



Nitrogen-Use Efficient, Water-Use Efficient and Salt Tolerant Rice Project

ANNUAL PROGRESS REPORT 2012

Message from the Project Coordinator



Rice consumption in Sub-Saharan Africa (SSA) has been growing by 6 percent per annum over the years. This is more than double the rate of population growth resulting to demands that far exceed local supply in SSA. The rising demand for the

commodity has been largely attributed to changing food preferences in both urban and rural areas coupled with high population growth rates and rapid urbanisation in Africa. This demand and consumption rate indicates that rice is an important staple food and a commodity of strategic significance across much of Africa, requiring specific interventions that target supply (production) constraints.

The Nitrogen-Use Efficient, Water-Use Efficient and Salt Tolerant (NEWEST) Rice project was launched in 2008 as a strategic pathway to addressing food insecurity in the face of many abiotic constraints facing rice production and the impending challenge of climate variability in Africa. The initiative strives to genetically transform some varieties of the New Rice for Africa (NERICA) using plant transformation technologies to improve their productivity in nitrogen-deficient soils, drought prone regions and in soils with high salinity. The goal is to provide smallholder rice farmers with higher yielding varieties that are well adapted to marginal agricultural production conditions that characterise many areas in Africa.

The NEWEST Project is at the product development stage

and all the required trait technologies have been negotiated and accessed. Financial limitations at AATF resulted in cutting out some project activities such as dropping transformation work on lowland rice and changing the original plans with Arcadia Biosciences and the National Agricultural Research Systems (NARS). Delays in approval of the confined field trial (CFT) applications in the countries also contributed to delayed implementation.

Despite the few challenges and delays, the Project has achieved significant milestones. In 2012, the product development partner in the project, Arcadia Biosciences, generated and characterised 12 transgenic NUE Nerica-4 T2 lines (pipeline 1 - NUE1.1 (pPIPRA543 + pARC321) & Pipeline 2 - NUE1.1 (pPIPRA543 + pARC163)-NPTII) ready for shipment to the partner countries in SSA for testing. Through negotiation by AATF, the project was granted technology license agreements for use of Agro bacterium-mediated plant transformation technology known as 'PureIntro' by Japan Tobacco. This will improve the efficiency in generating transgenic lines and thus save time in product development. The country partners Uganda and Ghana - completed land preparation and confined field trial (CFT) site construction in readiness for the tests. Uganda and Ghana were both granted approval by their national biosafety committees to conduct CFT's. These developments are encouraging and the project partners are gearing up for more field activity in 2013.

I would like to take this opportunity to thank the project partners and investors for their support in the implementation of these project activities. Special gratitude goes to each individual who has made contribution towards propelling the project to where it is now.

Dr Jacob H Mignouna

About the NEWEST Rice Project

The NEWEST Rice project aims to develop and deploy farmer preferred and locally adapted genetically improved African rice varieties with enhanced agronomic traits, specifically nitrogen-use-efficiency (NUE), water-use-efficiency (WUE) and salt tolerance (ST). Rice production in Africa has been characterised by low yield. Average grain yield is as low as 2.2mt/ha, compared to a potential yield of 7mt/ha. This low yield has been attributed to several abiotic factors. Drought and nitrogen deficiency have been cited as leading constraints to upland rice production. About 80 percent of African rice farms traditionally depend on rainfall as farmers are poor, and cannot afford to install irrigation systems or buy much fertilizer. High salinity is increasingly becoming a major problem in many rice growing areas of Africa. The NEWEST rice project offers a window of opportunity to addressing these constraints.

The project promises a rice variety (NEWEST rice) that is associated with high economic advantages as it is expected to reduce the amount of money spent on providing irrigation facilities and use of fertiliser thus lowering cost of



Mrs Florence Apochi a smallholder rice farmer in Osudoku Asutsuare, Ghana one of the NEWEST Rice Project countries

production. The variety can offer environmental gains as it promises a reduction in the use of chemical fertilisers (expected to reduce from 200kg/ha to 9kg/ha). It will also require less water and thrive on degraded soils (nitrogen depleted soils). The project is therefore very important for SSA as it has great potential to increase rice production, improve incomes for farmers and strengthen national and household food security.

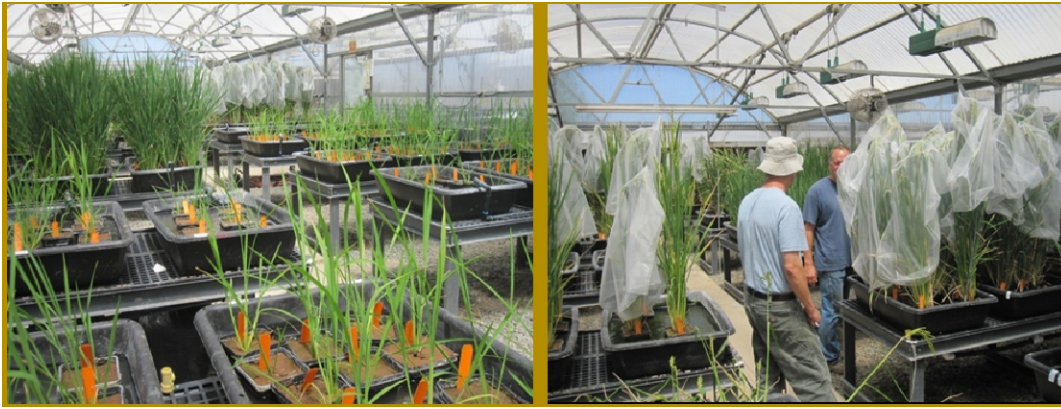
Project Activities and Key Milestones

The main project activities during the year included negotiating and accessing trait technologies, product development to produce transgenic plants, field testing for trait gain and the general management of the project:

Technology access and agreements

A royalty-free non-exclusive license that allows access to the licensor's *Agro bacterium*-mediated plant transformation technology known as 'PureIntro®' was granted to AATF by Japan Tobacco. This license was received in March 2012 after a series of negotiations with Japan Tobacco that began in 2011. The PureIntro® is an agrobacterium-mediated plant transformation technology that is recognised worldwide as the standard transformation system. The technology reduces development costs and time. PureIntro® has been licensed by Japan Tobacco to around 50 private and public entities worldwide for numerous crops including maize, rice, wheat, barley, sorghum, banana, sugarcane, switchgrass, miscanthus and turf.

Earlier on, transformation vectors from Arcadia Biosciences: pARC321 (gene conferring nitrogen-use efficiency); pARC609 (which carries the triple gene stack NUE-WUE-ST), and DNA constructs from The Regents of the University of California, Davis which include the binary transformation vectors, pPIPRA543 and pPIPRA549, had been negotiated and accessed. These licensed materials have been very useful in the transformation process.



One of the two greenhouses that was adapted especially for growing tropical rice. The improvements consisted of implementing re-circulating water system, fogging system, increased supplemental lighting and quality of water source. Left: tubs with re-circulating water system for growing Nerica plants. Right: bagging of flowering panicles to prevent pollen dispersion

Product development to produce transgenic plants

The transformation of rice using the licensed materials is being conducted at Arcadia Biosciences (the product development partner in the project). Significant progress has been achieved in developing transgenic events carrying the NUE, and the triple gene stacked NUE-WUE-ST traits. The transformation involves three pipelines and the achievements so far are as follows:

a) Transformation Pipeline 1 - NUE lines generated with pARC321/pPIPRA543 co-transformation. On this pipeline, molecular quality control on all T₂ lines coming out of this transformation pipeline has been finalised. Six Nerica-4 T₃ NUE lines have been generated and characterised. These are available for field observation and are awaiting shipment to the National Agricultural Research Organisation (NARO), Uganda, as soon as the seed import permit is received.

b) Transformation Pipeline 2 - Co-transformation with pARC321/pARC163 (repaired pPIPRA543) is phenotypically the same with pipeline 1, but derived differently using a combination of different vectors. The lab analysis work is progressing well, and T₂ seeds are available. T₃ seed production is in process. It is expected that 17 NUE lines will be ready by January 2013.

c) Transformation Pipeline 3 - Co-transformation with pARC609/pARC163 (TGS approach). Two methods of segregation are used - pipeline 3A and pipeline 3B. Pipeline 3A: From the co-transformations

performed between Feb 2011 - 2012, using pARC609/pARC163, over 1200 KanR Nerica-4 T₀ plants were generated and genotyped. In this group, 6 lines that are out-segregating the selectable marker (SM) and 9 in which the gene of interest (GOI) and SM cassettes are linked have been identified. These 15 lines are single copy insertion for both GOI

and SM. For part of these lines, T₂ seed has already been harvested; next step is zygosity analysis on T₂ plants. Six marker-free lines and nine marker-bearing T₂ lines are expected in the first quarter of 2013.



The CFT site in Uganda: up is the seed store, office and drying platform and below is the field with fully installed drip irrigation facility

Pipeline 3B: Transformations with pARC609/pARC163 done after February 2012 have generated some 600 T0 plants thus far. Some T1 seeds have already been harvested, while other shoots are still in tissue culture.

Field testing for trait gain

Parallel to product development, a number of activities were conducted at the NAR's in the two pilot countries of Uganda and Ghana in preparation for CFT's. These included application for regulatory approvals and compliance, field/site preparation, nitrogen depletion with maize and soils analysis.

Uganda: The CFT site construction has been concluded and inspection carried out by the National Biosafety Committee (NBC). The final decision document to conduct the CFT has also been issued by the NBC and the plant import permit application submitted. Planting of the first CFT is planned for December 2012.

Ghana: The CFT site development is in its final stages. Approval to conduct CFT was granted in November 2012. In preparation for the trials, a CFT Compliance Management training workshop was held for project staff to refresh on the theory and practice of regulatory compliance for GM crops in readiness for importation of seed and planting of the trial.

Project Management

The Project Advisory Committee (PAC) provides overall technical and strategic backstopping of the project implementation. During the year, the committee held monthly teleconferences to review project progress.

Major issues discussed during the teleconferences included product development, preparations for confined field trials and regulatory compliance.

The NEWEST Rice Project Partnership

Various parties are collaborating to achieve this goal:

- AATF is coordinating project activities throughout the entire product value chain (Intellectual property management, business development, project management, regulatory affairs and communication and outreach), ensuring that activities are carried out as planned
- Arcadia Biosciences is donating the NUE, WUE and ST trait technologies, producing transgenic plants and providing technical support;
- Public Intellectual Property Resource for Agriculture (PIPRA) is donating the enabling technologies for plant transformation;
- International Centre for Tropical Agriculture (CIAT) is carrying out seed multiplication and early agronomic trials
- National agricultural research partners of Ghana, and Uganda together with the International Center for Tropical Agriculture (CIAT) are involved in field testing for trait gain.
- The project is funded by the United States Agency for International Development (USAID).

Project Partners



African Agricultural Technology Foundation P. O. Box 30709-00100, Nairobi, Kenya Tel + 254 20 422 3700 via USA:
Tel +1650 833 6660 3700 Fax +1 650 6661 3701 Email: aatf@aatf-africa.org Web: www.aatf-africa.org

African Agricultural Technology Foundation, Nigeria Office: David Towers, Number 3, Idris Ibrahim Crescent (behind Mr. Biggs) Jabi, Abuja.
Tel +234 9 870 1684, +234 803 700 6483, +234 7067 775 048 Email: aatf@aatf-africa.org Web: www.aatf-africa.org