I. Abstract of Research and Capacity Strengthening Achievements

Common bean (*Phaseolus vulgaris* L.) is the most important grain legume consumed in Uganda and Zambia. The development of improved bean varieties and germplasm with high yield potential, healthy root systems, improved symbiotic nitrogen fixation (SNF) with resistance to multiple diseases, and sustained or improved water use efficiency under limited soil water conditions are needed to increase profit margins, and lower production costs. An improved understanding of plant traits and genotypes with resistance to multiple stresses from abiotic (drought) and biotic (root and foliar pathogens) sources will provide unique genetic materials for enhanced plant breeding methods and sources to study plant tolerance mechanisms in common bean. Improvements in current understanding of the physiology of drought and evapo-transpiration and the genetics of drought tolerance in common bean and the development of effective molecular and quantitative methods for the selection of drought tolerance are needed. The project will use QTL analysis and SNP-based genome-wide association mapping to uncover regions associated with drought tolerance, anthracnose resistance, enhanced SNF and shorter cooking time. Results of this project would contribute to improved yield, farm profitability and human resources in the host countries and indirect benefit to participating U.S. institutions and bean producers.

II. Project Problem Statement and Justification

Beans are the second most important food legume crop after ground nuts in Zambia and are a major source of income and cheap protein for many Zambians. Most of the bean crop (62%) is produced on 60,000 ha in the higher altitudes, cooler and high rainfall zones of the northern part of Zambia. Andean beans are predominant and land races are the most widely grown although a few improved cultivars are also grown as sole crops or in association mainly with maize. Bean production is constrained by several abiotic and biotic stresses that include diseases, pests, low soil fertility and drought. All the popular local landraces in Zambia are highly susceptible to pests and diseases that severely limit their productivity. This is reflected in the very low national yields ranging from 300 to 500 kg/ha that result in annual deficit...
of 5,000MT. To avert future food shortages and feed the growing population of 13M, there is critical need for increasing the productivity of most food crops including beans as Zambia ranks 164 out of 184 countries in the Human Poverty Index. Beans are an important crop in Uganda and are grown on over 660,000 ha of land and consumed throughout the country. Beans are a major source of food and income for the rural smallholder farmers especially the women and children. The majority of bean production in Uganda is dependent mainly on the use of inferior landrace varieties which are generally low yielding due to susceptibility to the major biotic and abiotic (drought, low soil fertility) stresses. These stresses gravely undermine the potential of the bean as a food security crop, a source of income, and as a main source of dietary protein for the majority of Ugandans. Drought affects 60% of global bean production and the severity of yield reduction depends on the timing, extent, and duration of the drought stress.

The development of improved varieties and germplasm with high yield potential, healthy root systems, improved SNF with resistance to multiple diseases, and sustained or improved water use efficiency under limited soil water conditions are needed to increase profit margins, lower production costs. The project will use QTL analysis and SNP-based genome-wide association mapping to uncover regions associated with drought tolerance, disease resistance, enhanced SNF and faster cooking time.

III. Technical Research Progress

Objective 1: Integrate traditional and marker-assisted selection (MAS) approaches to combine resistances to economically important foliar diseases, drought and improved symbiotic nitrogen fixation (SNF) and assess acceptability of fast cooking, high mineral content in a range of large-seeded, high-yielding red mottled, white and yellow Andean bean germplasm for the Eastern Africa highlands (Zambia and Uganda), and the U.S.

1.1. Evaluation of integrated nursery in Uganda

During this year, several nurseries have been evaluated and from these nurseries a number of lines have been selected as sources of resistance or for conducting yield trials and/or genetic studies. Some of the nurseries evaluated and their sources of origin are indicated below. A series of segregating populations and nurseries bred for drought tolerance have been received and together with lines generated within the country have been evaluated in the Ugandan environment. These include;

35 PIC drought tolerant large seeded breeding populations obtained South Africa (ARC-Grain Crops Institute- Potchefstroom) from which 421 individual plants have been selected. Initial selections made from these materials was based on adaptation, plant architecture, number of pods, seed size, seed color and yield, the attributes suitable for the Uganda market and consumers. This particular germplasm was unique in that we initially did not have any large seed drought genotypes in our breeding program. These plants have been planted to establish families which will further be screened for drought and other biotic stresses.

Adaptation and preliminary yield trials were conducted on-station at NaCRRI, Namulonge for a total of 169 newly acquired drought bean lines obtained from CIAT. In the preliminary yield trials conducted. From these 98 lines that showed promise (yielded ≥ 1500 kg/ha) were selected for further evaluation and utilization in Uganda drought breeding program.

Evaluation of ADP lines is being undertaken on two fronts, in the first evaluation, a panel consisted of 250 genotypes, of which 233 were from the global ADP panel and 17 local genotypes were evaluated for drought at end of the first season (May-August 2014) on-station at NaCRRI-Namulonge, targeting off-season planting. The field experiment consisted of two treatments (irrigated and non-irrigated), in two replications planted in the field. Also plans were underway to optimize the use of photosynq for measuring photosynthetic traits in common bean under field conditions. Unfortunately, during the off season planting, the station received unusual high rainfall and no drought stress was observed. These moist
conditions however led to multiple disease condition and data was collected on all agronomic traits and on major common bean disease such as ALS, rust disease, CBB and BMCV were observed that more than 80% of the ADP lines were seriously affected by the above named foliar diseases. Angular leaf spot and rust were noted as the major diseases challenging these lines. There were also some technical challenges and lack of trained field staff that did not allow for the use of the photosynq technology.

In the other experiment 23 ADP lines with faster cooking time with one local check, are being evaluated on-farm in participatory variety section using the mother-baby trial evaluation method. In total 9 mother gardens have established in four Ugandan districts (Hoima, Kamuli, Masaka and Rakai) with a total of close to 90 baby trials where farmers have been availed with 3 genotypes for own testing. The trials are directly engaging 326 farmers (women=230) within nine famer groups. This experiment is currently at R7 and we hope by at the end we will be able to identify at least one fast cooking line that has characteristic that are preferred by Ugandan famers.

Anthracnose and Pythium root rot: Use is being made of 43 breeding populations from earlier work where a series of crosses and selection assisted with the use of molecular markers were made and the result was lines that had been pyramided with 3 anthracnose and one root rot (Pythium) genes and are being advanced and evaluated in field condition on-station. A total of 144 individual plants were selected from the 43 population as indicated in Table 1 below. The selected lines have been established in yield trials to evaluation performance and advance promising families

1.2. Evaluation of Integrated Nursery in Zambia
A regional nursery Southern Africa Bean Evaluation Nursery (SARBEN) consisting of 100 genotypes of various market class and traits was evaluated at Misamfu Research Station in Zambia. The nursery was evaluated in single row plots of 4m long spaced at 60 cm between rows and 10cm intra row spacing. The lines were evaluated for major diseases and yield. The trial mean yield of 1602 kg/ha was obtained with the highest yielding genotype giving 3046 kg/ha and poorest yielded 633 kg/ha. The major diseases were ALS, CBB, ANT, Rust and BCMV. Among these diseases, CBB had high incidence compared to the other diseases. Most lines were found to be resistant/tolerant to ANT, BCMV and Rust. Genotype SDDT SS-C2 and CIM-SUG07-ALS-S1-3 had scores of 5 and 4 respectively for ALS. Genotype HIGH MIN P No 105 GR was found to be more susceptible to Common Bacterial blight with a score of 7 (on the CIAT scale 1-9). A number of lines were found to have combined resistance to most diseases that prevailed in the season.

1.3. Identification of resistance sources in Uganda
A series of efforts have been initiated in trying to identify resistance source for the different foliar diseases. Most notable is the screening of the acquired germplasm both in the field and some cases in the screenhouse to establish the reaction of the reaction of the different germplasm to the different pathogens. Below are some of the activities that have been undertaken in trying to find resistant sources for the respective diseases.

Rust resistance: To identify rust resistant lines, a germplasm collection of 143 lines made up of 30 landraces, 20 released and 93 introduced lines including the 12 rust differentials was screened using field. The reason for the selection of the introduced lines for inclusion in this germplasm set, was their linkage to having drought tolerance, anthracnose and possible rust resistance genes. Identification of rust resistant sources was done using both phenotypic and genetic characterization (conducted at MSU by PhD student) (Appendix 1) and identified lines including Mexico 309, CNC, P1181996, Mexico 235, Redland Pioneer, Oura Negro and Aurora. These have been utilized to introgress rust resistance into Ugandan germplasm.

Anthracnose and Angular leaf spot: For resistance to these two pathogens we will still rely on the differential genotypes like AB 136, G2333 for anthracnose and Mexico 54, for ALS, until we are able to obtain clean isolates and screen the available germplasm for new resistant sources.
**Common Bacterial blight (CBB) resistance:** To identify resistance sources of resistance to CBB, 132 genotypes comprising of 80 local collection and 32 imported germplasm (from Nebraska) have been screened using the most prevalent *Xanthomonas campestris pv. phaseoli (Xcp)* isolate “Kawempe 1” in Uganda. We have so far identified six genotypes CBB22, CBB34, CBB37, CBB2, CBB24 and CBB45 as the most resistant, all from the Nebraska CBB nursery. Resistance sources were identified in a number of nurseries that were under evaluation. These lines were identified on the basis of good seed size and color, upright architecture. These lines will be included on the list of lines that have been identified and used in the region for control of a number of diseases.

**Nebraska:** The ADP was also screened to common bacterial blight at the West Central Research and Extension Center, North Platte, NE, in an augmented replicated trial. The plot size will consisted of 1 row 3 m long spaced 0.56 m. The resistant XAN 159 line; the moderately resistant Neb 1 Sel. #27, and ABC-Weihing; and the susceptible Orion lines were used as a reference checks. At flowering, plants were sprayed with a bacterial solution of $3 \times 10^7$ cfu ml$^{-1}$ using a backpack sprayer with the CBB Nebraskan strains SC-4A and LB-2. The lines were evaluated at the pod filling stage using a 1-9 scale, where 1= immune and 9= very susceptible. H9659-21-1 had the lowest CBB score of 2.9 followed by Badillo with a score of 3.4. Njano-Dulea and OPS-R54 had a score of 3.6. Masusu, Uyole 96, OPS-R51, Red Rider, and AC Elk had a score of 4.1. Micran, H9659-27-10, Montcalm, and USCR-CBB-20 had a score of 4.4. Kijivu, Mrondo, Mkokola, and Kablanketi had a score of 4.6. The checks XAN 159, Neb 1 Sel. #27, ABC-Weihing, and Orion had a score of 2.4, 3.3, 3.9, and 8.3, respectively. On July 2015, a CBB Nursery was assembled and dispatched to Uganda and Zambia for being tested at both locations. Fifty entries were selected based on CBB screening at North Platte, NE during 2013 and 2014.

**Bean common Mosaic Virus resistance:** In Uganda a germplasm collection of 84 lines has been assembled and we are yet to screen for resistance and as such we have not yet identified resistance sources for BCMV.

**Drought tolerance:** We obtained over 150 lines tolerant drought, these have been screened for adaptation in Uganda and some have been even utilized in crosses. We intend to screen these in the field although so far we have failed due to the unpredictable weather conditions (rains off season). No new drought tolerant lines have been identified as yet but a five old CIAT lines (SEN 98, SEN 99, SCR 48, SCN 6 and SCN 9) have been utilized in making crosses and the progenies are already being evaluated for consumer preferred agronomic traits and yield.

A drought evaluation nursery was planted during the off-season (July-November) under irrigation for evaluation for drought in Zambia. The nursery consisting of 60 lines was planted in single rows in two blocks. One block was stressed (irrigation discontinued just before flowering -about 30 DAP) while the other block was not stressed. A number of lines were observed to show some tolerance while some were not tolerant to drought. The nursery is expected to be harvested within November 2015.

The ADP lines were also evaluated to terminal drought (irrigation was stopped at flowering stage) in western Nebraska. Yield and 100-seed weight was reduced by 41.2 and 9.6% when beans were stressed, respectively. VA-19 had the highest geometric mean, followed by Bilfa 4, Bukoba, A 800, and Krimson with values of 3035, 2418, 2349, 2100, and 1973 kg ha$^{-1}$, respectively. Bilfa 4 had the lowest yield reduction comparing normal and drought stress with a value of 6.5%. On July 2015, a drought trial was assembled and dispatched to Uganda and Zambia for being tested at both locations under normal and drought conditions. Sixty entries were selected based on experiments conducted at Scottsbluff, and Mitchell, NE 2013 and 2014.

A 25-entry uniform drought nursery was grown in Michigan in 2015. Weather conditions did not favor the development of drought and the local check varieties outyielded the drought tolerant lines. The nursery was coordinated by Drs. Urrea and Porch.
Field Evaluation of the Nutritionally Superior Common Bean Genotypes with Farmers in Three Agro-ecological Zones in Uganda

Iron and zinc deficiency are the most prevalent micronutrient deficiencies in the world. Biofortification has a potential to address micronutrient malnutrition especially in developing countries where plant based staples are widely grown and consumed. Common bean is an important source of micronutrients. The efficacy of biofortified crops to address human malnutrition can further be improved if genotypes with highly bioavailable minerals are developed. Andean bean genotypes with high iron and zinc concentration, and high iron bioavailability were identified from a set of diverse Andean bean germplasm using an *in vitro* caco-2 cell line assay. A subset of 23 of the best lines for mineral nutrition and also fast cooking are being evaluated in farmers’ fields along with local check genotypes in a participatory variety selection in Hoima, Kamuli, and Masaka. Nine farmer groups each comprised of about 40 farmers are participating in the research. There are three farmer groups per district and each group has a large garden planted with 23 experimental entries (genotypes) + 1 local check so the mother trial has 24 entries as two field reps under RCBD. Each genotype was planted (in August 2015) as a 5-row plot, the rows are 3.5 meters long, and between row spacing is 50 cm. The entire 5-row plot is planted with about 220 dry bean seeds. For each mother trial there are 8 farmers (or 8 baby trials), therefore there are 8*9 or 72 baby trials. Each baby trial has a subset of 3 genotypes, the farmers have been encouraged to include a local check in their baby trial plots. Baby trial genotypes are planted as single replicates. The farmers are from three districts representing three agro-ecological zones in Uganda that are important in both bean production and consumption. The study will help us identify which of the nutritionally superior genotypes might be well adapted for the Ugandan conditions and are preferable to the Ugandan farmers and consumers and what are the breeding needs for nutritionally superior fast cooking cultivars to be accepted by farmers.

1.4. Crossing and backcrossing resistance sources in Uganda

The project has embarked on the process of introgression the different resistance genes into the backgrounds of some of the most preferred Andean bean varieties in Uganda. Work on the introgression is in most cases at the initial stages. Below are some of the different crosses that have been initiated.

**Rust:** Using the seven identified rust resistant source, (Mexico 309, CNC, P1181996, Mexico 235, Redland pioneer, Ouro Negro and Aurora), crosses have already been initiated with Uganda market class bean lines including NABE 15, NABE 16, NABE 19 and NABE 21 to try and introgress rust resistance into their background.

**Drought:** To introgress drought tolerance into farmer and market bean cultivars, use was made of already known drought tolerant sources. Evaluations and selections are being made from the advancing of the progenies arising from 15 crosses that were have been made between Ugandan market class varieties K132, NABE 4 and NABE 15 with introduced drought tolerant germplasm that included SEN 98, SEN 99, SCR 48, SCN 6 and SCN 9. To date, nine (9) promising lines from nine different crosses have been selected and are currently undergoing preliminary yield trials (PYT) on-station at NaCRRI. Additional crosses are planned using selection that will be made from drought germplasm obtained from Nebraska.

**Common bacterial blight (CBB):** Cross have also been initiated between CBB resistant materials CBB22, CBB34, CBB37, CBB2, CBB24 and CBB45 with four Ugandan Andean susceptible market class varieties, NABE 15, NABE 16, NABE 17 and NABE 19 to try and introgress CBB resistance.

**Crossing and backcrossing resistant sources in Zambia:** Under the crossing program a number of lines an early generation trail of 60 selected lines from the F4s were constituted into a trial and was evaluated at Misamfu – Zambia. The lines performed very well with the highest line, Kalungu x Lyambai/3 yielding highest with 2342 kg/ha followed by CIM-ALS-FeZn08-16-6 (2088kg/ha), Chambeshi x NUA 45/1 (1900kg/ha) Chambaeshi x NUA 59/3 (1771kg/ha) and Chamabeshi x NUA 45/2 (1679 kg/ha). These lines combined high yield with resistance to major diseases (ALS, CBB, ANT, Rust). These lines were also large
seeded meeting the preference of the Zambian small scale farmers and end users. Forty-eight percent (48%) of the lines yielded above 1 t/ha. One line (Lusaka x NUA 45/4) did not germinate so no yield was recorded for this line. The lines are currently being advanced to F5 under irrigation. Part of the seed of the selected lines will be tested for levels of Iron and Zinc.

1.5. Evaluation of lines for SNF
Adaptation and evaluation trials for 68 low soil fertility and drought tolerant genotypes obtained from CIAT-Colombia were conducted on-station at NaCRRI. The low soil fertility and drought tolerant materials were evaluated and multiplied to generate enough seed to test in other sites. The indication is that most of these lines perform well under Ugandan conditions and it envisaged that good genotypes will be obtained from these materials. These genotypes are currently being evaluated again for yield and other stresses.

1.6. Population development for genetic studies
We have started generating population especially for the new crosses these include, populations for rust, CBB resistance, BCMV and stem maggot resistance and hope that will lead into formulate some genetic studies for resistance to the different stresses in relation to the Andean Ugandan market class genotypes. Based on 2013 and 2014 data collected in Nebraska (common bacterial blight, experiments on drought, and cooking time), 14 F1 combinations were initiated in 2015. A new set of hybrids is planned on December 2015.

1.7. Cooking time prediction in intact dry bean seeds using visible/near-infrared spectroscopy
Dry beans require long cooking times to become palatable and there is significant genetic variability for this trait. The objective of this study was to evaluate the utility of visible and near-infrared reflectance spectroscopy (Vis/NIRS, 400-2,498 nm) for predicting cooking time from intact dry seeds. A total of 475 bean samples of the ADP grown at the Montcalm Research Farm, Michigan with wide variations in cooking time (15 to 90 min) were used in this study. Spectral preprocessing methods coupled with a feature selection method were tested for improving the prediction of cooking time using partial least squares regression (PLSR) models. Overall, the two-wavelength ratio preprocessing approach produced more precise results (R_pred = 83.2%) than smooth (R_pred = 68.1%), 1st (R_pred = 73.0%) and 2nd (R_pred = 69.3%) derivatives, or continuous wavelength transform (R_pred = 74.8%). The Vis/NIRS technique appeared promising for predicting cooking time from intact dry seeds. Nonetheless, further tests using samples from different years and locations along with the application of additional spectral imaging techniques (e.g., multi- and hyper-spectral) and image processing methods should be considered for improving the prediction of cooking time. The ADP panel was also grown under normal and drought stress in western Nebraska and the panel were cooked in a Mattson cooker. Seeds were soaked overnight (16 hours) then, placed in the Mattson Bean Cooker, and beans were cooked when 80% of the weighted plungers dropped. On average, the beans under normal conditions cooked at 59 minutes and bean under drought stress at 68 minutes. Under normal conditions, Montcalm, Soya, Kablanketi, Soya, and USCR-CBB-20 had the lowest cooking time of 29, 37, 38, 39, and 40 minutes, respectively. Under drought stress, Moro, Njano Dolea, Kablanketi, Bukoba, Wallace 773-V98, RH No.12, and Soya had the fastest cooking time of 39, 46, 47, 47, 48, 49, 51, 52, and 52 minutes, respectively.

1.8. Seed multiplication
A number of promising lines including those from MSU were increased so as to prepare for inclusion of promising one into trails for the 2015/16 crop season in Zambia.
Objective 2: Characterize pathogenic and genetic variability of isolates of foliar pathogens collected in Uganda, and Zambia and identify sources of resistance to angular leaf spot (ALS), anthracnose (ANT), common bacterial blight (CBB), bean common mosaic virus (BCMV) and bean rust present in Andean germplasm.

2.1 Anthracnose and Angular Leaf Spot (ALS) characterization and screening in Uganda: Although plant diseases samples were collected for ANT and ALS, race characterization has not been completed due challenges of contaminations encountered during isolation. In the due course, samples were used up and as such we have embarked on further collections of the same within this season but we have continued to screen materials especially those that had been initially pyramided with anthracnose and root rot resistance genes in the field. Pathotype characterization for these two diseases will commence as soon as we are able to get our isolation protocol right. Considering that most of the disease samples have degenerated, we have embarked on collecting new diseased samples for these two pathogens.

2.2. Rust characterization and screening: Having failed to utilize the mobile nursery technique for the identification of rust pathotypes in the field, we collected and sent 136 rust diseased samples to Nebraska for isolation and identification. Unfortunately these samples were lost due to fluctuation in the moisture levels, only one spore was recoverable. We are currently collecting new samples which we hope to characterize genotypically. Samples received in UNL from Zambia were leaves that were collected from the rust differential lines (mobile nursery) set out to monitor for rust near Kasama. No rust was collected from differential lines Mexico 309 (Ur-5) or PI 181996 (Ur-11). Based on the reactions of the differential cultivars in UNL greenhouses to the Zambian rust, the Ur genes -4, -6, -7, -12, -13, and -9 do not provide protection. However, cultivars with Ur-3, -3+, -5, and -11 would provide resistance. The set of leaf samples from Uganda and collected on an air strip road near Kireka north of Kampala were not allowed to air dry before shipping. They also were packed very tightly into a plastic container and had a white mold growth on the leaves. From one sample Ur-3, -4, -6, -7, -9, -12, and -13 were susceptible and Ur-3+, -5, and -11 were resistant. We plan to determine race structure of new leaf samples. Having received two sets of leaves in conditions less than favorable for rust pathogen viability, we developed two protocols: one, to help with rust infected bean leaf collections and another for expedited shipment to UNL. We have shared them with our collaborators in Uganda and Zambia.

2.3. Common bacterial Blight (CBB) screening: In Uganda they obtained and utilized already available pathotypes “Kawempe 1” to screen the available CBB nursery for resistant sources and will use it to screen for resistance in the resulting progenies. During screening, they inoculated the second trifoliate leaf of an 18 day old plant using the razor blade method and two pods are inoculated per plant during pod filling stage using the multiple needle scratch method. Both leaf and pod inoculated should be on the same plant as recommended by Singh and Miklas, (2015). Disease severity was measured at 14, 21 and 35 days after inoculation, using the CIAT 1-9 rating scale. Data so far collected for the available germplasm is indicated in Table 2.

2.4. Compile data base of past pathogen collections in Uganda: Although still raw and rough, we have initiated a computer based data base for all collection so far made indicate areas of collection and numbers collected. We also intend to show the pathotypes found in the respective area as soon as the characterizations are completed. Long term storage of pathotypes is still a challenge though but we are leasing with CIAT-Uganda to find a solution.

2.5. Anthracnose race characterization, screening in Zambia
Low pressure of ANT was observed during the season and from the nurseries that were evaluated at Misamfu, screening for ANT was done using field infection. Due to dry spells that occurred during the season the screening for ANT was not up to date, though a number of lines were found to be resistant.

2.6. Angular Leaf Spot characterization, screening in Zambia
An ALS nursery consisting of 46 lines was evaluated at Misamfu. The lines were planted in an ALS hot spot so as to get a natural field infestation. The lines were planted in single row plots 4m long spaced at 60cm between rows and 10cm intra row. Data collected included Days to flowering, Stand at harvest, ALS, BCMV, ANT, CBB, Rust, 100 seed weight and Yield. The yield range was from 346 – 2342 kg/ha. The line G9282 was the highest yielder and was found to be resistant to ALS, BCMV and Rust but was intermediate or tolerant to CBB (Score 4). Line BM 12732-57 VEF 2000 121 was resistant to all the diseases that were prevalent during the season, but was small seeded, which might not be preferred by the Zambian small scale famers. This line could be used as source for resistance in the breeding program. High incidences of CBB were noticed during the season. ALS sources used in the region CAL 143 and AND 277 were found still resistant and were among the top yielders in this nursery with yield of 1792 and 1717 kg/ha respectively.

2.7. Rust characterization, screening
Rust characterization using the Rust differential (received from UNL) was done in collaboration with Dr. Jim Steadman of UNL. The results have since been shared with UNL for characterization.

2.8. Common Bacterial Blight Screening in Zambia
A regional Common bacterial blight nursery consisting of 49 lines were plated at Misamfu in single row plots. Most of the lines in the nursery had traceable parentage of VAX lines which are used in the region as CBB resistance sources. Data collected included Days to flowering, Stand at harvest, ALS, BCMV, Anthracnose, CBB, Rust, 100 seed weight and Yield. The mean yield for the nursery was 1519 kg/ha from a range of 683 – 2288 kg/ha. The top yielders included BRB 267/VAX3-14 (2288kg/ha), SEQ11/ RMX-19-4 (2162 kg/ha), BRB 267/VAX3-3, SAB 575/VAX 6-3 (both 2150kg/ha) BRB 267/VAX3-5 (2083 kg/ha). Line BRB 265/VAX3-1 though was found to be resistant to most diseases in the field it was susceptible to rust with a score of 7. Line BRB 265/VAX3-8 was the only line with highest CBB score of 5. The mean seed size (100 seed weight) for the lines was 29.0g.

2.9. Field ratings of rust and common blight in a root rot trial in Zambia
Data was taken on rust and common bacterial blight severity on the foliage of 12 bean lines selected from earlier trials of 362 (2013) and 60 (2014) bean lines including the Andean Diversity Panel and Nebraska select lines. The trial also had four local landraces and was a split/split replicated experiment with three reps of granular insecticide preplant treatment, fertilizer applied and no treatment or fertilizer. The stem maggot insecticide treated bean lines in some cases were less rust resistant than the untreated. Eight lines including three local landraces were resistant; four lines were moderately resistant, two lines were moderately susceptible and only one line was considered susceptible. The common bacterial blight reactions were very low intensity with only two lines moderately resistant (4, 3 rating) and the rest were resistant (2, 3 rating) with only an occasional 4 using the 1 - 9 CIAT scale. The best lines for resistance to both pathogens were Larga Commercial, PI 321094-D, NE 34-12-47, Local Mbbebereshi and Local Cim-Climb 03-48.

2.10. Mapping resistance genes for anthracnose
New sources of anthracnose resistance in a highly diverse panel of 226 Andean beans was screened with eight races of anthracnose to identify and map new sources of resistance using a genome-wide association study (GWAS) at MSU. Only one line Uyole 98 was resistant to all 8 races. Outputs from the GWAS indicated major QTL for resistance on three linkage groups: Pv01, Pv02, and Pv04 and minor QTL on Pv10 and Pv11. Candidate genes associated with the significant SNPs were detected on all five chromosomes. A QTL study with the black bean cultivar Jaguar, known to possess resistance to anthracnose race 73, was conducted to determine the basis of the anthracnose resistance commonly used in the MSU breeding program. Resistance to anthracnose was investigated in an F4:6 recombinant inbred line (RIL) population developed from a cross between Jaguar and Puebla 152 (landrace cultivar known to be susceptible to race 73). Resistance in Jaguar was determined to be conditioned by the single dominant gene Co-I. Using the Illumina BARCBean6K_3 BeadChip, the physical location of the Co-
A breeder friendly InDel marker was developed (50.2Mb) that was linked to the Co-1 locus as 3.1cM and could be used in selecting 4 of the 5 resistance alleles at the Co-1 locus. The genomic positions of the numerous resistance loci on Pv01, Pv02, Pv04 and Pv11 identified in Jaguar and in the Andean panel should prove useful for breeding programs interested in improving anthracnose resistance in cultivars using marker assisted selection.

Objective 3: Use single nucleotide polymorphism (SNP)-based genome-wide association mapping to uncover regions associated with drought tolerance, disease resistance, cooking time and BNF to identify QTLs for use in MAS to improve Andean germplasm.

To better understand the genetic architecture of SNF at the molecular level variability three studies were conducted at MSU: (i) genome-wide association study (GWAS), (ii) Quantitative Trait Loci (QTL) mapping study, and (iii) transcriptome profiling study. GWAS was conducted using an Andean Diversity Panel (ADP) comprised of 259 genotypes. The ADP was evaluated for SNF in both greenhouse and field experiments, and genotyped using an Illumina BARCBean6K_3 BeadChip with 5398 single nucleotide polymorphism (SNP) markers. A mixed linear model was used to identify marker-trait associations. The QTL mapping study was conducted using 188 F4:5 recombinant inbred lines (RILs) derived from cross of Solwezi and AO-1012-29-3-3A. These 188 F4:5 RILs were evaluated for SNF in greenhouse experiments, and genotyped using the same BeadChip. Transcriptome profiling was conducted on RILs SA36 and SA118 contrasting for SNF that were selected from the Solwezi x AO-1012-29-3-3A population used in the QTL mapping study. RNA samples were collected from leaves, nodules and roots of SA36 and SA118 grown under N fixing and non-fixing conditions, and sequenced using Illumina technology. Using GWAS, significant associations for nitrogen derived from atmosphere (Ndfa) were identified on chromosomes Pv03, Pv07 and Pv09. QTL mapping identified QTL for Ndfa on Pv02, Pv04, Pv06, Pv07, Pv09, Pv10, and Pv11. The GWAS peak identified on Pv09 for Ndfa overlapped with the QTL on Pv09 for Ndfa identified in QTL mapping study. Previous studies have reported QTL for Ndfa on Pv04 and Pv10. Genes encoding receptor kinases, transmembrane transporters, and transcription factors (TFs) were among differentially expressed genes (DEGs) between SA36 and SA118 under N-fixing condition, but not under non-fixing condition. Out of the 51 genes that were in 400 kb region surrounding the GWAS peak on Pv07, only four including Phvul.007G048000 encoding a MADS BOX TF were identified as expression candidates for SNF in the transcriptome profiling study. In the 400 kb region surrounding the GWAS peak on Pv09 there were 44 genes, but only Phvul.009G137500 encoding a WRKY TF was identified as an expression candidate gene in the RNA-seq study. Using GWAS, QTL mapping and transcriptome profiling, genomic regions and expression candidate genes for SNF have been identified. Once validated, these QTL and genes have potential to be used in marker-assisted breeding to circumvent challenges of phenotypic selection for SNF, and accelerate genetic improvement of common bean for symbiotic nitrogen fixation.

Objective 4: Develop phenometric approaches to improving the efficiencies of breeding for abiotic stress tolerance, especially drought

Much of the research focused on examining constitutive differences between drought tolerant and drought susceptible genotypes so that mechanisms contributing to drought tolerance might be discovered and further investigated. To that end, the morphology of a drought tolerant genotype, tepary bean (P. acutifolius), and a drought susceptible genotype, common bean (P. vulgaris) cultivar Jaguar, were examined. First, the leaf density of both genotypes was measured. Ecologists have observed that species growing in low precipitation areas have sclerophyllous leaves - leaves that are small, thick, and leathery, and because drought tolerant tepary bean exhibited smaller leaf sizes that susceptible Jaguar or common beans in general, it was hypothesized that tepary bean might also have
denser leaves. Multiple detached leaves of Jaguar and tepary were photographed, and their leaf areas determined with image analysis software. The detached leaves were also weighed, and from these two parameters, leaf density for both genotypes was determined. Jaguar and tepary were not significantly different from each other in leaf density; both had a leaf density of approximately 0.018 g/cm². Four bean genotypes contrasting in drought tolerance were also tested with assimilation vs. intercellular CO₂ (A-C) curves, which can give insight into various parameters that limit photosynthesis. Essentially, the photosynthetic rates of a plant are measured at different CO₂ concentrations, and the resulting data points can be measured with a simple tool (Sharkey et al., 2007). From the analysis, differences in maximum carboxylation rate of rubisco (V_{cmax}) and the electron transport rate (J) were found among the varieties (Table X1). V_{cmax} limits photosynthesis under low CO₂ conditions such as drought while J limits photosynthesis under high light conditions. Zorro is only moderately drought tolerant, but it is a highly productive cultivar, so its higher V_{cmax} and J could contribute to its higher productivity and ability to withstand intermittent drought. Tepary is extremely drought tolerant, and its higher J could allow it to withstand the higher light intensities coincident with drought stress without being damaged.

Also integral to this objective of the project is developing new methods of evaluating the stress resistance of different genotypes. In pursuance of this aim, a screening method utilizing heat stress was developed. Drought and heat tolerance share many physiological and genetic pathways, so it was hypothesized that heat could be used to indirectly screen for drought tolerance. Heat has the advantage of being easier and quicker to uniformly and reproducibly apply when compared with drought stress. Fifteen bean genotypes contrasting for drought tolerance were gathered: five used in previous research, and ten from a bean drought panel recommended by Carlos Urrea of University of Lincoln - Nebraska and Timothy Porch of the USDA Tropical Agriculture Research Station in Mayaguez, Puerto Rico. Successive replications of the 15 genotypes were grown in a growth chamber at 35°C, and then the temperature was raised to 40°C, and finally to 45°C. After a few days acclimation to the new temperatures, the plants were measured for parameters that respond to stress with a variety of methods: gas exchange, chlorophyll fluorescence, leaf temperature, and qualitative visual assessment. The results show that no significant differences were seen among these genotypes until the most severe stress of 45°C (Figure X1A-F). At that point, drought tolerant genotypes like tepary clearly separated out from susceptible phenotypes like Jaguar and 553. This heat screening method is a fast, effective, and reproducible way of testing many genotypes for drought tolerance specifically and abiotic stress tolerance generally.

Objective 5: Institutional Capacity Building and Training for doctoral student from Zambia (Kelvin Kamfwa) and Uganda (Isaac Dramadri), and the US (Jesse Traub), and one MS (Grady Zuiderveen) student from the US all in Plant Breeding, Genetics and Biotechnology.

1. Activities conducted by Kelvin Kamfwa in Michigan – listed under objective 3

Activities conducted by Isaac Dramadri. Mr. Dramadi planted the ADP panel consisted of 250 genotypes, 233 from the global ADP panel and 17 local genotypes and field evaluated during the months May-August, targeting the end of first rainy season at NaCRRI-Namulonge. The aim of the research was to use genomic and phenometric tools for improving selection and breeding for drought tolerance in the large seed bean. The field experiment consisted of two treatments (irrigated and non-irrigated), in two replications planted in the field at NaCCRI. In order to prepare and plan for future field experiments, field evaluations were necessary to assess the performance of the ADP under Ugandan conditions specifically targeting terminal drought stress, optimize the use of photosynq for measuring photosynthetic traits in common bean under field conditions, and multiply seed for the subsequent experiments. During the months of May-August, NaCRRI received unusual high rainfall and no drought stress was observed. These moist conditions however led to multiple disease condition and data was collected on all agronomic traits and on major common bean
disease such as ALS, Rust, CBB, and BMCV. In addition, the initial plans of setting field irrigation facilities to provide supplementary irrigation were not implemented or needed. The major finding from this initial trial is that another location in Uganda is needed for drought screening due to the unpredictability of rainfall even in the dry season at Namulonge and the lack of trained field staff prevented the use of the photosyn as most field workers are actual laborers and could not be trained to assist in using the devise.

3. Activities conducted by Grady Zuiderveen in Michigan – listed under objective 2.10

4. Activities conducted by Jesse Traub in Michigan are listed under objective 4.

IV. Major Achievements
The project in Uganda has made some significant achievement towards achieving the breeding objectives especially in the area of germplasm acquisition and utilization. We have also able to forge working relationships between NaCRRI and other institutes like Makerere University, Michigan State University, University of Nebraska, ARC-Grain Crops Institute- Potchefstroom- South Africa where we are able to have exchange visits for both students and researchers and also in the exchange of germplasm See Annex 3 for details.

The Bean Program conducted a Bean Research Methodology training Course at Misamfu research station from 29th March to 3rd April, 2015. The objectives of the training were;

- To share the standard evaluation of bean germplasm
- To identify and score bean diseases and pests in the field
- To learn how to set out experimental plots
- To learn how to manage Bean Trials
- To learn issues related to data collection and related information

Participants were drawn from ZARI stations of; Msekera, Mt Makulu, Kabwe, Mutanda, Mufulira, Mansa and Misamfu. The total number of 19 (15 male and 4 females) participated in the training

V. Research Capacity Strengthening
The collaborative research has enabled us to build research capacity at NaCRRI not only in terms of breeding activities but also in developing human resource capacity. In this year we were able to continue training and mentoring one PhD and two MSc students. We are also able to train a three research assistant and 5 technicians in Uganda on the use of modern technologies to capture field data and reduce on errors. Also the host country PI-Uganda, was facilitated to attend and participate in a common bean disease workshop on angular leaf spot and root rot where new insights and methods were shared on how to combat these two diseases. We also able to network with other renowned scientists and sharing research information and knowledge. For human capacity building, two short term trainings were organized for Research assistants and technicians in host country-Uganda. This was to strengthen their research capability in as far as data collection is concerned. There was training on the use of new data collection tools as part of breeding management system which tools are being utilized by the project.

VI. Human Resource and Institution Capacity Development

1. Short-Term Training
Two short training were conducted during this year. The first was on the use of Geographic information systems (GIS) with a special attention to the use of a GPS to capture and map location where data is collected. This was to help in the collection of diseases samples from within the country. This training was conducted by Dr. Michael Otim.
i. **Purpose of Training**: To introduce the Research Assistant and Technicians with the country’s legume program to GIS and its importance in data collection and mapping data point.

ii. **Type of Training**: Illustration using GPS and computer- and hands-on practicals

iii. **Country Benefiting**: Uganda

iv. **Location and dates of training**: NaCRRI – Namulonge, Uganda, 25-28th May 2015

v. **Number receiving training (by gender)**: Male-04 and Female-05 (Table 3)

vi. **Home institution(s) (if applicable)**: NaCRI

vii. **Institution providing training or mechanism**: NaCRI

The second training was on the use of the breeding management system for the integrated breeding platform also for Research Assistant and technician. This is a computer based tool that is used manage breeding data and also use of tablets for field data capture and reduce on errors. The training was conducted by Dr. Magni Bjarnason a consultant with the Integrated Breeding Platform who was assisted by Dr. Stanley Nkalubo, the Host country PI.

i. **Purpose of Training**: To introduce Research Assistant and Technician to the use of the Breeding management system (BMS) and use of computer based tools for field data capture to reduce on errors.

ii. **Type of Training**: Power point presentation and hands- on computer and field practicals

iii. **Country Benefiting**: Uganda

iv. **Location and dates of training**: Uganda; 06-09th July 2015.

v. **Number receiving training (by gender)**: Male-04 and Female-05

vi. **Home institution(s) (if applicable)**: NaCRI

vii. **Institution providing training or mechanism**: Integrated Breeding Platform

2. **Degree Training**

The PhD student (Ms. Blessing Adogwu), continued to undertake her research work on rust with the project. Through the Norman E. Borlaug Leadership Enhancement in Agriculture Program fellowship, she was also able to travel to MSU and University of Nebraska to undertake hands-on training of the use of molecular markers for screening purposes. In addition two other MSc. students have been taken on by the project to undertake their researchers on under some of our project objectives. The first MSc. students is looking at breeding for resistance to common bacterial blight disease (CBB) while the second student is conducting research on the bean common mosaic virus (BCMV) disease. It hoped that the three students will make positive contribution towards new discoveries and also gain experience in research implementation. Details for the students are given below;

i. **Name of trainee**: Blessing Odogwu

ii. **Country of Citizenship**: Nigeria

iii. **Gender**: Female

iv. **Host Country Institution Benefitting from Training**: University of Port Harcourt, Nigeria

v. **Institution providing training**: Makerere University/NaCRI

vi. **Supervising CRSP PI**: Prof. Jimmy Kelly

vii. **Degree Program**: PhD

viii. **Field or Discipline**: Plant Breeding and Biotechnology

ix. **Research Project Title**: Breeding for rust resistance in common beans in Uganda

x. **Start Date**: January 2014

xi. **Projected Completion Date**: December 2017

xii. Is trainee a USAID Participant Trainee and registered on TraiNet?: No
Training status: Active

Student 2

i. Name of trainee: Boris Mahulé Elysé Alladassi
ii. Country of Citizenship: Benin
iii. Gender: Male
iv. Host Country Institution Benefitting from Training: University of Abomey-Calavi, Benin
v. Institution providing training: Makerere University/NaCRRI
vi. Supervising CRSP PI: None
vii. Degree Program: Masters Degree
viii. Field or Discipline: Plant breeding and seed systems
ix. Research Project Title: Genetic Analysis of Resistance to Common bacterial blight and association of candidate SNP markers of common bean (Phaseolus vulgaris L.) in Uganda
x. Start Date: December 2014
xi. Projected Completion Date: September 2016
xii. Is trainee a USAID Participant Trainee and registered on TraiNet?: No
xiii. Training status: Active

Student 3

i. Name of trainee: Basil Evarist Kavishe
ii. Country of Citizenship: Tanzania
iii. Gender: Male
iv. Host Country Institution Benefitting from Training: Sokoine University of Agriculture, Tanzania
v. Institution providing training: Makerere University/NaCRRI
vi. Supervising CRSP PI: None
vii. Degree Program: Masters Degree
viii. Field or Discipline: Plant breeding and seed systems
ix. Research Project Title: Resistance to bean common mosaic virus and its inheritance in selected Ugandan bean genotypes
x. Start Date: December 2014
xi. Projected Completion Date: September 2016
xii. Is trainee a USAID Participant Trainee and registered on TraiNet?: No
xiii. Training status: Active

Student 4

i. Name of trainee (First and Last Name): Kelvin Kamfwa
ii. Citizenship: Zambian
iii. Gender: M
iv. Training Institution: MSU
v. Host Country Institution Benefitting from Training: University of Zambia
vi. Supervising Legume Innovation Lab PI: James D. Kelly and Karen A. Cichy
vii. Degree Program for training: Doctorate
viii. Program Areas or Discipline: Plant Breeding, Genetics and Biotechnology
ix. Thesis Title/ Research Area: Genetic dissection of biological nitrogen fixation in common bean using genome-wide association analysis and linkage mapping.
x. Start Date: August 2008
xi. Projected Completion Date: November 2015
xii. Is trainee a USAID Participant Trainee and registered on TraiNet? Yes
xiii. Training Status: Active

Student 5:
 i. Name of trainee (First and Last Name): Grady Zuiderveen
 ii. Citizenship: US
 iii. Gender: M
 iv. Training Institution: MSU
 v. Supervising Legume Innovation Lab PI: James D. Kelly
 vi. Degree Program for training: Masters
 vii. Program Areas or Discipline: Plant Breeding, Genetics and Biotechnology
 viii. Host Country Institution to Benefit from Training: US
 ix. Thesis Title/ Research Area: SNP marker development for major resistance genes
 x. Start Date: August 2013
 xi. Projected Completion Date: September 2015
 xii. Is trainee a USAID Participant Trainee and registered on TraiNet? No
 xiii. Training Status: Active

Student 6:
 i. Name of trainee (First and Last Name): Jesse Traub
 ii. Citizenship: US
 iii. Gender: M
 iv. Host Country Institution to Benefit from Training: US
 v. Training Institution: MSU
 vi. Supervising Legume Innovation Lab PI: Wayne Loescher
 vii. Degree Program for training: Doctorate
 viii. Field or Discipline: Plant Breeding, Genetics and Biotechnology
 ix. Thesis Title/ Research Area: Physiological differences among Phaseolus vulgaris cultivars differing in drought tolerance.
 x. Start Date: August 2013 on Legume Innovation Funding
 xi. Projected Completion Date: November 2015
 xii. Is trainee a USAID Participant Trainee and registered on TraiNet? No
 xiii. Training Status: Active

Student 7:
 i. Name of trainee (First and Last Name): Isaac Dramadri
 ii. Citizenship: Uganda
 iii. Gender: M
 iv. Host Country Institution to Benefit from Training: Makerere University
 v. Training Institution: MSU
 vi. Supervising Legume Innovation Lab PI: James D. Kelly and Wayne Loescher
 vii. Degree Program for training: Doctorate
 viii. Field or Discipline: Plant Breeding, Genetics and Biotechnology
 ix. Thesis Title/ Research Area: Physiological studies on drought tolerance in Andean beans.
 x. Start Date: August 2013 on Legume Innovation Funding
 xi. Projected Completion Date: September 2017
 xii. Is trainee a USAID Participant Trainee and registered on TraiNet? Yes
 xiii. Training Status: Active, Partial -BHEARD Fellowship from USAID Mission, Kampala.

VII. Achievement of Gender Equity Goals
For all activities we have undertaken under the project in Uganda we have ensure that women are actively represented at NaCRRRI, this has also been shown in all our activities with the farmers and short term training. We have achieved more than the 30 percent women representation that has been set during project planning.

In Zambia we have identified NGO’s that we can partner with for outreach and technology dissemination for female farmers which are: Kusefya pa Ngw’ena Women’s Farmer Group, Shangila Seed Growers Association (SSGA) in Mpika and the Participatory Village Development in Isolated Areas (PaViDIA) in Mporokoso and Luwingu. PaViDIA is working towards empowering women in communities in Income Generating Activities (IGA) and seed and grain production for market sales to elevate income and reduce poverty. In Uganda the NGOs include: Community Enterprise Development Organization (CEDO), Integrated Seed Sector Development (ISSD)-Uganda, CARE, ADRA, SHUPO, SASAKAWA Global 2000; Nyakatozi Growers Cooperative Union, Appropriate Technology (Uganda); Seed companies such as (Pearl, Victoria, NASECO, East African Seed, FICA seed). Many organizations have as objectives to increase women’s agriculture skills and leadership roles as well as access to credit for sustainable and profitable farming.

VIII. Explanation for Changes
There are no changes to set out activities but a few delays may be due to loss of diseases samples due to contaminations. We are currently relying on field screening but we hope that by mid next years all the new isolations would have been completed and characterization completed.

IX. Self-Evaluation and Lessons-Learned
Apart from the setbacks in the poor isolation and characterization, we believe that that project is fairly on course. Considering the unpredictable rainfall patterns, we have learnt we cannot rely on off-season planting to conduct drought trails anymore. We may have to think of other innovative ways of evaluating drought germplasm in the field. We may need to start thinking of constructing rainout shelters in the field.

X. Scholarly Accomplishments – See Annex 1

XI. Progress in Implementing Impact Pathway Action Plan
The project is on track toward implementing the impact pathway. All activities listed under step 4.1 of the impact pathway have been met with the exception of disease characterization in country and those activities will be conducted during FY16.

XII. Data Management
The project is on track toward compliance with USAID standard provisions to make public all data bases generated as part of the project. All published data is available as supplemental files at the journals listed under item X, scholarly achievements
Annexes: Scholarly Accomplishments


Presentations, Dissertations and Awards:


4. Reports submitted in the NARO annual report in Uganda
Annex 2. Literature Cited


Annex 3. Research achievements in Uganda obtained are inclusive but not limited to the following;

- Through the project we have been able to obtain and are utilizing four (4) nurseries including that of rust, CBB, and 2 sets of Drought nurseries and two (2) sets of differentials (Rust and ALS) from collaborating partners.
- We have identified 6 and 5 tentative resistance bean sources for rust and CBB diseases respectively.
- Have made a germplasm collection of close to 500 accessions which are utilizing to identify resistance sources for the different bean foliar diseases.
- Made over 60 different crosses to try and introgress different diseases resistances into the susceptible Uganda market class bean varieties.
- Made selection for over
- We have determined the incidences and severities of the different bean folia pathogen within major bean growing regions of Uganda.
- We have built capacity for 4 research Assistant and 5 technicians in form of data collection and management of breeding system also training and mentoring 3 students (1 PhD and 2 MScs) in breeding methodologies.
- We have engaged 326 farmers in on-farm trials that are evaluating and making selections for the utilization of fast cooking bean varieties.
- One PhD student (Blessing Adogwu) from NaCRRI -Uganda visiting Michigan State University and University of Nebraska to acquire skills in use of marker assisted selection and two PhD from Michigan State University (Dennis Katuramu and Isaac Dramadri) conducting their researchers with NaCRRI-Uganda.
- One poster paper “Implications of Fungal Pathogens to Bean Production in Uganda” presented at the common bean disease workshop on angular leaf spot and root rot from 20-23 July 2015, Kruger Gate Skukuza, South Africa.
- Several sources of common bacterial blight resistance were identified. A common bacterial blight nursery of 50 entries is being tested in 2015 at Uganda and Zambia.
- Several sources of drought tolerance were identified. A drought trial of 60 entries is being tested in 2015 at Uganda and Zambia under normal and drought stress conditions.