Selecting Partial Inbred Clones and Exploring Heterosis in Cassava

We started exploring the heterosis in cassava and made selfing of 23 clones, of which 20 clones have high dry matter, and three have high beta-carotene. Even though cassava is a clonal propagation crop, we started with selfing to explore heterosis. Compared with using conventional breeding material, selfing provided at least three advantages, 1) selfing is efficient to purge recessive deleterious mutations; 2) selfed clones show smaller within family variation and larger among family variation, which facilitates determining GCA; 3) inbred or semi-inbreed progenitors will make trait introgression in cassava as efficient as that in corn and rice, so breeding program can respond quickly to farmers'needs.

In 2019 October, we planted 1147 seeds from 23 progenitors, and these plants were transplanted into the field in Palmira in January 2020 (**Table 1**). Selections were made based on their vigor, plant type, dry matter, yield, and disease resistance. From each famlily, 1-5 clones were selected for self-pollination to purge genetic load and for cross-pollination to test GCA. In total, 51 clones were selected and planted in a crossing nursery, and 104 clones were selected and planted in yield trials for further performance evaluation (**Table 2**).

Table 1. The S1 families planted in Palmira in Jan 2020.

cross	parent		num_plant	cross	parent		num_plant
AM1570	AM1253-6	beta-carotene	36	AM1608	SM3110-15	dry matter	50
AM1590	AM1451-4	beta-carotene	32	AM1609	SM3134-5	dry matter	47
AM1592	AM1572-22	beta-carotene	36	AM1612	SM3196-1	dry matter	47
AM1427	SM3150-17	dry matter	45	AM1618	HB60	dry matter	28
AM1597	GM579-13	dry matter	53	AM320	TAI8	dry matter	34
AM1598	GM957-11	dry matter	48	AM329	SM805-15	dry matter	36
AM1599	GM1067-28	dry matter	49	AM332	SM1411-5	dry matter	96
AM1600	GM1070-17	dry matter	47	AM405	SGB765-4	dry matter	43
AM1602	GM1406-12	dry matter	52	AM578	SM2629-36	dry matter	44
AM1603	GM1484-8	dry matter	47	AM583	SM2792-31	dry matter	40
AM1605	SM2828-28	dry matter	90	AM889	GM273-57	dry matter	94
AM1607	SM3060-34	dry matter	53				

Table 2. The number of selected progeny from each parent for crossing nursery or yield trial.

Parent	crossing nursery	yield trial
SM3110-15	2	6
GM1406-12	5	25
GM273-57	4	8
GM1067-28	3	5
GM957-11	5	15
SM2828-28	2	2
SM3134-5	4	6
GM1070-17	3	5
SM3150-17	2	2
GM1484-8	2	2
SM1411-5	2	5
HB60	2	3
SGB765-4	3	5
SM3196-1	1	3
SM2792-31	1	2
TAI8	2	2
SM3060-34	1	1
GM579-13	1	1
SM2629-36	1	1
SM2775-4	2	2
AM1572-22	1	1
AM1253-6	1	1
AM1432-2	1	1

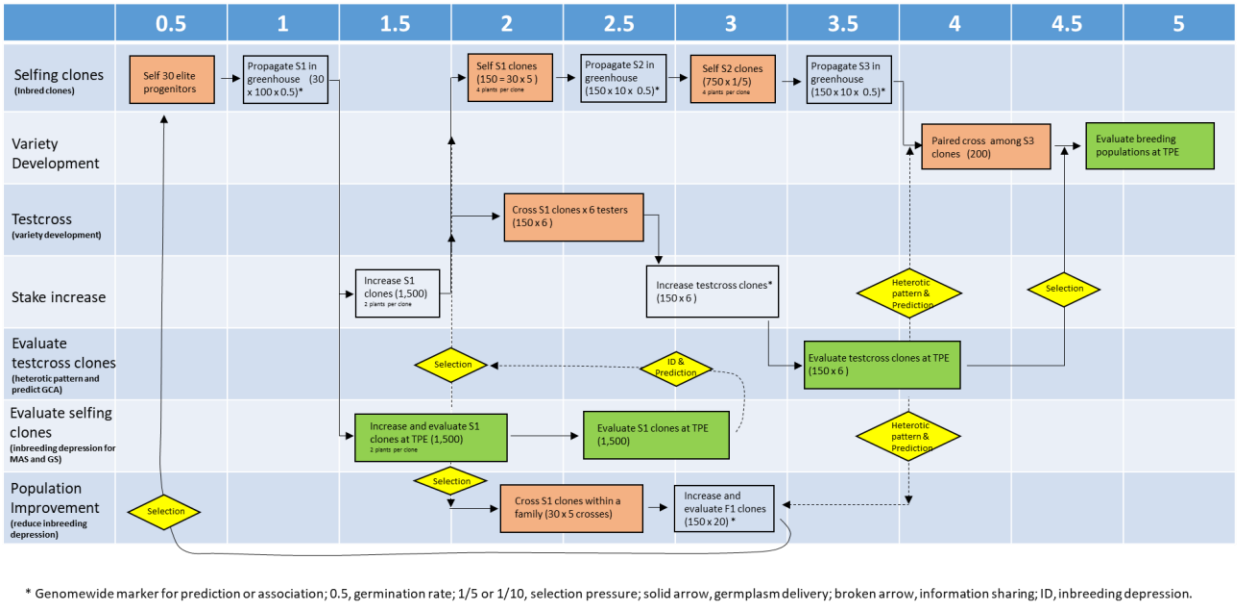
At the same time, we keep discussing hybrid breeding with Excellence in Breeding. In 2021, we proposed the inbred progenitor-based hybrid cassava breeding (**Figure** 1). We will start with elite progenitors with the preferred traits required in the product profile. Because little selfing has been done in cassava, deleterious mutations have been hidden and accumulated in the cassava genome, even in the elite progenitors (Ramu *et al.* 2017). Since previously observing significant low germination of seeds or low vigors in some selfing families, we assume large-effect deleterious mutations might be present in some elite progenitors. Thus, we will evaluate the S1 populations in the target environment to understand the genetic architecture of inbreeding depression and identify the large-effect deleterious mutations for MAS in the next breeding cycles. On the other hand, during the development of selfing clones, we will use genome-wide markers to monitor homozygosity status and select the selfing clones with high rate of homozygous loci for further development selfing. With the assistance of genome-wide markers, we expect to obtain S3 clones with more than 90% homozygosity. We will select the S3 clones with high homozygosity, the complementary deleterious mutations, and favorable alleles for preferred traits for hybrid variety development.

For hybrid breeding, one critical component is to determine the heterotic pattern, either identifying or creating heterotic pools. We will perform testcross to understand the heterotic pattern of the progenitors' S1 families. We will select the testers from the released varieties with all the preferred traits. These testers also need significant genetic distance and the most complementary deleterious mutations (Mezmouk and Ross-Ibarra 2014; Yang et al. 2017). The

evaluation of testcross will guide us to determine the heterotic pools, cross S3 clones for hybrid variety development, and perform the next cycle of within-pool population improvement.

Reciprocal recurrent selection is an effective method for population improvement. Before we identify or create the heterotic patter, we will start crossing selected S1 clones within a family to produce F1 clones for a next selection cycle. After we determine heterotic pools, we will cross S1 among families within a pool for population improvement.

Now, it is pollination season at CIAT. We are selfing elite progenitors to initiate this hybrid cassava breeding project. Within five year, we aim to understand the genetics of inbreeding depression, determine heterotic pools, optimize hybrid breeding scheme and produce hybrid variety candidates.



* Genomewide marker for prediction or association; 0.5, germination rate; 1/5 or 1/10, selection pressure; solid arrow, germplasm delivery; broken arrow, information sharing; ID, inbreeding depression.

Figure 1. The flow chart about hybrid cassava breeding. The orange boxes show the crossing nurseries; the green ones for yield trials; the yellow diamonds for decision making; and the white one for seed increase.

Reference:

Mezmouk, S., and J. Ross-Ibarra, 2014 The pattern and distribution of deleterious mutations in maize. G3 Genes, Genomes, Genet.

Ramu, P., W. Esuma, R. Kawuki, I. Y. Rabbi, C. Egesi *et al.*, 2017 Cassava haplotype map highlights fixation of deleterious mutations during clonal propagation. Nat. Genet.

Yang, J., S. Mezmouk, A. Baumgarten, E. S. Buckler, K. E. Guill *et al.*, 2017 Incomplete dominance of deleterious alleles contributes substantially to trait variation and heterosis in maize. PLoS Genet.