Transgenics in crop improvement research at IITA

Leena Tripathi (l.tripathi@cgiar.org)

Harvested bunch of transgenic bananas, Kampala, Uganda. Photo by L. Tripathi.

Biotechnology has opened unprecedented avenues for exploring biological systems. Transgenics is one of the key techniques particularly useful for the genetic improvement of crops that are not amenable to conventional breeding, such as those that are vegetatively propagated. In IITA, transgenic technologies are being used for improving banana/plantain (*Musa* sp.), cassava (*Manihot esculenta*), and yam (*Dioscorea* sp.).

Genetic transformation platform

An efficient protocol for plant regeneration and transformation is a prerequisite for the successful use of transgenic technologies. Despite the technical difficulties in transforming monocot species, efficient transformation protocols that are embryogenic cell suspension based and *Agrobacterium* mediated have been established for many cultivars of banana/plantain. This system, however, is a lengthy process and cultivar dependent. Therefore, a transformation protocol using meristematic tissues was also established which is rapid and genotype independent. These protocols have paved a way for the genetic manipulation of banana/plantain by incorporating agronomically important traits such as those conferring resistance

L. Tripathi, Biotechnologist, IITA, Nairobi, Kenya.

to diseases or pests as well as tolerance to abiotic stress factors.

Agrobacterium-mediated transformation protocols for three popular cassava varieties preferred by African farmers were established through somatic embryogenesis. A regeneration and transformation protocol is also established for yam (*Dioscorea rotundata* and *D. alata*) using nodal explants, but transformation efficiency needs to be improved. A transformation protocol using somatic embryogenic callus for yam is under development.

Development of disease- and pest-resistant transgenic crops

Banana *Xanthomonas* wilt (BXW), caused by the bacterium *Xanthomonas campestris* pv. *musacearum* (Xcm), is the most devastating disease of banana in the Great Lakes region of Africa. In the absence of natural host plant resistance, IITA, in partnership with NARO-Uganda and the African Agricultural Technology Foundation, has developed transgenic banana by constitutively expressing the Hypersensitive Response Assisting Protein (*Hrap*) or plant ferredoxin-like protein (*Pflp*) gene from sweet pepper (*Capsicum annuum*). The transgenic plants have exhibited strong resistance to BXW in the laboratory and screenhouse tests. The best 65 resistant lines were planted in a confined field trial at the National Agricultural Research Laboratories (NARL), Kawanda, Uganda, for further evaluation.

Based on results from mother plants and their first ratoon plants, 12 lines were identified that show absolute resistance. The plant phenotype and the bunch weight and size of transgenic lines are similar to those of nontransgenic plants. These lines will be further tested in a multilocation trial in Uganda. They will be evaluated for environmental and food safety in compliance with Uganda's biosafety regulations, risk assessment and management, and procedures for seed registration and release, and are expected to be released to farmers in 2017.

Cassava brown streak disease (CBSD) has emerged as the biggest threat to cassava cultivation in East Africa. As known sources of resistance are difficult to introgress by conventional methods into the cultivars that farmers prefer, the integration of resistance traits via transgenics holds a significant potential to address CBSD. Of the available transgenic approaches, RNA silencing is a very promising strategy that has been successfully employed to control viral diseases. IITA, in collaboration with Donald Danforth Plant Science Centre (DDPSC), USA, is developing CBSD-resistant cassava for East Africa.

Nematodes pose severe production constraints, with losses estimated at about 20% worldwide. Locally, however, losses of 40% or more occur frequently, particularly in areas prone to tropical storms that topple the banana plants. IITA, in collaboration with the University of Leeds, UK, has generated transgenic plantain using maize cystatin that limits the digestion of dietary protein by nematodes, synthetic peptide that disrupts chemoreception, or both of these traits. These lines expressing the transgenes were challenged in a replicated screenhouse trial with a mixed population of the banana nematodes, *Radopholus similis* and *Helicotylenchus multicinctus*.

Many lines were significantly resistant to nematodes compared with nontransgenic controls. The promising transgenic lines showing high resistance will be planted in confined fields in Uganda for further evaluation in mid-2012.

Transgenic technologies for abiotic stress tolerance

Cassava roots undergo rapid deterioration within 24–48 hours after harvest, the so-called postharvest physiological deterioration (PPD), which renders the roots unpalatable and unmarketable. IITA, in collaboration with the Swiss Federal Institute of Technology (ETH) Zurich, is developing cassava tolerant of PPD through the modification of ROS (reactive oxygen species) scavenging systems. The potential is being assessed of various ROS production and scavenging enzymes, such as superoxide dismutase, dehydroascorbate reductase, nucleoside diphosphate kinase 2, and abscisic acid responsive element-binding protein 9 genes, to reduce the oxidative stress and the extent of PPD in transgenic cassava plants.

Future road map

Efforts at IITA over the last 10 years to establish transformation protocols for all the IITA crops have been paying off and have led to the establishment of a genetic transformation platform for cassava, banana/plantain, and yam—the three most important food crops in sub-Saharan Africa. These technologies have contributed to significant advances in incorporating resistance to pests and diseases in banana and cassava. Some of these technologies have the potential to offer additional benefits. For instance, the transgenic technology to control *Xanthomonas* wilt may also provide an effective control of other bacterial diseases of banana (*Moko*, blood, and *bugtok* diseases), and of bacterial blight in other crops such as cassava and cowpea.

Transgenic technologies provide a platform for controlling diseases in banana, cassava, and cowpea. Photo by IITA.