

# Africa Biosciences Challenge Fund



*A new initiative empowering African scientists  
to solve Africa's agricultural challenges*

biosciences  
eastern and central africa

**ILRI**  
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## What is the ABCF?

Established in 2010 by the Biosciences eastern and central Africa - International Livestock Research Institute (BecA-ILRI) Hub, the Africa Biosciences Challenge Fund (ABCF) is a new and innovative way of building African biosciences capacity and leadership while tackling important agricultural constraints in food production, nutrition and animal health (<http://hub.africabiosciences.org/about-abcfl>).

ABCF is a competitive fund that provides capacity building and research opportunities at the BecA-ILRI Hub for African scientists: (1) Hands-on training workshops to acquire relevant research skills; and (2) ABCF Fellowships which enable African scientists to spend up to 6 months at the Hub to address key agricultural constraints through research while building their research leadership skills.

ABCF is supported by the following major donors: the Australian Agency for International Development (AusAID), the Bill & Melinda Gates Foundation (BMGF), and the Swedish Ministry of Foreign Affairs through the Swedish International Development Agency (Sida). ABCF is co-supported by a number of other initiatives and organisations, including the Syngenta Foundation for Sustainable Agriculture (SfSA), African Women in Agricultural Research and Development (AWARD), the Association for Strengthening Agricultural Research in Eastern and Central Africa (ASARECA) and the United Nations Educational Scientific and Cultural Organisation (UNESCO).

## ABCF training workshops:

### Providing relevant skills for research

The ABCF has enabled the Hub to strengthen regional biosciences capacity through short

term training, workshops and scientific conferences. Four annual training workshops are held under the ABCF programme:

- Introduction to molecular biology and bioinformatics
- Advanced bioinformatics
- Scientific research paper writing
- Introduction to principles in laboratory management & equipment maintenance

## ABCF Research Fellowships: Addressing agricultural constraints and building African research leadership

Between September 2010 and December 2011, ABCF Fellowships supported 17 African scientists; and in 2012 at least 50 African scientists will benefit from this programme. To date, the Fellowships have supported scientists from 17 African countries: Burundi, Cameroon, Central African Republic, Cote d'Ivoire, DR Congo, Eritrea, Ethiopia, Kenya, Madagascar, Nigeria, Republic of the Congo, Senegal, Somalia, South Sudan, Sudan, Tanzania, Uganda.



Field work being undertaken to improve the understanding of African swine fever (ASF). ABCF recipient Charles Masembe was able to sequence the genome of the ASF virus, contributing to a broader research project addressing ASF and its movement between African countries.

## ABCF Research Fellows (2010-2012)

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### Vincent Were

Research Associate, BecA-ILRI Hub, Nairobi Kenya  
Sept 2010-Jan 2011

#### **Characterizing aflatoxin accumulation and susceptibility factors in Kenyan smallholder farmer**

The UN's Food and Agriculture Organization (FAO) estimates that 25% of the world's agricultural crops are contaminated with mycotoxins. Among these, of major concern in the East African food systems are fumonisins and aflatoxins. Due to a lack of management strategies, appropriate sources of resistance and surveillance/diagnostics, it is estimated that ~4.5 billion people in the developing world are chronically exposed to unchecked, high levels of aflatoxins. Chronic exposure to mycotoxins causes or is correlated with immunosuppression, cancer, reduced nutrient absorption and is associated with fetal and infant growth retardation. Acute exposure can lead to death. While there is a small but growing body of information about aflatoxins in smallholder farmer-produced maize in Kenya, the presence of fumonisins is essentially unknown.

The objective of this work was to determine whether fumonisins are a significant problem in smallholder farmer maize in Kenya. Vincent's work continued ongoing characterization of aflatoxin levels in smallholder farmer maize and also added another dimension by conducting analysis of fumonisins. Approximately two hundred samples, from three regions of Kenya and previously screened for aflatoxin levels, were analysed for fumonisin content; samples spanning the range of aflatoxin content were included. Fumonisins were actually more prevalent than aflatoxins in the western Kenya samples: 32% of the samples tested have fumonisin levels above the commonly recognized legal limits (>1000 ppb).

The discovery that this second class of mycotoxins are present in high levels is critical for effective targeting of future research. Vincent's work has directly contributed to a research project funded by AusAID as part of the BecA-Commonwealth Science and Industrial Research Organization (CSIRO) partnership focused on reducing aflatoxin levels in maize in East Africa.

**Partners:** BecA-ILRI Hub; Cornell University, USA.



## Charles Masembe

Senior Lecturer, School of Biological Sciences, Makerere University, Uganda

8 Nov-6 Dec 2010; 5 Jun-28 Jul 2011

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### **Pig diseases and food security: Next-generation DNA sequencing of African swine fever virus (ASFV) in Uganda**

Uganda has the largest and most rapidly growing pig production in eastern Africa, with a current pig population of 3.2 million. Seventy-five per cent of pig keeping is found in the rural areas and is mostly practiced by women. African Swine Fever (ASF) is a devastating viral disease that is endemic in Uganda. It is a major constraint to pig production in the country, periodically killing 90–100% of affected animals in outbreaks and has neither treatment nor vaccine.

In this study, 454 high-throughput sequencing of the P54 and P72 genes was used on field materials to determine viral diversity in individual samples and relationships between different ASF outbreaks. The clinical samples were collected from the districts of Gulu, Moyo, Mpigi and Mityana. There were no significant ASFV variants within individual samples. However there were fewer (5%) nucleotide differences among Ugandan ASFV across time and space; and more (50%) when compared to the rest of ASF sequences from other parts of the world. These results show the absence of multiple strains within the various epidemics in the country and confirm the presence of a single genotype during recent ASF outbreaks in Uganda. The results also showed similarity between epidemic ASFV and the virus carried by healthy pigs. This suggests a possible similar source of the virus. The low genetic variation and lack of genetic differentiation in space and time between outbreaks has important implications on the spread and control of ASF.

An additional component of the study was pathogen discovery in pigs. Analysis of ‘shotgun’ sequence data demonstrated that domestic pigs are a potential reservoir of the *Ndumu virus* (NDUV). Previous studies have shown that NDUV is transmitted by mosquitoes, but this is the first time the virus has been found in a vertebrate host and represents a potential zoonotic agent. This is an important finding, because the intensification of pig production also leads to increased contact between humans and animals with possible public health consequences.

**Partners:** Department of Biological Sciences, Makerere University, Uganda; Ministry of Agriculture, Animal Industry and Fisheries, Uganda; Swedish University of Agricultural Sciences (SLU); ILRI Biotechnology Theme; BecA-ILRI Hub; African Insect Science for Food and Health (*icipe*).



## Felix Meutchieye

Lecturer, Department of Animal Science, University of Dschang, Cameroon

Nov-Dec 2010; 1 Aug-27 Oct 2011

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### **Molecular characterization of Cameroon indigenous goats**

Indigenous goat breeds adapted to harsh environments produce meat, milk and hides which serve urban and rural markets of developing countries in Africa and Asia. In Cameroon, knowledge on the genetic diversity of these breeds is limited and this has led to unsatisfactory breeding strategies and loss of diversity. For decades goats have had negligible attention in livestock development programmes and indigenous breeds are poorly described. Breeding strategies can only be effective if relevant documented characterization data are available.

The main objective of this project was to assess the genetic diversity of indigenous goats in Cameroon. The work used 12 SSR markers (microsatellites) and the generation of mitochondrial sequences. The genetic relationships amongst 179 adult goats from eight ecotypes were evaluated. All markers were polymorphic with a PIC of 0.39. The mean number of alleles was 5.08 (2 to 8),  $F_{ST}$  value overall ecotypes was 0.453, and expected heterozygosity ranged from 0.130 to 0.338. AMOVA confirmed a high variation among (33.24%) and within (47.92%) ecotypes. The PCA and NJ tree classified the goats into four major clusters that relate to the agro-ecological zones in Cameroon. This study has shown that Cameroon indigenous goats have interesting potential and the genetic data can aid the rational development, utilization and conservation of these animal genetic resources.

As a result of the studies carried out under this project a significant two-year project “Harnessing genetic diversity for improving goat productivity in Cameroon” has been funded by the Swedish government through the Swedish International Development Agency (Sida).

**Partners:** University of Dschang; BecA-ILRI Hub; ILRI Biotechnology Theme.



## Joseph Ndunguru

Principle Agricultural Research Officer, Mikocheni Agricultural Research Institute (MARI), Tanzania

1 Dec - 31 Dec 2010

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### **Molecular characterization of a wide set of cassava brown streak virus (CBSV) isolates**

*Cassava brown streak virus* (CBSV) has become an increasing problem in most cassava growing areas of sub-Saharan East Africa, and is considered by many to be the most significant threat to food security in the region. There are currently no improved varieties of cassava that have been found to be resistant to CBSV and most promising lines possess only tolerant traits. Distribution of clean planting materials must rely heavily on the availability of reliable diagnostics for CBSV detection. To do this, CBSV sequence data from a wider set of isolates must be generated.

The main symptom of cassava brown streak disease (CBSD) is root necrosis, the premature death of cells and living tissue, which causes tissue in the root to discolour and harden. CBSD is difficult to detect because root necrosis is not visible until the root is harvested. Also, where other symptoms are detectable during production, they are weakly correlated with the disease.

Ndunguru worked with BecA-ILRI Hub research assistant Martina Kyalo to amplify and sequence the coat protein gene from 95 Tanzanian CBSV isolates. Sequence analysis is now enabling Joseph to design strain-specific diagnostic tests. These diagnostics will be disseminated to the cassava research and breeding community and used to further characterize the spread of this disease and in development of resistant varieties and clean planting materials. Joseph plans to continue using the Hub, in addition to his own laboratory at Mikocheni Agricultural Research Institute (MARI), Tanzania, to continue this work.

**Partners:** Mikocheni Agricultural Research Institute (MARI); BecA-ILRI Hub.



## Esther Kanduma

Assistant Lecturer, Department of Biochemistry, University of Nairobi, Kenya

1 Jan – 30 Apr 2011

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### **Understanding the genetic diversity of *Rhipicephalus appendiculatus*, the tick vector for East Coast fever, using microsatellite markers**

The ixodid tick *Rhipicephalus appendiculatus* is the main vector of *Theileria parva*, the causative agent of East Coast fever (ECF) in cattle. ECF is considered to be the most economically important tick-borne disease of cattle in East, Central and Southern Africa. It is associated with high levels of mortality, especially in exotic and cross-bred cattle and is a major constraint to improvement of livestock production across this part of Africa. The genetic diversity and population structure of *R. appendiculatus* remains unknown due to lack of appropriate markers. The objective of this study was to develop a panel of microsatellite markers for *R. appendiculatus* and use them to determine the population structure of the tick in Kenya and in ILRI laboratory stocks.

Twenty nine microsatellite markers were developed and applied to genotyping 25 tick populations belonging to *R. appendiculatus* and five other rhipicephaline tick species. Five markers were found to be useful for intra-species discrimination while a different panel of markers are more useful for inter-species discrimination within the rhipicephaline complex. Genetic differentiation was highest in field ticks which showed higher diversity and admixing of genotypes compared to laboratory stocks. Diversity and population structure of *R. appendiculatus* at 10 field sites in Kenya and 15 laboratory tick stocks was determined. The markers have been applied in identification of related tick species that may transmit *T. parva*.

These markers represent the first molecular tools for defining *R. appendiculatus* genotypes, their distribution, and degree of geographic differentiation in Kenya. They have provided molecular tools that can be used for standardisation of the ITM (Live) *T. parva* vaccine. The markers should prove useful for studies on the spread of acaricide resistance in ticks, the relationship of the tick genotype to the epidemiology of ECF, understanding the epidemiology of ECF as an emerging disease in new areas such as South Sudan, and as tools for quality control of genotypes of *R. appendiculatus* ticks used in live vaccine production.

**Partners:** University of Nairobi; BecA-ILRI Hub; ILRI Biotechnology Theme; University of Nottingham.



## Salma Hassan

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1 Jan - 30 Apr 2011

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### New diagnostic tools to understand the epidemiology of sheep theileriosis in northern Sudan

*Theileria lestoquardi* is a protozoan parasite pathogenic to sheep and goats. It is transmitted by the tick *Hyalomma anatolicum anatolicum* and causes a disease called malignant theileriosis. The disease occurs where the vector tick species is found, in northern Africa (including northern Sudan), southern Europe and the Middle East. Sudan has a population of 50 million desert sheep, a major breed for the production of mutton and milk. However, malignant ovine theileriosis causes massive losses among the sheep in northern Sudan. Outbreaks occur with mortality rates of ~46-100%.

In the laboratory, identification of the parasites in infected animals is based on microscopical examinations of smears of blood or lymph node aspirates. In general, serological tests such as immunofluorescence antibody test (IFAT) and enzyme linked immunosorbent assay (ELISA) are more suitable as surveillance tools. An antibody detection ELISA based on Clone 5 recombinant antigen has been developed (Bakheit et al. 2006). A number of PCR-based methods to detect parasite DNA such as simple PCR and the Reverse Line Blot (RLB) (Schnittger et al. 2004) have also been developed, and recently, loop-mediated isothermal amplification of DNA (LAMP), based on the Clone 5 gene, has been shown to be a promising tool to detect *T. lestoquardi*.

Although both the Clone 5 ELISA and LAMP have shown promise, Clone 5 has features typical of a highly polymorphic protein of theileria. A significant level of Clone 5 sequence polymorphism could compromise the performance of the ELISA and LAMP assays.

The objectives of this study were to clone, sequence and analyse the diagnostic Clone 5 gene from four stocks of Sudanese *T. lestoquardi* schizont-infected lymphocyte cells lines.

Clone 5 sequence data indicate significant sequence and size heterogeneity at both the DNA and protein level. The extent of the variation observed could compromise the performance of ELISA and LAMP assays based on Clone 5, and further research is required.

**Partners:** BecA-ILRI Hub; Central Veterinary Research Laboratory (CVRL), Sudan; ILRI Biotechnology Theme; Research Center Borstel, Germany.





## Hashim M. Mangosongo

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15 Jan – 21 Apr 2012

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### The genetic diversity of wild rice (*Oryza longistaminata*) in selected areas of Tanzania

*Oryza longistaminata* is a wild species found in Africa that is closely related to cultivated rice (*O. sativa* L.). It has abundant genetic diversity and contains various valuable traits such as bacterial blight resistance, high pollen production, long stigmas and drought resistance, which could be introduced into cultivated rice through introgression. The spatial distribution of genetic diversity of a plant species is influenced by environmental variables such as climate, soils, vegetation and elevation. The selection and use of genetic resources from this wild species to improve rice productivity requires a detailed understanding of its genetic diversity.

The objective of this study was to determine the intra-specific genetic diversity of *O. longistaminata* and establish how the variation is partitioned within and between populations from different geographic locations in selected areas of Tanzania by using microsatellite (SSR) markers. The information obtained will be useful for identifying areas with high genetic diversity of *O. longistaminata*, for development of appropriate strategies for conservation of the species and for future use in rice breeding. Areas with higher genetic diversity of *O. longistaminata* or those with unique genetic resources will be given conservation priority.

A hundred and fifty four leaf samples were collected from four study sites in Tanzania: Bagamoyo, Kibaha, Kilombero and Mbarali, which were selected because they harbor high abundance of *O. longistaminata*, are important rice cultivation areas and they have different environmental conditions. Preliminary results show that *O. longistaminata* samples from Mbarali were the most diverse followed by those from Kilombero, and the Kibaha samples showed least diversity. Moreover, there was more diversity within populations than between populations.

This information on the genetic diversity of *O. longistaminata* will be useful for the development of appropriate strategies for conservation. Areas with higher or unique genetic resources will be given conservation priority. Rice breeders and other researchers relying on efficient utilization of genetic resources will be informed about these results, so that they can incorporate *O. longistaminata* into their cultivated rice improvement efforts and hence increase rice productivity.

**Partners:** University of Dar es Salaam; Africa Rice Center (AfricaRice); BecA-ILRI Hub.



## Thomas Bazarusanga

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Sheikh, Somaliland

31 Jan – 27 Feb 2011

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### East coast fever investigation in Somaliland cattle

East Coast fever (ECF) is a severe tick-borne disease of cattle in eastern, central and southern Africa, caused by the parasite *Theileria parva*. Occasional ECF-like symptoms have been observed in Somaliland cattle, but no confirmed cases of ECF have been reported, nor has the most common tick vector of *T. parva* (*Rhipicephalus appendiculatus*) been identified in the country. In 2009, cattle in Somaliland were tested using a *T. parva* antibody ELISA, and 20% were positive. If *T. parva* infection in Somaliland cattle is confirmed, it would have significant implications for disease control and cattle productivity, especially if improved, susceptible breeds are imported into the country for milk production.

The objective of this project at the BecA-ILRI Hub was to follow-up the earlier serological study to determine the species of parasites infecting cattle. Additional cattle blood and serum samples and ticks were collected and included in the study. All samples were tested by two PCR assays: (1) nested 18s gene PCR assay which can detect all *Theileria* and *Babesia* species, and (2) a nested p104 gene PCR assay which is specific for *T. parva*.

The results show that *T. parva* is not present in the Somaliland cattle and ticks samples tested, which indicates the *T. parva* ELISA may be prone to some false positives with sera from Somaliland cattle. There was a high prevalence of *T. mutans* and *T. velifera*, which are considered to be benign in cattle. A report from Uganda suggested *T. mutans* and *T. velifera* could occasionally cause ECF-like symptoms in cattle. Further studies are required to identify the pathogen(s) that cause ECF-like symptoms in Somaliland cattle.

At the Hub Dr Bazarusanga was accompanied by two laboratory technicians from STVS, Ms. Awo Ibrahim Isman and Mr. Abidirazak Alkalid, who received short-term training in serological and molecular diagnostics and assisted with the project. Funding for the training was provided by STVS.

**Partners:** Sheikh Technical Veterinary School; BecA-ILRI Hub; ILRI Biotechnology Theme; Terra Nuova.



## Selamawit Getachew Bedane

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26 Jul - 28 Oct 2011  
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## Molecular characterization of enset collections in some selected regions of Ethiopia using banana SSRs

Enset (*Ensete ventricosum*) is a staple food crop in the south, south-western and central parts of Ethiopia and its morphology resembles that of banana (*Musa accuminata*); both species are members of the Musaceae family. Enset has remained largely unimproved and there is need to study the genetic diversity of current accessions and build on it to develop new cultivars/varieties with desired agronomic traits (disease resistance, adaptability to different environments, yield etc.). The development of markers for enset breeding is therefore an important step to set the stage for the improvement of enset. Transferability of 71 SSR loci from banana to enset was investigated using 220 enset accessions collected from eight different regions and a live enset gene bank in Ethiopia.

Of the 71 SSRs tested, 28 (39%) were successfully transferable to enset. Of these 11 were shown to be polymorphic and were subsequently used to examine the enset diversity. In total, 61 alleles were detected by the 11 SSR loci. Six markers were highly polymorphic with mean allelic numbers per locus  $> 5.5$  and polymorphic information content (PIC)  $> 0.5$ . The mean observed heterozygosity was 0.67. The inbreeding coefficient for all loci was negative; the mean was  $-0.2750$ . There was high gene flow rate between populations ( $Nm = 5.18$ ) and the coefficient of genetic differentiation ( $F_{st}$ ) between populations was 0.46, which was in accordance with the results of AMOVA analysis that indicated enset accessions are not genetically different by zone of collection. The transferability of banana SSR's to enset has been confirmed by this study. Sequencing results indicated that there is no correlation between taxonomic classification and number of repeats found. There was no distinct clustering by zone, a result supported by the observed high rate of gene flow which indicates the existence of a high rate of exchange of planting materials among and between zones, and a large within - compared to between-group variation. This implies that conservation of genetic diversity should focus on identifying distinct groups within a zone.

**Partners:** Haramaya University; Ethiopian Institute of Agricultural Research; BecA-ILRI Hub.



## Diaeldin Ahmed Salih Hassan

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15 Aug – 14 Nov 2011

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### Genotyping of *Theileria parva* in South Sudan to support control of East Coast fever

East Coast fever (ECF) is a serious tick-borne disease of cattle in eastern, central and southern Africa caused by the parasite *Theileria parva*. In many countries *T. parva* is controlled by reducing infestation with the tick vector, *Rhipicephalus appendiculatus*, using acaricides. However, this has major drawbacks, including the development of tick resistance and concerns over food-safety and environmental contamination. An alternative method of ECF control is immunization with a live sporozoite vaccine, but this has not yet been deployed in South Sudan. Following the signing of comprehensive peace agreements in Sudan in 2005, there has been extensive movement within South Sudan of people and their livestock, with concomitant reports of ECF spreading to more northern areas that were previously free of the disease.

The objective of this project is to determine genotypes of *T. parva* circulating in South Sudan, and compare with the vaccine strain. The results will support a rational approach to vaccine deployment, providing data on the *T. parva* in South Sudan before vaccination programmes begin. Activities in the project include genotyping using microsatellite markers, and sequencing of two *T. parva* CD8 cytotoxic T-cell (CTL) antigen genes (Tp1 and Tp2).

The preliminary results reveal diversity of the parasite in South Sudan, but principal component analysis (PCA) suggests the parasites may belong to one population. Some Tp1 sequences in field isolates are identical to the CTL epitopes found in the vaccine strain, and no new CTL epitopes were identified beyond those previously reported in Kenya. Data analysis is ongoing.

Efficient disease control will result in increased income for pastoralists in areas that have been affected by civil war in South Sudan, which may strengthen the peace process by assisting with settlement of refugees and job creation.

**Partners:** VRI, Sudan; BecA-ILRI Hub; ILRI Biotechnology Theme; Ministry of Animal Resources and Fisheries, South Sudan.



## Mame Nahé Diouf

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### Genetic characterization of Senegalese N'dama cattle

Animal trypanosomiasis is a tsetse-transmitted disease that is endemic in 37 African countries and remains a major constraint to livestock production. In the absence of a vaccine, the control of this disease has for long been focused on expensive and unsustainable means such as chemotherapy and tsetse fly eradication. During the last decade, strategies to combat the disease have been oriented towards a better understanding of trypanotolerance of cattle.

Different trypanotolerant cattle breeds exist in West and Central Africa. Among these breeds, the 'N'dama taurine' is the most widely represented (more than 45% of cattle) and several subpopulations have been reported in many agro-ecological zones. In south Senegal, farmers recognize different breeds, including Petit, Moyenne and Grande N'dama, which exhibit different levels of trypanotolerance, with Petit N'dama being the most trypanotolerant. The question remains as to whether there is a genetic basis for these subpopulations.

The main objectives of this study were to (1) estimate the level of genetic diversity and population structure of Senegalese N'dama subpopulations (Petit, Moyenne, Grande, CRZ Kolda) using a FAO-recommended panel of 20 microsatellite markers, and (2) determine the level of *Bos indicus* introgression in the subpopulations and correlate with trypanotolerance.

The results show that genetic variability within subpopulations is higher than variability between subpopulations. The level of *Bos indicus* introgression in the subpopulations was inversely correlated with trypanotolerance. For example, CRZ Kolda, the least trypanotolerant, had the largest introgression of indicus, while Petit N'dama- the most trypanotolerant- had the least introgression of *Bos indicus*.

It is recommended that breeding and management programmes are established to conserve the farmer-recognized trypanotolerant N'dama breeds (Petit, Moyenne, Grande) and limit uncontrolled mixing between N'dama (*Bos taurus*) and *Bos indicus*, especially Petit N'dama. Conservation of Petit N'dama is particularly important for the south of Senegal where tsetse challenge is high.

**Partners:** Institut Sénégalais de Recherches Agricoles (ISRA); BecA-ILRI Hub; ILRI Biotechnology Theme.



## **Bombom Alexander Jr.**

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16 Sept – 16 Dec 2011; 13 Feb – 14 Jun 2012; 2 July – 30 Sept 2012  
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### **Development and characterisation of maize x sorghum hybrids**

In plant breeding, access to adequate genetic variation and identification of unique and desirable traits is a key objective in crop improvement. The conventional approach to generating new and increased variation is by compatible interspecific and inter-generic hybridisation. Successful wide crosses have been previously reported in cereals, such as with wheat and rye leading to the development of 'triticale'. Efforts to develop maize x sorghum hybrids however have not been successful and in cases where plants were obtained, the progeny could not be confirmed as true crosses. In 2009, a successful cross between maize and sorghum was observed while pursuing a PhD at Makerere University. The cross has led to fertile progeny. The precise factors responsible for this success are yet to be determined.

The objective of this project is to characterise progeny from the maize x sorghum cross using maize and sorghum microsatellite markers. The main activity in this project is the characterisation of maize x sorghum progeny to confirm hybrids as true crosses. Preliminary genotyping results reveal a mixture of fingerprints for the progeny. Though not conclusive, the results suggest possible phenomena such as genome deletion and/or rearrangement. Cytological characterisation of parents and progeny is also being attempted. In addition to genotyping, a second major activity that includes repeating the initial cross was carried out. Seed has been observed in some crosses with maize as female. These include new  $F_1$  progeny and backcrosses using  $F_1$  pollen from the initial cross.

Outputs from this project will include (1) maize x sorghum hybrids confirmed from genotyping data, cytology, and genotype by sequencing results, and (2)  $F_1$  and backcross seed from the repeat hybridisation experiment thus providing evidence that crosses can be achieved with other maize and sorghum cultivars. Due to the novelty of this study, extra funding is being sought to scale up the project and provide proof of concept for existence of the maize x sorghum cross. Upon invitation, a proposal was drafted and submitted to the Bill & Melinda Gates Foundation for funding and is currently under review. It is anticipated that the results, techniques and knowledge generated as a result of this funding will contribute to development of other maize x sorghum hybrids for specific traits hence impacting on agricultural research and development.

**Partners:** Makerere University; BecA-ILRI Hub; International Maize and Wheat Improvement Center (CIMMYT).



## Ruth Wanyera

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23 Nov 2011 - 10 Aug 2012

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Co-supported by AWARD

### Molecular characterization of wheat stem rust races and Ug99 lineage in Kenya

Wheat is an important cereal that contributes significantly to food security in Kenya; among the cereals, it ranks second after maize. The crop has greater potential because it is grown in varied agro-ecological zones and the current area under production is estimated at 150,000 ha with annual production estimated at 300,000 metric tonnes (MT). Currently, the demand in Kenya is for 900,000 MT per annum and the deficit is imported. The demand for consumption is driven by population growth, increased urbanization and changing diets.

Wheat production faces many challenges, including wheat stem rust disease whose casual pathogen is *Puccinia graminis* f. sp. *tritici* (Pgt). It causes up to 100% crop losses and has overcome many sources of resistance previously protecting against wheat stem rust. The disease is now a major concern in Africa, since the detection of Pgt race Ug99 in Uganda and its subsequent spread to the wheat growing areas of Kenya, Ethiopia, Yemen, Iran, Sudan, Eritrea, Tanzania, South Africa, Zimbabwe, Mozambique and South Africa. It is predicted to spread towards North Africa, Middle East, Asia and beyond raising serious concerns of major epidemics that could destroy the wheat crop in various parts of the world. More variants of race Ug99 show that Ug99 is evolving. Regular monitoring, sampling and identification of the races provide knowledge on the pathogen population dynamics and evolution.

In this study, the genetic diversity of 455 wheat stem rust isolates collected in 2011 from the four major wheat growing areas of Kenya was investigated at the BecA-ILRI Hub using 11 microsatellite markers. The broad objective was to characterize Kenyan stem rust races, including isolates of the Ug99 lineage, to determine whether Pgt consists of a single population or if there are discrete populations in different regions. The results showed that there are two major populations in Kenya and there are potentially new races that require further (genetic and phenotypic) characterization. The information generated is useful to both the Durable Rust Resistance Wheat Project and the KARI wheat breeding effort. The characterized isolates will be used to incorporate relevant sources of stem rust resistance into the KARI wheat breeding programme so that resistant varieties can be available to farmers in the future.

**Partners:** KARI; U.S. Department of Agriculture - Agricultural Research Service (USDA-ARS) Cereal Disease Laboratory; BecA-ILRI Hub.



## Rasha Ali Mohamed Ali

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20 Nov 2011 - 20 Feb 2012

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### **Genotyping of BC<sub>4</sub>F<sub>1</sub> population with SSR markers associated with striga resistance in sorghum**

During the last two decades, considerable progress has been made in the development of molecular markers and their use in mapping/tagging of genes/quantitative trait loci (QTLs) controlling important agronomic traits in all major crops. As a result, molecular markers closely associated with desirable traits are being utilized to increase the efficiency and effectiveness of conventional breeding by indirect selection of the desirable plants in segregating populations. Molecular markers have been used in breeding programmes to transfer novel genes, accelerate backcrossing, and pyramid resistance genes. They have also been used to estimate genetic relatedness and in comparative mapping. The use of MAS in breeding programmes for striga resistance in sorghum is a new strategy that offers great opportunities for enhancing the efficiency and cost effectiveness of striga resistance breeding strategies.

This project focused on development of striga-resistant sorghum varieties nearing readiness to undergo National Performance Trials and subsequent release in Sudan. The genotyping is part of developing superior striga resistant lines using Marker Assisted Backcross (MAB). The objectives of the research include:

- Identification of QTLs associated with striga resistance using the foreground markers (SSRs)
- Screening of generated BC<sub>4</sub>F<sub>1</sub> with the background markers
- Advancing superior lines with 2 or more major QTLs associated with striga resistance

Rasha is a researcher at the Agricultural Research Cooperation in Khartoum, Sudan where her research mainly focuses on striga host-parasite interactions and management. She is also a PhD student at the University of Khartoum studying molecular breeding. Rasha hopes that this research will be a contribution towards poverty reduction through enhancing household food security in the semi-arid regions of Sudan, neighboring parts of eastern Africa and other parts of the world where striga is endemic. There are currently no effective striga-resistant sorghum lines available to farmers in Sudan.

**Partners:** Agricultural Research Cooperation, Khartoum, Sudan; International Crops Research Institute for the Semi-Arid Tropics (ICRISAT; BecA-ILRI Hub).





## Sheila Ommeh

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2 Jan -2 Jul 2012

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### **Genetic diversity and population structure of chicken populations from Lamu and Turkana regions in Kenya**

In Kenya, chicken meat and eggs are increasingly an important source of quality protein. There are three main types of poultry production systems. The intensive commercial systems focusing on commercial breeds, the semi-intensive systems is mostly on hybrids as seen in areas close to urban centres, and lastly the backyard poultry farming which is the most predominant accounting for 70% of the approximately 25 million chickens in Kenya. The latter population is usually indigenous chickens kept by smallholder rural farmers who are mostly women and children. The indigenous populations of chicken are currently facing a risk of genetic dilution from various exotic cockerel exchange programmes introduced by the government in several districts in Kenya to boost meat and egg production.

A recent study has classified the indigenous chickens in Kenya into eight phenotypes present in seven agro climatic zones. From published studies, most of these phenotypes have been characterized genetically. However, phenotypes in arid and semi-arid agro-climatic zones have not been characterized mainly due to difficulty in accessing the areas in which they exist. The first are Kuchi chicken found in Lamu District of Kenya, but also exist on Pemba Island in Tanzania. Although at high risk of genetic dilution, this breed is very hardy, resistant to diseases like Newcastle Disease and has a high weight at maturity. The second are indigenous chickens kept by pastoralists in the remote Turkana region of Kenya that are well adapted to drought conditions.

The main objective of this work was to study the genetic diversity and population structure of indigenous chicken samples from Lamu and Turkana. The approach used was simple sequence repeats (SSRs) using 24 FAO-recommended markers. Ninety-one chicken samples were analysed, which grouped into six populations. The results show there is different clustering for two ecotypes and a distinct genetic diversity in Turkana remote populations. There is no bottleneck evident in both ecotypes. Cross breeding and not inbreeding is the real threat to Lamu Kuchi chickens and there is also crossbreeding in Turkana “town populations”. The results pave the way towards the conservation of these two ecotypes.

**Partners:** BecA-ILRI Hub; ILRI Biotechnology Theme; Combating Infectious Diseases in Livestock (CIDLID); Rural Development Agency, Korea.



## Gladness Elibariki

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### Protecting Tanzanian cassava landraces against Cassava Mosaic Disease using RNA interference technology

According to FAO, cassava (*Manihot esculenta* (Crantz)) is the fourth most important source of carbohydrate in Africa. In Tanzania, cassava ranks second to maize, and the crop is grown by subsistence farmers for local consumption or as a cash crop for sale in local markets. Although the majority of cassava landraces grown in Tanzania are potentially high yielding, they are susceptible to viral diseases. Cassava mosaic disease (CMD) accounts for losses of up to 47%. Efforts to combat CMD in Tanzania, which include phytosanitation, conventional breeding and currently, marker assisted selection breeding, have had little success. The Mikocheni Agricultural Research Institute (MARI) has a project to generate farmer-preferred cassava that is resistant to CMD through RNA interference (RNAi) technology, and part of this research was carried out at the BecA-ILRI Hub.

The research activities at the Hub comprised (1) preparing RNAi plasmid constructs containing *East African cassava mosaic virus* and *African cassava mosaic virus* genes, suitable for agrobacterium-mediated transformation of cassava, and (2) assessing the genetic diversity and genetic relationships of 20 cassava landraces to facilitate selection for transformation. Two double-stranded RNAi plasmid constructs were made and confirmed by DNA sequencing. Microsatellite genotyping revealed high genetic diversity between the selected landraces. Landraces will be selected from different 'clusters' for transformation.

At MARI, the two RNAi constructs were used to transform *Agrobacterium tumefaciens* bacteria strain LBA4404. New somatic embryos have been induced from three cultivars. In the longer term, several cassava cultivars will be transformed with RNAi constructs, and then tested for resistance to CMD.

**Partners:** University of Dar es Salaam; Mikocheni Agricultural Research Institute; BecA-ILRI Hub.



## Christian Tiambo Keambou

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15 Jan – 26 May 2012

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### Molecular polymorphism of phenotypes and ecotypes of Cameroon indigenous chicken (*Gallus gallus*)

The productivity of local chicken in Cameroon must be improved to help alleviate food insecurity and poverty of farmers. This study is focused on understanding the different physical and genetic traits of indigenous chicken which can make them better performers in terms of disease resistance and meat and egg production. Indigenous chickens are a very important livestock in rural Africa, as they are the most widespread type of livestock in Africa, found in over 80% of the rural homesteads; they are easy to breed, ready source of nutrition and income and mostly managed by women and children. The research at the BecA-ILRI Hub was to undertake the molecular characterization of Cameroon local chicken, and to link molecular data to the phenotypic data previously collected in Cameroon. This will constitute the basis for efficient decision making for conservation and genetic improvement.

The activities at the Hub were genotyping using FAO-recommended microsatellites and mitochondrial DNA sequencing of a sample of ecotypes and genetic types of Cameroon local chicken. From the results, three main clusters were shown in the Cameroon chicken population, and sequence data show the existence of 18 haplotypes, of which 12 are new haplotypes in Cameroon local chicken populations. These results offer the basic step towards rational decision making prior to the genetic improvement and conservation programmes. At least two centres of conservation of local chicken are proposed in Cameroon, one in the northern and the other in the western highlands of Cameroon. The next steps are to complete the chicken adaptability characterization, propose a breeding, conservation and development programme. This work will be implemented through selection of targeted phenotypes/ecotypes for goal oriented (meat or egg) development.

**Partners:** BecA-ILRI Hub; ILRI Biotechnology Theme; University of Buea, Cameroon; University of Dschang, Cameroon; University of N'Djamena, Chad; CIAR, Central African Republic; University of Sciences and Technics of Masuku, Gabon; Centre National de Recherche Scientifique et Technologique (CENAREST), Gabon.



## Patrick Jolly Ngoni Ema

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14 Feb-15 Jun 2012

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### Molecular characterization of Cameroon indigenous cattle

Cattle breeds fall into two main types: *Bos indicus* cattle, also called zebu, and *Bos taurus*. Both are found in Cameroon, mostly in three of its five agro-ecological zones. Unfortunately, due to the pastoral systems in place, the flow of genes between *Bos indicus* and *Bos taurus* cattle breeds is uncontrolled. Therefore, should this trend continue, the loss of genetic diversity will increase with potential consequences such as the disappearance of trypanotolerant taurine cattle. A detailed understanding of the genetic diversity of indigenous cattle is crucial for rational crossbreeding systems in Cameroon. Until now, the diversity of indigenous cattle in Cameroon has been mainly investigated by phenotypic characterisation. The aim of this study was to assess genetic diversity and relationships within the Cameroon indigenous cattle using mitochondrial DNA displacement loop (mtDNA D-Loop) sequences and microsatellite markers.

Twenty FAO-recommended microsatellite markers were used to assess the genetic diversity of four cattle populations in Cameroon (Arab Shuwa, Goudali, White Fulani, and Namchi). This study observed a moderate genetic differentiation among the populations while intra-population genetic differentiation was high. Analysis of haplotype diversity indicated that 6% and 94% of the variability was due to differentiation among and within populations respectively. The samples formed two genotypic clusters indicating that two main populations are present in Cameroon, and several minor sub-populations. mtDNA sequence analysis identified two main clades, implying distinct origins of Cameroonian cattle, further confirming the well-known dichotomy between *Bos taurus* and *Bos indicus*.

In general, cattle populations under the study shared many alleles regardless of geographical location, meaning a great admixture between them. The genetic differentiation among the breeds was moderate. These factors may reduce the amount of genetic variation and consequently result in lowered fitness and reduced potential for future adaptation. Information generated by this study will form a valuable resource for the national cattle breeding programme in Cameroon. This study has formed the basis of a new project aimed at conserving genetic resources, particularly the trypanotolerant Namchi or Gudali cattle breeds in Cameroon.

**Partners:** University of Ngaoundere; ILRI Biotechnology Theme; BecA-ILRI Hub.



## Adey Feleke Desta

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Funded by Bio-resources Innovations Network for Eastern Africa  
Development (Bio-Innovate)

### Microbial diversity in an experimental tannery wastewater treatment plant in Ethiopia

Environmental pollution is an inevitable consequence of economic development. Ethiopia has a long tradition of processing and exporting leather and leather products but the leather industry is often associated with the generation of high amount of liquid wastes constituting high salt content, organic load, inorganic matter, dissolved and suspended solids and synthetic chemical pollutants. To alleviate the problem of pollution, cost-effective treatment and reuse of the effluents by using biological wastewater treatment plants have been developed. However, most biological wastewater treatment processes are prone to instabilities and failure of specific functions mainly because they have been managed as 'black box' systems and their functioning is controlled by using only empirical knowledge of the processes other than the use of microorganisms in treatment. The use of DNA-based techniques has revolutionized the methods of characterizing the microbial populations that modify sludge environments and their interaction within, thus providing a way to maximize performance of the treatment processes.

The current study reports the structure and diversity of the microbial community in the sludge of the different reactors as well as the root area of a constructed wetland that treats tannery effluent in Modjo, Ethiopia. Bacterial phylogenetic analysis utilizing the 16S ribosomal RNA gene sequence data were performed for each of the selected reactors. The results revealed a total of 31 different phylotypes with the dominant members belonging to *Firmicutes*, *Bacteroidia*, *Proteobacteria*, *Sphingobacteria* and *Synergistia* in the anaerobic and aerobic reactors. In the constructed wetland, members of the group *Cyanobacteria* were also identified in addition to *Firmicutes*, *Bacteroidia* and *Proteobacteria*.

The presence of members of groups that are associated with reduction of sulfate and removal of chromium, nitrate and degradation of synthetic aromatic compounds implicates them in the current performance of the treatment system. Furthermore, physico-chemical data (total nitrogen, total carbon, pH, sulphate, nitrate) have been recorded in the anaerobic, aerobic and constructed wetlands. Statistical analyses of the data will elucidate the key factors structuring the observed microbial communities. These abiotic-biotic correlations will help in designing strategies to harness the complex microbial communities for process optimization for a more stable and efficient pollutant removal.

**Partners:** Addis Ababa University; Modjo Tannery – Ethiopia; BecA-ILRI Hub.



## Eric Maina Magembe

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5 Mar – 5 Sep 2012

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### **Genetic variation and aflatoxigenicity analysis in *Aspergillus flavus* isolates from soils and maize kernels at pre- and post-harvest in eastern Kenya**

Maize is vulnerable to contamination by mycotoxigenic fungi belonging to the *Aspergillus*, *Fusarium*, and *Penicillium* genera. Aflatoxins are carcinogenic metabolites produced by several *Aspergillus* species. Globally, 4.5 million people are chronically exposed to aflatoxins, while acute exposure can lead to death. Contamination of maize also leads to impairment of trade. Sustainable mitigation approaches have so far not been implemented due to several factors including: (1) poor understanding of the maize -*A. flavus* pathosystem, (2) genetic diversity of *A. flavus* and factors that lead to aflatoxin accumulation, (3) high costs associated with grain contamination surveillance, and (4) inadequate regulation of food systems. The most sustainable aflatoxin accumulation mitigation approach would be to introduce genetic resistance to *A. flavus* infection into maize varieties grown by farmers. The objectives of this study were to assess the genetic variation of *A. flavus* isolates from kernels at pre- and post-harvest, and to compare the aflatoxigenicity of pre- and post-harvest isolates and thus determine which isolate or strain contributes to aflatoxin accumulation.

*A. flavus* was isolated from mature corn ears from farmers' fields in two counties in eastern Kenya (Kiboko and Makindu) that are considered high risk for aflatoxins. Maize ears were sampled at pre-harvest and two months post-harvest during storage. DNA was extracted from cultured, identified isolates and eleven microsatellite markers were used for genetic diversity analysis. Aflatoxin-producing potential of the isolates was assessed by ELISA. Preliminary analysis of *A. flavus* diversity has shown genetic distinction between pre- and post-harvest isolates from eastern Kenya. We hypothesize two distinct niches of *A. flavus*, and these niches are occupied by genetically distinct strains or isolates.

This study provides information on the pre- and post-harvest genetic variation, aflatoxin accumulation and distribution of *A. flavus*. The results from this study will be useful for identifying *A. flavus* strains for breeding towards resistant/tolerance to farmer preferred varieties and in biological control. These interventions will lead to maize produce that is free of aflatoxins. The study will also provide the first glimpse into whether different *A. flavus* populations are better suited to these niches, reflecting the two main stages of fungal colonization and aflatoxin production in maize. As such, it can significantly influence global aflatoxin research.

**Partners:** BecA-ILRI Hub; Cornell University.



## Harriet Angwech

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26 Mar – 28 Aug 2012

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### **Molecular epidemiology and characterization of trypanosome infections of domestic livestock in the post conflict districts of Amuru and Nwoya, northern Uganda**

Livestock trypanosomiasis is an important constraint to livestock productivity in sub-Saharan Africa. It is caused by protozoan blood parasites of the genus *Trypanosoma* and transmitted by tsetse flies of the genus *Glossina*. Livestock in endemic foci are constantly under threat of infection, especially where there is close proximity with wildlife - as in the study area of Amuru and Nwoya Districts, Northern Uganda. Knowledge of the host range, distribution pattern and prevalence of trypanosome species and host preference/feeding behaviour of tsetse flies is essential to understand the epidemiology of trypanosomiasis. It also provides an important foundation on which to establish disease control strategies.

This study was conducted to determine the prevalence of trypanosome species infecting tsetse and livestock in the study area; the role of domestic livestock in the epidemiology of animal and human African trypanosomiasis; and the host preference of tsetse flies. The study used mitochondrial DNA barcoding for host-preference identification, and PCR using the internal transcribed spacer (ITS) of the rDNA gene for trypanosome detection.

The results show significant heterogeneity of trypanosome infections in various cattle age categories and species of livestock sampled in the study. Infection rates were 41.6% and 23.5% in domestic livestock and tsetse flies respectively. Of the infections, 81.7% are due to *Trypanosoma vivax* and this is expected in areas where animals are not routinely treated with trypanocides. The study also reveals that tsetse flies in the area feed on a wide range of hosts including cattle, bush buck, warthog, human, and Nile monitor lizard. The feeding behaviour of the tsetse flies appears to depend on the availability, abundance and behaviour of the host animals. It is therefore important that other animals besides cattle are considered as baits in tsetse and trypanosomiasis control programmes, and that the age of cattle should be considered when treating the animals (baits) with insecticides for tsetse control for effective results. Also, in the study area, chemotherapy could form an integral part of control of livestock trypanosomiasis.

This project is helping to build capacity at Gulu University in the use of molecular tools for trypanosomiasis epidemiological studies. The project complements the ABCF project of Robert Opiro on 'The characterization of trypanosome infections in tsetse flies in the post-conflict West Nile Districts of Adjumani and Moyo, Uganda.

**Partners:** Gulu University; BecA-ILRI Hub.



## Robert Opiro

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26 Mar - 28 Aug 2012

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### **The characterization of trypanosome infections in tsetse flies in the post-conflict West Nile districts of Adjumani and Moyo, Uganda**

African trypanosomiasis is an infectious disease of humans and animals, caused by protozoan parasites of the genus *Trypanosoma* that multiply in blood and tissue fluids of mammalian hosts. Trypanosomes are transmitted by the bite of infected tsetse, and the distribution of trypanosomiasis in Africa corresponds to the range of tsetse flies. Currently, there is a lack of accurate and comprehensive data on tsetse fly distribution and trypanosomes species in the study area of West Nile Districts of Adjumani and Moyo, Uganda on which to base intervention strategies. Due to a civil war that lasted for over 20 years, people in this area were displaced from their homes and trypanosomiasis control programmes were interrupted. Since 2008 the communities resettling into their original homesteads face problems of tsetse and trypanosomiasis which is likely to compromise human health and agricultural production in the area.

The broad objective of this project is to generate comprehensive reference data on the incidence and distribution of trypanosome species in the tsetse vector and the identification of tsetse host preference for feeding which to base control strategies. Mitochondrial DNA barcoding was used for host preference identification, and trypanosomes species were detected using PCR of the Internal Transcribed Spacer (ITS) regions of the ribosomal DNA gene. Two hundred and seventy two tsetse samples were collected from the field for analysis, and included 34 blood-fed flies for host preference identification.

A diversity of hosts were identified as tsetse blood meal sources in the study areas including cattle, humans, Nile monitor lizard, turtles, elephant and domestic pig. Cattle and humans were the most important hosts, representing 65% of all host bloodmeals. *Trypanosoma vivax* was the most common trypanosome species (59.6%), followed by *Trypanosoma brucei* (13.5%), *Trypanosoma simiae* (11.5%) and *Trypanosoma congolense* (9.6%). Of 272 tsetse examined, 52 were positive for one or more of the different trypanosome species giving an overall tsetse infection rate of 19.1%.

The *T. brucei* identified in this study should be reanalysed to determine if it represents agents of Human African Trypanosomiasis, and control mechanisms should be established accordingly. Disease management tools using an integrated approach that targets both the vectors and reservoirs will offer a more sustainable option for trypanosomiasis control.

This project is helping to build capacity at Gulu University in the use of molecular tools for trypanosomiasis epidemiological studies. The project complements the ABCF project of Harriet Angwech on 'Molecular epidemiology and characterization of trypanosome infections of domestic livestock in the post conflict districts of Amuru and Nwoya, Northern Uganda'.

**Partners:** Gulu University; BecA-ILRI Hub.





## Dora Chao Kilalo

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Apr 16–Oct 16 2012

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### Validation and transfer of diagnostic tools for viruses causing passion fruit woodiness disease

Passion fruit woodiness disease (PWD) is known to be caused by three potyviruses, *cowpea aphid-borne mosaic virus* (CABMV), *passion fruit woodiness virus* (PWV) and *East Asian passiflora virus* (EAPV), in various parts of the world. PWD is a major biotic stress of passion fruit worldwide. It is also a problem in Kenya and in the East African region, where the disease is caused by CABMV and PWV. Promotion and production of passion fruit has recently been increased to help alleviate poverty among small scale farmers in Kenya, however increased PWD incidence has also been observed. Unfortunately, there is no sensitive indexing tool to avail virus-free planting material.

During a placement at BecA-ILRI Hub through AWARD in 2010, PCR primers to CABMV and PWV were designed to the coat protein gene sequences that were available in databases. They were tested on leaf samples collected from farmers in the main growing areas in Kenya. CABMV was detected in 45% of samples using the specific primers. PWV was not detected in any sample. The CABMV and universal potyvirus PCRs are more sensitive than commercial ELISAs. PCR using universal potyvirus primers also detected an additional 20% of samples not positive with the CABMV test, indicating another passion fruit potyvirus in Kenya. Diversity of CABMV in Kenya was characterized with the coat protein sequences. The diagnostic test was used to confirm that 100% of samples collected from the Kenya Agricultural Research Institute (KARI) motherblock (for seed production for distribution) were positive for CABMV.

The current study objectives are to: (1) validate the CABMV-specific and universal-potyvirus PCR diagnostic tools for virus indexing of passion fruit, (2) to transfer the tool to Kenya Plant Health Inspectorate Services (KEPHIS), KARI and local universities to avail virus-free passion fruit planting material to Kenyan farmers, and (3) to identify the unidentified potyvirus. Research work is ongoing with the validation of the PCR tools, measuring the incidence of virus infection in passion fruit from local nurseries, and identifying the unknown passion fruit virus. The expected output from this project is a highly sensitive molecular diagnostic tool to be used by Kenyan organisations to identify virus-free planting materials for distribution to farmers, which will lead to greater yield and quality of product, giving higher incomes to farmers.

**Partners:** BecA-ILRI Hub; University of Nairobi.



## Benius Tukahirwa

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16 Apr – 15 Oct 2012

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### Characterizing type III secretion system (TTSS) genes implicated in hypersensitive response (HR) induction and pathogenicity of *Xanthomonas campestris* pv. *musacearum*

Banana is an important household food security crop and source of income in East Africa. Production is currently threatened by an emerging bacterial disease: the Banana *Xanthomonas* wilt (BXW) caused by a gram-negative bacterium *Xanthomonas campestris* pv. *musacearum* (Xcm). BXW is considered the biggest threat to banana production in the region, and there are no resistant banana genotypes to be employed against the disease. Pathogenicity of bacterial phytopathogens is mainly dependent on type III secretion system (TTSS) genes encoded by the chromosomal *hrp* gene cluster. These enable successful bacterial multiplication and disease development in susceptible host plants and the hypersensitive response (HR) in resistant and non-host plants. As of now there is no knowledge on the pathogenicity mechanism of the pathogen Xcm upon host banana infection.

This study is investigating the virulence mechanism of the banana wilt pathogen (Xcm). This will be achieved by identifying the TTSS (*Hrp* and Effector) genes expressed and perhaps responsible for bacterial multiplication in planta for infection in host banana and HR in non-host maize plants. The progressive multiplication of the bacteria *in vitro* and *in planta* will be assessed, after which TTSS genes, which are expressed at different stages of the infection, will be identified using qPCR. Data obtained from this study will guide on further studies leading to generation of gene-based approaches to control the disease.

Preliminary results showed that bacteria multiplied in inoculated banana leaf tissues to about 109 CFU/cm<sup>2</sup> after 20 days post inoculation (dpi) from an initial density of about 104 CFU/cm<sup>2</sup> (at point of inoculation) and reaching maximum at 28 dpi when the inoculated leaves have completely wilted. Bacteria inoculated into the non-host maize leaves were unable to multiply and the density declined to about 103 CFU/cm<sup>2</sup> from the initial density of about 104 CFU/cm<sup>2</sup> in 10 dpi. The decline in bacteria population in non-host maize could be the restriction of bacterial growth by the HR at the point of inoculation, and this is currently being investigated. Currently, RNA samples from inoculated plants are being analysed by qPCR.

Comparison of the expression of these genes in host and non-host will help to elucidate mechanism behind host colonization and infection, and subsequently to generation of gene-based approaches to control the disease. After this project, the TTSS genes implicated in host infection will be functionally characterized to establish roles they play either individually or in combination for successful bacterial multiplication *in planta* and hence disease development. These genes will be potential targets in developing new methods to control the BXW disease.

**Partners:** NaCRRI/National Agricultural Research Organization (NARO); International Institute of Tropical Agriculture (IITA)-Nairobi; Makerere University, Uganda.



## Esther Kanduma

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Co-supported by AWARD

### Development and laboratory evaluation of a lateral flow test (LFT) for the serodiagnosis of *Theileria parva* infection in cattle

East Coast Fever (ECF) is caused by the haemoprotozoan parasite, *Theileria parva*, and is considered to be one of the most important cattle diseases in East, Central and Southern Africa. It is a major constraint to livestock productivity by causing high mortalities and morbidity, especially in exotic or cross-bred cattle. ECF control is normally achieved by acaricide treatment of cattle. An effective live vaccine against ECF is available, but currently is not widely used.

Conventional diagnosis of *T. parva* is based on the microscopic demonstration of parasites in blood. An ELISA based on a recombinant antigen is available for the detection of *T. parva* antibodies in cattle and is the method of choice for epidemiological studies and large-scale investigations. Several molecular tools have also been developed for *T. parva* detection. The ELISA and molecular tools are not widely used outside of well-equipped laboratories.

There is a need for affordable rapid 'point-of-care' diagnostic test of *T. parva* infection in cattle. A simple antibody test would be useful in ECF surveys and for monitoring control programmes including vaccination, especially in resource-poor countries with little laboratory support, such as South Sudan where ECF is an emerging disease.

The objective of the current project is to develop and validate a rapid, simple lateral flow test (LFT) for the detection of *T. parva* antibodies in cattle. The research activities will include the expression and purification of a *T. parva* recombinant diagnostic antigen and the production of rabbit polyclonal antibodies to the antigen. Using the recombinant antigen and antisera, a commercial company in Europe will then manufacture the LFT devices, which will be evaluated at the BecA-ILRI Hub using defined cattle infection sera. To date, the antigen gene has been cloned and sequenced.

The expected output from this research will be a *T. parva* serodiagnostic LFT for use in the field to monitor ECF, and to support control programmes including the deployment of a live vaccine against ECF. The LFT will have particular application to countries with limited laboratory facilities.

**Partners:** University of Nairobi; BecA-ILRI Hub; ILRI Biotechnology Theme; Research Center Borstel, Germany.



## Nehemie Donfagsiteli Tchinda

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1 May– 31 Oct 2012

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### Genetic diversity of wild and cultivated *Dacryodes edulis* (African plum) in Cameroon

*Dacryodes edulis*, otherwise called African plum, is a multipurpose fruit tree known for its dietary uses and economic importance. The tree is a source of edible oils and nutritious fruits. In central Africa, cultivated forms are integrated in cropping systems and consist of heterogeneous genetic material, resulting from empirical selections in different agro ecological regions. *D. edulis* is grown in a region still dominated by subsistence agriculture that is the centre of diversity for the species. Some populations grown are wild but the species has evolved very little over time. Thus there is need to enhance genetic richness of this important crop. The aim of this project is to assess genetic diversity, population structure and gene flow of *D. edulis*. This will help in identification, germplasm collection, improvement and conservation.

Leaf samples were collected from 25 wild *D. edulis* trees in Mbakwa Supe region and 50 cultivated trees in Yaounde and Santchou regions in Cameroon. Six available microsatellite markers were used for genotyping. Genotyping revealed polymorphism and allelic richness within the populations, helping to group individuals presenting with similar characteristics. This will provide a baseline survey of the diversity of this species in Cameroon. For a good practical system of molecular characterization of *D. edulis*, additional microsatellite markers are required. 454 pyrosequencing technology will be used to identify additional microsatellites.

The results will contribute to a better knowledge of population structure within and between cultivated and wild forms. New microsatellite markers will be of great value for germplasm evaluation and improvement programmes for *D. edulis* in Cameroon and Africa.

**Partners:** Medicinal Plants and Traditional Medicine Research Centre, Cameroon; BecA-ILRI Hub; ILRI Biotechnology Theme; International Centre for Research in Agroforestry (ICRAF), Nairobi.



## Monde Godefroid

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1 May – 31 Aug 2012

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### **Molecular epidemiology of cassava mosaic and cassava brown streak virus pandemics in North-eastern Democratic Republic of Congo**

Cassava is known as a staple food in sub-Saharan Africa. Both cassava leaves and roots nourish > 60% of population in Democratic Republic of Congo (DRC). However, two pandemic viruses are the most economically important biotic constraints of cassava cultivation in DRC: Cassava Mosaic Disease (CMD) caused by a ssDNA Begomovirus (Geminiviridae) and suspected cases of Cassava Brown Streak Disease (CBSD) caused by a ssRNA<sup>+</sup> Ipomovirus (Potyviridae). Both viruses are persistently or semi-persistently transmitted by the same whitefly insect vector, *Bemisia tabaci* (Aleyrodidae; Hemiptera). Transportation of infected propagation materials is strongly pointed to contribute to CMD and CBSD spread.

Cassava affected by CBSD produces non-edible tuberous roots that cannot be marketed, making CBSD the most economically damaging viral disease for cassava since it affects the quality and quantity of roots. CBSD-like symptoms identified in DRC are not yet confirmed by any virus diagnostic technique; this justifies the necessity of molecular diagnostics to confirm the presence (CBSV) and begin to understand the distribution pattern of these cassava viruses.

The objectives of this project were (1) confirm the presence of CBSD in DRC using molecular techniques, (2) determine the genetic diversity CMD- and CBSD-associated viruses in cassava cultivated in DRC using molecular techniques, and (3) determine the distribution of the virus strains in agro-ecological zones in order to contribute to global control strategy.

Field samples from north-eastern DRC were analyzed for the presence of CMV and CBSV using PCR and RT-PCR respectively. The incidence of CMV as determined by PCR was 59% while the observed field incidence of CMD was 47%. Two species of CBSV were identified in the samples: *Cassava brown streak virus* (CBSV) and *Cassava brown streak Uganda virus* (CBSUV), with a total incidence of 43% as determined by RT-nested PCR. This is the first confirmation of CBSV in DRC. Interestingly, the observed molecular incidence of CBSV (43%) was far higher than the field incidence of CBSD (5%). This suggests that some plants are non-symptomatic carriers of CBSV and highlights the importance of developing reliable molecular diagnostics tests for the virus to facilitate the implementation of effective CBSD control measures. Furthermore, there may be sources of tolerant material in DRC.

**Partners:** Agriculture Institute of Yangambi; BecA-ILRI Hub.



## Tony Bakelana Zeyimo

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7 May - 30 Sept 2012

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### **Prevalence of cassava viral diseases in different agro-ecological zones in the western part of Democratic Republic of Congo (DRC)**

The most widespread crop disease in Africa, in terms of economic importance, is arguably cassava mosaic disease (CMD), transmitted by the whitefly *Bemisia tabaci*. CMD is known to be caused by viruses belonging to nine different begomovirus species in the African continent and the Indian subcontinent. During the 2000s, DRC experienced an outbreak of the CMD Ugandan variant (EACMV-UG) which was detected in many areas in the country and resulted in significant crop losses by smallholder farmers. Cassava brown streak disease (CBSD) is another viral disease also transmitted by *B. tabaci* whose causal agent is a potyvirus. CBSD has been a problem in the East African coastal cassava growing areas for more than 70 years. CBSD has been reported in various countries including Kenya, Malawi, Mozambique, Tanzania and Uganda. Other countries are suspected to be affected, including DRC, Angola, Congo Brazzaville, Gabon and Cameroun. CBSD-like symptoms were reported for the first time in DRC in 2002. Since 2002, several diagnostic investigations have been undertaken on affected plant samples (leaves and roots) from the western part of DRC in an attempt to find out the causative agent; none have been successful to date.

The objectives of this project are (1) to identify viruses causing CMD- and CBSD-like symptoms in western DRC using molecular techniques, (2) determine the spatial distribution of the viruses within western DRC. As the most likely causative agents are CMV and CBSV initial efforts focused on attempting to identify these viruses in the samples. Nucleic acids were extracted from the cassava samples and analyzed for CMV and CBSV by PCR using published primers. CMV was detected in 86% of samples with 9% being ACMV positive, 11 % being EACMV-UG positive and 66% with mixed infections (ACMV and EACMV-UG). The sequencing of CMV PCR products is ongoing and should provide valuable data on the CMV present in the region. All samples tested negative for CBSV using published East African species specific primers and universal potyvirus primers. This suggests that another causative agent is present in these samples.

Future work – Diversity analysis of CMV sequences from western DRC is ongoing. Deep sequencing of freshly collected infected cassava leaves using next-generation sequencing of polyA+RNA and small RNAs should help to elucidate the causes of CBSD-like symptoms in western DRC. This also has the potential to discover novel viruses, in addition to being the most in depth analysis of cassava pathogens in DRC. Data collected from deep sequencing can be used to develop molecular diagnostic assays for the causative agents.

Partners: INERA; Kinshasa University; BecA-ILRI Hub; International Institute of Tropical Agriculture (IITA); FAO.



## Parfait Kouadio Kouakou

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15 May – 15 Oct 2012

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### Genetic diversity of the domestic cavy in Cote d'Ivoire

The domestic cavy (guinea pig, *Cavia porcellus*) is a promising “micro-livestock” species for increasing food and nutritional security and income generation in many parts of rural Africa. It requires little investment, feeds from kitchen wastes, and yields a cheap, high quality meat. The cavy has rapid growth, high reproductive rate with up to 5 litters per year, and is less prone to diseases than chickens, rabbits and pigs. Despite its potential, and its widespread distribution across west, central and east Africa, the cavy is a neglected livestock species in the continent. Little is known about local husbandry practices, body weight, feeds, or existing breeds. It is suspected that the cavy has a very narrow genetic base in Africa, with little potential for improvement through breeding.

The objective of this ABCF project was to determine the genetic diversity of cavies in Côte d'Ivoire. Two key areas were of interest: a) the genetic diversity and inbreeding levels in the populations, b) the population structure. 140 cavy blood samples were collected from north, central and south Cote d'Ivoire. Phenotypic data and information on cavy farming systems were also collected. Sixteen SSR (microsatellite) markers were used in the genetic analysis.

The analysis of the genotypes revealed considerable similarities between the three populations. This applied for both sub populations and for the total population. Although the three populations were considered as different genetic entities, the populations presented only minor changes in the allele frequency. There were no clear differences in the population structure with only 2.6% variation among the three populations and 22% variation among individuals within a population. There were high rates of inbreeding in all the three populations. It is difficult to select non-related animals and thus control inbreeding in the target populations or select for particular traits of interest. These results can be attributed to the non-random mating systems practiced by the farmers. The cavies are reared in small confined areas with only one male in the herd for the purposes of breeding. With such a narrow genetic base between the populations it is likely that the population could show an increase in the negative aspects associated with inbreeding.

This study complements two other ongoing cavy projects by increasing the number of genotyped populations of domestic cavies in East and Central Africa: (1) the project of ABCF Fellow, Bertin Bisimwa, who is conducting a similar study on cavies from DRC, and (2) the BecA-CSIRO FANS project: ‘Harnessing Guinea Pig Husbandry for Alternative and Rapid Access to Income and Food in Cameroon and DRC’.

**Partners:** BecA-ILRI Hub; University of Dschang, Cameroon; Catholic University of Bukavu; International Center for Tropical Agriculture (CIAT); University of Abobo Adjame, Cote d'Ivoire.



## Laude Abel Ouakou

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21 June - 21 Oct 2012

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### Identification and characterization of Cassava Mosaic Viruses and Cassava Brown Streak Viruses at Bateke-plateau in Republic of Congo using molecular tools

Cassava is grown in most parts of the Republic of Congo (ROC), where 95,700 ha are under cultivation with a total production of 861,500 t. Per capita consumption in ROC is the highest on the continent (FAO, 2003). Production of cassava in Africa is greatly constrained by pests and diseases, particularly cassava mosaic disease (CMD) and cassava brown streak disease (CBSD). CMD is caused by cassava mosaic geminiviruses (*Cassava mosaic viruses*; CMV) transmitted by the whitefly vector, *Bemisia tabaci*, and through the infected cuttings used for vegetative propagation. Viruses causing CBSD (*Cassava brown streak viruses*; CBSV) belong to the genus *Ipomovirus*, family *Potyviridae*, and are also transmitted by *Bemisia tabaci* whiteflies.

The objective of this project are to (1) determine, using published PCR methods and newly developed methods where necessary, if the CBSD-like symptoms in cassava from ROC are actually caused by known CBSD associated viruses, (2) to further confirm the presence of CMD associated viruses by PCR and their diversity by sequencing, (3) to establish the incidence of co-infection in field samples, and (4) provide training in molecular diagnostic tools that can be transferred to ROC. Results from this study will help researchers in ROC to identify improved diseases resistant cultivars, to identify clean planting materials and to identify and monitor the diseases affecting cassava. This will improve the quality and productivity of cassava and through their adoption, improve food security.

To date molecular tools have been used to confirm that CMD symptoms observed throughout ROC, resulting in 60-80 % crop loss, were indeed caused by CMV. Two variants of CMV were detected, namely ACMV and EACMV-UG, this is the first confirmation of CMV in ROC using molecular techniques. CBSD symptoms also occur in ROC, but the samples tested negative for CBSV using published PCR primers. This suggests that the CBSD-like symptoms observed these samples may be due to unknown variants of CBSV or potentially a novel as yet uncharacterized virus. Work will continue using genus-wide potyvirus primers which may help to detect CBSV variants more distantly related to those previously characterized in East Africa. Alternatively, deep sequencing of RNA may provide the most comprehensive analysis of the pathogens present in the CBSD symptomatic samples.

**Partners:** Marien Ngouabi University; BecA-ILRI Hub.





## Christopher Mukasa

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26 Jun – 22 Nov 2012

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### **Harnessing genetic diversity for conservation, resistance to disease and improving productivity of goats in Uganda and Nigeria**

Indigenous goats are an important resource for African farmers, providing meat, milk, manure, fibres and hides, and satisfying various cultural and religious functions. In Africa, the survival of many indigenous goat populations is threatened by diseases, adverse climatic conditions, civil strife, pressure of economic development, abandonment of traditional farming practices, and more importantly through cross-breeding or replacement with animals from the developed world. Many gaps still exist in our understanding of productivity, adaptation and disease resistance traits, as well as the genes controlling these traits. The characterization and mapping of genes controlling such traits – “quantitative trait loci” (QTL) and the subsequent use of this information in selection and breeding programmes, should make it possible to facilitate significant increases in small ruminant productivity

The current study is being carried out to obtain detailed information on the phenotypic and genetic diversity and differentiation of the indigenous goat populations of Uganda and Nigeria. The objectives of this study are to assess phenotypic and genetic variability, and detect population structure, which are vital for conservation and utilization of goat breeds in the two countries.

The indigenous goats were classified phenotypically into three types in Nigerian (Sahel, Red Sokoto and West African Dwarf) and into three types in Uganda (Mubende, Small East Africa and Kigezi). The phenotypic parameters recorded were: chest (heart) girth, body length, height at the withers and pin bone length. Information on common disease occurrence, reproduction and production were also recorded.

At the BecA-ILRI Hub, the populations are being assessed for genetic diversity using 20 microsatellite markers and comparison will be made with the phenotypic data. Standard statistical procedures will be employed to analyze the phenotypic and molecular data.

The genetic data will be used to help breeders in Uganda and Nigeria to implement rational decisions for conservation and improvement of valuable germplasm. This study is a starting point and critical component of a much larger goat project to generate a haplotype map (HapMap) of East and West African goat for genetic improvement.

**Partners:** National Animal Genetic Resource Centre and Data Bank, Entebbe, Uganda; Ahmadu Bello University, Zaria, Nigeria; BecA-ILRI Hub.



## Diaeldin Ahmed Salih Hassan

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1 Jul – 30 Nov 2012

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### Genotyping of *Theileria annulata* in Sudan to support control of tropical theileriosis

Tropical theileriosis is a serious disease of cattle in tropical and sub-tropical regions of the world, including Sudan. The disease, caused by the tick-borne protozoan parasite *Theileria annulata*, is often fatal in calves, and meat and milk production is reduced in older cattle. More productive European cattle breeds are most susceptible to the disease and this prevents improvement of livestock in endemic regions.

Tropical theileriosis is endemic in the north and central parts of the Sudan. Due to changes in the distribution of the tick vector, *Hyalomma anatolicum anatolicum*, outbreaks of the disease have been reported in east and west parts of the country, outside the known endemic areas. To control the disease, a live attenuated vaccine based on a Sudanese strain of the parasite, has been developed. Before the vaccine can be deployed there is a need to genotype both the parasite in the field and the vaccine strain. This is very important data to enable monitoring of the vaccine after deployment for any breakthrough or breakdown. Such data may also be useful for identifying the origins of new outbreaks. Very little data is currently available regarding the genotypes of this *Theileria* parasite in Sudan.

In the current study, a PCR diagnostic assay was carried on 139 cattle samples collected from several regions of Sudan. A total of 78 samples were positive for *Theileria annulata*, of which 45 samples were from known endemic areas in the north and central parts of Sudan, while the remaining were from 'new areas' in the east and west of the country. These positive samples, and the vaccine strain, will now be subjected to genotyping using microsatellite markers.

With the data, the government of Sudan will be in a good position to deploy the vaccine in the first quarter of 2013, confident that vaccine performance can be monitored through the use of molecular tools, which will be transferred to the Veterinary Research Institute in Khartoum.

**Partners:** Veterinary Research Institute, Khartoum; BecA-ILRI Hub; ILRI Biotechnology Theme.



## Wani Loku Marcellino

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1 Jul – 30 Nov 2012

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### **Distribution of ticks and tick-borne diseases of cattle in South Sudan, with particular emphasis on East Coast fever, an emerging disease**

East Coast fever (ECF) is a serious disease of cattle caused by the protozoan parasite *Theileria parva*, and transmitted by the tick *Rhipicephalus appendiculatus*. ECF is a major challenge to development of the cattle industry in most of Southern, Central and Eastern African countries. ECF is an emerging disease in South Sudan, and is having a devastating impact on cattle production. Epidemiological studies in the 1980s showed that *R. appendiculatus* and ECF was confined to Central and Eastern Equatoria region of South Sudan. However, after the signing of a comprehensive peace agreement (CPA) in January 2005, there has been extensive movement within South Sudan of people and their livestock, with concomitant reports of ECF spreading to more northern areas that were previously free of the disease, including 2012 outbreaks in Warrap and Jonglei States. There is, therefore, an urgent need to conduct epidemiological studies to determine the extent of the disease in the country. Data from these studies will form the basis of integrated control strategies for South Sudan.

The objective of this project is to determine the distribution of ticks and TBD of cattle in South Sudan by tick identification (by microscopy and DNA barcoding) and serological and molecular tools, with particular emphasis on determining the northern limit of *R. appendiculatus* and *T. parva* in South Sudan.

The areas included in the study were Central Equatoria and Jonglei States. A total of 1636 ticks were collected from these states, of which 250 were identified as *R. appendiculatus* from areas as far north as Bor. To date, the results of serology and molecular analysis show confirm that *T. parva* is present in Central Equatoria, and also in Jonglei State north to Bor along the Nile. This is the first laboratory confirmation of *R. appendiculatus* and *T. parva* north of Juba.

The results of this work will assist the South Sudan government in developing a strategy on ECF control, and where to focus control measures. ECF control, which is key to the health of cattle in the region, will be directly reflected in animal production and consequently in improving livelihoods. In addition, the project may ultimately enable introduction of foreign breeds and cross-breeding to improve smallholder income which may strengthen the peace process by assisting with settlement of refugees and job creation.

**Partners:** Ministry of Animal Resources and Fisheries, South Sudan; BecA-ILRI Hub; ILRI Biotechnology Theme; Veterinary Research Institute, Sudan.



## Awadia M. Ali Mousa

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1 Jul - 30 Nov, 2012

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### **Genotyping of *Theileria lestoquardi* from sheep in Sudan to support control of Malignant Ovine Theileriosis, an emerging disease**

*Theileria lestoquardi* is a tick-borne protozoan parasite transmitted by Hyalomma ticks and causes malignant ovine theileriosis (MOT) in sheep. The disease causes high mortality and severely impacts on the livelihood of resource-poor farming communities in Sudan. MOT is endemic in some parts of Sudan, but outbreaks in new areas also occur. Currently, management of MOT in Sudan is through control of the tick vector using acaricides, although there are concerns with increased acaricide resistance and food safety. Control of MOT can be achieved by immunization of sheep with attenuated *T. lestoquardi* schizont-infected ovine cells. An attenuated vaccine against MOT has been carried out successfully in Iraq and Iran. A live vaccine based on a Sudanese isolate of *T. lestoquardi* (Atbara strain) has been developed for the control of MOT in Sudan, but not yet deployed in the field.

This project will provide background information to support the deployment of the live vaccine in Sudan. The objectives are to define the genotype of the live vaccine and *T. lestoquardi* isolates in the field. There is need to compare the diversity in the field with the vaccine genotype before a decision is made to deploy the vaccine in Sudan, as there is a concern that vaccination with live parasites could introduce new and potentially more virulent genotypes into new areas. Knowing the genotypes of vaccine and field strains will also help understand the nature of any vaccine failures or breakthroughs that occur. Genotyping information will also help to reveal the possible origins of the rapidly spreading *T. lestoquardi* outbreaks in Sudan.

Preliminary results using a molecular diagnostic test on sheep samples from the field show an infection rate of 35%, confirming that *T. lestoquardi* is endemic in the north part of the Sudan. Infected samples are now being genotyped using microsatellites and antigen gene sequencing.

The results of this work will assist in deciding on the best control policy for control of MOT in Sudan, which is key to the health of sheep in the region and will be directly reflected in animal production and consequently in increasing income of farmers. In addition, the project may ultimately enable the upgrading of sheep breeds to improve smallholder incomes.

**Partners:** University of Khartoum; BecA-ILRI Hub; ILRI Biotechnology Theme; Veterinary Research Institute (VRI), Sudan; Research Center Borstel-Germany.



## Gaspard Ndarubayemwo

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2 Jul – 2 Dec 2012

### Identification of fungi infecting taro in Burundi

Burundi is a predominantly agricultural country. Food crops are the main source of income for rural people. The agricultural sector is, however, facing serious problems or deficiencies. Most of the crop species grown locally are suffering from degenerative diseases caused by bacterial, fungal or viral infections.

Taro is an important crop that feeds many families during the dry season in Burundi. It is actually suffering from degeneration (from 1992 to 2010 taro production decreased from 140,000 tons to 18,000 tons). The suspected causes include soil fungal diseases. Taro is susceptible to at least twenty-three fungal pathogens among the soil fungi (e.g. *Pythium*, *Fusarium*, *Macrophomina*). Farmers are currently facing an acute shortage of good planting material. To overcome this situation, this project aims to identify and characterize the fungal diseases, to produce clean planting material using tissue culture and then transfer clean plantlets to private laboratories for multiplication and subsequent distribution. Given the nature of fungal diseases, the identity of the fungi infecting taro in Burundi will inform the wider community about which management strategies might help, as well as help in future identification of sources of resistance.

To achieve these objectives, corms of *Colocasia esculenta* and *Xanthosoma sagittifolium*, the two taro species grown in Burundi, were collected from different agro-ecological zones of Burundi. Fungi were cultured, isolated and identified from the corms using direct plating and macro and micro observation. Purification is ongoing and molecular analysis (PCR and sequencing) will be used to confirm isolate identity and characterize overall diversity. So far, using direct plating, we have successfully isolated various fungi belonging to different taxonomic groups. These include (i) *Fusarium spp.*, (ii) *Penicillium spp.*, (iii) *Pythium spp.*, (iv) *Bissoclamyces spp.*, (v) *Aspergillus flavus* and *A. niger*. We have analyzed the incidence of these fungi in each region from which samples were collected. For tissue culture, we have established plantlets of *C. esculenta* and *X. sagittifolium* on multiplication medium.

This project complements the taro projects of Donatien Bigirimana and Niyonzima Pierre which focus on virus identification and tissue culture for the production of clean taro planting materials for Burundi; and ABCF Fellow Dawit Beyene from Ethiopia focusing on virus diagnostics.

**Partners:** University of Burundi, Bujumbura; Burundi Agriculture Research Institute-ISABU; Laboratoire de l'Agro-biotechnologie (AGROBIOTEC); Laboratoire de Biotechnologie des Plantes du Burundi (PHYTOLABU); Provincial Directorate of Agriculture and Livestock (DPAE); Ethiopian Institute of Agricultural Research (EIAR); BecA-ILRI Hub.



## Donatien Bigirimana

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2 Jul – 2 Dec 2012

### Virus identification, diagnostic development and tissue culture for clean taro multiplication in Burundi

Taro is an herbaceous and perennial plant of the aroid family. Its high carbohydrate content makes it an important energy food. It is an important crop in Burundi on which many resource-poor subsistence farmers and consumers rely during the dry season. In spite of its importance, taro culture in Burundi is threatened by several diseases, mainly fungal and viral. From 1992 to 2010, taro production in Burundi decreased from 140,000 tons to 18,000 tons (FAOSTAT, 2011). Actually, five taro viruses are known in the world and similar viral symptoms are observed on taro in Burundi. However, no research on taro viruses has been done in Burundi or the region to date. To overcome this situation, the project aims to identify and characterize the causal agents of taro viral diseases in Burundi, to develop diagnostics, to produce cleaned planting material using tissue culture.

To achieve these objectives, leaves from different agro-ecological zones of Burundi were collected. Viral diagnostics are being conducted using ELISA, PCR/RT-PCR and characterization by sequencing. At the end of this research, taro viruses in Burundi will be known, a diagnostic test will be developed, viral disease distribution in different agro-ecological zones in Burundi will be characterized and clean taro planting material will have been produced and transferred back to Burundi for multiplication and distribution to farmers.

So far, ELISA results show that, for 102 samples tested, 35 are infected by potyviruses. This translates to 34.3% of samples collected across Burundi are infected by these viruses. The identity of the virus(es) will be confirmed using further diagnostics and sequencing.

This project will produce the first information about taro viruses in Burundi (and the region). The identity of the virus(es) and appropriate diagnostic tests will be transferred back to the University of Burundi, ISABU and two private tissue culture laboratories that are already distributing millions of plantlets of other crop species to NGOs, for distribution to farmers across Burundi.

This project complements the taro projects of ABCF Fellows Pierre Niyonzima and Gaspard Ndarubayemwo from Burundi, which focus on tissue culture and diagnostics for the production of clean taro planting materials, and Dawit Beyene from Ethiopia, who is focusing on virus diagnostics.

**Partners:** University of Burundi, Bujumbura; Burundi Agriculture Research Institute-ISABU; BecA-ILRI Hub; Laboratoire de l'Agro-biotechnologie (AGROBIOTEC; Laboratoire de Biotechnologie des Plantes du Burundi (PHYTOLABU); Provincial Directorate of Agriculture and Livestock (DPAE); Ethiopian Institute of Agricultural Research (EIAR).



## Ann Nanteza

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### **Major Histocompatibility Complex I heterogeneity of the Ankole cattle in western Uganda: a marker of protection against East Coast fever?**

East Coast fever (ECF) of cattle, which is caused by *Theileria parva* infection, results in enormous economic losses to farmers and the livestock industry in East, Central and Southern Africa.

Improved *Bos taurus* breeds are more prone to severe ECF, unlike the local Ankole cattle breeds in Uganda that remain apparently healthy despite harbouring potentially lethal *T. parva* infections. The MHC I plays an important role in the response of cattle to *T. parva* infections and it is possible that particular sequence variants give rise to the CD8+ immune-protection against ECF.

In this study, the MHC I sequences of Ankole cattle will be determined and compared to available cattle MHC sequence data. This will provide insights into the differences amongst the various Ankole cattle breeds, which may be related to the *T. parva* infections. These variations could be correlated with ECF survival traits for the different Ankole cattle breeds, which would be useful for conservation or breeding for ECF resistance.

**Partners:** Makerere University, Uganda; ILRI Biotechnology Theme; BecA-ILRI Hub.



## Elisa Daniel Mwega

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2 Jul – 30 Nov 2012

### Genotyping of *Theileria parva* in cattle in Tanzania to support ECF vaccination

East Coast fever (ECF) is a lympho-proliferative disease of cattle caused by the protozoan parasite *Theileria parva*. It is a tick transmitted disease which kills a million cattle every year and devastates the livelihood of those who depend on cattle for survival. ECF devastates indigenous cattle, but is an even greater threat to improved cattle breeds and is therefore limiting livestock development, particularly affecting smallholder dairy production. The disease is endemic in Eastern, Central and Southern Africa. Acaricide treatment of cattle to control the tick vector is currently the main ECF control method, but this is not a sustainable approach. A live sporozoite vaccine was recently deployed in pastoral systems in Northern, Eastern and Central part of Tanzania, which great success. Despite this, many scientists have accepted that the widespread deployment of the live sporozoite vaccine has been hindered by several concerns including the possibility of introducing new strains through vaccination. Initial reports indicate that parasite populations are changed, at least in the short-term, after vaccination. There is, therefore, a need to understand the population genetics of *T. parva* in the field.

The aim of this project is to investigate the diversity of *T. parva* isolates present in cattle in eastern and southern part of Tanzania, including areas that have had ECF vaccination programmes. Blood samples were collected from 123 cattle. A sensitive *T. parva* PCR assay was used on the samples, and 39 were found to be infected with *T. parva*. All positive samples are currently being genotyped with 14 minisatellite and microsatellite markers. In addition, two CD8 antigens (Tp1 and Tp2), which are potential components of a new recombinant vaccine, are being sequenced to understand the antigenic diversity of *T. parva*.

The information generated from this study will increase our understanding of the diversity of the parasite existing in the field. This has very important implications for the sustained use of live sporozoite vaccine into wider areas in eastern and southern part of Tanzania. The results obtained from this project will be disseminated to policy and decision makers. The technologies and knowledge gained from this placement will be transferred to the home institute, SUA, in Tanzania.

**Partners:** Deutsche Forschungsgemeinschaft (DFG) project German-African Cooperation Projects in Infectology, “Molecular epidemiology network for promotion and support of delivery of live vaccines against *Theileria parva* and *T. annulata* infection in Eastern and Northern Africa” (AH 41/7-1); Nelson Mandela African Institute of Science and Technology, Tanzania; Sokoine University of Agriculture; BecA-ILRI Hub; ILRI Biotech Theme.





## Dawit Beyene

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16 Jul – 14 Dec, 2012

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### Viruses infecting the food crop Taro (*Colocasia esculenta* L.) in Ethiopia

Taro (*Colocasia esculenta* L.) is a tropical plant grown primarily for its edible starchy corms. The majority of the population in south and southwest Ethiopia solely depend on the root crops Enset (false banana), taro, potato and sweet potato for daily food consumption. There is a report that shows the production of taro in Ethiopia has declined significantly, in large part due to viral disease (personal communication, Root Crop Research Coordinator, Areca Agricultural Research Centre, Ethiopia). Taro, being a vegetatively propagated crop, is prone to viral infection. The objective of this project is to determine the identity and incidence of viruses associated with taro in Ethiopia. This information will support the implementation of diagnostic tools and management of germplasm movement, production of virus free planting materials through tissue culture and distribution to farmers. This will improve the productivity of taro and play a major role in poverty alleviation and food security.

The viral diseases of taro in East Africa have yet to be confirmed. For this project ELISA, PCR and LAMP-based disease diagnosis tools will be used to test for the following viruses, which have been shown in previous studies to infect taro in other parts of the world: Taro bacilliform virus (TaBV), Taro vein chlorosis virus (TaVCV), *Dasheen mosaic virus* (DsMV), *Colocasia bobone disease virus* (CBDV) and *Taro reovirus* (TaRV).

Two hundred and ninety five symptomatic and non-symptomatic taro leaf samples were collected from major growing areas of south and southwest Ethiopia. The results to date by ELISA show that 36.9% of the taro samples found to be positive for potyviruses and 26.8% react positively to DsMV specific antibodies. PCR and sequencing analysis is ongoing, but preliminary results from Badnavirus PCR have identified a DNA sequence with only 75% identify to *Dioscorea bacilliform virus* sequences, which might represent a novel virus variant. Further investigation is confirming the identity of this potentially novel virus, and others identified in the sample materials.

This project complements the taro projects of ABCF Fellows Pierre Niyonzima, Gaspard Ndarubayemwo, and Donatien Bigirimana, which focus on tissue culture and diagnostics for the production of clean taro planting materials for Burundi.

**Partners:** Ethiopian Institute of Agricultural Research (EIAR); BecA-ILRI Hub; Areca Agricultural Research Centre – Ethiopia; University of Burundi, Bujumbura; Burundi Agriculture Research Institute-ISABU.



## **Blaise Arnaud Hako Touko**

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20 Jul – 20 Dec 2012

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**Exploring disease resistance by evaluation of the genetic diversity of the major histocompatibility complex (MHC-B) in Cameroon local chicken populations**

In Cameroon, the local ('native') chicken is the major livestock species used by rural households in low input and dual purposes production systems. Therefore, conservation and productivity improvement will be of great importance in poverty alleviation and food security for poor families. However, the local chicken is subject to serious production constraints. Diseases are a major cause of production loss, of which Newcastle disease (ND), with 70 - 100 % mortality, is by far the most devastating.

Previous studies have been carried out mainly on phenotypic characterisation of the local chicken and production systems. However, the genetic potential of the Cameroon local chicken cannot be efficiently improved without also considering the genetics. The general objective of this project is to contribute to the genetic improvement of the local chicken, which will lead to increase in income for rural householders. More specifically, this project aims to assess the genetic variability of the chicken Major Histocompatibility Complex (MHC), correlating MHC-B polymorphism with QTLs associated with ND resistance or susceptibility and ND antibody response. The MHC-B and QTL polymorphism will be studied using microsatellite markers on 380 chicken blood samples collected according to phenotype, ecotype and Cameroon agro-ecological zone. At the end of the project, genotypes associated with resistance will be linked to in situ community-based management of native chicken genetic resources.

This project complements two other ABCF projects investigating local chicken diversity in Central Africa: (1) Christian Keambou who studied the genetic diversity of the local chicken, and identified three chicken groups in Cameroon, and (2) Célestine Bembide who is currently studying the local chicken diversity in the Central African Republic.

**Partners:** University of Dschang; Institut National Supérieur d'Agronomie (INSAB), Gabon; BecA-ILRI Hub; ILRI Biotechnology Theme; Institut Centrafricain de la Recherche Agronomique (ICRA), Central African Republic.



## Tajelser Idris Badri

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23 Jul – 23 Nov 2012

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### **Development of a thermostable vaccine against Peste des petits ruminants (PPR)**

Peste des petits ruminants (PPR), is an acute, highly contagious transboundary disease caused by a virus of the same family as the rinderpest virus of cattle. It affects mainly sheep, goats and occasionally small ruminants in the wild, causing mortality up to 100%. It occurs in East and Central Africa, Middle East and the Indian Subcontinent, where it is a major threat to the small ruminant industry, and directly impacts upon poor farmers, the main owners of sheep and goats. It also limits trade and export in sheep and goats.

Current control of the disease includes animal isolation and disinfection of the contaminated area, and vaccination of animals with a live-attenuated vaccine. The Nigerian 75/1 PPR vaccine strain is commonly used for vaccination. However, the vaccine is heat sensitive and requires a cold chain to maintain vaccine efficacy during transportation. Maintenance of cold chain is difficult in tropical and subtropical countries. A thermostable vaccine would avoid the need for a cold chain, and would help enable mass vaccination of sheep and goats in endemic countries for PPR eradication.

This ABCF research project at the BecA-ILRI hub directly links with the PPR project of the partnership between BecA and Commonwealth Scientific and Industrial Research Organisation (CSIRO), under the supervision of Dr Jeff Mariner of the ILRI Biotechnology Theme. Activities at the Hub include the production of new batches of thermostable vaccine. Production will involve virus inoculation of cell cultures, vaccine lyophilization using protocols established within the BecA-CSIRO project, sterility, identity, and stability tests, and determining the residual moisture in vaccine batches. Research will also be conducted to identify new protocols for thermostable PPR vaccine production.

Batches of PPR vaccine prepared at the BecA-ILRI Hub will be used in a field trial in an endemic area in Sudan to assess the efficacy and potency of the vaccine against the natural infection in a hot climatic environment. This new vaccine will reduce the cost of transportation and improve control and eradication of PPR.

**Partners:** ILRI Biotechnology Theme; BecA-ILRI Hub; CSIRO; Kenya Agricultural Research Institute (KARI); Ministry of Animal Resources, Fisheries and Ranges, Sudan.



## Pierre Niyonzima

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23 Jul - 21 Dec 2012

### Taro (*Colocasia esculenta*) tissue culture and virus diagnostics for the production of clean planting materials

Bacterial, viral and fungal diseases of taro cause reduction in quality and yield. Tissue culture has assumed considerable importance as a method of producing clean taro plants, however information on diseases and appropriate diagnostic tests are lacking in Burundi and the region as a whole.

The project aims are to identify and characterize taro viral diseases and their causal agents in Burundi, and to produce clean planting material through elimination of these viruses through tissue culture and diagnostics. To achieve those objectives, 102 leaf samples from lowland, midland and highland agro-ecological zones of Burundi were collected and brought to the BecA-ILRI Hub.

To identify virus-positive samples (and clean materials), leaves were tested by antigen-coated plate (ACP) ELISA using polyclonal antibody specific for potyviruses. Results to date show that 35/102 of the plant materials collected from Burundi are infected with potyvirus, of which a subset were positive by Dasheen mosaic potyvirus (DsMV) ELISA. The identity of the potyviruses in the DsMV negative samples will be determined by RT-PCR and sequencing. Other virus groups will be identified using universal and specific PCR and sequencing. Primers will be designed to novel virus sequences for the development of new diagnostic assays that will be applied in the production of verified clean planting materials in Burundi, for distribution to farmers.

The clean planting materials produced at the BecA-ILRI Hub will be transferred and maintained in the plant biotechnology laboratory at the Institut des Sciences Agronomiques du Burundi (ISABU), the Burundi national agriculture research institution. Diagnostic technologies will also be transferred to ISABU, as well as two private tissue culture companies in Burundi currently distributing millions of plantlets (of other crop species) to farmers annually, through NGO partners. Clean plantlets will also be passed to Laboratoire de l'Agro-biotechnologie (AGROBIOTEC) and Laboratoire de Biotechnologie des Plantes du Burundi (PHYTOLABU), the private laboratories, for multiplication. ISABU will also distribute the clean plants to nurseries and farmers via its collaboration with the Provincial Directorate of Agriculture and Livestock (DPAE).

**Partners:** ISABU, Burundi; BecA-ILRI Hub; AGROBIOTEC; PHYTOLABU; Provincial Directorate of Agriculture and Livestock (DPAE).



## Célestine Bemvide

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23 Jul -21 Dec, 2012

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### Phenotypic and molecular characterization of local chicken populations (*Gallus gallus*) in Central African Republic

The 'local' ('native') chickens found in most villages in Central African Republic (CAR) are associated with household food security as they are often found in poorer households that are unable to keep other livestock. These chickens have not been subjected to selection in formal research or breeding programmes nor have they been included in national conservation programmes. Also, there is no published data on CAR chicken classification based on phenotype, geographical location or genotype. To begin to understand the population structure, a phenotypic characterization of local chickens in different agro ecological zones in CAR was carried out. These studies found significant differences within local breeds of chickens suggesting a reservoir of genetic diversity that could contribute to the definition of breeds and improved production of the various chicken lines. The genetic diversity found in local breeds confers on them the ability to adapt and survive in the challenging environmental and ecological conditions associated with farming in CAR.

To complement the phenotypic characterization, the current project will use molecular markers (microsatellites) to investigate the genetic diversity and population structure in the CAR chicken, the genetic distance between the different phenotypes observed and to identify unique markers for breed characterization. This analysis will facilitate the development of strategies for genetic improvement and conservation of CAR genetic resources in support of food security and poverty reduction.

Samples for genotyping were collected from 205 local chickens from two agroecological zones in three Divisions in CAR (savannah and forest in Lobaye and Ombella Mpoko Divisions; savannah in Ouham Division). Different phenotypes were identified based on several criteria, including feather structure and distribution.

A number of FAO-recommended microsatellite markers are available for genotyping. In this study 28 markers will be used to investigate the genetic diversity and population structure of local chickens in CAR, and to correlate with phenotyping data, which should lead to clarification of parameters for breed differentiation and genetic conservation of this valuable resource.

This project complements the African local chicken projects of three other ABCF Fellows: Christian Keambou, Arnaud Hako and Sheila Ommeh.

**Partners:** ICRA; BecA-ILRI Hub; ILRI Biotechnology Theme.



## Jibril Lubega

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1 Aug – 28 Sept, 2012

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### **Establishment of genetic transformation of East Africa Highland Banana for resistance against biotic stress using meristematic tissues**

Banana bacterial wilt caused by *Xanthomonas campestris* pv. *musacearum*, is a vascular disease that results in permanent wilting and eventual death of the plant. All banana cultivars and genome groups are susceptible to the disease which has been found to be very destructive with incidence of 70-80% in many plantations and yield losses of 100% have been reported. The development of transgenic banana varieties resistant to banana *Xanthomonas* wilt would be of great value in the fight against the disease and to save livelihoods in the Great Lakes region. Regenerable banana embryogenic cells by which genetic transformation could become a viable solution to the above constraint has been suggested. However, establishing cell suspensions from East African Highland (EAH) bananas particularly is cultivar dependent and not yet achieved. Hence there is urgent need to genetically transform EAH banana using meristematic tissues.

The overall objective of the research is to utilize and optimize an efficient genetic transformation protocol of EAH Banana using meristematic tissues that can be applied in generation of transgenic banana with resistance to *Xanthomonas* wilt. Transgenic EAH Bananas will be generated through meristematic genetic transformation, using the *gusA* gene as a reporter and *nptII* gene as selection marker. GUS histochemical assay for transient expression of *gusA* gene and molecular analysis will be carried out in transformed explants of banana to determine the efficiency of transformation. Results have already been obtained on culturing of buds on media to generate scalps for transformation, testing of scalps and intercalary meristems for regeneration on media before transformation, culturing of agro-bacterium to be utilized in transformation process, transformation of scalps, testing for transient expression of *gusA* gene in the transformed scalps with expression being observed and culturing on the selective media.

Expected outputs include a highly efficient banana meristematic transformation protocol that can result in the establishment of transgenic EAH Banana with potential resistance against banana constraints.

**Partners:** International Institute of Tropical Agriculture (IITA); National Agricultural Research Laboratories (NARL).



## Peter Akoll

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1 Aug – 30 Oct 2012

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### Pathogenic bacterial diversity in catfish hatcheries in Uganda and the development of rapid diagnostics

Disease outbreaks in catfish (*Claris gariepinus*) hatcheries cause an estimated loss of 20-100%. This has significantly constrained development and sustainable production of catfish in Uganda. Despite the outbreaks, information on fish pathogens to guide knowledge-based management of diseases in Uganda is limited. Most fish bacterial pathogens require special growth media for laboratory culture, which takes time, resulting in loss of fish fry before the aetiology can be determined for appropriate treatment. Further, prevailing environmental conditions usually aggravate infections. Therefore, understanding the predisposing factors and causative agents for sudden outbreaks of bacterial infections is paramount in designing of disease management schemes.

The aim of this study is to investigate fish pathogenic bacteria and relate infection dynamics to water quality in catfish hatcheries, to generate knowledge for the development of rapid culture-independent diagnostic tools for selected bacterial infections. The project will also build capacity in molecular genomics and bioinformatics dedicated to aquatic animal health research and development in the region. During the study, molecular techniques will be used to characterize bacteria in catfish and culture water in hatcheries. Specific/target sequences that will contribute to the development of rapid, culture-independent diagnostic tools, e.g. LAMP, will be identified for selected fish pathogenic bacteria. Also, the influence of water quality conditions (e.g. pH, DO, nutrients, conductivity) on occurrence and diversity of pathogenic bacteria will be assessed.

DNA sequences identified for selected bacteria pathogens will contribute to the development of specific and rapid diagnostic tools. Such tools will reduce catfish losses due to delayed and/or misidentification of the aetiology thus reducing the impact of disease resulting in good quality and quantity seed production and subsequently increased fish production. The knowledge generated on the relationship between ecology and bacterial infections in aquaculture systems will enhance the understanding of pathogen dynamics and will guide the designing of management schemes for catfish hatcheries in Uganda. Overall, the study will contribute significantly to the development of aquaculture in Uganda.

**Partners:** Makerere University, Uganda; BecA-ILRI Hub.



## Fredrick Kabi

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4 Aug – 4 Dec 2012

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### Population genetics of *Theileria parva* in the different agro-ecological zones of Uganda

East Coast fever (ECF) is an acute disease of cattle in the eastern, central and southern Africa (ECA). It is caused by a haemoprotazoan parasite, *Theileria parva*, and is transmitted by the tick *Rhipicephalus appendiculatus*. ECF causes a mortality of up to 100% among exotic and crossbred cattle, which are preferred by governments and farmers within the region for increased production and poverty alleviation. The mortality rate among naïve indigenous adult cattle and calves can be as high as 30%, however the morbidity rate can be 100%. ECF is the most important impediment to the development of the dairy industry in Uganda. For many years the control of ECF has relied on the use of acaricides targeting the tick vector, and treatment of clinically sick cattle. However, continuous use of acaricides has resulted into tick resistance, contamination of the environment subsequently the food chain. The costs of acaricides and chemotherapeutic drugs are high which reduces farmers' profits. A live vaccine against ECF is available, and has been tested in small scale trials in Uganda. The Government of Uganda is seeking more research information prior to wide scale deployment.

This study is investigating the genetic diversity of *Theileria parva* from cattle among all the different agro-ecological zones of Uganda. This information is necessary in order to provide a deeper understanding of the epidemiology of ECF within this region to support effective control, and to support future wide-scale deployment of a live ECF vaccine. This data will be useful to assess any breakthrough infections following the deployment of the live vaccine. The research project will also provide training in molecular diagnostics and genotyping techniques which can be transferred to NaLIRRI to support the ECF project, and other animal disease research projects at NaLIRRI.

To date genomic DNA has been extracted from 200 cattle samples. A *T. parva* diagnostic PCR will be carried out on the extracted DNA samples; positive samples will then be genotyped using microsatellites and antigen sequencing.

This project complements other *Theileria* research projects currently being undertaken by ABCF fellows: Anne Nanteza (Uganda), Wani Marcellino (South Sudan), Dia Hassan (Sudan), Awadia Mousa (Sudan) and Elisa Mwegu (Tanzania).

**Partners:** National Livestock Resources Research Institute (NaLIRRI), Tororo, Uganda; Makerere University; BecA-ILRI Hub; ILRI Biotechnology Theme; Veterinary Research Institute, Sudan.





## Cécile Annie Ewané

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3 Aug – 20 Dec 2012

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### Improving knowledge on diversity to enhance the conservation and promotion of bananas and plantains for food security in West and Central Africa

Bananas and plantains are a major staple food in West and Central Africa (WCA) and other parts of Africa. Plantains play a vital role in contributing to food security for more than 250 million people in WCA as indicated by the very high per capita consumption in Gabon and Cameroon (159 and 126 kg/person/year respectively). Plantain production as a staple food or for sale on local markets is done by small holder poor farmers and in home gardens. These are characterised by low productivity resulting in a demand that outweighs supply, provoking very high prices for this commodity. It is therefore critical to develop technologies to contribute to the increase of productivity from 10 tons per hectare per year to 30 tons per hectare per year.

The African Research Centre on Bananas and Plantains (CARBAP) in Cameroon conduct various research programmes focused on improving productivity of banana. To work towards achieving its objective, CARBAP hosts, conserves and relies on the world's second largest collection of *Musa* reference materials. It is a field collection of more than 700 accessions including a unique set of 150 plantain varieties. CARBAP's breeding programmes have led to the development of a number of improved plantain varieties (hybrids) with consistently high yields and resistance to diseases and pests, and have the potential to significantly contribute to increasing global productivity in the region if disseminated. However, it is important to develop a certification process to ensure the safe movement of plant material in the sub region, and accurate information identification of genotypes of materials is an important prerequisite.

The use of molecular tools to genotype the field collection will help rationalise the materials so that only essential accessions are maintained. This project will characterize a total of 148 *Musa* spp. accessions from the collection using 26 microsatellite markers. Samples to be characterized were selected based on the importance of the accession for breeding and also for direct food uses. A total of 3848 data points will be generated and analyzed with various computational packages to determine the diversity and structure of the population.

The data generated from this project will form the basis of a project proposal to obtain funding to continue the molecular characterization of the entire banana collection in CARBAP for comprehensive rationalisation.

**Partners:** CARBAP, Cameroon; University of Yaoundé I, Cameroon; University of Liège, Belgium; BecA-ILRI Hub; French Agricultural Research Centre for International Development (CIRAD).



## Bertin Bisimwa

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6 Aug – 30 Nov 2012

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### Molecular characterization of cavies from South Kivu Province, eastern Democratic Republic of Congo

Recent armed conflict in the Kivu region, Democratic Republic of Congo (DRC), has led to a heavy toll on livestock development. This has particularly affected cattle, small ruminant and swine production. Consequently, the many years of war have led to a rapid decline in the nutritional status of people in the region. To cover their protein requirements, the rural and peri-urban population in the South Kivu region has turned to raising cavies (*Cavia porcellus*; guinea pig). For many families these rodents are now not just a vital element of their food security, but also an essential source of income. The popularity of the cavy in the region can be explained by several factors: cavies grow very fast and multiply rapidly (four to five litters a year); they rarely suffer from diseases even when raised under minimally hygienic conditions; they are a source of low-fat, protein rich meat; they supply good-quality organic fertilizer for crops; minimum start-up capital is required; feeding them is very cheap (kitchen waste etc.) and do not compete for food with humans and other livestock animals; and they can be easily transported in the event of displacement of communities. There is great potential to improve the production of cavies by the small holder farmers through improved management practices and breeding. As a pre-requisite to breeding, information on the current status of the breeds and genetic diversity is required.

The objectives of this study are to determine the genetic diversity of cavies found in the territories of Walungu, Kalehe and Kabare in the South Kivu Province in eastern DRC. Blood samples from 204 cavies were collected from these areas, and associated phenotyping and farm-related data was also collected. This project is currently at an early stage, DNA is now being extracted from the blood samples, and genotyping methods are being optimized. Using 16 molecular markers (microsatellites), this study will provide baseline data regarding the genetic diversity of cavies in these areas, and genetic data will be compared with phenotypic data that was collected at the time of sample collection. The data generated will inform breeding programmes that will allow the selection of desired traits, and thus help the small holder farmers to improve their livelihood and health with more income and more meat in the pot.

This study complements two other ongoing cavy projects by increasing the number of genotyped populations of domestic cavies in East and Central Africa: (1) the project of ABCF Fellow, Parfait Kouakou, who is conducting a similar study on cavies from Cote d'Ivoire, and (2) the BecA-CSIRO project: 'Harnessing Guinea Pig Husbandry for Alternative and Rapid Access to Income and Food in Cameroon and DRC'.

**Partners:** BecA-ILRI Hub; University of Dschang, Cameroon; Catholic University of Bukavu; International Center for Tropical Agriculture (CIAT); University of Abobo Adjame, Cote d'Ivoire.



## Zelalem Fisseha Gebreegziabher

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15 Aug – 15 Dec, 2012

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### **Molecular diversity of haricot bean (*Phaseolus vulgaris* L.) germplasm from Ethiopia**

Haricot bean (*Phaseolus vulgaris* L.) is among the most important legumes cultivated in Ethiopia and export value for the crop has been increasing over the last 4-5 decades. Beans are grown in Ethiopia in diverse environments and socio-economic conditions. They are the primary source of dietary protein for many people. Nonetheless, bean production in Ethiopia is constrained by low yield of existing cultivars, lack of improved varieties, and biotic/abiotic production constraints.

Determining the genetic diversity of landraces is important to help establish a sustainable breeding/improvement strategy. Identifying elite genotypes, which can solve the clear and present constraint of lower national bean yields, is difficult without periodic assessments of the genetic diversity of germplasm resources available. Importantly, there are reported cases of duplications and losses of germplasm collections in Ethiopian legume gene bank stocks, which can be avoided with knowledge of the morphological/molecular diversity of entries.

The objective of this study is to assess the molecular diversity of haricot bean landrace germplasm from Ethiopia using 33 microsatellite markers. This information will be used to document the clustering/classification of germplasm, so that it can be used in further breeding and conservation endeavours. One hundred and twenty one haricot bean samples from 15 population groups were collected. The activities being conducted at the BecA-ILRI Hub include: planting bean samples in the greenhouse and subsequent collection of leaf samples for genotyping, and detailed diversity analysis. Genotyping results will be correlated with that from the morphological characterization, which is currently underway in Ethiopia.

Expected outputs from the research include knowledge on the diversity among Ethiopian common bean germplasm, and clustering genotypes to different centres of domestication/diversity. Combined with the morphological information, this will serve as essential reference information for establishing a sustainable national breeding strategy.

**Partners:** Addis Ababa University; BecA-ILRI Hub.

## Find out more about the ABCF

Each year, the BecA-ILRI Hub seeks applicants who have exceptional ideas for short-term projects (3-6 months) related to food and nutritional security or animal health, to use the advanced biosciences capacity available at the Hub.

### Applicant requirements:

- Working knowledge of English
- PhD or MSc in any agricultural bioscience area
- Currently affiliated with an African national agricultural research programme or university
- From one of the BecA countries: Burundi, Cameroon, Central Africa Republic, Congo Brazzaville, Democratic Republic of the Congo, Equatorial Guinea, Eritrea, Ethiopia, Gabon, Kenya, Madagascar, Rwanda, São Tomé and Príncipe, Somalia, South Sudan, Sudan, Tanzania and Uganda.

## ABCF Investors

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