Kirkhouse Times

No. 8 November 2018

# The KT-funded dry bean project at KALRO Kakamega makes a difference *by Dr Reuben M. Otsyula*

**Highlights**



**An overview**

The mandate of Kenya Agricultural and Livestock Research Organization (KALRO) is to conduct agricultural research in Kenya. The KALRO Non-Ruminant Institute is based at Kakamega, 500km west of Nairobi. Apart from conducting non-ruminant research, the institute works on various food crops grown in its mandate region. The region is characterized by high population density ranging from 261 to 1044 persons per hectare and annual bimodal rainfall of 1650 to 2200mm. The mandate region is divided into a number of distinct agro-ecological zones, namely, lower highlands (LH2-LH4), medium altitude (UM1-UM4) and lower midlands (LM1-LM4). The region is dominated by subsistence-oriented mixed crop-livestock farming in which the major food crops are maize, beans, cassava, sweet potato, banana, sorghum and finger millet, along with a number of both local and exotic vegetables. Although the region has high to medium potential agricultural productivity, soil fertility is declining due to continuous cultivation on small pieces of land. These conditions have encouraged the build-up of crop pathogen inoculum, resulting in a high disease incidence in both beans and other crops. Despite low levels of productivity, farmers do rely on the income derived from selling food crops. A significant proportion of households are not self-sufficient in food and rely on the market to alleviate food shortages. Local poverty is driving the export of labour out of the region, particularly adult males, resulting in a high prevalence of female-headed households (40%). Since female-headed households tend to have less access to land, labour, capital and information than male-headed ones, there is a need to develop technologies which are cost effective and do not overly rely on manual labour.

**Figure 1.** KALRO—Non Ruminant Research Institute, Kakamega, Kenya.

For social and climatic reasons, the region provides a suitable environment for screening crop germplasm for resistance to both biotic (diseases and insect pest) and abiotic stresses (low soil phosphorus and low soil nitrogen). The environment suits the model sought by KT for funding research aimed at developing multiple disease resistant bean varieties able to be cost effectively grown. This aim is consistent with the overall goal of the KALRO bean breeding programme which seeks to "increase and sustain productivity per unit area through the development and selection of improved bean varieties and other production technologies in order to improve food security, nutrition and household income, and hence contribute to poverty alleviation among resource-constrained farmers while conserving the natural resource base".


**Figure 2.** Commercial varieties resistant to bean root rot but infected by other diseases in farmers fields.

Over several years, conventional breeding carried out at KALRO Kakamega has succeeded in releasing a number of high yielding commercial varieties resistant to bean root rot and well suited to the requirements of the home consumer, the grain market and the processors. However, these varieties are typically susceptible to anthracnose, common bacterial blight (CBB) and bean common mosaic (necrotic) virus (BCM(N)V). The co-existence of multiple diseases on farmer's fields has created the

*Dr Reuben M. Otsyula is the Head of Grain Legume Research, KALRO, Kakamenga, PI KT funded project KALRO dry bean project.*

need to introduce genes encoding resistance to these pathogens into a market-class bean variety. With this in mind KT is supporting the KALRO programme by sponsoring the use of marker assisted selection to accelerate the introduction of resistance against the major diseases. To facilitate this, KT has created a molecular laboratory at KALRO Kakamega for marker genotyping, complemented by carrying out disease evaluations in the screen house and the pathology laboratory equipped by KT. The lab is used to culture the important pathogens of bean, and is considered to be the best such facility in western Kenya.

Some of the outputs of phase one of the KT funded bean breeding program are:

**1. Plant pathology research supported KT at KALRO Kakamega**

This project was designed to include a strong component of crop protection and especially plant pathology. Since KALRO Kakamega had only one in house plant pathologist, responsible for more than 12 crops, KT was persuaded to support an MSc pathology student as part of its contribution to capacity building and to service the KT-funded breeding programme. The agreement of KT to fund a full scholarship was received by KALRO with delight: the Centre's director mentioned during the 12th annual ABC meeting at Kakamega (June 17th - 23rd, 2018) that "we are an aging population of scientists at the national program and anything that KT has contributed in terms of training our young people who would replace us is incredibly appreciated".

Yona Masheti is pursuing an MSc in plant pathology at the University of Nairobi. He has been characterizing 18 bean varieties for their disease reaction and adaptation to different micro environments. He reports: "A management method which reduces the need for inputs is both environmentally friendly and advantageous to smallholder farmers." The diseases reported from his study were anthracnose, scab, angular leaf spot (ALS), rust, floury leaf spot, CBB, halo blight, bacterial brown spot and the viral diseases yellow mosaic and BCM(N)V. This is the first time the importance of scab disease (caused by the fungus *Elsinoe phaseoli* pv. Jenkins) relative to other major diseases has been highlighted. The study also identified varieties Red 16 and KKCal 33 as the best adapted to various environments.

**2. The KT-funded molecular laboratory at KALRO Kakamega**

The KT lab at KALRO Kakamega has now been established. In the words of Dr. Stanley Nkalubo, the bean programme coordinator in Uganda, when visiting during the 12th ABC meeting "The lab is small and cute". Since its establishment, it has been used to implement various markers in the bean breeding programme. We have hired Shadrack Odikara to act as the laboratory technician as well as a laboratory manager.

**3. Pyramiding disease resistance genes into a market acceptable bean varieties through MAS (**Allan Shivachi, PhD student)

My project aims to introgress resistance to anthracnose, BCMV/BCMNV and CBB into the root rot resistant variety KKCal194, using the parallel backcrossing strategy. The target genes for CBB are marked by SU-91 and SAP-6, the anthracnose resistance gene *Co-4*[2] by SBB-14, and the BCM(N)V resistance gene *bc-3* by ROC-11 and ElF4E. So far, $BC_4F_1$ progeny have been generated for each target, and are being combined into a single population to allow for the selection of multiple disease resistant lines. Some sample gels used in the marker assisted selection procedure are shown in figure 3.

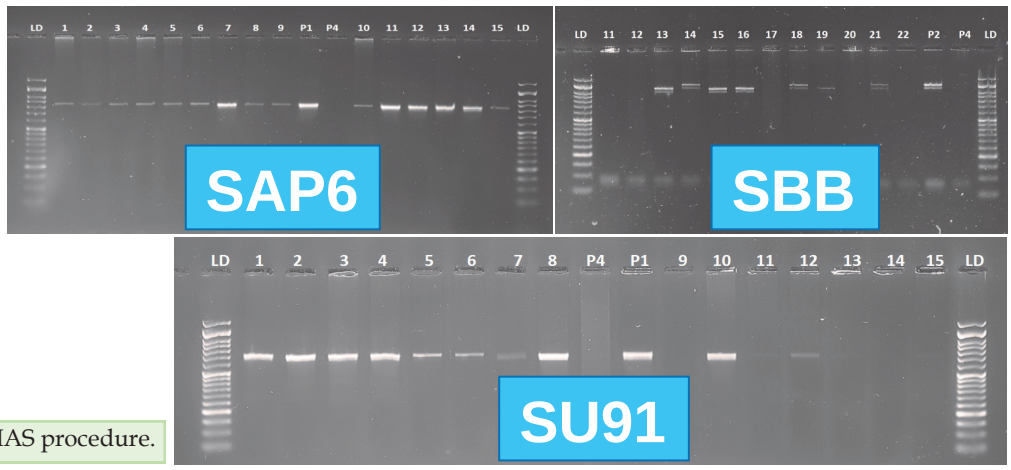



**Figure 3.** Gels used in MAS procedure.

Yona Masheti is an MSc in Plant pathology at the University of Nairobi, Kenya.

Allan Shivachi is a PhD student at the University of Nairobi, Kenya.

## Visit to One Acre Fund *by Dr Becky Lockyer*

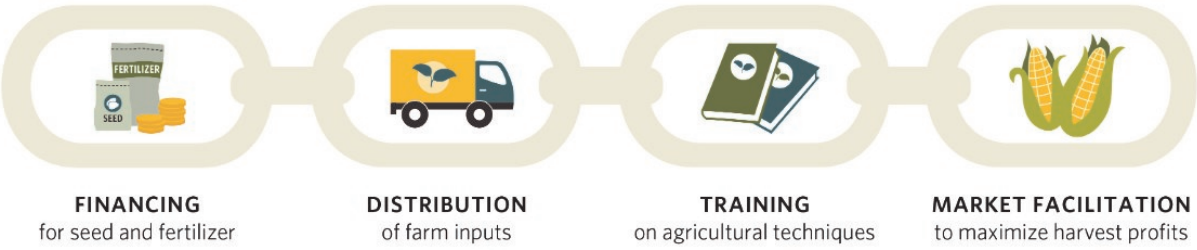
**Figure 1:** HQ building.

**Figure 2:** Products on offer at reception.

One Acre Fund is a non-profit organisation which supplies smallholder farmers in East Africa with inputs, financing and training, enabling them to increase their yields, becoming more prosperous, and subsequently helping to reduce poverty and hunger. In June 2018, ABC PIs, KT consultants and staff visited the new headquarters of the One Acre Fund in Kakamega, Kenya, which was opened in 2017 to accommodate the organisation's rapid growth, moving from their former location in Bungoma (two hours drive away).

In Kenya, One Acre Fund distributes a basic package of maize and fertilizer to farmers. Farmers are then free to choose their own "add-ons", which can include bean seed, onion seed, collard greens ('sukuma wiki' in Swahili) seed, PICS bags, cooking stoves and crop insurance. The predominant bean variety currently distributed in Kenya by One Acre Fund, KK8, was initially bred by the ABC PI Dr Reuben Otsyula, who is based at the Kenya Agricultural & Livestock Research Organization (KALRO) Non-Ruminant Research Institute in Kakamega.

On 16th June, we were met in the Kakamega Headquarters auditorium by Jakob, Joseph and Joel, who (on a Saturday morning, no less) kindly talked us through the work carried out by One Acre Fund and answered questions from curious bean breeders and KT consultants.




*One Acre Fund Model (oneacrefund.org/what-we-do/our-model/)*

The One Acre Fund model is centred on market-based strategies and has four key steps:

**Asset-Based Loans.** Farmers receive high-quality seeds and fertilizer on credit, and are offered a flexible repayment system to pay back their loans.

**Delivery.** One Acre Fund aim to deliver inputs to locations within walking distance of the farmers they serve.

**Training.** Farmers receive training throughout the season in modern agricultural techniques.

**Figure 3:** Meeting smallholders.

**Market Facilitation.** One Acre Fund offer crop storage solutions and teach farmers about market fluctuations, so that they can time crop sales to maximize profits.

The money farmers repay is channelled back to the farmers; by 2025 One Acre Fund hopes to be self-sustaining, without the need for further donor funding. Very few farmers default on their loans – farmer groups are sanctioned if members default,

## Awards

- Congratulations to **Distinguished Professor Paul Gepts** on receiving the 2018 Chancellor's Award for International Engagement at UC Davis on 2 March 2018. Paul Gepts is a '**Distinguished Professor**' in Plant Sciences.

- Congratulations to **Dr Idah Sithole-Niang** of the University of Zimbabwe, on being elected as a fellow of The World Academy of Sciences (TWAS) for the advancement of science in developing countries, as of 1 January 2018.

## *Cont'd… by Dr Becky Lockyer*

**Figure 4.** Hand thrashing and its products.

which leads to community policing (if a farmer group does not collectively repay its outstanding credit, none of the individual members will be eligible to re-enrol in One Acre Fund the following season).

We were delighted to meet with a group of smallholders who have benefited from working with the Fund. Indeed, in Kenya an average of $125 of profit was gained per farmer in 2016. The farmers talked to us about the benefits of working with One Acre Fund and demonstrated a traditional (and very energetic!) technique for threshing beans. They also generously supplied us with tea and sandwiches, which were gratefully received! The ABC PIs were interested to note how the farming preferences and methods employed in Western Kenya compared to those of their own countries.


**Figure 5.** (*Top*) Research site bean varieties. (*Middle*) Mike, Luseko, Annuarite, Reuben, Stanley and Kelvin survey the beans. (*Bottom*) KK8 variety.

Our group were also fortunate to be able to visit the One Acre Fund research site in Ekero, an hour outside of Kakamega. They are currently experimenting with four varieties: "Iron Rich Beans" from Rwanda, and the KALRO lines KKCAL194, KK8 and Red16, all bred by Dr Otsyula. KK8 is currently the most popular locally grown variety, preferred because it is high yielding and resistant to root rot. Demand is created by popularity – the local seed companies promote the varieties they are selling, and if the farmers like them they will request them from One Acre Fund, who in turn investigate how they can source them, leading them back to the seed companies. KKCAL194 was established in 2014 but will not be commercially released to farmers until next season, when it has completed the seed release process. Red 16 was created in 2016 so similarly will undergo testing for a further few seasons prior to its commercial release. In yield trials, KKCAL194 outperforms KK8, so it is hoped that this variety will quickly become popular once it is more widely available.

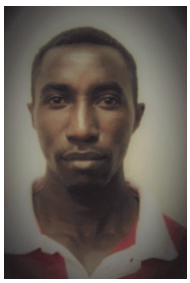
**Figure 6.** Paul Gepts in his natural habitat (looking at beans).

The One Acre Fund was started in Kenya but they also operate in Rwanda, Burundi and Tanzania. Additionally, they are currently running trials in Uganda, Ethiopia and Malawi. When the One Acre Fund was founded in 2006 they began with a pilot group of 38 farmers in Kenya. In 2017, they served 217,000 clients in Kenya alone, worked with 615,000 farm families across East Africa, and employed 6,900 full-time staff. In 2017, they boasted a 65% increase in farmer income (when compared with the income of "likely-to-enrol" or "newly enrolled" farmers to ensure a comparable control group) and a 98% level of loan repayment. If farmers are dissatisfied with the service, they can opt out of the programme by not repaying their loan; as such, the rate of loan repayment is an encouraging indicator of farmer satisfaction.

We are extremely grateful to the One Acre Fund Kakamega team for devoting so much time to educating us on the organisation's background and operations, and taking the time to answer all of our wide-ranging questions. The staff and farmers were endlessly patient, knowledgeable and welcoming; we would like to again extend our thanks to them all for being such generous hosts.

*Dr Becky Lockyer is the Project Administrator for the Trust's seed systems projects.*

## Review of the training workshop for plant breeding students held in Embu, Kenya, and feedback for the African Bean Consortium held in Kakamega, Kenya *by Kuwabo Kuwabo*



I would like to begin by expressing my gratitude for the opportunity afforded me by the Kirkhouse Trust by including me in the afore mentioned training. It was of great value to me. I liked particularly how well the resource personnel delivered their lessons such that I gained fundamental knowledge in plant breeding and genetics even though I am yet to undertake my masters' studies. I also appreciated the exchange of knowledge and ideas through the interaction amongst the students.

The training has made three major impacts on me. First, I have gained more skill and knowledge that has made me better at my part in our breeding program here.

## *Cont'd... by Kuwabo Kuwabo*

Second, from the intensive training, I have acquired the skill to organize and analyze research data and even how to properly interpret results from research experiments via the use of various software packages such as R and Rstudio, Tassel, structure and SAS. This has enabled me to be of help to some students with their data analysis.

Most importantly, I feel that the training has played a key role in preparing me for the Masters' studies in Plant Breeding and Seed Systems I am about to undertake in the coming year. I will be a better student and researcher thereafter because of the training.

I extend my gratitude to the Kirkhouse Trust and all personnel involved that made this training possible. Should other such trainings take place in the future, I sincerely hope to be afforded the opportunity to attend.

As someone attending the ABC for the first time, I found it very interesting and highly informative. There was a great deal of knowledge shared amongst the different breeding programs, from various countries that attended. I learned a lot about how to properly present one's reports from the many presentations made.

The journal club that was conducted during the course of the meeting was a major learning experience. One thing I learned was that it is not enough to just read frequently but one must read critically so as to develop skill in writing a paper for publishing. Another lesson learned was how to write a research paper. All this knowledge will come in handy as I undertake further studies and need to write my research paper.

My experiences at both the training workshop in Embu and the ABC in Kakamega were most informative and invaluable, and for that I am deeply thankful.

*Kuwabo Kuwabo is an MSc student at the University of Zambia.*

## Laboratory Highlights (PCR pre-mix) – *by Ann Lonie*

The Trust supplies the PCR pre-mix 'GE illustra PuReTaq Ready-To-Go PCR Beads'. Although this product is expensive, the advantage of the beads is that they are extremely stable at room temperature due to a stringent lyophilisation process in their manufacture. However, once opened the strips of tubes must be kept in a desiccator with active silica gel.
Each tube contains a bead which is made up to 25 µl but it is possible to divide the mix into two 12.5 µl PCR reactions using extra PCR tubes supplied without any loss in sensitivity.

Protocol to reconstitute a PCR bead to a final "half" volume of 12.5 µl:

**Figure 1.** GE illustra PuReTaq Ready-To-Go PCR Beads.

- Add 17 µl of water to dissolve the bead.
- Measure out 8.5 µl (17÷2) of the mixture and dispense in a separate sterile PCR tube giving you two separate tubes, each containing 8.5 µl of pre-mix.
- Prepare 5 µM / µl of primer stock by performing a 1:20 dilution (i.e. 5 µl of 100 µM stock + 95 µl 1X TE buffer) on each forward and reverse (F&R) primer.
- Add 1 µl each of the F&R primer. This makes up a total of 2 µl of the primers in the PCR mixture. This volume is added to each of the tubes containing the split pre-mix.
- Add 2 µl of the template DNA to each tube containing the split pre-mix and mix well.
- The reaction mixture is now ready for PCR amplification.

*Ann Lonie is the Dispatch and Procurement Officer for the Trust.*

## Graduations

- Congratulations to Dr Yayis Rezene on successful completion of a PhD thesis entitled "Genome wide marker trait association study, molecular characterization and pathogenic variability in *Phaeoisariopsis griseola* (Sacc.) Crous & Braun, Isolates the causal agent of angular leaf spot (ALS), in Ethiopia and response of common bean germplasm to different races of pathogen" in June 2018.

- Congratulations to Misgana Mitiku on successful completion of an MSc thesis on "Genotypic and Virulence Characterization of *xanthomonas axonopodis pv.phaseoli and xanthomonas axonopodis pv.phaseoli var.fuscans,* and the Reaction of Bc4 Common Bean Population to the identified strains of common bacterial Blight " in June 2018.

## *Cont'd... by Ann Lonie*

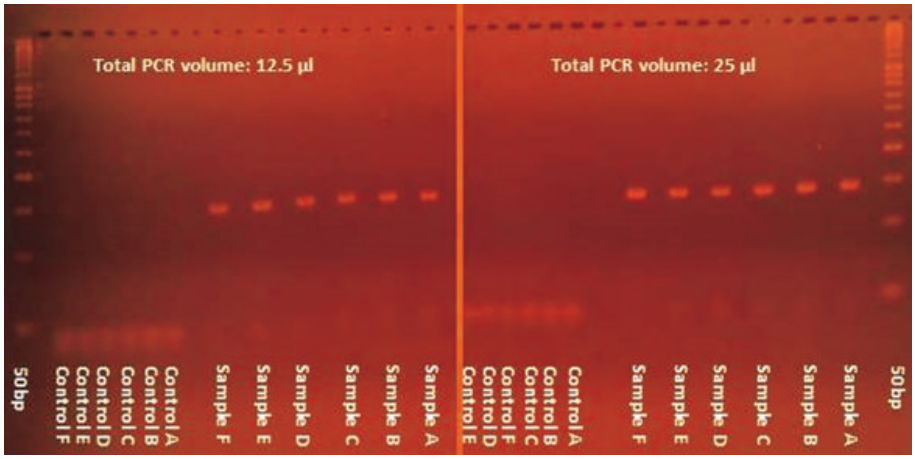
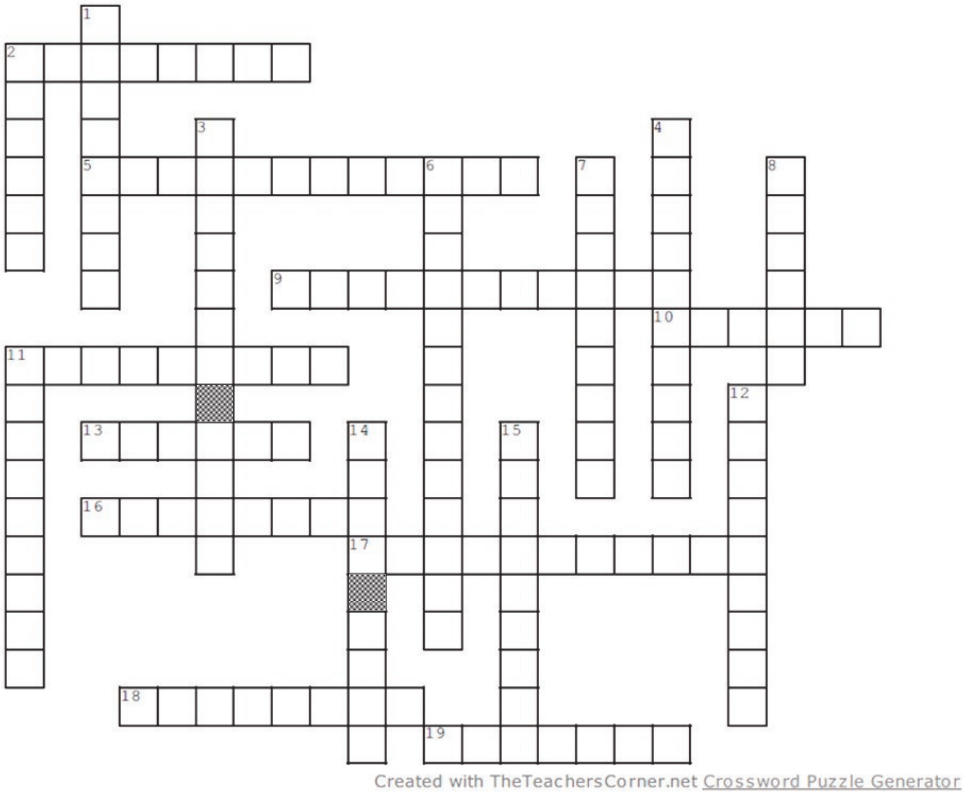



**Figure 2.** Comparison of amplified samples on 6% acrylamide gel (at 120 V for 3 hours) using 12.5 µl and 25 µl PCR starting volumes.

## The Kirkhouse Trust Crossword *provided by Colin Dexter (answers on website)*




**Across**

**2.** One of the two alternative schemes used to introduce multiple genes by backcrossing (8 letters)
**5.** The proportion of the total variance shown by a trait which is due to genetic variation (12)
**9.** Introducing unrelated genetic material into a breeding line (11)
**10.** One possible form of a gene (6)
**11.** Describes a trait which is controlled by many genes (9)
**13.** A mixture of DNA fragments of known size used to estimate the size of fragment after electrophoresis (6)
**16.** Achieving the homozygous state for a particular gene (8)
**17.** A technical word to describe the widespread outbreak of a plant disease (11)
**18.** A section of DNA to be replicated (8)
**19.** One of the two parasitic weeds which attacks cowpea (7)

**Down**

**1.** A type of seed weevil which destroys stored seed (8)
**2.** A short piece of DNA used to ensure the PCR amplification of a target sequence (6)
**3.** What happens when non selected genes move with selected genes (7,4)
**4.** The PCR step in which the target fragments are lengthened by the addition of nucleotides (10)
**6.** Movement of a gene from one species into another by repeated backcrossing (13)
**7.** Expression of one allele over another (9)
**8.** A solution designed to provide a stable pH (6)
**11.** Controlled by two or more genes (9)
**12.** What needs to be done a few times to recreate a genetic background (9)
**14.** Transfer of alleles from one population to another (4,4)
**15.** Variant of a infective bacteria or fungus (9)

*Colin Dexter is the Project Administrator for the Trust's West African Cowpea Consortium (WACC) projects.*