Enhancing Biological Nitrogen Fixation (BNF) of Leguminous Crops Grown on Degraded Soils in Uganda, Rwanda, and Tanzania

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Abstract of Research Achievements and Impacts
Loss of soil fertility is recognized as the most important constraint to food security in sub-Saharan Africa. Enhancing the natural capacity of legume crops, such as common beans, for biological nitrogen fixation (BNF) has been shown to help to overcome this constraint, but an optimum combination of variety, inoculant, and crop management has not been established. To this end, this CRSP program will identify production systems that enhance BNF, develop germplasm that benefits most from symbiotic inoculation, and aggressively share this new information with small landholder farmers in sub-Saharan Africa whose health and well being depend heavily on legume production.

This first report encompasses research achievements since the program was formally initiated in August 2010. Although the program was intended to begin January 1 of this year, funding allocation to the lead US institution was delayed and sub-contracts to partner and host country institutions were finalized only in August and September. While US institutions initiated some aspects of the program objectives prior to funding, the host country partners began first field trials with the second planting season in September. As a result, this report describes the preliminary investigations initiated this summer at US institutions, and the design of field trials and initial collection at the host-country research sites.

Although the project activities have been underway for only 2-3 months, all the 6-month benchmarks outlined for Objectives 1 and 2 in the revised FY10 workplan have been accomplished. These include: Identifying the genotypes and research demonstration sites to be examined at HC institutions, Quantifying soil physical and chemical characteristics at all test sites, Obtaining experimental and adapted common bean germplasm for genetic marker analyses, and Increasing seed of existing mapping populations for QTL analysis. A

number of 12-month benchmarks are being addressed and are ahead of schedule for completion in FY11. While no funding was allocated to conduct activities outlined under Objective 3, a number of initial steps were taken to ensure progress on this objective during FY11.

All HC institutions have identified graduate students or undergraduate interns and have initiated their research activities. Students from partner countries have begun their graduate study or are slated to begin study in January 2011 at US institutions.

This project is in its earliest stages with the first field trials just reaching flowering and first major sampling activities. Harvest is anticipated in early December, which will provide initial results to evaluate for the potential impact of advanced inoculant technologies on BNF. Initial field evaluation of bean germplasm for genetic marker analyses also have yet to be analyzed and need to be repeated under controlled conditions. Planning is underway for a workshop to bring together all BNF-CRSP program PIs to develop synergies among these complementary programs.

Project Problem Statement and Justification

Loss of soil fertility is the most important constraint to food security in sub-Saharan Africa (CIAT 2002, Bationo 2004). Low levels of nitrogen and phosphorous are the primary fertility constraints (Ndakidemi et al 2006). Because soils are increasingly becoming degraded, an affordable means of improving soil fertility and productivity of nitrogen-accumulating crops is critical. Numerous studies have shown the potential of improving legume productivity by enhancing nodulation through proper use of biological inoculants (e.g. Ndakidemi et al 2006, Silver and Nkwiine 2007). Yet field trials have provided mixed results (Nkwiine 1999, Musdandu and Joshua 2001). Potential reasons for failure include poor quality of inoculants, failure to compete with local rhizobia, inhibition by indigenous microbial flora, or failure of the inoculants to survive in low pH and droughty soils (Graham, 1981). Modern inoculant formulations designed to deliver a synergistic suite of biological and chemical enhancements for biological nitrogen fixation under stressful soil conditions have been made available to our collaborative research project by Becker Underwood, Inc. (see letter of collaboration). Becker Underwood's **BioStacked®** inoculant technologies for legume crops consist of well stabilized *Rhizobium* bacteria, a biological fungicide, plant growth promoting rhizobacteria, and other biologically derived proprietary biostimulant technologies which promote plant growth and overall plant health. These stacked inoculants decrease chemical fertilizer use in crop rotations, increase legume yields, suppress root diseases, and improve rhizosphere conditions for root growth. And they are suitable for use on a variety of legume crops such as soybean, common bean, cowpea, and pigeon pea. We anticipate they will be particularly effective under degraded soil conditions encountered on small-landholder farms in Uganda, Rwanda, and Tanzania.

To optimize BNF, it is essential to identify germplasm with greatest capacity for this trait (Bliss et al 1989, Diouf et al 2008). Although common bean has the potential for BNF, it is reported to have the lowest percent N_2 derived from N fixation among legumes (Martinez-Romero 2003). Genetic variation for BNF has been reported and lines with superior BNF have been identified (Bliss, 1993; Graham et al., 2003). Superior BNF lines such as Puebla 152 and BAT 477 (Vadez et al., 1999; Miklas et al., 2006) have been used as parents in crosses to generate populations for genetic studies and breeding for improved BNF. Few breeding lines with improved BNF,

however, have been developed. Low heritability estimates for BNF and related traits indicate that BNF traits are quantitatively inherited and influenced by environment. The optimal selection environment for BNF is under low soil N since application of nitrogen fertilizer reduces N fixation capacity (Schulze 2004). Marker-assisted selection under such conditions is highly sought after as a means to facilitate breeding for improved BNF because of its low heritability.

There have been few molecular mapping studies conducted for BNF in legumes. But there are many available recombinant inbred mapping populations within the bean breeding community that are ideal for a BNF-QTL study. Molecular mapping in combination with germplasm screening and marker assisted selection (MAS) would be a powerful way to improve locally adapted germplasm for BNF in a host country. Recombinant inbred populations are ideal for tagging and mapping genes that influence quantitative traits (QTLs). These populations provide segregating inbred lines that can be replicated over space and time and maintained for many years, which is ideal for characterizing traits conditioned by many genes and influenced by environment. Few QTLs associated with BNF have been identified to date, and those identified have not been validated. Therefore, identification and subsequent validation of QTL conditioning enhanced BNF would represent a major contribution to the scientific community, and represent a major step toward generating capacity for marker-assisted selection for BNF.

Our CRSP program objectives address the need to identify production systems that enhance BNF, develop germplasm that benefits most from symbiotic inoculation, and aggressively share this new information with small landholder farmers in sub-Saharan Africa whose health and well being depend heavily on legume production.

Results, Achievements and Outputs of Research

<u>Objective 1:</u> Improve BNF and seed yields of common beans significantly using superior seed inoculants such as Becker Underwood's BioStacked® inoculant through farmer-based experimentation and adoption of innovative production techniques.

1a. Evaluate effectiveness of Biologically Stacked Inoculants on Local And Improved Germplasm

- 6 month benchmark: Identify genotypes and research demonstration sites at HC institutions

Trial sites on research stations have been established in Uganda, Rwanda, and Tanzania. Similar protocols were followed in all there HCs based on collaborative discussions among HC PIs. Varieties and specific field designs vary based on local adaptation and production preferences.

<u>Germplasm</u>

In Uganda, there are three common bean varieties with market preference considered i.e. K 132, Kanyebwa (local cultivar) and K131 (V_1, V_2, V_3, respectively). Kachwekano was selected for climbing bean and three varieties namely NABE10C, NABE12C and local cultivar (V_1, V_2 and V_3 respectively) were selected and planted under the same treatments. Figure 1 shows the general outline of the field study.

In Tanzania at SUA, a total of 20 local and improved germplasm lines for the experiments have been collected from National Agricultural Research (NARs) Institutes and CIAT for evaluating the effectiveness the inoculants (both local and the Becker Underwood's BioStacked® inoculant). Seeds are now being increased at the station (SUA) to get adequate seed for planting.

In Rwanda, two improved climbing bean varieties: ISAR-CB-105 and ISAR-CB-107 (Type IV) and two bush: ISAR-SCB-102 (Type IIA) and RWR 1668 (Type I) were selected among the newly released bean varieties in Rwanda (Table 1). The varieties were chosen for their adaptability in the low altitude zones of eastern Rwanda, and for their culinary and marketable attributes that were appreciated by the farmers during the participatory variety selection trials. The climbing varieties were earlier maturing compared to usual climbers.

Field sites

In all cases, field research and demonstration sites are on national or university research stations. This was done to ensure control of field operations and uniformity of treatment applications.

In Uganda these are located in three agro-ecological zones identified in cooperation with Dr. Tenywa and two masters students (Ms. Martha Abwate and Mr. Peter Ssenyonga at Makerere University) in central Uganda at Namulonge (NaCRRI) and southwestern Uganda at Mbarara ZARDI and Kachwekano ZRDI research stations. Treatments include three rhizobia types sourced from USA (Becker Underwood), and from Universities of Nairobi and Makerere. The latter two were considered as indigenous for comparison purposes.

In Rwanda, two sites were selected at ISAR Nyagatare research station and at the farmers' field in Nyakigando sector of Nyagatare district. Nyagatare lies within 30°20E and 1°20S. The mean altitude is 1450 masl and 700 - 900 mm and 22.4C for the rainfall and temperature respectively. The soils are generally sand (41 - 68%), clay (20-38% and loam 8 - 27%). Silt content is very low while percolation is moderate and evapotranspiration is high. Nyakigando site was selected for research but also for demonstration and training purposes of the members of the farmers' cooperatives and other farmers in the area.

In Tanzania, the field sites are located at the research stations at Morogoro, Mbeya and Arusha. Details of the field location and plot design to be provided in the next semi-annual report.

Additional field management details

At the NaCCRI stations, phosphorus being the most constraining nutrient in the soils of east Africa and yet very crucial to effective BNF, was considered as a treatment (0 and 40 kg P ha^{-1}) to evaluate to what extent the imported rhizobia can withstand the limited P supply in the soil. The factorial combinations culminated into a total of 72 treatment plots. The spacing was 10cm x 50cm for common bean varieties in Namulonge and Mbarara and 20cm x 50cm for climbing bean varieties in Kabale (Kachwekano ZARDI). The project has also planted 130 lines of bush beans at NaCRRI for multiplication to be shared with other countries in the coming season.

At ISAR, four varieties (V1, V2, V3 and V4) x rhizobia (Ru, Rn with or without: Ro) x P fertilizer at 2 levels (with and without) were applied in all combinations to give a total of 24 treatments (Table 2). Rhizobia inoculants were applied at 20 g per kg of seed, while P was

sprinkled in planting rows at 20 kg per ha. Inoculation was done separately in plastic basins using hand grooves to avoid cross contamination. Eight 4m long rows were planted per plot of 4m x 4m with 4 replications at the on-farm site of Nyakigando. The plot size was 6m x 6m at the on station site of Nyagatare. Randomization was of treatments was done for each replication at planting. Planting was done after rains in moist soils.

1b. Quantify Genotype by Environment Interactions and Constraints to Enhancing BNF of Inoculated Plants

- (6 mo) Quantified soils physical and chemical characteristics at all test sites

Soil samples were collected for chemical, physical analysis which analysis has already been carried out for all three sites. Additional soil samples will be collected for DNA extraction and further analysis is scheduled when plants reach flowering and will coincide with biomass sampling and assessments of nodule number and activity. Standard weather data will be collected throughout the growing season.

Data on crop development related to N2 fixation to be collected at flowering: Nodulation potential at 10 – 20% flowering, number of effective nodules (based on legheamoglobin pigment status), leaf area index (LAI) at flowering, visual chlorotic symptoms (green vs. yellowness), vegetative biomass, total plant N, and petiole ureide concentration.

Harvesting for grain yield and total plant N is anticipated two months after planting dates for each site.
Harvest data will include final seed weight, pod number, seed number, seed size, and seed nitrogen content.

<u>Objective 2</u>: Examine the inheritance of genetic and environmental variation in BMF in common bean, and to identify molecular markers associated with QTL conditioning for enhanced BNF.

2a. Identify Parental Materials for Inheritance Studies of BNF.

- 6 month benchmark: Obtained experimental and adapted common bean germplasm

Michigan State University: Parental materials were for inheritance studies were identified based on previous knowledge of BNF capacity. One line, Puebla 152, was identified as BNF efficient and a RIL population exists with Eagle (snap bean) as the other parent. Additional genotypes were planted that are parents of other available RIL populations. Ninety two genotypes were planted in low nitrogen (25 lbs/acre) in Frankenmuth, MI. These materials included Eagle, Puebla 152, 72 Eagle x Puebla RILs, a no nod mutant, and 17 additional genotypes. The materials were planted under two treatments: 1) plus Becker Underwood 'Nodulator' inoculant, 2) no inoculant.

2b. Phenotype Existing Mapping Populations for Bnf Response, Populate With Molecular Markers, and Conduct QTL Analysis.

- 6 month benchmark: Increased seed of existing mapping populations for QTL analysis

Washington State University: A BNF experiment was conducted in the field in WA in 2010 at two separate locations, Prosser and Paterson. The objective was to survey bean genotypes for biological nitrogen response under low soil N conditions. There were three treatments: i) NT=no nitrogen or rhizobium inoculum, ii) BS=Biostacked rhizobium inoculum only, and iii) N=75 lbs of additional N only in the form of urea (46-0-0).

The Prosser trial site is a Warden Silt Loam and is used for selecting bean lines under multiple stresses (low fertility, soil compaction, drought, and root rot diseases). Historically the residual N for this trial has been about 25 lbs/A; however, this year 75 lbs/A residual N was detected in the trial ground right after planting. The high residual soil N appears to have compromised examination of the BNF response for the 23 genotypes tested at Prosser. Therefore, results from the Prosser trial will not be interpreted in detail at this time (supplementary Table).

The Paterson trial site is a Quincy Sand. Low residual N (25lbs/A) was confirmed for this trial prior to planting. The same set of genotypes plus five more (28 genotypes total) was tested in Paterson. Soil and plant samples obtained from specific treatments and genotypes at harvest maturity were recently transferred to the lab in Pullman (LCB) for examination of N levels but have not been analyzed yet. Soil total, available, and mineralizable N will be analyzed. Plant %N and 15/14N ratio will be analyzed. This information combined with yield data will be used to quantify the proportions and total amounts of BNF by these genotypes and treatments.

The non-nodulating genotypic check R99 had 43% more seed yield in the N (3460 kgha-1) than the NT (1966 kgha-1), which suggests that response to supplemental N in the absence of nodulation was detectable in this field trial (supplementary Table). Across all the genotypes tested however there was no significant difference between NT and N treatments suggesting that most of the genotypes included in the study are quite efficient for BNF. There was a significant effect for the BS inoculant treatment, which unexpectedly resulted in 7 and 8% less yield than the NT & N treatments, respectively**.** Perhaps the added Rhizobia were less effective than endemic strains.

Note that nodules for typing Rhizobium strains were not collected from the WA trials this season, but will be collected and characterized across treatments from select genotypes for both WA test locations next season. Procedures to analyze nodule and soil rhizobia will primarily use full community pyrosequencing of nifH, nifD, and (for nodules) 16SrDNA genes. Pyrosequencing is available through the WSU Core Molecular Biology laboratory. This method bypasses isolation of individual strains and cloning, and determines not only the nitrogen-fixing organisms present, but their relative proportions in soils and nodules of the various soil and bean genotype treatments. Where individual treatments are of particular interest for very high BNF, individual strains will be isolated from nodules for pure culture.

Preliminary greenhouse trials were undertaken in July – October to optimize growth conditions for P. vulgaris in the WSU-Pullman greenhouse facilities. A perlite-vermiculite mixture was found to reduce seedling growth but increase nodulation and final biomass as compared to perlite alone. Inoculated plants supplied Hoagland solution (N-free recipe after wk 2) twice weekly produced more biomass and nodules than plants fertilized once weekly. A severe infestation by thrips caused some defoliation in September; diligent observation and control of thrips will be

undertaken for successful genotype and strain trials. Fifty lines including the 28 genotypes tested in the field and 23 additional lines were sent to Pullman for analysis of BNF response in the greenhouse (supplemental Table). The greenhouse studies will commence in early November 2010.

QTL analyses: Development of genetic populations for mapping and QTL analysis has not commenced yet because suitable parental genotypes with clear differential BNF efficiency responses have not been identified. An existing RIL population (Eagle/Pueblo-152) was increased in the greenhouse for 2011 field planting but lack of adaptation of the parents for this population in 2010 WA field trials indicated that this population may only be useful for greenhouse examination of BNF response.

Michigan State University: Plant samples of the BNF trial were taken at mid pod fill for each genotype/treatment/rep. The sample consisted of all above ground biomass of 2 plants for each entry. Samples were oven dried and ground to a fine powder. These samples are being analyzed for N15 via natural abundance analysis at the UC Davis Stable Isotope Facility. Results are expected by December 15, 2010. Nitrogen from fixation will be estimated relative to the nitrogen accumulated by a non-nodulating bean line included as a control in the experiment.

SSR screening was conducted on Eagle and Puebla 152 to identify polymorphic markers. The BNF trial, including Eagle x Puebla RILs and additional 18 lines, was harvested on Oct 1, 2010. The Eagle x Puebla population did not do well in the field in MI. Many of the lines were very late maturing and did not have desirable growth habit. Marker analysis for the entire RIL population has yet to be conducted pending yield evaluation from the field plots.

<u>Objective 3</u>: Improve the productivity, profitability, and sustainability of agricultural systems on degraded soils through effective dissemination of new information and technologies to small-landholder farmers through on-farm demonstrations, mass media, field schools, and local forums.

1. Improve farmer awareness of inoculation technologies
2. Conduct on-farm demonstrations comparing inoculant strategies
3. Strengthen farmers' collective capabilities to purchase inoculants and incorporate them into a profitable and sustainable system for small landholders.

- **No funding allocated or benchmarks for this period.**

On-station research and demonstration trials have been initiated in which benefits of inoculants on the different varieties are being compared. These sites will serve as initial demonstrations for farmer field days. Initial contacts were made with the collaborating farmers that offered their fields for experimentation and demonstration. Training sessions followed by site visits and field days will be organized during this growing season.

PI Westgate met with the Chair of PELUM Uganda and Communications Coordinator for VEDCO, Agnes Kirabo, to initiate outreach activities with participating farmer organizations in

PELUM Uganda, PELUM Kenya, PELUM Tanzania, and PELUM Rwanda. Strategy meeting among PELUM country coordinators to initiate dissemination activities is scheduled for November.

Objective 4: Institutional capacity building

Three laptops (Dell) and one printer HP LaserJet P1006 were purchased by Makerere University for a total cost of US$ 5369 through the procurement system of the University. This equipment is being shared by two graduate students and the PI (Tenywa) at Makerere University.

Degree Training

Iowa State University
First and Other Given Names: **Mercy**
Last Name: **Kabahuma**
Citizenship: Ugandan
Gender: Female
Degree: MSc
Discipline: Crop Production and Physiology
Host Country Institution to Benefit from Training:
Training Location: Iowa State University
Supervising CRSP PI: Mark Westgate
Start Date of Degree Program: August 2010
Program Completion Date: August 2012
Training Status during Fiscal Year 2010: Just starting
Type of CRSP Support (full, partial or indirect): Full

Makerere University
First and Other Given Names: **Martha**
Last Name: **Abwate**
Citizenship: Ugandan
Gender: Female
Degree: MSc
Discipline: Soil Science
Host Country Institution to Benefit from Training: Makerere University
Training Location: Makerere University
Supervising CRSP PI: Steven Tenywa and Michael Ugen
Start Date of Degree Program: September 2010
Program Completion Date: August, 2012
Training Status during Fiscal Year 2010: Just starting
Type of CRSP Support (full, partial or indirect): Full

Makerere University
First and Other Given Names: **Peter**
Last Name: **Ssenyonga**
Citizenship: Ugandan
Gender: Male

Degree:. MSc
Discipline: Soil Microbiology
Host Country Institution to Benefit from Training: Makerere University
Training Location: Makerere University
Supervising CRSP PI: Steven Tenywa and Michael Ugen
Start Date of Degree Program: September 2010
Program Completion Date: August, 2012
Training Status during Fiscal Year 2010: Just starting
Type of CRSP Support (full, partial or indirect): Full

Sokoine University
First and Other Given Names: **Charles**
Last Name: **Komba**
Citizenship: Tanzanian
Gender: Male
Degree:. MSc
Discipline: Agronomy
Host Country Institution to Benefit from Training: Sokoine University of Agriculture (SUA)
Training Location: SUA
Supervising CRSP PI: Susan Nchimbi-Msolla
Start Date of Degree Program: September 2010
Program Completion Date: September, 2012
Training Status during Fiscal Year 2010: Just starting
Type of CRSP Support (full, partial or indirect): Full

Sokoine University
First and Other Given Names: **Beata**
Last Name: **Khafa**
Citizenship: Tanzanian
Gender: Female
Degree:. MSc
Discipline: Plant Breeding
Host Country Institution to Benefit from Training: Sokoine University of Agriculture (SUA)
Training Location: SUA
Supervising CRSP PI: Susan Nchimbi-Msolla
Start Date of Degree Program: September 2010
Program Completion Date: September, 2012
Training Status during Fiscal Year 2010: Just starting
Type of CRSP Support (full, partial or indirect): Full

Washington State University
First and Other Given Names: **Acceptance to program pending for January 2011**.
Last Name: Pending acceptance
Citizenship:
Gender: Female
Degree: MSc

Discipline: Plant Genetics and Plant Breeding
Host Country Institution to Benefit from Training: Washington State University
Training Location: Washington State University
Supervising CRSP PI: Lynne Carpenter-Boggs
Start Date of Degree Program: January 2011
Program Completion Date: December 2012
Training Status during Fiscal Year 2010: Not on site
Type of CRSP Support (full, partial or indirect): no support

Short Term Training

Two undergraduate students were recruited for attachment to the project to undertake field data collection at ISAR in Rwanda. 1. **Emma Uwera** 2. **Justin Tuyisenge** from Umutara Polytechnic University in Rwanda. The recruitment of a third undergraduate is being concluded. Their training on laboratory and field BNF techniques is underway.

Explanation for Changes

The project was approved and funding acquired by HC collaborators after July 2010. This was towards the end of the first growing season that started in March and ended in the same June/July 2010. Any activities not implemented as per performance indicators were due to this off-season arrangements. The main growing season for the implementation of the project started this September/October, 2010.

Due to an abrupt change from Professor Bekunda Matete to Dr. J.S. Tenywa at Makerere University, which followed shortly after the funds were transmitted, project activities started in August with selection of students. Field activities started mid September. As such data collection is underway and therefore we cannot present realist datasets at the moment.

The short-term training visit planned for Mr. Hamisi Tindwa of SUA to Washington State University to learn soil microbiology techniques from Dr. Lynne Carpenter-Boggs did not take place in this funding period due to late disbursement of funds from the MO. This training will take place in FY11.

Networking and Linkages with Stakeholders

Obtained germplasm from the NARs in Tanzania and from CIAT.

Dr. Tenywa travelled to Rwanda in September 2010 to discuss project activities with Dr. Augustine Musoni. Both travelled to Nyagatare where the then proposed site was evaluated for suitability for the BNF demonstrations. Soil sampling was conducted and the study design and treatments adopted for the NaCRRI sites were also considered for the Rwanda sites to permit regional comparisons.

A Material Transfer Agreement was established between ISU and CIAT Columbia to obtain germplasm with potential application to this project.

Plans to conduct a workshop among BNF-CRSP PIs directing Phase II and Phase III projects currently being planned for early in FY11.

Leveraged Funds

ISAR is a partner in the N2Africa program led by the University of Wangengen and CIAT investigating effects of inoculants on yields of improved bean germplasm. Funding from this program supports training for one PhD and MSc student in Rwanda. ISAR is also part of the AGRA Soil Health Program project investigating interactions between inoculants, varieties, and soil conditions. Complementation among these projects leverages results on germplasm sources and inoculants developed locally, regionally, and from the US.

Scholarly Activities and Accomplishments

No publications, technical reports, or theses submitted during this funding period.

Tables/Figures

Figure 1. General field map for one replicate of the inoculant evaluation trials. R_0 = No Rhizobium strain Inoculated, R_M = Rhizobium strain from Makerere University Bio-fix, R_N = Rhizobium strain from Nairobi Bio-N-fix, R_U = Rhizobium strain from Underwood BioStacked®, P_0 = No phosphorus fertilizer applied, P_+ = Phosphorus fertilizer (TSP) applied at 40 kg P ha^{-1}. Varieties V1-3 varied by location.

3x5 m^2	1	2	3	4	5	6	7	8
1	$V_1R_0P_0$	$V_1R_NP_+$	$V_1R_0P_0$	$V_1R_NP_0$	$V_1R_UP_0$	$V_1R_MP_+$	$V_1R_MP_0$	$V_1R_UP_+$
	$V_1R_0P_+$	$V_1R_UP_0$	$V_1R_NP_0$	$V_1R_NP_0$	$V_1R_NP_+$	$V_1R_MP_+$	$V_1R_0P_0$	$V_1R_UP_+$
	$V_1R_0P_+$	$V_1R_UP_0$	$V_1R_UP_+$	$V_1R_MP_+$	$V_1R_MP_0$	$V_1R_NP_+$	$V_1R_MP_0$	$V_1R_0P_+$
2	$V_2R_UP_0$	$V_2R_NP_0$	$V_2R_0P_0$	$V_2R_NP_0$	$V_2R_0P_+$	$V_2R_MP_+$	$V_2R_MP_0$	$V_2R_MP_0$
	$V_2R_0P_+$	$V_2R_NP_+$	$V_2R_NP_+$	$V_2R_MP_+$	$V_2R_NP_0$	$V_2R_UP_+$	$V_2R_NP_+$	$V_2R_0P_+$
	$V_2R_UP_0$	$V_2R_MP_0$	$V_2R_MP_+$	$V_2R_UP_0$	$V_2R_0P_+$	$V_2R_UP_+$	$V_2R_0P_0$	$V_2R_UP_+$
3	$V_3R_MP_0$	$V_3R_0P_0$	$V_3R_NP_+$	$V_3R_UP_0$	$V_3R_MP_0$	$V_3R_UP_+$	$V_3R_MP_+$	$V_3R_UP_0$
	$V_3R_UP_+$	$V_3R_MP_+$	$V_3R_UP_+$	$V_3R_NP_+$	$V_3R_MP_+$	$V_3R_0P_0$	$V_3R_0P_0$	$V_3R_NP_+$
	$V_3R_NP_0$	$V_3R_0P_+$	$V_3R_0P_+$	$V_3R_NP_0$	$V_3R_0P_+$	$V_3R_NP_0$	$V_3R_UP_0$	$V_3R_MP_0$

Table 1. High yielding climbing and bush beans varieties selected for the bionitrogen fixation trial in Nyagatare district (1200 – 1500 masl) of Rwanda in 2010.

Variety	Plant type	Market class	Maturity (M)	Mean Yield (t/ha)	Special attributes
ISAR-CB-105	IVA	Calima/mottled	3.0	3.0	Heat & drought tolerant; extra early; rust &CBB resistant, marketable
ISAR-CB-107	IVA	Calima/mottled	3.0	3.5	Heat & drought tolerant; extra early; BMV, rust resistant, marketable and taste
ISAR-SCB-102	IIA	Small red	2.5	2.5	Drought resistant, marketable, taste
RWR 1668	I	Kidney	2.5	2.0	Multiple resistance, marketable and culinary

Literature Cited

Bationo, A. 2004. Managing Nutrient Cycles to Sustain Soil Fertility in Sub- Saharan Africa. *TSBF-CIAT.* Academy Science Publishers, Nairobi

Bliss, F.A., Pereira, P.A.A., Araujo, R.S., Henson, R.A., Kmiecik, K.A., McFerson, J.R., Teixeira, M.G., and daSilva, C.C. 1989. Registration of five high nitrogen fixation common bean germplasm lines. Crop Sci. 29: 240-241.

Bliss, F.A., 1993. Breeding common bean for improved nitrogen fixation. Plant and Soil 152: 71-79.

CIAT 2002. Soil fertility degradation in sub-Saharan Africa: Levering lasting solutions to a long lasting problem. Rockefeller Bellagio Study. www.ciat.cgiar.org/news/pdf/tsbf_bellagio.pdf

Diouf, A., Abdoulaye Diop, T., and Gueye, M. 2008. Nodulation in situ of common bean (Phaseolus vulgaris L.) and field outcome of an elite symbiotic association in Senegal. Res. J. Agric. Biol. Sci. 4: 810-818.

Graham, P.H. 1981. Some problems of nodulation and symbiotic nitrogen fixation in Phaseolus vulgaris L.: A Review. Field Crops Research 4: 93-112

Graham, P.H. 2003. Addressing edaphic constraints. Field Crop Research 82:179-192.

Martinez-Romero, E. 2003. Diversity of Rhizobium-Phaseolus vulgaris symbiosis: overview and perspectives. Plant and Soil 252: 11-23.

Miklas, P. N., Kelly, J. D., Beebe, S. E., and Blair, M.W. 2006. Common bean breeding for resistance against biotic and abiotic stresses: from classical to MAS breeding. Euphytica 147:105-131.

Musdandu, A.A.O., and Joshua, O.O. 2001. Response of common bean to rhizobium inoculation and fertilizers. J. Food Tech. Africa 6:121-125.

Ndakidemi, P.A. , Dakora, F.D. , Nkonya, C. , Ringo, D., and Mansoor, D. 2006. Yield and economic benefits of common bean (Phaseolus vulgaris) and soybean (Glycine max) inoculation in northern Tanzania. Aust. J. Exp. Agric. 46(4) 571–577.

Nkwiine, C. 1999. Increase of crop yields by beneficial organisms: a case of rhizobia use in Uganda. Proc. Soil Sci. Soc. East Africa. 17th Conf. p 174-6.

Schulze, J. 2004. How are nitrogen fixation rates regulated in legumes? J. Plant Nutr. Soil Sci. 167:125-137.

Silver, M.C.R., and Nkwiine, C. 2007. A review of work on rhizosphere microbiota in Uganda. African J. Ecol.. 45(2):20-26.

Vadez, V., Lasso, J.H., Beck, D.P., and Drevon, J.J. 1999. Variability of N_2-fixation in common bean (*Phaseolus vulgaris* L.) under P deficiency is related to P use efficiency: N_2-fixation tolerant to P deficiency. Euphytica 106: 231-242.

Contribution to Gender Equity Goal

Of the eight graduate and undergraduate students formally involved in training activities at US and Host Countries thus far, five (62.5%) are female. It is our plan to involve where possible at least 50% women to participate in field demonstrations and on-farm trials.

Dry Grain Pulses CRSP

Research, Training and Outreach Workplans

(October 1, 20010 -- September 30, 2011)

FY 2011 PERFORMANCE INDICATORS-SUA

for Foreign Assistance Framework and the Initiative to End Hunger in Africa (IEHA)

Project Title: Enhancing biological nitrogen fixation (BNF) of leguminous crops grown on degraded soils in Uganda, Rwanda, and Tanzania

Lead U.S. PI and University: Mark E. Westgate, Iowa State University
Host Country(s): Rwanda, Tanzania, Uganda

Output Indicators	2011 Target (October 1, 2010-Sept 30, 2011)	2011 Actual (October 1, 2010-Sept 30, 2011)
Degree Training: Number of individuals enrolled in degree training		
Number of women	4	4
Number of men	3	4
Short-term Training: Number of individuals who received short-term training		
Number of women	50	4
Number of men	50	4
Technologies and Policies		
Number of technologies and management practices under research	1	1
Number of technologies and management practices under field testing	1	1
Number of technologies and management practices made available for transfer	1	1
Number of policy studies undertaken		
Beneficiaries:		
Number of rural households benefiting directly	1000	0
Number of agricultural firms/enterprises benefiting	6	3
Number of producer and/or community-based organizations receiving technical assistance	50	0
Number of women organizations receiving technical assistance	3	0
Number of HC partner organizations/institutions benefiting	6	3
Developmental outcomes:		
Number of additional hectares under improved technologies or management practices	1000	20

Dry Grain Pulses CRSP

Report on the Achievement of "Semi-Annual Indicators of Progress"

(For the Period: October 1, 2010 – September 30, 2011)

This form should be completed by the U.S. Lead PI and submitted to the MO by October 1, 2011

Project Title: *Enhancing biological nitrogen fixation (BNF) of leguminous crops grown on degraded soils in Uganda, Rwanda, and Tanzania*

Abbreviated name of institutions

	ISU			MSU			WSU			VEDCO			NaCCRI			SUA			Makerere			ISAR		
	Target	Achieved		Target	Achieved		Target	Achieved		Target	Achieved		Target	Achieved		Target	Achieved		Target	Achieved		Target	Achieved	
Benchmarks by Objectives	#######	Y	N*	#######	Y	N*	#######	Y	N*	#######	Y	N*	#######	Y	N*	#######	Y	N*	#######	Y	N*	#######	Y	N*
	(Tick mark the Yes or No column for identified benchmarks by institution)																							
Objective 1																								
Field Trial 1 established at HC sites													x	Y		x	Y					x	Y	
Quantified yield advantage of inoculation in US and HC field trials				x	Y		x	Y					x	Y		x	Y					x	Y	
Established project website for data storage/sharing	x		N																					
Secured non-nodulating lines for field studies							x	Y					x	Y										
Analysis of soil/weather data completed	x												x	Y		x	Y		x	Y		x	Y	
Common varieties to test at all HC sites identified													x	Y		x	Y					x	Y	
Finalized protocols for field sampling and controlled environment work	x	Y					x	Y																
Phenotypic responses to inoculant x genotype x environment identified	x	Y		x	Y		x	Y					x	Y		x	Y		x	Y		x	Y	
Soil from field sites tested for indigenous rhizobia strains	x		N	x		N	x		N				x	Y		x		N	x		N	x		N
Established indigenous rhizobia levels associated with environment							x		N															
Initial characterization of soil rhizobia populations conducted							x		N															
Trial inoculants evaluated for rhizobia titre	x		N				x		N															
Objective 2																								
Identified/collected bean lines for BNF diversity panel				x	Y		x	Y																
Collected/increased seed of non-nodulating adapted lines	x	Y		x	Y		x	Y																
Screened selected lines for BNF in low soil N +/- inoculants in HC field trials				x	Y		x	Y																
Greenhouse screening trials initiated on selected lines for BNF response				x	Y		x	Y																
Identify potenial RIL parental lines for BNF response	x	Y		x	Y		x	Y																
Phenotyping of selected lines for BNF response at US site initated				x	Y		x	Y																
Plant samples for SNP analysis collected from CAP and SA Core collection				x	Y		x	Y																
Mechanism to correlate field and GH reponses established	x		N	x		N	x		N				x		N	x		N	x		N	x		N
Iniated search for BNF responsive genes in silico				x	Y		x	Y																
Selected RILs advanced to F2				x	Y		x		N															
Objective 3																								
Created awarenes among Extensionists at HC institutions in benefits of BNF										x	Y		x	Y		x		N				x		N
Evaluated farmer Knowledge, Practices, Attitudes on inoculation										x		N				x		N				x		N
Prepared for on-farm trials by identifying potential farmer cooperators										x		N	x		N	x	Y					x	Y	
Prepared for on-farm trails by training cooperators on field methods										x		N	x		N	x	Y					x	Y	
Initiated training materials on BNF and seed inoculation for extansionists,	x	Y								x	Y								x		N			
Initiated production of training materials to disseminate through PELUM	x		N							x		N							x		N			
Objective 4																								
Graduate students identified																x	Y							
Graduate research programs initiated																x	Y							
Undergraduate student interns identified																								
Undergraduate student projects intiated																								
Team members trained in mehods of SNP analysis							x		N															
Conducted short-term training of graduate students on BNF protocols	x	Y					x		N															

Name of the PI reporting on benchmarks by institution	Westgate	Cichy	Miklas	Musoke	Ugen	Mchimbi	Tenywa	Musoni

Name of the U.S. Lead PI submitting this Report to the MO #######

Signature **Date**