Title: Improving sorghum adaptation in West Africa with genomics-enabled breeding	
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2. Executive Summary

Crop production on smallholder farms in West Africa is extremely vulnerable to unfavorable weather and soil conditions and depredation from pests and disease. Since smallholder farming is the basis of economic activity and food production in this region, crop yield losses due to abiotic and biotic stressors are a key limitation to economic development and threat to food security. A major goal for crop breeders in National Agricultural Research Systems (NARS) is to develop new varieties that maintain, and ultimately increase, crop yields in the face of abiotic and biotic stressors. However, the development of more resilient varieties has been slow because the same poor and unpredictable growing conditions that limit crop production also limit crop breeding.

One approach to address this challenge is marker-assisted breeding, which enables phenotyping efforts to be distilled to inexpensive genetic markers that can be selected for or against without further phenotyping. After many years of investment, marker-assisted breeding is now a viable strategy to meet the challenges of crop improvement in West Africa. Here we propose use new genomic tools to accelerate marker-assisted breeding and expand its impact in West Africa, with six integrated objectives: (1) Genomic characterization of Senegalese and Nigerien landraces and breeding lines to connect West African germplasm to global sorghum breeding efforts; (2) Development of simplified genomics toolkit to provide optimized marker sets for marker-assisted breeding in West Africa; (3) Development of multi-parent populations for more efficient trait mapping and breeding and combine traits from locally-preferred varieties and elite global lines; (4) Improved mapping of stress resistance/tolerance traits to generate more effective trait-associated markers; and (5) Implementation of Marker Assisted Recurrent Selection to develop more resilient locally-preferred varieties; (6) Long-term and short-term training on genomics-enabled breeding for West African crop scientists. The project represents a collaborative effort of crop breeders and scientists at INRAN, CERAAS/ISRA, CIRAD, ICRISAT, and KSU, integrating several existing collaborations and bringing in new partners.

The proposed project to accelerate sorghum breeding will have numerous outcomes that support the relevant USAID country strategies and FtF objectives, as the sorghum value chain has been identified as target area by bilateral USAID missions in West Africa and sorghum is major component of the diet of many of sub-saharan Africa's poorest rural people. In particular, the proposed project will directly address the USAID strategy for climate-smart agriculture in West Africa by accelerating the development of sorghum varieties with increased resilience to abiotic and biotic stressors. Improving the productivity, resilience, and quality of cereal crops is a major leverage point for development in WA because of the potential for impacts in regional trade, rural food security, and the health of women and children. As the starting point for a major agriculture value chain, enhanced sorghum varieties with greater yields, and improved yield stability, can support agricultural and economic development at regional scale.

3. Introduction and Objectives

3.1 The role of molecular breeding in the genetic enhancement of sorghum

A primary goal for crop improvement programs in West Africa is to stabilize and increase yields in the context of biotic and abiotic stressors (Waddington et al. 2008). At that the same time, breeding programs must maintain, or preferably enhance, locally-preferred processing and end-use traits so that improved varieties will be adopted. Unfortunately, accurate phenotyping of yield, adaptive traits, and grain quality is challenging and time-consuming, so only a small fraction of available germplasm is evaluated for potential useful traits, and few useful alleles can be bred into locally-preferred varieties. For instance, even though drought is a common stressor, on any given year a drought may be too severe, or not severe enough, for effective phenotypic selection by breeding programs. This principle is true for many stress resistance traits. While marker-assisted approaches do not circumvent the challenges of phenotyping in variable and marginal production environments, they do allow us to capture more value from investments in phenotyping, by distilling phenotyping efforts into trait-associated markers.

Genomics-based approaches to crop improvement are accelerating genetic gain in breeding programs for major crops and have the potential to do so for orphan crops (Heffner et al. 2009; Varshney et al. 2012). To facilitate genomics-enabled breeding for Sub-Saharan Africa, Genotyping-By-Sequencing (GBS) has been developed as a lower cost high-throughput approach for genome-wide SNP discovery and genotyping (Elshire et al. 2011). In sorghum, it has recently been demonstrated that GBS is an effective tool for mapping agroclimatic and grain quality traits (Morris et al. 2013; 2013b). By directly integrating genomics-enabled breeding into existing conventional and participatory breeding efforts, GBS could accelerate the genetic enhancement of locally-improved sorghum varieties. Some previous efforts to bring global elite germplasm to smallholder farmers saw limited adoption of released materials (McGuire et al. 2008; Curran and Cook 2009). The combination of multi-parent population development and marker-assisted recurrent selection can generate new varieties that are more likely to be adopted, by capturing transgressive segregation of introgressing alleles for yield and stress tolerance into locally-preferred genetic background.

3.2. Sorghum improvement in Senegal: Context and breeding targets

Sorghum is cultivated in Senegal for human and animal consumption. Both require good grain quality. Recently varieties without a pigmented testa that meet these requirements have been released. Most of the sorghum acreage in Senegal is cultivated in the Soudano-Sahelian and Soudanian zones (82%). Varieties released in these areas mature in 90-105 days after planting. In these conditions sorghum is subjected to periodic rainfall during flowering and grain development and would develop grain mold. One of the most damaging aspects of this disease is the loss of germination of infected kernels. Grain mold can be controlled by growing high-tannin (pigmented testa), colored-grain cultivars that resist mold infection. However, such grain is not generally used for food and there is a need to identifying and utilizing mold resistance traits that are compatible with local preferences.

Many species of fungi are involved with the deterioration of sorghum grain. The most important and commonly reported are *Fusarium thapsinum* and *Curvularia lunata*. Other have separated the damage caused by these saprophytic fungi and other microorganisms such as those associated with grain weathering (Frederiksen and Odvody, 2000). However, there are many other fungi that colonize sorghum grain after it matures. These organisms are numerous saprophytes and under warm humid conditions, digest sorghum

grain as the crop matures in the field. Consequently, grain mold in food-quality grain production can be a major economic problem (Forbes et al. 1989). Developing higher levels of resistance to grain mold remains one of the great challenges in sorghum improvement.

The development of elite sorghum varieties with improved resistance to grain mold has been impeded by the complexity of the trait. Grain mold resistance involves multiple mechanisms including unfavorable digestibility seed traits (pigmented testa, red pericarp color), other seed and panicle characteristics (kernel hardness, pericarp thinness, corneous endosperm, increased glume coverage, open panicle structure (Thakur et al. 2006; Menkir et al. 1996; Esele et al. 1993; Bandyopadhyay et al. 2000; Katile, 2007; Esele et al. 1993; Sharma et al. 2010; Audilakshmi et al. 1999) as well as constitutive and inducible physiological mechanism such as the accumulation of antifungal proteins (β-1,3-glucanases, chitinases, sormatins, ribosome-inhibiting protein (RIPs), and permatins (Kumari et al. 1994; Krishnaveni et al. 1999; Rodriguez-Herrera et al. 1999). This deserves to be clearly addressed with adequate phenotyping methods and molecular tools. Understanding the molecular basis of mold resistance and grain quality will help developing high yielding varieties, adapted to the agro-ecological areas of Central and Eastern Senegal with improved mold tolerance and digestibility.

3.3 Sorghum improvement in Niger: Context and breeding targets

Niger is a landlocked country in the West African semi-arid tropics, with 2/3 of its area being desert. The climate is typically Sahelian, with two seasons: a long dry season of eight months and a short rainy season of four months which usually starts in May or June. The rainfall is low, variable and undependable. The cropping area is limited to the region that has a length of growing period (LGP) of 75 to 150 days, which is classified as semi-arid (CEEPA, 2006). Most rainfall occurs for the three months of July, August, and September, which account for 90% of total annual precipitation. Annual precipitation in this area is about 150-900 mm (CEEPA, 2006). Crops are grown between June and October. Rains are most often irregular, badly distributed and insufficient to ensure satisfactory levels of food crops production (CEEPA, 2006). Most soils have sandy texture, so water holding capacity is also generally low.

In Niger agriculture is practiced only in the southern part and is essentially rainfed characterized by harsh environmental conditions – high temperature, low rainfall and low soil fertility caused by increased climate degradation and demographic pressure (CEEPA 2006). The majority of the population is composed of farmers and their survival is dependant only upon subsistence agriculture. Most of the cereal produced is for household consumption and is grown under rainfed conditions. Agricultural activities determine the household income and nutritional conditions, which are below the FAO minimum standards. Sorghum is the second staple food after pearl millet and 85% of the crop is produced by subsistence farmers, who often use local landraces that provide low, but stable yields under marginal conditions. Yields are low, however, with national average of 500 kg/ha, and a range of 150 kg/ha to 4000 kg/ha depending on the growing conditions and use of inputs (Adamou et al. 1985). More land depletion, low soil fertility, the low level of external input, and low investment in irrigated agriculture, drought and *Striga* are major constraints on sorghum production in Niger.

Despite the importance of sorghum as a food source, increased production through increased yield per unit land area in recent years has been rather modest at best, as abiotic and biotic stresses limit potential grain yield. However, stable, high-yielding sorghum varieties have been recently developed through breeding and improvement programs utilizing sorghum landrace varieties from Africa, India and China. This has

involved selecting traits such as photoperiod insensitivity, reduced height (to reduce lodging), drought tolerance, and pest and disease resistance (Reddy et al. 2006). Improved sorghum varieties adapted to smallholder farmers' conditions in West-Saharan Africa, have been developed (Obilana, 1998; Obilana et al. 1997) and sorghum yields have substantially increased with the introduction of hybrids, facilitated by the development of cytoplasmic-genetic male sterility (Axtell et al. 1999). Efforts to diversify parental lines could increase the rate of yield improvement.

Striga spp. (*Striga hermonthica* (Del.) Benth. and *S. asiatica* (L.) Kuntze) is an important parasite of sorghum, millets and other cereals in tropical Africa and Asia. This parasitic weed is a major biotic constraint to sorghum production, especially in the infertile semi-arid areas of Africa. According to Kim et al. (1998), two out of three fields cropped in cereals are estimated to be infested by *Striga* spp. in 17 sub-Saharan African countries. Yield losses from damage by *Striga* are often very significant. In West Africa, *Striga* infests over 64% (17 million ha) of the land planted to cereals, resulting in significant yield losses that range from 10 to 100% depending on crop and cultivar (Doggett, 1988, Obilana and Ramaiah, 1992, Gressel et al. 2004). Depending on the species and the environmental conditions for plant development, one *Striga* plant may produce 40,000 to 90,000 seeds (Ejeta et al. 1997) that can remain viable for as long as 20 years (Doggett, 1988). Yield losses of sorghum due to *Striga* infestation, coupled with poor soil fertility, low rainfall, and lack of production inputs, all contribute to survival difficulties for subsistence farmers. The control of *Striga* in cereals has been a major challenge to peasant farmers because of its adaptation to its environment and the complexity of the host-parasite relationship (Ejeta 2007). Use of resistant crop cultivars is the only practicable and economically feasible *Striga* control measure (Ekeleme et al. 2011). While breeding for resistance may offer future solutions to *Striga*, currently, only SRN 39 has a good level of resistance to *S. hermonthica* in sorghum accessions in Niger. What the subsistence farmer needs is a cheap option, easy to manage, durable and economically feasible.

3.4 Project objectives

The proposed project consist of six specific objectives, with the overall workflows outlined in Figure 1.

3.4.1 Specific Objectives

The project activities are organized into five specific objectives:

Obj. 1: Genomic characterization of Senegalese and Nigerien landraces and breeding lines to connect West African germplasm to global sorghum breeding efforts.

Obj. 2: Development of simplified genomics toolkit to provide optimized marker sets for marker-assisted breeding targets in West Africa.

Obj. 3: Development of multi-parent populations to combine locally-preferred varieties and elite global lines for more efficient trait mapping and breeding.

Obj. 4: Higher-resolution mapping of key resistance traits to produce more effective trait-associated markers.

Obj. 5: Implementation of Marker Assisted Recurrent Selection to develop more resilient locally-preferred varieties.

Obj. 6: A training and support network that will allow the national programs to take advantage of, and contribute to, new technologies and knowledge for crop improvement.

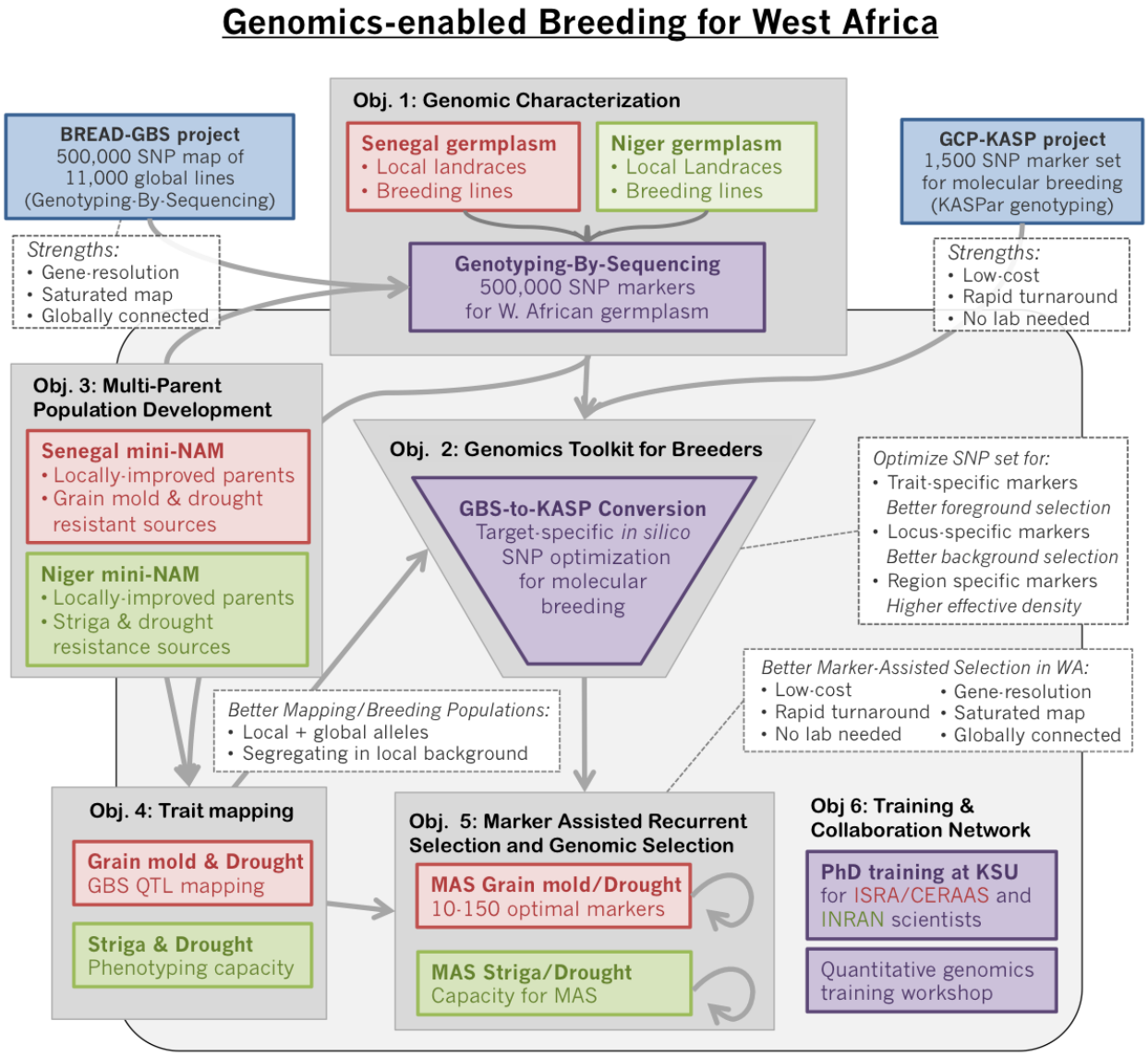

Figure 1: Overview of a genomics-enabled breeding platform for West Africa. Blue boxes summarize previous projects to advance SNP marker platforms that form the basis of the current proposal. Purple boxes denote general resource and capacity development, while red and green boxes denote specific activities for Senegal and Niger, respectively. Dashed boxes summarize rationales for the given objectives.

4. Testable Hypothesis

4.1. Core hypotheses

The core hypotheses underlying the proposed work are:

(1) The allelic variation required for transformative gains in yield and stress resistance traits is already available in regional and global germplasm (e.g. Vaksmann et al. 2008);
(2) The key limit to genetic gains is the excessive time and effort required to:
(i) identify beneficial alleles and the germplasm that harbors them,
(ii) and incorporate beneficial alleles into preferred varieties without disrupting other desired traits.

To test these core hypotheses we will implement genomic approaches that accelerate identification and incorporation of beneficial alleles, and determine whether genetic gains are indeed accelerated as proposed.

4.2: Specific hypotheses

Based on these core underlying hypotheses, we also derive the following specific hypotheses on the implementation of genomics-enabled breeding:

(1) A major opportunity for accelerating genetic gain is by leveraging existing phenotyping efforts through the development of genotype-phenotype model (trait-associated markers), that leverage phenotype data across locations and growing seasons.
(2) Marker-assisted recurrent selection will allow more rapid and specific introgression of useful alleles into locally-improved varieties of West African programs.

We predict that marker-assisted recurrent selection (MARS) will accelerate improve genetic gains, especially for difficult-to-phenotype, environmentally-dependent traits as compared to observed gains (historical and concurrent) from conventional breeding (Varshney et al. 2012).

5. Research Strategy

5.1 Genomic characterization of West African germplasm (Objective 1)

5.1.1 Germplasm

US: In the US National Plant Germplasm System (NPGS) there are 5,108 accessions collected from West Africa which will form a reference data set on sorghum genomic variation in West Africa. A small proportion (382 of 5,108) of these accessions have been genotyped, either as part of global core collections (Morris et al. 2013a) or source-identified collections (Morris et al. in prep), but most have never been genotyped. Since material held at NPGS is readily available to the global plant breeding and genetics community through the Germplasm Resources Information Network, we will genotype these accessions to provide a collaborative platform for genomics-enabled breeding for West Africa. Once genotyped, breeders and geneticists will be able to use any subset of these 4800 accessions for genome-wide association mapping of traits, without any further molecular laboratory work needed. Together this data will form the basis of a

haplotype map for West African germplasm, a detailed catalogue of genetic variation shared among landraces and breeding lines which will facilitate germplasm utilization and allele mining.

Senegal: In addition to characterizing Senegalese germplasm available from GRIN, we will characterize several hundred Senegalese lines held by CERAAS/ISRA. We will characterize 500 breeding lines to determine which genomic regions contributed to the elite material, particularly the four varieties recently released (Nguinthe, Faourou, Darou and Nganda). In addition we will characterize 100 local landraces held by CERAAS/ISRA.

Niger: To build a foundation for genomics-enabled breeding in locally preferred varieties, we will genotype all the local landraces of the Nigerien 2003 collection (Deu et al. 2008). This will help to inform the design of breeding and mapping populations and link findings from Niger to worldwide studies on global sorghum germplasm. Two reference studies (Deu et al. 2008; Deu et al 2010) were published by CIRAD researchers and can be used to look for useful alleles in landraces. A full inventory of genetic material in the INRAN breeding program will be necessary to determine which lines will be used for genotyping, but it will include the best performing locally-improved varieties and the twenty (20) local varieties used during the McKnight Foundation project ["Farmer-participatory improvement of sorghum and pearl millet genetic resources for increased adaptation to diverse production environments in West Africa", Haussmann (ICRISAT) and Souley (INRAN)].

West Sahelian Association Panel:

Based on input from collaborators and stakeholders at the SMIL West Africa Inception and Planning Meeting we will assemble a West Sahelian Association Panel, as a community resource for West African sorghum improvement. This panel will include locally-improved varieties and landraces harboring useful characteristics for the Sahelian zone. Bassirou Sine (Ecophysiologist, CERAAS) and Sophie Bouchet (KSU) are coordinating the assembly of the panel. Co-PI Cisse and Bassirou Sine are providing the germplasm from Senegal. Co-PIs Mamadou and Abdou are providing germplasm from Niger. Clarisse Kondombo-Barro (Sorghum Breeder, INERA) is providing material from Burkina Faso and Niaba Teme (Sorghum Breeder, IER) is providing material from Mali. We will assemble a preliminarily panel of ~500 for evaluation in year one in order to build a final panel of at least 400 accessions with comparable maturity. CERAAS will increase seed so that the panel will be available as a reference germplasm set for phenotypic screening and GWAS QTL mapping in the Sahelian region.

5.1.2 Genotyping-by-Sequencing

We will use genotyping-by-sequencing (~500,000 SNPs) to characterize genomic variation (SNP diversity, population structure, and haplotype structure) in breeding material. DNA extractions will be carried out at CERAAS for the Senegalese material, and at the ICRISAT Sahelian Center (Sadoré, Niger) in collaboration with INRAN for the Nigerien material. GBS will be carried out at KSU with 384X sample multiplexing (cost ~$10 sample) using the ApeKI restriction enzyme for reduced representation, as done for 11,000 sorghum accessions previously. To build on the strengths identified in the Senegal and Niger consultation reports, we will also genotype the national germplasm collections described in 5.1.1. The GBS SNP maps for Nigerien and Senegalese landraces and breeding lines will be integrated with a published GBS SNP map for global sorghum diversity (Morris et al. 2013), a newly-developed GBS database for 2,000 source-identified African and Asian sorghum landraces (Morris et al. in preparation),

and GBS SNP maps for six ICRISAT biparental mapping population (Ramu et al. in preparation).

5.1.3 Population genomic analysis

Using the GBS SNP map for West African germplasm the PhD trainees and post-doc will carry out population genomic analysis to identify the genomic regions and accessions that harbor alleles that are adaptive for particular regions or environments. This will include analysis of population structure, haplotype structure, fixation index (Fst) genome scans, and environment-of-origin genome scans.

5.2 Genomics-to-breeding toolkit (Objective 2)

5.2.1 GBS-to-KASP Strategy

The development of rapid, cost-effective marker systems to enable sorghum molecular breeding has been an active area of research for over two decades. While marker technology has improved dramatically in terms of cost and easy of use as SNP-based assays have matured, the impact on breeding has been slower coming. Recently, a major effort was undertaken through the Generation Challenge Program (GCP) to make SNP marker technology practical for molecular breeding in developing countries. This project, developed as part of the Integrated Breeding Platform (IBP) has delivered a practical approach for molecular breeding using a KASP (Kompetitive Alelle Specific PCR) SNP genotyping service [https://www.integratedbreeding.net/snp-marker-conversion]. This approach, hereafter referred to as the GCP-KASP approach, has been validated on sorghum in several projects conducted in Mali in collaboration with IER sorghum breeding programs and led by Eva Weltzien (ICRISAT), Niaba Teme (IER), and Jean-Francois Rami (CIRAD). It has several key strengths: The low cost per marker and per line (about $0.1 per data-point); the rapid turnaround time from sample collection to line selection (4-6 weeks); no molecular biology laboratory is required since small unrefrigerated leaf samples to be sent to the service lab using a kit (desiccants contained in the kit dry the material in transit and nullify the requirements of phytosanitary certificates by local participants); and the quantity of data is easily manageable with even basic computational resources (e.g. a laptop).

However, the current implementation of GCP-KASP approach, with a single global set of 1,500 marker to cover all traits and all regional gene pools, has key limitations. While in principle 1,500 markers is sufficient to cover all recombination bins within a breeding program, in practice, a single global 1,500 marker set will perform poorly in a number of respects: For any given trait of interest, there is little chance that a perfectly-linked SNP will be assayed (e.g. a SNP in causative gene); for any given locus of interest, the marker density will be low, so efficacy of background selection will be limited; and, for any given regional breeding program, a large fraction of the global markers will be non-segregating, so uninformative. Importantly, the BREAD-GBS approach complements the GCP-KASP approach: The high-resolution GBS SNP maps are capable of identifying SNPs perfectly linked to causative polymorphisms for a trait; the high SNP density means that the map will effectively be saturated at almost any locus in the genome; and many segregating SNPs are available across the genome for any regional gene pool. However, the BREAD-GBS approach is much slower (months), the costs are higher ($10-20 per line), and high-quality molecular and computational resources are required (beyond those available most NARS). Therefore, we propose here to develop a workflow, and necessary bioinformatic tools, to combine the strengths of both approaches, with no additional cost over the GCP-KASP approach beyond the initial investments in West Africa targeted GBS (Objective 1).

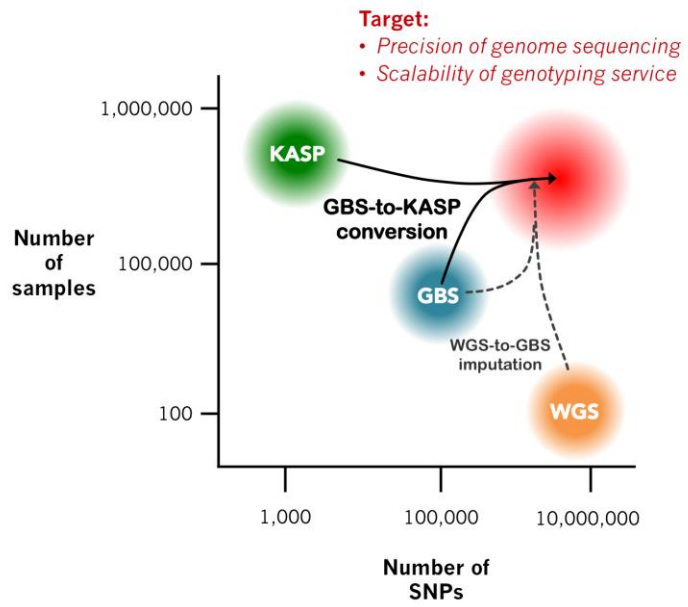

Figure 2: Genomics toolkit for molecular breeding in West Africa. By optimizing SNP marker sets for a particular breeding target, the data sets (Objective 1 & 4) and bioinformatic tools (Objective 2) we develop will provide the precision of genome sequencing (resolution, function) with the scalability of genotyping service (low cost, rapid turnaround, and ease of use). The GBS-to-KASP conversion will allow us to take advantage of parallel work by ICRISAT (S. Deshpande, T. Shah), DAAFQ (E. Mace) and KSU (Morris), to impute functional polymorphisms from whole-genome sequence (WGS-to-GBS imputation).

5.2.2 GBS-to-KASP Implementation

Given that an optimized set of 1,500 markers will be sufficient for foreground and background selection, we will develop a bioinformatics pipeline that identifies (from >500,000 GBS SNP markers) a targeted set of 1,500 markers for the KASP assay (trait, locus, and region specific). Addressing each of the GCP-KASP limitations identified previously, the marker inclusion criteria will be based on (1) high SNP-trait associations from GWAS and linkage mapping; (2) high-density given locus- and gene pool-specific recombination rates and linkage disequilibrium patterns (Morris et al. 2013a; Ramu et al. in prep); and (3) high coverage given regional population structure and haplotype distributions. Importantly, in the KASP system (unlike some chip-based SNP assays, for instance) there is essentially no incremental set-up cost associated with using a novel 1,500 SNP marker set, so each regional breeding program can be using a fully-optimized SNP set for each trait or trait combination under selection. We will implement the GBS-to-KASP conversion in open source software (R code) and validate it in the following proposed molecular breeding activities (Objective 5). Once validated, we will make the tools and data available to the molecular breeding community through the Integrated Breeding Platform (https://www.integratedbreeding.net).

5.3 Multi-parent population development (Objective 3)

5.3.1 New NAM populations for Senegal and Niger

<u>Senegal:</u>

A mini-NAM cross design will be used from existing populations already developed between Nganda, an elite variety recently released by ISRA, and 3 parental lines: two known sources of mold resistance (Sureno and Macia) and one source of grain quality and early flowering (CSM 63E). Populations are currently available as F3 families with respectively 627, 385 and 422 individuals.

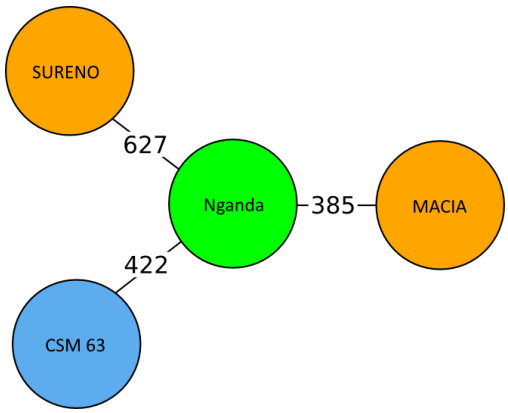

Figure 3: A mini-NAM for marker assisted breeding in Senegal. Populations recently developed by ISRA and available as F4 families. Figures represent the number of progenies available for each cross.

These 3 populations will be used to conduct a multi-parental MARS (see 5.5.1). For this, F4 seeds will be produced on single seed descent derived F3 plants during 2014 cropping season. F3 seeds will be treated to avoid negative mold effect on seedling and consecutive potential bias in the populations. During off-season (sowing December 2014), F4 rows of at least 10 plants will be grown for the three populations and selfed. The harvested seeds will be bulked and used for further phenotyping during 2015 cropping season. At the same time, the whole population will be advanced up to F7 generation to provide recombinant inbred lines.

5.3.2 New NAM populations for Niger

Niger: Make a multi-parent crossed population mostly of local varieties (introduce *Striga* and drought resistant/tolerant lines to cross with local varieties). The Single Seed Descent method will be used to develop varieties resistant/tolerant to drought and *Striga*). Marker-assisted backcross can be used to introgress useful alleles from global sorghum germplasm into local varieties. Some varieties such as N'Gabiri kime, MDK, Bagoba and L28 are supposed to harbor stay-green genes. From the material we have from GCP (A. Borrell), we can add these to the local ones to screen and cross those which have the stay-green gene and proceed to produce a dual purpose varieties.

In Niger we will develop a small NAM using Mace De Kunya (MDK) as a common parent. Malick Ba (ICRISAT-Niamey) and a WACCI PhD student are generating RIL family (currently at F3 generation) between MDK and ICSV88-032, a source of midge resistance (Tao et al. 2003). To complement this midge resistance RIL family, we will generate two more RIL families with MDK as a common parent. The first will use SRN39 as source of Striga resistance. Field inheritance studies have demonstrated SRN39 is an effective source of Striga resistance in Niger (Hess and Ejeta 1992). The second will use L153-5, an early, moderate height, white/tan/non-tannin caudatum, to provide a source of early season drought tolerance.

Figure 4: Phenotyping capabilities at CERAAS for grain mold and drought. Shown are inoculated tests for grain mold susceptibility, demonstrating multiple levels of infection, and drought trials with irrigated and none irrigated treatments.

5.4. Trait mapping for abiotic and biotic stress tolerance (Objective 4)

5.4.1 Phenotyping and trait mapping strategy

Identification of novel sources of mold resistance and grain quality in a panel of diverse landraces and improved germplasm from Senegal, Niger, Mali, and Burkina Faso.

A collection of breeding and landrace-derived material from Niger and Senegal will be constituted (*Objective 1*). From this collection a set of genotypes showing suitable grain quality and yield traits will be defined and phenotyped in trials in Senegal for traits related to yield, grain mold (Senegal), drought. In Senegal, phenotyping experimental will be performed in Bambey (Central). In Niger, phenotyping experimental will be performed in Konni, Madaoua and Maradi.

The mini-NAM populations developed for trait mapping in Senegal will be genotyped using GBS facility developed by KSU. A total of 1100 individuals (370 per population) will be genotyped together with the parental lines using ApeK1 384-plex GBS protocol.

5.4.2 Drought adaptation

Terminal drought: In the Sahelian zone drought stress iscommon on both sandy and heavier soils. Drought screening is possible under field condition during the offseason with early October sowing (for photoperiod sensitive materials) and possibly in January for insensitive materials. Drip irrigation capacity available, but system for screening has not been developed for routine observations. Drought screening is also possible in offseason at CNRA-Bambey, where an irrigation system with oscillating ramp is available for providing irrigation supply. It is also possible in rainy season by delaying the sowing dates in August notwithstanding the photoperiod sensitivity of genotypes. This will allow multi-site testing. In addition, recessional farming provides a large testing area through the Senegal valley. Equipment exists at CERAAS-Thies to measure soil water stock and climatic parameters to the field. Biophysical equipment is also available to observe physiological traits. In case where the number of genotypes is higher than twenty, the most observed traits are the leaf temperature, the fluorescence, the LAI and the SPAD, obtained easily and faster. Yield stability and some of agro-morphological and phonological traits are used as drought adaptation criterions. Drought

stress index is also calculated from some of these traits. Pot experiments are common at CERAAS-Thies. They allow to apply controlled stress and to have access to the root system. Moreover a rainout shelter is being built at CNRA-Bambey and will increase our phenotyping capacity.

Early drought: In Sahelian zone drought periods also occur frequently during the early stage at the starting of the rainy season due to the irregularity of rain. This is increased by the dry sowing practice of some of the farmers. The early drought could cause lethal injury or depressed yield. Experiments to assess early drought are sown at early October during the offseason. Pot trials are also suitable. The early vigor is one of the major traits assessed.

5.4.3 Grain Mold

For each locality, climatic conditions such as temperature and relative humidity will be recorded daily. The collection will be evaluated for days to flowering, plant height, panicle length, peduncle length, kernel weight per panicle, thousand-kernel weight, number of kernel per panicle, panicle compactness, seed color, glume length, endosperm texture. In Senegal, grain mold will be rated at seed physiological maturity using the '1 to 5' scale (Thakur et al 2006) where 1= percentage of molded grains on a panicle < 1% and 5 = percentage of molded grains on a panicle > 50%.

For grain nutritional quality, the total protein and sugar contain as well as Fe and Zn contain will be assessed. For these specific traits a clear linkage will be made with IER Mali and ICRISAT who have developed strong phenotyping capacities for these traits.

5.4.5 Genome-wide Association Studies

With breeding materials and other diverse germplasm that is being screened for key traits we will map the loci underlying the traits using genome-wide association studies (GWAS). The PhD-trainee from INRAN and CERAAS/ISRA will work with the post-doc at KSU to identify genetic markers that are associated with grain mold, *Striga* and drought resistance. We will use genome-wide association analysis to identify trait-associated SNP markers in diverse materials and in multi-parent populations, implemented in the open-source R software GAPIT (Lipka et al. 2013). In the multi-parent populations we will also conduct joint-linkage analysis (Li et al. 2011).

5.4.5 Phenotyping in Niger

In Niger, our focus will be to improve phenotyping capacity for abiotic and biotic stress tolerance, laying the groundwork for future trait mapping efforts using the populations developed under Objective 3. Using support from SMIL, we will phenotype our breeding material for high and stable yield, terminal drought resistance, and *Striga* resistance and tolerance. Our goals are to develop Tillabéri site for drought phenotyping and the Konni site for *Striga* phenotyping.

Tillabéri drought phenotyping: Drought is not predictable, so for drought resistance strong irrigation system is needed at Tillabéri where the trial will be conducted at the INRAN station. The INRAN research farm at Tillabéri is a 26 ha site located on the bank of the Niger river, 115 km northwest of Niamey. For accurate drought tolerance phenotyping it is important to be able to apply an appropriate level of water limitation and also have well-watered control treatment (Vadez et al. 2011), so we will first focus on strengthening this capability for the sorghum breeding program. Currently at Tillabéri there is flood (furrow) irrigation

available on several hectares, using an electric pump to access river water. Based on recent installations the research farm manager estimates that it costs $4,000 per hectare to add irrigated acreage from the existing irrigation infrastructure. Therefore, for the current project we add 2 ha of irrigated plots for controlled water-limited and well-watered treatments. To refine and validate the phenotyping methods at the Tillabéri site we will concentrate our efforts on a small (n = 20) highly-replicated (10X) panel of known drought tolerant and drought susceptible lines for at least the first two growing seasons. Once it is clear based on the positive and negative control lines that accurate drought tolerance phenotypes can be obtained, our focus will then shift to obtaining phenotypes for trait mapping in the West Sahelian Association Panel, most likely in year 3 or 4. At that point we will be able to compare drought phenotyping and mapping results from Niger with those from Senegal.

Konni Striga phenotyping: We will grow a small panel (n = 20) of control lines (*Striga* resistant and susceptible) in a *Striga* infested field. During the first year, some samples of *Striga* inoculum will be collected in the infested regions surrounding Konni. In the second year, varieties will be submitted to artificial *Striga* infestation. Seed of varieties will be sown at the time with *Striga* inoculum in the same hill into two to three replications prior to evaluation. The number of *Striga* plant in each hill will be counted sixty days and ninety days after sowing. Varieties resistant to drought and *Striga* will be used as control.

5.5 Marker-Assisted Recurrent Selection and Genomic Selection (Objective 5)

5.5.1 Implementing Marker-Assisted Breeding

Senegal: Multi-parental MARS design

One of two marker-assisted breeding strategies will be implemented, depending on the number of QTL detected and their related effects especially for mold resistance QTL.

In case of few large effect QTLs for mold resistance and limited number of favourable alleles for other traits from the donor parents, a MABC strategy will be implemented in order to improve marginally the elite parental line Ndanga for mold resistance. We will use marker-assisted selection and genomic background to precisely introgress the most promising QTL-alleles into locally-improved varieties. Other locally-improved elite varieties will be also considered as potential recipient lines. Genotyping of the BC1 plants for foreground and background will be performed using selected SNP on carrier and non-carrier chromosomes. The best BC1 plants will then be crossed again with their recurrent parents. BC2 plants will be genotyped again for foreground and background selection and the best ones will be selfed to deriving lines carrying large effect mold resistance QTL in a homogeneous recurrent parent genetic background. In parallel, for all traits, a MARS strategy will be used to cumulating most of the detected QTLs with favourable effects.

Alternatively, in case of a more complex genetic determinism of mold resistance and/or if we detect favourable QTLs for other traits contributed by the donor parents, we will implement a multi-parental MARS approach in order to cumulate favourable alleles from the 4 parents toward completely redesigned ideotypes. From the QTL results, a target genotype will be constructed. The best 18 F4 families accross the three populations will be selected based on QTL results and sown during 2016 cropping season with approximately 40 plants per family (total 700 plants). These plants will be genotyped on key QTLs using KASP assays (50 loci anticipated) and the best combinations will be determined to conduct intercrossing

of progenies. Depending on the number of QTLs to cumulate, up to three cycles will be conducted following the same approach. At each cycles, the best crosses based on a genotypic index constructed from one or more target genotypic ideotype will be identified from genotyping data of individual lines. In addition, use of genomic estimated breeding values as established from Objective 1 will be explored as a complement to QTL based models to predict best crossing combinations.

5.5.2 Evaluating genomic selection

Using genotype (*Objective 1*) and phenotype (*Objective 4*) data already collected, we will estimate allelic effects for at ~500,000 SNP markers and use to predict genomic estimated breeding values (GEBVs) for genomic selection. Building from phenotyping efforts from the previous objectives, we will estimate GEBVs for biotic stress tolerance (*Striga*, grain mold), abiotic stress tolerance (drought), and yield. In the current proposal, we will use GEBVs to guide selection, but to test *in silico* through cross-validation whether genomic selection approaches are likely to be effective in the context of West African breeding programs.

6. Theory of Change Statement

Strengthening the network for West African sorghum breeding and genetics:

We feel that a sustained and concerted effort is required to achieve the goal of accelerating breeding in Africa, so have made every effort to build directly on recent collaborative projects.

The methods and capacity developed for sorghum in the current proposal will transfer to millet in next ~2 years as genomic resources in millet become available (reference genome sequence and genotyping-by-sequencing data). In particular, this will be facilitated by interaction with Tom Hash, the lead pearl millet breeder for ICRISAT in West Africa and member of the Pearl Millet Genome Sequencing Consortium, who will be well-positioned integrate successful approaches from the proposed work into the pearl millet breeding programs.

7. Gender Issue Planning

Since sorghum is primarily grown as a food crop in Sub-Saharan Africa, and is often thought of as a woman's crop, the genetic enhancement of sorghum may have many direct impacts on women and indirect impacts on smallholder families. In a report prepared for the Bill & Melinda Gates Foundation, Curran and Cook (2009) conclude that women farmers stand to be the primary beneficiaries of increased sorghum yields if threats from *Striga*, pests, and drought are alleviated. In some cases, women's sorghum plots in West Africa are especially vulnerable to environmental stressors because of gender dynamics of land use. For instance, in some parts of West Africa women farmers are more likely to have flood-prone plots, and therefore place more emphasis of flood tolerance in participatory breeding activities (Haussman et al. 2012).

Some mechanisms to increase crop yield or resistance may have unfavorable consequences for agronomic traits and post-harvest processing traits. For instance, increasing grain hardness and glume coverage reduce grain mold, but may make processing more difficult. Since women are primarily responsible for crop maintenance and post-harvest processing, we will favor resistance mechanisms that do not increase the time and effort required of women farmers for maintenance and processing.

We feel that genomics-enabled breeding approaches have the potential to enhance gender inclusion by facilitating participatory breeding programs. For instance, following participatory selection efforts with women smallholder farmers, the preferred varieties can be rapidly screened for key resistance traits with trait-associated markers. ICRISAT-Niger has implemented such an integrated program that combines conventional, participatory, and genomic approaches, and uses each approach for the traits where they are most effective.

8. Human and Institutional Capacity Development Strategy

8.1. Nature of collaboration among institutions

The proposed project is a collaborative effort to implement genomic-enabled breeding in West Africa, with the national programs bringing germplasm and phenotyping resources, the KSU team bringing genomics and bioinformatics resources, the ICRISAT and CIRAD collaborators providing a regional scope and experience in successful integration of marker-based technologies in national breeding programs in Senegal and Mali.

In the Niger Consultation Report, the participants identified computational hardware ("Matériels informatiques") and molecular lab resources ("Laboratoire de biologie moléculaire") as major weaknesses in the current research infrastructure. At ISRA/CERAAS, where greater lab and phenotyping resources exists, our focus will be to develop bioinformatics and data analyses capacities and to supplement and enhance the utilization of existing capacity through increased training and collaborative experience. Since capacity and international collaborations are most developed at ISRA/CERAAS, they will contribute additional linkages with Niger, Mali, and Burkina Faso. CERAAS has capacity in phenotyping for drought resistance and will assist Niger in evaluating its population. Genotyping capacities have also been developed at CERAAS and serve as training facility for the region.

8.2 Long-term training activities (Objective 6):

The major long-term training goal will be PhD training (4 years) in genomics-enabled breeding at KSU for two crop scientists that are currently affiliated with the host institutions. The PhD trainees from Senegal and Niger have been identified by the host institutions in consultation with the KSU team, and are well prepared for genomics-enabled breeding training at KSU. The PhD trainee from Senegal will be Jacques Martin Faye, a crop geneticist currently finishing a Master's degree in Plant Biotechnology at University Cheickh Anta Diop of Dakar and working at CERAAS with co-PIs Cisse and Fonceka. The PhD trainee from Niger will be Fanna Maïna, a crop geneticist with a Master's degree in "Biodiversity and Management of the Soudanian and Sahelo-Saharian Environment" from University of Maradi, who is currently employed as a researcher at INRAN-Niamey.

The PhD trainees will be advised by the PI Morris (sorghum genetics and genomics) and co-advised by KSU sorghum breeders Tesfaye Tesso and Ramasamy Perumal. The resulting increased human capacity in molecular biology and bioinformatics will facilitate future transfer of technologies and knowledge between breeding programs in West Africa and those in the Americas, Europe, and Australia. To help the PhD trainees build their professional network, each year they will attend and present at an international crop genetics meeting, either Plant and Animal Genome or Crop Science Society of America. At the same, the long-term visits of the PhD trainees will be an opportunity for them to educate the KSU collaborators, and

the U.S. breeding and genetics community more broadly, on the context for crop improvement in Niger and Senegal. For instance, while in the U.S., the PhD trainees will present webcast presentation on crop improvement in the host countries through the *Center for Sorghum Improvement* seminar series.

8.3 Short-term training activities (Objective 6):

In the third year of the project, co-PI Morris, Rami, and Fonceka will lead a training workshop in at CERAAS for plant breeders and geneticists focused on experimental design for genomics-enabled breeding and open-source tools for quantitative genomics (population genomics, GWAS, and GS). The training course will be based on a WAAPP-sponsored course held in 2012 by CERAAS and CIRAD, co-led by co-PIs Fonceka and Rami, on molecular markers for crop diversity and breeding. We will emphasize effective use of available GBS resources and GBS-to-KASP conversion so that programs without molecular biology capacity can benefit. We will coordinate our quantitative genomics course with the functional genomics course that will be led by SMIL PI Mitch Tuinstra,

Co-PIs Weltzien and Mamadou will coordinate *Striga* phenotyping training at ICRISAT-Samanko (Mali) for a technician based at INRAN-Maradi who will contribute to phenotyping at Konni.

8.4 Institutional capacity building:

Niger: In the *Niger Consultation Report*, the participants identified computational hardware ("Matériels informatiques") and molecular lab resources ("Laboratoire de biologie moléculaire") as major weaknesses in the current research infrastructure. We will purchase computer hardware to address computational capacity and strengthen connections to ICRISAT Sahelian Center and global genotyping service laboratories to build capacity to use molecular tools. Other capacity building focuses on the breeding materials and phenotyping. Co-PI Mamadou has identified cold storage for breeding materials as a key need so we will provide support to refurbish an existing non-functional unit. We will also build phenotyping capacity through the installation of additional irrigation systems, and the purchase of vehicle to support phenotyping at multiple sites.

Senegal: At ISRA/CERAAS, where laboratory and phenotyping resources are relatively well developed, our focus will be to develop bioinformatics and data analyses capacities and to supplement and enhance the utilization of existing capacity through increased training and collaborative experience. The purchase of vehicle will facilitate travel from Thies (laboratories/offices) to Bambey (field site).

9. Communication Planning

To build working relationships among project participants and finalize implementation details, we will carry out a project meeting at CERAAS in late April or early May prior to the cropping season. In addition, the KSU team will organize quarterly conference calls among project participants to review progress and address contingencies.

For archiving and distribution of raw data from genotyping-by-sequencing (Objective 1), we will deposit Illumina sequencing reads to the NCBI Short Read Archive (http://www.ncbi.nlm.nih.gov/sra). Since the raw data requires computationally-intensive processing prior to use, we will also make ready-to-use genotype data sets available in standard formats through our CMS and the Dryad Digital Repository

(http://datadryad.org).

A content-management system (CMS) website to facilitate internal communication (sharing of methods and data), and open access to results. As development of the Integrated Breeding Platform progresses, we will migrate the resources we produce to that platform to facilitate broader access and integration with other resources.

To ensure the broad accessibility of our findings, we will publish all manuscripts arising from the proposed work in open access journals, or using the open access options of traditional journals. (Accordingly, additional funds have been budgeted for publication costs.)

10. Conflict of Interest Statement

This project does not duplicate activities from any current projects. No part of this has been or will be considered for funding form other donors.

11. Detailed Budget, Budget Justification, and Contingency Planning

11A. Detailed Budget

Attached

11.2 Budget Justification

11.2.1 KSU Primary award (Lead Institution)

Personnel $49,770:

We request funds of $49,770 ($37,142 salary and $12,628 fringe benefits) to support one month summer salary for the primary investigator in years 1 - 4. The PI will oversee the project and coordinate all work efforts of the postdoc and graduate students. PI requested salary is $8,878 in year 1, $9,144 in year 2, and $9,419 in year 3, and $9,701 in year 4. Fringe benefits are calculated at 34%.

Travel: $73,340:

We request funds for once-per-year travel between KSU and West Africa for PI Morris to facilitate collaborative project management and the sharing of findings, and to carry-out short term training efforts. Additionally, we request funds for twice-per-year travel between KSU and West Africa for two PhD students, to allow graduate training at KSU and field-based research in WA. Travel at approximately $2,594 per person per trip includes airfare, per diem, lodging and miscellaneous travel to include but not limited to ground transportation, baggage fees, tolls.

Funds are requested for student travel to the winter nurseries in Puerto Rico and Mexico in years 3 and 4. One student per nursery at two trips per year for 13 days each. Expenses for Puerto Rice include but not limited to airfare $609 at per trip, lodging at $100 per night and per diem of $46 per day. Expenses for Mexico include but not limited to airfare $659 at per trip, lodging at $100 per night and per diem of $69 per day.

Supplies $98,100:

We request funds of $98,100 ($35,000 in year 1, $25,000 per year in years 2 and 3, and $13,100 in year 4) for supplies to include but not limited to shipping supplies, genotyping and molecular biology supplies, and computers and computer supplies.

Training $206,437:

Funds of $206,437 ($24,672 per year per grad student in years 1 – 4) are requested to support two graduate students to carry out trait mapping and molecular breeding activities. A 3% annual increase is included in years 2 – 4.

Other direct costs $20,000:

Publication costs: Funds of $5,000 per year in years 3 and 4 is requested for publication charges.

Winter nursery: Funds of $5,000 per year in years 3 and 4 is requested for winter nursery fees.

Indirect Costs $116,388:

Indirect costs are calculated at the university approved rate of 26%. Indirect costs are calculated on KSU direct costs only. Collaborative Institutions (INRAN, CERAAS, ICRISAT and CIRAD) are calculated as pass through funds.

Contractual $526,058:

Pass through funds are requested for collaborative institutions agreements to INRAN ($199,640), CERAAS ($195,054), ICRISAT ($37,197) and CIRAD ($94,167), as detailed below.

11.2.2 CERAAS sub-award:

Personnel Costs: ($73,612)

Funds of $15,000 ($3,750 per year) are requested for a staff scientist to assist in the scientific objects of the project as needed. Funds of $6,000 ($1,500 per year) is requested for specialized technicians to carry out population development and phenotyping. Funds of $22,612 ($13,153 per year) is requested for additional labor to support phenotyping efforts in Years 1-4.

Travel: ($41,000)

To support domestic travel required for population development, phenotyping activities, and sample collection, we request $31,000 for years 1-3 for per diem, lodging, communications and vehicle gas/oil.

Funds of $10,000 ($2,500 per year) are requested to support international travel including but not limited to airfare, lodging and per diem.

Equipment: ($25,000)

Funds are requested to purchase a vehicle to allow for multi-site phenotyping.

Supplies: ($30,000)

Funds of $15,000 ($3,750 per year) for field supplies to include but not limited to fertilizer, pesticides and paper bags.

Funds of $15,000 ($3,750 per year) are requested for genotyping costs necessary for molecular breeding activities.

Indirect costs ($25,442) are calculated at 15%.

11.2.3 INRAN sub-award:

Personnel Costs: ($56,600)
Salary: Funds of $6,000 (1,500 per year) is requested for specialized technicians to carry out population development and phenotyping. Funds of $50,600 is requested for temporary labor to support phenotyping efforts in Years 1-4.

Travel: ($41,000)
To support domestic travel required for population development, phenotyping activities, and sample collection, we request $31,000 for years 1-4 for per diem, lodging, communications and vehicle gas/oil.

Funds of $10,000 ($2,500 per year) are requested to support international travel including but not limited to airfare, lodging and per diem.

Equipment: ($38,000)
To safeguard breeding material developed in the Nigerienne activities, we request $5,000 in Year 1 to repair cold storage equipment that is existing but not currently functional.

To build capacity for drought-tolerance phenotyping, we request $8,000 for irrigation systems Year 1.

Funds of $25,000 are requested in year 1 to purchase a vehicle to allow for multi-site phenotyping.

Supplies: ($38,000)
To build capacity for bioinformatics and general computation we request $8,000 in Year 1 for computers (5) and computer accessories (printer, scanner, projector, and an uninterruptible power supply).

Funds of $15,000 ($3,750 per year) for field supplies to include but not limited to fertilizer, pesticides and paper bags.

Funds of $15,000 ($5,000 per year for years 1 - 3) are requested for genotyping costs.

Indirect costs ($26,040) are calculated at 15%.

11.2.4 ICRISAT sub-award:

Personnel Costs: ($15,845)

Funds of $15,845 are requested for salary for Dr. Eva Weltzien ($3,787 in year 1, $3,901 in year 2, $4,018 in year 3 and $4,139 in year 4. Salaries include an annual increase of 3%). Eva Weltzien will contribute breeding and source material from the regional program, contribute to training activities, eventually linkage to the seed project, to facilitate also targeting of priority traits for future variety development. She is expected to contribute to the expansion of the project to Mali.

Supplies: ($3,000)

Funds are requested for supplies to complete DNA sample preparation ($1,000 per year in years 1 - 3).

Travel: ($13,500)

Funds of $9,000 ($2,250 per year in years 1-4) are requested to support domestic travel required for population development, phenotyping activities, and sample collection, to include but not limited to per diem and lodging.

Funds of $4,500 ($1,125 per year in years 1-4) are requested to support international travel including but not limited to airfare, lodging and per diem.

Indirect costs ($4,852) are calculated at 15%.

11.2.5 CIRAD sub-award:

Personnel Costs: ($53,884)

Funds of $53,884 are requested for salary for Drs. Fonceka ($6,440 in year 1, $6,633 in year 2, $6,832 in year 3, and $7,037 in year 4. Salaries include an annual increase of 3%) and Rami ($6,440 in year 1, $6,633 in year 2, $6,832 in year 3, and $7,037 in year 4. Salaries include an annual increase of 3%)

Personnel costs are requested to cover 8% full-time equivalent of Dr. Fonceka and 5% full-time equivalent of Dr. Rami. Dr. Fonceka will be involved in coordinating CERAAS activities in Senegal especially population development, QTL mapping and marker assisted selection. Dr Rami will be involved in coordinating CIRAD activities in Senegal especially designing and implementing marker-assisted selection.

Fringe benefits: the salary amount is for both salary and FBs

Travel: ($28,000)

Funds of $18,000 ($4,500 per year) are requested support domestic travel required for population development, phenotyping activities, and sample collection, to include but not limited to per diem and lodging.

Funds of $10,000 ($2,500 per year) are requested to support international travel including but not limited to airfare, lodging and per diem.

Indirect costs ($12,283) are calculated as 15% of salaries.

Cost sharing: ($21,541)

Cost sharing includes salary ($5,621) corresponding to 2% full-time equivalent of Dr. Fonceka, expatriation cost of Dr. Fonceka based on 10% full-time equivalent ($12,097) and 68% overhead on salary ($3,822).

11.3 Contingency planning

We know that the traits we have selected present challenges for phenotyping. In fact, this is part of the reason we have selected them, since the greatest value for marker-assisted breeding is obtained when marker-based selection can be substituted for difficult phenotype-based selection. There is a possibility that conditions during the project period will not be favorable for phenotyping, with abiotic and biotic stressors either too severe (*e.g.* all genotypes are affected) or too mild (*e.g.* no genotypes are affected). To hedge against this, we have proposed multi-location trials and irrigation resources for INRAN, and have discussed coordination of phenotyping efforts with other SMIL applicants (Bonnie Pendleton and Gary Peterson). In addition, several of the Objectives (1-3) can be carried out even if phenotyping is delayed due unfavorable environmental conditions.

If funds were to be reduced by 10% we would limit the phenotyping activities to a single target trait per country, and reduce the later stage Genotyping-By-Sequencing activities since they are meant to build on phenotyping efforts for trait mapping and molecular breeding implementation.

If funds were to be increased by 10% we would increase the investment in phenotyping activities in order to further leverage the proposed investments genomic and population resources.

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OPERATIONAL PLAN

Project name: **Improving sorghum adaptation in West Africa with genomics-enabled breeding**

Objective 1: Genomic characterization of West African germplasm														
Institution or Person Responsible		***(Note: Year 1 / Period 1 is April 01 – Sept. 30)***	**Yr 1**	**Yr 2**		**Yr 3**		**Yr 4**		**Yr 5**	
			1	**1**	**2**	**1**	**2**	**1**	**2**	**1**	**2**
	Anticipated Research Result 1: Genotyping-by-Sequencing SNP map for West African germplasm										
	Key Activities										
CERAAS	1.1	Seedling growth and DNA extraction (Senegalese germplasm)	**x**	**x**							
INRAN & ICRISAT	1.2	Seedling growth and DNA extraction (Nigerien germplasm)	**x**	**x**							
KSU	1.3	Library preparation and Illumina sequencing for Genotyping-By-Sequencing	**x**	**x**		**x**		**x**			
KSU	1.4	SNP discovery and typing from Genotyping-By-Sequencing		**x**	**x**	**x**	**x**	**x**	**x**		
	Anticipated Research Result 2: Identification of adaptive genomic regions in West African germplasm										
	Key Activities										
KSU	1.1	Genomic analysis of population structure and haplotype structure of West African germplasm			**x**	**x**	**x**	**x**	**x**	**x**	
KSU	1.2	Genome-wide scans for locally-adaptive genomic regions associated with environmental variables			**x**	**x**	**x**	**x**	**x**	**x**	

Objective 2: Build genomics-to-breeding toolkit												
Institution or Person Responsible		***(Note: Year 1 / Period 1 is April 01 – Sept. 30)***	**Yr 1**	**Yr 2**		**Yr 3**		**Yr 4**		**Yr 5**	
			1	**1**	**2**	**1**	**2**	**1**	**2**	**1**	**2**
	Anticipated Research Result 1: Release of open-source GBS-to-KASP conversion tools											
	Key Activities											
KSU & CIRAD	1.1	Development and testing with existing GBS data for West African germplasm	**x**	**x**								
KSU & CIRAD	1.2	Implementation with new GBS data and trait associations from this project			**x**	**x**						
KSU	1.3	Release R scripts for optimized GBS-to-KASP conversion					**x**	**x**				

Objective 3: Multi-Parent Population Development												
Institution or Person Responsible		***(Note: Year 1 / Period 1 is April 01 – Sept. 30)***	**Yr 1**	**Yr 2**		**Yr 3**		**Yr 4**		**Yr 5**	
			1	**1**	**2**	**1**	**2**	**1**	**2**	**1**	**2**
	Anticipated Research Result 1: Senegal NAM population											
	Key Activities											
CERAAS	1.1	F3 plants DNA sampling of NAM population	**x**									
CERAAS	1.2	Generation advance of NAM population F_3 > F_4	**x**	**x**								
CERAAS	1.3	Generation advance of NAM population F_4 > F_5 Seeds multiplication of NAM population F_4 > $F_{3:5}$		**x**	**x**							
CERAAS	1.4	Generation advance of NAM population F_5 > F_6				**x**	**x**					
	Anticipated Research Result 2: Niger NAM population											
	Key Activities											
INRAN	2.1	DNA sampling and KASP marker testing to confirm pedigrees	**x**		**x**		**x**					

Objective 4: Trait-mapping for grain mold, Striga, and drought											
Institution or Person Responsible	***(Note: Year 1 / Period 1 is April 01 – Sept. 30)***		**Yr 1**	**Yr 2**		**Yr 3**		**Yr 4**		**Yr 5**	
			1	**1**	**2**	**1**	**2**	**1**	**2**	**1**	**2**
	Anticipated Research Result 1: Phenotypes for										
	Key Activities										
CERAAS	1.1	Phenotyping of West Sahelian Association Panel for grain mold and drought (Bambey)	**x**		**x**		**x**				
INRAN	1.2	Phenotyping of control lines for Striga (Konni) and drought (Tillaberi)	**x**		**x**		**x**				
CERAAS	1.3	Phenotyping of NAM (F3:5 families) for grain mold and drought		**x**	**x**						
INRAN	1.4	Phenotyping of mapping populations for Striga (Konni) and drought (Tillaberi)					**x**				
	Anticipated Research Result 2: Trait-associated markers for grain mold, Striga, and drought										
	Key Activities										
CIRAD/ CERAAS/KSU	2.1	Genotyping-by-sequencing of NAM F3 populations from Senegal		**x**	**x**						
CIRAD/ CERAAS/KSU	2.2	Quantitative Trait Locus mapping for grain mold and drought in NAM F3 populations from Senegal			**x**	**x**					
KSU & INRAN	2.3	Quantitative Trait Locus mapping for Striga and drought							**x**	**x**	

Objective 5: **Marker Assisted Recurrent Selection and Genomic Selection**																
Institution or Person Responsible		***(Note: Year 1 / Period 1 is April 01 – Sept. 30)***	**Yr 1**	**Yr 2**		**Yr 3**		**Yr 4**		**Yr 5**	
			1	**1**	**2**	**1**	**2**	**1**	**2**	**1**	**2**
	Anticipated Research Result 1: Develop local germplasm with improved resistance traits with MARS															
	Key Activities															
CIRAD/CERAAS	1.1	MARS Cycle 1 or MABC (BC1>BC2)					**x**				
CIRAD/CERAAS	1.2	MARS Cycle 2 or MABC (BC2>BCF2)						**x**			
	Anticipated Research Result 2: Evaluation of genomic selection models for West African breeding															
	Key Activities															
KSU	2.1	Cross-validation testing of genomic selection models							**x**	**x**	

Objective 6: **Long and short term training**																
Institution or Person Responsible	*(Note: Year 1 / Period 1 is April 01 – Sept. 30)*		**Yr 1**	**Yr 2**		**Yr 3**		**Yr 4**		**Yr 5**	
			1	**1**	**2**	**1**	**2**	**1**	**2**	**1**	**2**
	Anticipated Research Result 1: Long term training for two PhD trainees) in genomics-enabled breeding										
	Key Activities										
KSU	1.1	PhD training for Nigerien student, Fanna Maina	**x**	**x**	**x**	**x**	**x**	**x**	**x**	**x**	
KSU	1.2	PhD training for Senegalese student, Jacques Martin Faye	**x**	**x**	**x**	**x**	**x**	**x**	**x**	**x**	
	Anticipated Research Result 2: Short term training workshop on quantitative genomics for crop improvement										
	Key Activities										
CERAAS/ CIRAD/KSU	2.1	A training workshop on quantitative genomics approaches and resources for genetic diversity studies and breeding				**x**					