgression of traits from wild species into prebreeding material within a reasonable time frame is a great challenge.

In this study, we evaluated early flowering transgenic apple lines overexpressing the BpMADS4 gene of silver birch with regard to tree morphology in glasshouse conditions. Based on the results obtained, line T1190 was selected for further analysis and application to fast breeding.

The DNA sequences flanking the T-DNA were isolated and the T-DNA integration site was mapped on linkage group 4. The inheritance and correctness of the T-DNA integration were confirmed after meiosis. A crossbred breeding programme was initiated by crossing T1190 with the fire blight-resistant wild species *Malus fusca*. Transgenic early flowering F(1) seedlings were selected and backcrossed with 'Regia' and 98/6-10 in order to introgress the apple scab Rvi2, Rvi4 and powdery mildew Pl-1, Pl-2 resistance genes and the fire blight resistance quantitative trait locus FB-F7 present in 'Regia'.

Three transgenic BC'1 seedlings pyramiding Rvi2, Rvi4 and FB-F7, as well as three other BC'1 seedlings combining Pl-1 and Pl-2, were identified. Thus, the first transgenic early flowering-based apple breeding programme combined with marker-assisted selection was established.

New Phytologist, (2011) 192(2):364-377.

BIOFORTIFICATION OF TOMATO (*SOLANUM LYCOPERSICUM*) FRUIT WITH THE ANTICANCER COMPOUND METHYLSELENOCYSTEINE USING A SELENOCYSTEINE METHYLTRANSFERASE FROM A SELENIUM HYPERACCUMULATOR

Brummell DA, Watson LM, Pathirana R, Joyce NI, West PJ, Hunter DA, McKenzie MJ

Methylselenocysteine (MeSeCys) is an amino acid derivative that possesses potent anticancer activity in animals. Plants that can tolerate growth on soils with high Se content, known as Se hyperaccumulators, do so by converting inorganic Se to MeSeCys by the enzyme selenocysteine methyltransferase (SMT). A cDNA encoding the SMT from a Se hyperaccumulator was overexpressed in tomato (*Solanum lycopersicum*). Transgenic plants were provided with selenite or selenate to the roots during fruit development, and liquid chromatography-mass spectrometry was used to show that MeSeCys accumulated in the fruit but not in the leaves. Depending on the transgenic line and Se treatment, up to 16% of the total Se in the fruit was present as MeSeCys. MeSeCys was produced more effectively from selenite on a percentage conversion basis, but greater accumulation of MeSeCys could be achieved from selenate due to its better translocation from the roots. MeSeCys was heat stable and survived processing of the fruit to tomato juice.

Journal of Agricultural and Food Chemistry, (2011). Sep 26. [Epub ahead of print]

CALENDAR OF EVENTS

Event	Organized by	Date and Venue	Website
INDIA			
Winter School on Introduction Molecular Breeding and Marker Assisted Selection in Rice Improvement	Central Rice Research Institute	November 1 - 21, 2011 Cuttack, Orissa	http://www.crri.nic.in/ or http://ow.ly/6W9N8
World Cotton Research Conference 5	International Cotton Advisory Committee and Indian Society for Crop Improvement under the aegis of Indian Council of Agricultural Research	November 7 - 11, 2011 Mumbai	http://www.wcrc-5.com/WCRC5_Circular.pdf
National Agricultural Innovation Project Sponsored Training Programme "Molecular Diagnostics for Risk Assessment and Management of Genetically Modified Crops"	National Bureau of Plant Genetic Resources	November 8 - 21, 2011 New Delhi	http://www.nbpgr.ernet.in/
3rd Global Conference on Plant Pathology for Food Security	Indian Society of Mycology and Plant Pathology and Maharana Pratap University of Agriculture and Technology	January 10 - 13, 2012 Udaipur	http://isompp.org/3gc.html
8th International Safflower Conference Safflower Research and Development in the World: Status and Strategies	Indian Society of Oilseeds Research and Indian Council of Agricultural Research	January 19 - 23, 2012 Hyderabad	http://www.dor-icar.org.in/downloads/Conference1.pdf
International Conference on Plant Biotechnology for Food Security: New Frontiers	Society for Plant Biochemistry and Biotechnology, National Research Centre on Plant Biotechnology	February 21 - 24, 2012 New Delhi	http://www.spbbindia.org/
INTERNATIONAL			
Workshop on Capacity-Building for Research and Information Exchange on Socio-economic Impacts of Living Modified Organisms Under the Cartagena Protocol on Biosafety	Secretariat of the Convention on Biological Diversity and Ministry of Environment and Forests (MoEF)	November 14 – 16, 2011 New Delhi	http://bch.cbd.int/protocol/meetings/
Asia-Pacific Regional Workshop on the Nagoya-Kuala Lumpur Supplementary Protocol on Liability and Redress to the Cartagena Protocol on Biosafety	Secretariat of the Convention on Biological Diversity and MoEF	November 17 – 18 2011 New Delhi	http://bch.cbd.int/protocol/meetings/
The International Conference of GM Crops	Faculty of Agriculture, Cairo University	November 20 - 23, 2011, Cairo University, Egypt	http://www.icgmc2011.com/
Asia Sub-regional Training of Trainers' Workshop on the Identification and Documentation of Living Modified Organisms	Secretariat of the Convention on Biological Diversity and Ministry of Environment and Forests (MoEF)	November 21 – 25, 2011 New Delhi	http://bch.cbd.int/protocol/meetings/
Regional Workshop on Field Trials and Post-Release Monitoring of GMOs	Ministry of Culture	December 5 - 7, 2011 Zagreb, Croatia	http://www.icgeb.org/meetings-2011.html
5th International Botanical Conference -- Climate Change and Biodiversity: Role of Plant Scientists	Bangladesh Botanical Society	December 9 – 11, 2011 Department of Botany, University of Dhaka, Bangladesh	www.bdbotsoc.org or http://www.dhakai.com/botany/Circular.pdf

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