Feed the Future Innovation Lab for
Collaborative Research on Grain Legumes
(Legume Innovation Lab)

FY 2017 Annual Project Technical Progress Report
(October 1, 2016 – September 29, 2017)

Project Code and Title:

SO1.A2 Improving Photosynthesis in Grain Legumes with New Plant Phenotyping Technologies

Lead U.S. Principal Investigator and University:

David M. Kramer Biochemistry and Molecular Biology and Plant Research Lab, Michigan State University

Collaborating Host Country and U.S. PIs and Institutions:

Kelvin Kamfwa, University of Zambia
Kennedy Muimui, ZARI, Zambia
Wayne Loescher, MSU
James Kelly, MSU
Tim Close, U.C. Riverside
Phil Roberts, U.C. Riverside
Maren Friesen, MSU, Plant Biology

I. Abstract of Research and Capacity Strengthening Achievements

(A succinct narrative on the technical progress of the project, including key research and capacity strengthening achievements and outcomes, during the FY 2017 performance and report period. 1200 character limit.)

In the previous year we met all of our objectives for FY 2017. The first critical step was the successful manufacturing of the next generation MultispeQ instrument, which has improved capabilities over the initial Beta MultispeQ. This allowed us to achieve our goals of enhancing the capabilities of our Feed the Future (FtF) research partners to ‘bring the lab to the field’ using advanced phenotyping technologies. We distributed new MultispeQ’s v1.0 instruments and training to students, research technicians and faculty at the University of Zambia and Makerere University. Additionally, we renovated the irrigation system at the University of Zambia research farm. These capacity building activities enabled us to identify photosynthetic efficiency QTL’s under drought conditions in the field at the University of Zambia.

Another key research goal was to bring the field to the lab. Using two different advanced phenotyping technologies, DEPI chambers and the PhotosynQ platform, we were able

identify key QTL's for heat and cold tolerance. We identified comparable heat tolerant QTL's using both advanced phenotyping platforms and identified cold tolerance QTL's in two different RIL populations using PhotosynQ, and evaluating the lipid membrane and ATP synthase activity.

II. Project Problem Statement and Justification

To avert food shortages and feed its growing population, there is critical need for increasing the productivity of grain legumes in Zambia, which ranks 164 out of 184 countries in the Human Poverty Index. Grain legumes are important crops in Zambia constituting both critical sources of protein and income. Bean production is constrained by its low inherent photosynthetic efficiency which is highly sensitive to abiotic and biotic stresses, including diseases, pests, low soil fertility hear and drought.

To achieve major gains in yield, we need to improve both the robustness and the efficiency of photosynthesis. This is a complex problem requiring the combined application of advanced genomics and high throughput phenotyping approaches. We will take a critical step in this direction by establishing a base of phenotyping technologies and advanced genetics and genomics approaches to identify quantitative trait loci (QTLs) that condition more efficient and robust photosynthesis and productivity in cowpea and common beans. We will also test the ability of a newly developed research platform, PhotosynQ, to enable researchers and farmers to conduct plant phenotyping experiments, analyze data and share results, and thus allow improvements in breeding and management on local to global scales.

In previous years of the project we identified environmental conditions that aid in the identification of QTL's in common bean and cowpea. Furthermore, we successfully deployed PhotosynQ under field conditions in Zambia. However, a lack of infrastructure was constraining the development of a robust phenotyping center at the University of Zambia.

III. Technical Research Progress

Objective 1: Probing photosynthetic responses in RIL and GWAS lines.

During the past year we have successfully mapped QTL's in common bean and cowpea using two advanced phenotyping platforms; DEPI chambers and the PhotosynQ platform (Fig. 1). Using both platforms were we able to achieve comparable results for a RIL cowpea population (developed by the University of California Riverside) under both heat stress and normal growth conditions. Furthermore, we have identified a QTL for relative chlorophyll content and photoprotection (qEt) that coincided with biomass production under heat stress but not control conditions.

Additionally, we were able to map QTL's in 2 different RIL populations (provided by UC Riverside) for cold tolerance. These traits can be used to enable earlier planting in order to avoid heat stress. Additionally, we mapped the lipid membrane and ATP synthase to identify QTL's for photosynthetic efficiency, which allowed us to explore the mechanisms behind cold tolerance.

In the field we were able to identify key QTL's for higher photosynthetic efficiency under moisture stress conditions such as drought. This allows us to identify germplasm that will serve as parents in making crosses to develop varieties with preferred seed types that are tolerant to drought. Furthermore, we were able to identify the same QTL's under drought stress in multiple field trials, validating the PhotosynQ approach to identifying key QTL's.

Objective 2: Develop a data management plan to improve the communication of ideas, results, and analysis to a large network of connected scientists.

The PhotosynQ platform is set up to allow for rapid communication of ideas, results, and analyses. All data collected on the PhotosynQ platform is open, making it easy for scientists from around the world to connect and share results and analyses. All data collected with the support of the Legume Innovation Lab can be easily searched on the PhotosynQ platform (www.photosynq.org) using the hashtags #LIL and #LegumeInnovationLab.

Objective 3: Increase the capacity, effectiveness and sustainability of agriculture research institutions which serve the bean and cowpea sectors in the target FTF countries by establishing an African-USA community of networked scientists, extension agents, students and growers to address field-level research and production questions.

In FY 2017 we made major strides towards achieving this objective. One of the capacity development goals in this project is to turn the University of Zambia and Makerere University into cutting edge phenotyping centers. To that end, we have distributed 6 next generation MultispeQ v1.0 instruments to the University of Zambia and 5 MultispeQ v1.0 instruments to Makerere University. Dr. Kamfwa trained 4 research technicians from the Zambia Agricultural Research Institute and 1 Masters Student at UNZA on the PhotosynQ platform and MultispeQ instrument. Additionally, students and research technicians have collected over 19,000 phenotyping measurements on the common bean Andean Diversity Panel. PhD student Isaac Dramadri has trained students at Makerere University who have collected over 14,000 measurements on cowpea, groundnuts, and maize

In addition to training and instrumentation, we have made changes to the PhotosynQ website that improve the connectivity of phenotyping data to advanced analytics. This includes developing R and Python packages allowing users to directly import data from www.photosynq.org into these platforms for analysis. We have also increased the number of tutorials, help modules and analytical tools available on PhotosynQ to allow anyone, with minimal training, to pick up a MultispeQ instrument, create a robust PhotosynQ project, collect high quality data, and properly analyze the data. These tools will greatly enhance the ability of users at UNZA and Makerere University to harness the full potential of the PhotosynQ platform.

IV. Major Achievements

(*Present a* ***list of significant research achievements and/or technical advances resulting from project activities during the FY 2017 performance period****. The description of each achievement need not be more than three sentences long. Quantitative information on or a technical description of the research achievement would be appreciated because it adds credibility to the importance of the achievement*.)

1. Dissemination of the next generation MultispeQ v1.0 instrument. 6 new MultispeQ's were provided to the University of Zambia and 5 MultispeQ's to Makerere University in Uganda. In total, 500 next generation MultispeQ's were manufactured.
2. Identified QTL's related to cold tolerant photosynthetic efficiency using the PhotosynQ platform and evaluation of the lipid membrane and ATP synthase.
3. Identified comparable heat tolerance QTLs' using DEPI and PhotosynQ data in climate controlled growth chambers.
4. Identified drought tolerant QTL's under field conditions in Zambia.
5. Trained several graduate students at both UNZA and MSU, as well as research technicians from ZARI and students at Makerere University.
6. Development of new PhotosynQ-guided experimental protocols.

V. Research Capacity Strengthening

(*Describe how collaborative research activities supported by the project during FY 2017 have contributed to the strengthening of institutional capacity to carry out multidisciplinary research on grain legumes and to solve the problems facing the legume sectors in host countries and regions. Appropriate capacity strengthening items to present in this section include research equipment procured (>$5,000), laboratory and analytical facilities developed, participation in professional meetings or other networking activities, etc. Please also identify in this section the activities completed and equipment procured during the past fiscal year with supplemental Institutional Capacity Strengthening funds received by host country institutions in the respective project.)*

We have made progress in several areas of research capacity building.

First, Several LIL participants are currently using the PhotosynQ platform as a part of their LIL-supported work, including Kelvin Kamfwa (University of Zambia), Isaac Dramadri (Makerere University), Isaac Osei-Bonsu (in the Kramer lab at MSU) as well as several students working with collaborators Tim Close (U.C. Riverside), Phillip Roberts (U.C. Riverside) and Phil McLean (NDSU).

Second, we are actively working towards setting up cutting edge phenotyping centers at the University of Zambia and Makerere University in Uganda. At UNZA, faculty and staff now have access to 6 new MultispeQ v1.0 instruments and we are actively involving the UNZA team in the design of the new online analytical capacity, which will both lead to publications and help nucleate a phenotyping center at UNZA. In addition, two research technicians from the Common Bean Breeding National Program and two research technicians from the Wheat Breeding Program, both part of the Zambia Agricultural Research Institute (ZARI) in Zambia have received training on PhotosynQ. Over the past year, this has resulted in 9 PhotosynQ projects being created in FY 2017 and 19,783 measurements being collected on common bean and wheat. Similarly, we have provided 5 MultispeQ v1.0 instruments to Makerere University, which are available for faculty and staff to use, resulting in the initiation of 9 separate research projects in which over 14,000 measurements have been made on cowpea, groundnuts, and maize.

The irrigation capacity at the University of Zambia research farm in Lusaka, Zambia has been upgraded using supplemental Institutional Capacity Strengthening funds. Purchase and installation of irrigation equipment was completed, increasing the research capacity at UNZA to conduct drought experiments.

Finally, it is important to emphasize that all of the research accomplishments are purposely and directly connected to capacity strengthening. For example, the development of the instrumentation and PhotosynQ-guided experimental protocols were guided by the research goal of identifying the genetic bases of photosynthetic responses, but are also incorporated in the PhotosynQ platform to enable future work.

VI. Human Resource and Institution Capacity Strengthening

1. **Short-Term Training**
   (*Provide the following information for each short-term training activity completed. If a training was repeated in several places, each training must have a separate entry. Short term training is defined as a minimum of two consecutive class days or more in duration, or 16 contact hours or more scheduled intermittently*.)
   i. Purpose of Training: Train research technicians at the Zambia Agricultural Research Institute on how to use the PhotosynQ platform to collect data from field experiments.
   ii. Type of Training: Short-term training
   iii. Country Benefiting: Zambia
   iv. Estimated USAID funding for activity[1]

[1] Instruction expenses include costs directly incurred to convey knowledge or impart training, such as books, equipment, supplies, course handouts; seminar/conference/workshop registration fees and published academic tuition and fees. Participant expenses include costs directly incurred to meet personal needs and program requirements including per diem, medical exams, visa fees; health and accident insurance premium; and federal state and local taxes. Travel expenses include those costs directly incurred transporting the participant from the home country to the training country and back, as well as costs within the country.

a. US$ 400 for Instruction
b. US$ 750 for Participants
c. US$ 300 for Travel

v. Location and dates of training: University of Zambia (05/15/2017-05/17/2017)
vi. Number receiving training (by gender): Two females and two males
vii. Home institution(s): Zambia Agricultural Research Institute
viii. Institution providing training or mechanism: University of Zambia

2. Degree Training in the US or elsewhere

i. Name of trainee: Susan Chipandwe
ii. Country of Citizenship: Zambia
iii. Gender: Female
iv. Host Country Institution Benefitting from Training: University of Zambia
v. Institution providing training: University of Zambia
vi. Supervising LIL PI: Kelvin Kamfwa
vii. Degree Program: Masters
viii. Field or Discipline: Plant Breeding and Seed Systems
ix. Research Project Title (if applicable)Estimated USAID funding for activity if not conducted in US
    a. US$ 2,000 for Instruction
    b. US$ 11,000 for Participants
    c. US$ 200 for Travel
x. Estimated funding from other sources for activity if not conducted in US (see footnote 1)
    a. Provider of Funds
    b. US$ for Instruction
    c. US$ for Participants
    d. US$ for Travel
xi. Start Date: September, 2016
xii. Projected/Actual Completion Date: September, 2018
xiii. Is trainee a USAID Participant Trainee and registered on TraiNet?
xiv. Training status: Active

i. Name of trainee: Isaac Osei-Bonsu
ii. Country of Citizenship: Ghana
iii. Gender: Male
iv. Host Country Institution Benefitting from Training: CSIR-Crops Research Institute
v. Institution providing training: Michigan State University
vi. Supervising LIL PI: David Kramer
vii. Degree Program: Doctorate
viii. Field or Discipline: Plant Physiology
ix. Research Project Title: Heat Stress Effects On Photosynthesis in Legumes

x. Estimated USAID funding for activity if not conducted in US (see footnote 1)
   d. US$ for Instruction
   e. US$ for Participants
   f. US$ for Travel

xi. Estimated funding from other sources for activity if not conducted in US (see footnote 1)
   e. Provider of Funds
   f. US$ for Instruction
   g. US$ for Participants
   h. US$ for Travel

xii. Start Date: August, 2015

xiii. Projected/Actual Completion Date: 2019

xiv. Is trainee a USAID Participant Trainee and registered on TraiNet?

xv. Training status: Active, indirect support

VII. Achievement of Gender Equity Goals

A critical component of accomplishing our gender equality goals is to ensure that PhotosynQ technologies are equally accessible to women and men. In FY 2017 we have made progress towards that goal by 1) training 2 female research technicians at ZARI and 2) supporting a female graduate student who is pursuing a Master's Degree in plant breeding and seed systems at UNZA using the PhotosynQ platform, with Dr. Kamfwa as her PI.

VIII. Implementation of Data Management Plan
(*Describe efforts to implement of project's Data Management Plan, including a summary of data sets that have been submitted to Open Data Access sites, including USAID DDL. Indicate your Open Data Access sites for future submissions as well.*)

The PhotosynQ platform is set up to allow for rapid communication of ideas, results, and analyses. All data collected on the PhotosynQ platform is open, making it easy for scientists from around the world to connect and share results and analyses. All data collected with the support of the Legume Innovation Lab can be easily searched on the PhotosynQ platform (www.photosynq.org) using the hashtags #LIL and #LegumeInnovationLab.

IX. Scholarly Accomplishments

(*Identify all* ***publications****, theses and/or dissertations, presentations, professional recognitions, awards, patents, and Plant Variety Protection Certificates that evidence scholarly accomplishments by U.S. and Host Country scientists as well as degree trainees during the performance period. Please send electronic copies of publications to the MO for sharing with USAID and include URL or DOI information where available.)*

Hoh D, Osei- Bonsu I, Cruz J, Roberts PA, Huynh BL, Tessmer OL, Savage L, Hall D, Kramer DM. 2017. Determination of cold tolerance QTLs via high throughput photosynthetic phenotyping photosynthesis. Poster, Phenome 2017. Tucson, Arizona

Hoh D, Osei- Bonsu I, Cruz J, Roberts PA, Huynh BL, Tessmer OL, Close T, Savage L, Hall D, Kramer DM. 2017.The genetic and mechanistic bases of photosynthetic cold tolerance in Cowpea (*Vigna unguiculata (L.)* Walp.*)* via high throughput environmental phenotyping. Poster, 43rd Annual Midwest/Southeast Photosynthesis Conference. Marshall, IN

Hoh D, Osei- Bonsu I, Cruz J, Roberts PA, Huynh BL, Tessmer OL, Close T, Savage L, Hall D, Kramer DM. 2017. Determination of cold tolerance QTLs in Cowpea (*Vigna unguiculata (L.) Walp.)* via high throughput photosynthetic phenotyping. Feed the Future Legume Innovation Lab Grain Legume Research Conference 2017. Ouagadougou, Burkina Faso.

Hoh D, Osei- Bonsu I, Cruz J, Roberts PA, Huynh BL, Tessmer OL, Close T, Savage L, Hall D, Kramer DM. 2017. Determination of cold tolerance QTLs in Cowpea (*Vigna unguiculata (L.) Walp.)* via high throughput photosynthetic phenotyping. Feed the Future Legume Innovation Lab Grain Legume Research Conference 2017. Ouagadougou, Burkina Faso.

Osei-Bonsu I, Hoh D, Loescher W, TerAvest D, Kramer D. (2017). Chlorophyll fluorescence and leaf gas exchange based phenotyping of heat stress tolerance in tepary, common bean and cowpea at the seedling and juvenile stages. Poster presented at the Phenome 2017 Conference, 10 - 14 February 2017, Tucson, Arizona.

Osei-Bonsu I, Hoh D, TerAvest D, Cruz J, Savage L, Huynh B-L, Roberts PA, Kramer D. (2017). Assessing tolerance of photosynthesis to high night temperature stress in cowpea genotypes and mapping of QTLs associated with photosynthetic traits. Poster presented at the Legume Innovation Lab Conference 2017, 13 - 18 August 2017, Ouagadougou, Burkina Faso.

Osei-Bonsu I, Hoh D, TerAvest D, Cruz J, Savage L, Huynh B-L, Tessmer O, Roberts PA, Close TJ, Kramer D. (2017). Thermo-tolerance of photosynthesis in grain legumes: mapping QTLs for photosynthetic traits in cowpea under heat stress by high throughput photosynthesis phenotyping. Presented at the 43 Midwest/Southeast Photosynthesis meeting 2017, 27 - 29 October 2017, Marshall, Indiana, USA.

Osei-Bonsu I, Hoh D, TerAvest D, Cruz J, Savage L, Huynh B-L, Roberts PA, Kramer D. (2017). Thermo-tolerance of photosynthesis in legumes: Mapping QTLs for photosynthetic traits in cowpea under heat stress. Presented at the Legume Innovation Lab Conference 2017, 13 - 18 August 2017, Ouagadougou, Burkina Faso.

TerAvest D, Mnthambala F, Kamfwa K, Yohane E, Kramer D. (2017). What do we learn from Phenotyping Tools that Bridge the Gaps between the Lab and the World? Presented at the Phenome 2017 Conference, 10 - 14 February 2017, Tucson, Arizona.

TerAvest D, Mnthambala F, Siyeni D, Ngwira A, Kramer D. (2017). PhotosynQ: Empowering Collection of Direct Crop Physiological Measurements on Smallholder Farms. Presented at the Legume Innovation Lab Conference 2017, 13 - 18 August 2017, Ouagadougou, Burkina Faso.

X. Achievement of Impact Pathway Action Plan

(At the project planning and workplan development stage, each project team prepared an Impact Pathway identifying major research outputs, users of these outputs, a vision of success, and necessary steps to achieve the vision of success. In the Impact Pathway worksheet, your project also identified strategies and an action plan to be undertaken by the project team over the 4.5 year life of the project to translate the outputs into outcomes. Please provide an update on your team's efforts in implementing the action plan and progress towards achieving the 'vision of success' as laid out in the Impact Pathway strategy. Discuss any constraints encountered and steps taken to overcome them.)

Outputs:

Output #1. Provide advanced scientific instrumentation for developing countries: The project will produce 20 MultispeQ instruments, 16 of which will be delivered to labs in Zambia and Uganda. Qualitatively, these instruments will immediately allow researchers in Africa to perform cutting edge research, enabling them to perform the work described in the proposal. In addition, we expect the capabilities of the instruments to enable researchers in HCs to initiate new research projects.

In FY 2017, we provided 9 next generation MultispeQ v1.0 instruments to project partners at the University of Zambia and Makerere University. Between the first generation MultispeQ Beta and revised MultispeQ v1.0 instruments, faculty and graduate and undergraduate students at Makerere University and the University of Zambia have contributed significant data to the PhotosynQ platform. Specifically, at Makerere University 6 different project leads contributed 4,339 and 14,315 MultispeQ beta and v1.0 measurements, respectively. At UNZA, 2 project leads contributed 7,586 and 12,197 MultispeQ beta and v1.0 measurements, respectively.

Output#2. Capacity building by internet-enabled data and communications platform. We will develop and deliver to the community a new platform for discovering QTL's to enhance the productivity of grain legumes.

We are currently developing a new automated data analysis pipeline within the PhotosynQ platform that will allow PhotosynQ users to rapidly connect in-field phenotyping data to genotypic data to identify QTL's in grain legumes. A beta version of this platform is expected to be completed in December, 2017.

Output #3. Capacity building through advanced phenotype-driven identification of QTLs for improving the efficiency and resilience of photosynthesis in grain legumes.

We have successfully identified phenotype-driven QTL's using both controlled conditions in DEPI chambers and using MultispeQ under true field conditions for cold, heat and drought tolerance.

Output #4. Capacity building through education: Another essential component of the project is to enable researchers in HC to take possession of both the technolog and the educational efforts required to use it effectively. To achieve this, we plan to engage students in the process of developing and disseminating the educational materials, giving them ultimate control of the platform and process.

We have made progress towards this output goal through the following of activities: 1) training of undergraduate and graduate students at Michigan State University, the University of Zambia and Makerere University and 2) developing training videos, tutorials and educational modules that are available on www.photosynq.org so any user on the platform can learn how to use the PhotosynQ platform to answer research questions.

ANNEXES

Annex 1. Tables, Figures, and Photos Cited in the Report

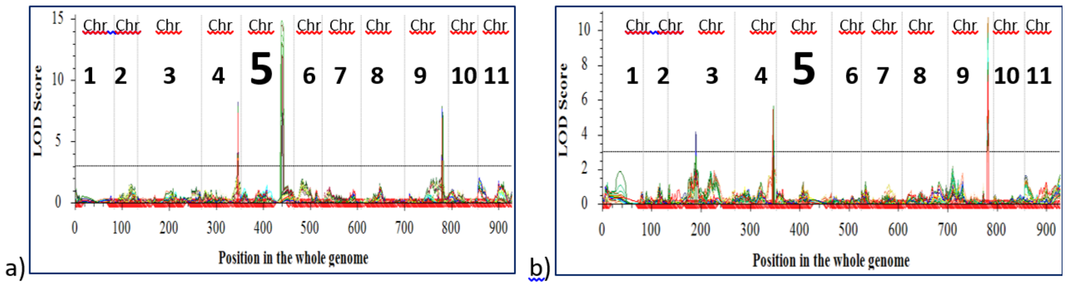

Fig 1: QTLs photosynthetic efficiency (Phi2) under a) heat stress and b) control growth conditio
The QTL band on chromosome 5 is apparently specific to the heat stress whereas those on chromosome 4 and 9 are constitutively expressed.