# ROOT CROPS PROGRAMME

## Application for Release of Three Cassava Varieties: NAROCASS 3, NAROCASS 4, and NAROCASS 5











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#### Application for Release of Cassava Varieties

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#### Area of Specilaisation

Plant Breeding and Genetics Plant Breeding and Genetics **Quantitative Genetics** Gender and Plant Breeding Quality Trait Profiling Plant pathology Vector entomology Agricultural Economics Socio Economics Gender **Cassava Breeding** Cassava Entomology **Cassava Breeding** Data Science Cassava Pathology **Tissue Culture** Cassava Breeding Cassava Pathology

Species:	Manihot esculenta (Crantz)
Origin of Parents:	Uganda and Tanzania.
Mode of Generation of Test Materials:	Natural Sexual Recombination
Locations of Evaluation & Selection:	Kigumba (mid-western region), Lira (northern), Arua (west Nile), Serere (eastern), Kaberamaido (eastern), Pallisa (eastern), Kamuli (eastern), Kasese (western) Mityana (central) and Namulonge (central), Luwero (central), Mityana (central), Dokolo (eastern), and Tororo (eastern)
End Use:	Human (boiled roots and flour-based meal) and Industry
Check Varieties:	NAROCASS 1

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#### Acknowledgment

In 2015, two cassava varieties *NAROCASS 1* and NAROCASS 2 were officially released. This application for three new varieties namely NAROCASS 3 (Mkumba); NAROCASS 4 (UG120156) and NAROCASS 5 (UG120193), comes after six years of undertaking field and laboratory performance evaluations. This significant milestone was made possible through support by National Agricultural Research Organization (NARO) together with its development partners.

Notable of these partners were the "Next Generation Cassava Breeding Project" a globally implemented cassava project supported by Cornell University. This progress has also been possible through the regionally implemented project "New cassava varieties and clean seeds to combat CMD and CBSD project" commonly referred to as the "5CP project" through which the cassava clone Mkumba was accessed from Tanzania.

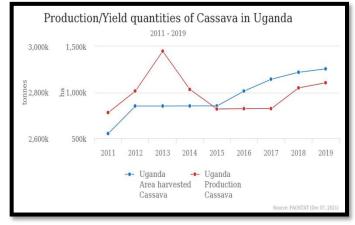
Support of Root Crops Programme staff, District Agricultural Officers (DAOs) and sub-county agricultural officers (SAO) is greatly appreciated. Farmers that participated in the evaluations across different geographies in Uganda: Kigumba (mid-western region), Lira (northern), Arua (west Nile), Serere (eastern), Kaberamaido (eastern), Pallisa (eastern), Kamuli (eastern), Tororo (eastern), Dokolo (eastern), Kasese (western), Mityana (central) and Namulonge (central), is greatly appreciated.

We also thank the National Agricultural Research Organisation (NARO), which provided us with conducive research environment and other logistical support.

#### 1.0 Background

Following the previous official cassava variety release in 2015, production trends In Uganda have had a revealing pattern. For example, in 2015, cassava was cultivated on only 852,000 ha from which 2.7 million MT were harvested. However, in 2019, cassava acreage had increased to 1.2 million hectares from which 2.8 million MT were harvested. Officially released cassava varieties and/or elite clones namely NASE14, NAROCASS1, NASE3 and TME14, occupy a significant market share, with local varieties namely (Bao, Nyaraboke, Bamunanika and Omo) occupying a minor market share. These realities present both exciting opportunities and challenges that variedly impact rural communities. For example, while local varieties are popular owing to their desirable food attributes, they will increase cassava brown streak disease

(CBSD) inoculum in communities. This hampers the useful previously deployed tolerant varieties i.e. NASE14, NAROCASS1 and/or NAROCASS2. It's such discrepancies that justify continued varietal replacements to with an aim of sustaining cassava's competitiveness in Uganda. Satisfying customer requirements is considered as a desiderata for the success of the cassava value chain. It's very likely that cassava producers will quickly adopt and keep a variety if it offers: cost reduction; risk mitigation; yield enhancement; simplicity; price benefit; and emotional needs.



It's for these reasons that NARO has for the past 12 years undertaken a purposeful search for cassava varieties to replace NAROCASS 1 and NASE 14, which have currently become less productive owing to their degeneration to CBSD. Previously, NAROCASS 1 was considered to be a highly tolerant to CBSD and thus highly recommended for cultivation especially in regions with high CBSD pressure. However, NAROCASS 1 has started to succumb to CBSD owing to increased viral load. Further, NAROCASS 1 is susceptible to *Bemesia tabaci*, the virus vector. Mindful of these limitations, cassava breeding has since then been modelled to focus on priority product profiles that meet specific market segments. Priority products profiles for cassava in Uganda are boiled cassava, flour-based meal and industrial use. i.e. cassava for brewing. Thus, herein, we report the evaluation and selection of candidate cassava varieties destined for food and/or industrial use.

#### 2.0 Methodology

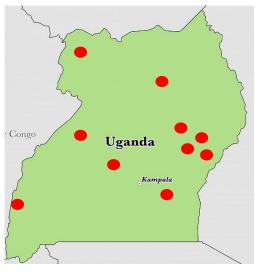
The breeding methodology adopted for cassava involves selection of parents (based on complimentary traits), crossing (via controlled or open pollinations) and simple phenotypic selection of individual clones based on *per see* performances. Cassava's vegetative nature allows for fixation of genotypes. Early stages of evaluation and selection are usually unreplicated and thus emphasis is placed on traits of high heritability, while late stages that have adequate replication, emphasis is placed on low heritability traits. Excellent reviews of hybridization, evaluation and selection, which we adopted for this work, have been documented (see Kawano, 2003). Hereafter, details undertaken at each stage to generate, evaluate and select the candidate clones are highlighted.

#### 2.1 Cycle-zero (C<sub>0</sub>) population from which candidate clones were selected

Owing to documented customer needs and the unrelenting CBSD challenge in Uganda, an initiative was undertaken in 2009 to assemble genetically diverse germplasm to enable systematic cassava genetic improvement and hence genetic gain deployment on-farmers' fields. Accordingly, germplasm was introduced from International Centre for Tropical Agriculture (CIAT), International Institute of Tropical Agriculture (IITA), and from Tanzania's cassava program. Germplasm from Tanzania was received as botanical seed, whereas germplasm from CIAT and IITA were introduced as tissue culture plantlets. Consequently, hybridizations were made among 52 parents selected on *per see* performance. From the progenies generated 395 clones were selected to constitute cycle zero (C<sub>0</sub>) from which the two candidate clones (UG120193 and UG120156) were identified. The candidate clone Mkumba was introduced as tissue culture material under the 5 CP project in 2016.

#### 2.2 Early stage evaluation trials

A total of 150 C<sub>0</sub> clones were subjected to an extensive field evaluation across 10 locations in Uganda, and in different seasons (1<sup>st</sup> rains and 2<sup>nd</sup> rains). The evaluation geographies represented different cassava growing regions and thus included: Kigumba (mid-western region), Lira (northern), Arua (west Nile), Serere (eastern), Kaberamaido (eastern), Pallisa (eastern), Kamuli (eastern), Kasese (western) Mityana (central) and Namulonge (central). These trials were planted in an augmented design with five checks at each of the trial sites. Each plot was represented by 10 plants. Three different trials were planted. First, were trials planted during first rains of 2015, this coincided with the period May 2015 to May 2016. Second were trials planted during second rains of 2015, this coincided with the period October 2015 to October 2016, and finally, the trials planted in first rains of 2016, this coincided with the period May 2016 to May 2017.



**Figure 1**: Target geographies for cassava evaluation

At each site, foliar CBSD severity (degree of infection on each plant) was scored on a 1–5 scale, where 1 = no symptoms; 2 = mild symptoms (1–10%); 3 = pronounced chlorotic mottle and mild stem lesion (11–25%); 4 = severe chlorotic mottle and stem lesions (26–50%) and 5 = very severe symptoms (>50%). Cassava mosaic disease severity (CMD) was assessed at six months after planting (MAP) using a scale of 1–5, where: 1 = no symptoms; and 5 = severe mosaic symptoms. At harvest, which coincided with 12 MAP, all plants in a plot were uprooted and all roots individually assessed for CBSD necrosis. This was done using the 1–5 scale, where 1 = no necrosis; 2 = mild necrotic lesions (1–10%); 3 = pronounced necrotic lesions (11–25%); 4 = severe necrotic lesions (26–50%) with mild root constriction and 5 = very severe necrotic lesions (50%) with severe root constrictions. Further, storage roots were bulked, counted and weighed to obtain both root weight per plant and root weight per plot. Datasets generated from these with these trials informed selection decisions for advancing clones to uniform yield trial stage in 2017.

#### 2.3 Late stage evaluation trials

Based on the extensive multi-location trials highlighted above, the top 22 outstanding C<sub>0</sub> clones were selected and advanced for uniform yield trial (UYT) evaluations at four locations: Arua (west Nile), Serere (eastern), Tororo (eastern) and Namulonge (central). UYT were laid out in completely randomized block design with two replications per location. Each plot comprised of 49 plants that were established at spacing of 1 m x 1 m. Agronomic data was collected from UYT as described earlier. All UYT were established in May 2017 and terminated in May 2018. Additional late stage evaluations were conducted by evaluations undertaken within operations of the 5CP project (<u>https://www.iita.org/news-item/iita-led-5cp-project-reports-great-strides-regional-exchange-improved-cassava-varieties/</u>) and African Cassava Whitefly Project (<u>http://www.cassavawhitefly.org/</u>). In both these trials the introduced clone 'Mkumba" was evaluated alongside NAROCASS 1 in trials established in central (Luwero, Namulonge) and/or eastern (Kamuli, Pallisa, and/or Serere) regions.

#### 2.4 Establishment of on-farm trials using TRICOT methodology

In order to satisfy end-user requirements, NARO adopted a novel approach triadic comparison of technologies (TRICOT). This approach hinges on crowd sourcing i.e. citizens in science approach and thus addresses hitches of conventional participatory variety selection. Overall, TRICOT involves large numbers of men and women farmers conducting simple, small trials on their land. TRICOT allows for participatory evaluation with over 1000 men and women farmers and thus sex disaggregated data generated to inform selection decisions. Accordingly, TRICOT methodology was used to evaluate candidate clones (selected from UYT) with an aim of identifying clones that possess end-user preferred trait preferences than those exhibited by the benchmark variety, NAROCASS 1.

#### 2.4.1 Recruitment of men and women

Purposive sampling was done to have a fair representation of men and women cassava producers at parish level. This was done with the intention to have a diverse sample of men and women from regions with high cassava production and utilization. Of priority were regions where cassava is destined to be consumed as a "boiled" or 'flour-based meal' product. Accordingly, three regions were prioritized and hence selected: Northern, Eastern and Central. Two districts were selected per region and thus making a total of six districts. Accordingly, Mityana and Luweero districts were selected to represent central Uganda; Dokolo selected to represent northern Uganda; Arua district selected to represent west Nile region, while Serere and Kaberamaido districts, were selected to represent eastern Uganda.



*Figure 2*: Awareness and recruitment of farmers for TRICOT evaluations

Upon selection of districts, key informant interviews were conducted with district agricultural officers (DAOs) to purposively select two sub-counties per district. Again, selection of sub-counties was based on high cassava production, existence of farmer groups and high consumption of a "boiled" or 'flour-based meal' products. A key informant guide was used to interview DAOs. At the end, DAOs were asked for contact information of the agricultural officers of the selected sub-counties.

Thereafter, key informant interviews were conducted with sub-county agricultural officers (SAO) to select two parishes per sub-county. Selection depended on existence of at least two farmer groups and processing patterns putting high emphasis on boiled cassava or cassava flour. In the end, a total of 240 cassava farmers were selected from central, northern and eastern regions. Collectively, farmers evaluated 13 cassava clones, with each farmer evaluating only three randomly assigned clones.

#### 2.4.2 Deployment of TRICOT on farmers' fields and data collection

NARO provided 12 candidate cassava varieties for TRICOT evaluation. In addition, NAROCASS 1 was added as a benchmark variety. The TRICOT project was designed using climmob software which generated packages for all participants. Each package had a unique identifier number and consisted of three clones inscribed with letters A or B or C. Each farmer received a TRICOT package to plant i.e. a total of 240 packages were generated and thus deployed on 240 farmers' fields. At each farmers' field, plant spacing of 1 m X 1 m within and between rows was adopted, while a 2 m alley separated the three candidate varieties at each farmers' field.

Data on agronomic-related traits were collected at four points. Firstly, at three MAP on vigor, pest and disease resistance (CMD and CBSD). Secondly, at six MAP on branching and stem appearance. Thirdly at nine MAP on height. And Fourthly, at 12 MAP on root shape, root size, yield and disease resistance (CBSD). At each data collection point, men and women were asked to rank the overall best performing clone i.e. best and worst clone. Furthermore, at 12 MAP, consumer testing was done on boiled and flour-based meal products processed and prepared at household (HH) level from harvested roots. Men and women who hosted TRICOT fields invited two members of their HH to participate in consumer testing. At each HH labelling with letters A, B and C was consist to avoid clone mixing.



*Figure 3*: Variety rankings done at critical stages during crop growth

Upon harvesting, cassava roots were processed following local processing procedure into boiled or flour-based meal. Briefly for boiled cassava in Luwero and Mityana, roots from each candidate variety were peeled, sliced and wrapped in banana leaves and placed in labeled sourcepans laid with banana sheath. All three source pans (A, B and C) were simultaneously placed on fire wood stoves to steam cassava for one hour. In Dokolo, Kaberamaido, Serere and Arua, cassava roots from each candidate variety were peeled, washed, sliced and placed in labeled source pans which were half- filled with water. Likewise, source pans were simultaneously placed on lit fire wood stoves to boil cassava roots for 45 minutes.



Figure 4: Variety ranking done at harvest

On the other hand, flour-based meal was processed in Dokolo, Kaberamaido, Serere and Arua. To process flour, cassava roots were peeled, washed, sliced and sun-dried on tarpaulin labeled with a corresponding letter (A, B, and c). Local motor and pestle were used to pound and thereafter, a wire meshed sieve was used to process fine cassava flour which was used to make the flourbased meal. The flour-based meal was prepared by adding flour to boiling water and mixing until a thick paste was formed. During consumer testing, the product (boiled or flour-based meal) was placed on a labeled plate and presented to three members of the same household to taste and evaluate.



Figure 5: Variety ranking done on processed food product

One member tasted the product from three candidate varieties and there after ranked the best and worst variety depending on the attribute. Water was used to rinse the mouth after tasting the product of each candidate variety. Boiled/ steamed cassava roots were evaluated for easy of peeling, cooking time, taste, softness. The flour-based meal product was evaluated for ease of mixing, stickiness of the paste, taste and texture.

#### 2.5 Distinctiveness, uniqueness and stability

Datasets generated collected across different locations enabled the quantification of genotype-byenvironment interactions and thus, the stability of the candidate varieties. Specifically, this was done using the UYT trials established at: Arua (west Nile), Serere (eastern), Tororo (eastern) and Namulonge (central). On the other hand, upon termination of TRICOT trials, observational trials were established in TRICOT maintenance sites located at (Namulonge, Serere, Kaberamaido, Arua and Dokolo) to enable measurements and/or observation of key morphological traits of the candidate varieties.

#### 2.6 Data Analysis

For the clonal trials, data analysis was based on single row plots with locations considered as replications, with the following model:  $y_{ij} = \mu + E_j + C_i + e_{ij}$ , where  $y_{ij} =$  plot measurement;  $\mu =$  grand mean;  $E_j =$  location effect;  $C_i =$  clone effect and  $e_{ij} =$  residual. For the UYT trial, data were analyzed as RCBD (randomized complete block design). The following model was used:  $y_{ij} = \mu + C_i + \beta_j + e_{ij}$ , where  $y_{ij} =$  plot measurement,  $\mu =$  grand mean;  $C_i =$  clone effect;  $\beta_j =$  effect of the replication; and  $e_{ij} =$  residual. For TRICOT data the *Plackettluce* package was used to generate favorability ranking of candidate varieties for attributes evaluated before harvest. Further, correlation analyses were done to determine the relationship between measured attributes (https://www.rdocumentation.org/packages/stats/versions/3.6.2/topics/cor). Using the late-stage evaluation data (i.e. national performance trial), stability analysis was undertaken (https://www.rdocumentation.org/packages/ammistability/versions/0.1.2/topics/ammistability) to get insights on variety stability.

#### 3.0 Results and Discussion

#### 3.1 Early stage field performances

At the clonal stage, a total of 179 clones were evaluated across the 10 locations. Evidently, the evaluated clones varied significantly in performance for all evaluated traits: cassava brown streak disease foliar resistance, cassava mosaic disease resistance, cassava brown streak disease root resistance, root dry matter content and fresh root yield per plant (Table 1). Of interest however, are clones for cultivation, which when deployed in farmers' fields are able to be resilient and thus meet customer specification.

Accordingly, candidate varieties (Mkumba, UG120156 and UG120193) outperformed the check variety (NAROCASS 1 syn UG110017) in almost all evaluations undertaken at different locations in Uganda (Tables 3 and 4). The candidate clones notably "Mkumba" consistently expressed higher levels of whitefly resistance as compared to NAROCASS 1 (Table 5 and Figure 6). This further underpins the superiority of the candidate clones to the key biotic stresses in Uganda. The increasing susceptibility of NAROCASS 1 to both CBSD and damage by *Bemesia tabaci*, make these varieties appropriate replacements and/or complimentary cultivars. Additional datasets on performances of candidate varieties can be accessed from cassavabase (<u>https://cassavabase.org/</u>), the central repository of all cassava breeding data.

#### 3.2 Late-stage field performances

Again, at uniform yield trial stage when truly outstanding clones are evaluated under high plot capacities i.e. 49 plants per plot with two replicates per site, the evaluated clones exhibited significant difference in performances for cassava brown streak disease root severity resistance, root dry matter content and fresh root (Table 2). Environmental effects were non-significant for CBSD response, while genotype-by-environments effects were non-significant for root dry matter content (Table 2). With exception of fresh root yield, all candidate varieties were superior to NAROCASS 1; this was evident in CBSD resistance (<8%), and high root dry matter content i.e. up to 38% for UG120193, and 42% for Mkumba as compared to the 35.5% for NAROCASS 1 (Tables 4 and 6). Additional datasets on performances of candidate varieties at late evaluation stage can be accessed from cassavabase (https://cassavabase.org/).

While NAROCASS 1 can yield up to 30 t/ha, its increasing being observed that higher CBSD root incidences (i.e. >30%), especially in hotspot areas, greatly undermine its fresh root yield attribute. Although fresh root weight is appreciable upon harvesting NAROCASS 1, processed roots aren't fit-for-purpose (i.e. have severe root necrosis with scores of up to 5), which greatly reduces the competitiveness of products processed from NAROCASS 1. We predict that degeneration of NAROCASS 1 could reach up to 50% within next five to six years as community phytosanitation is limited or non-existent in some regions. It's these deficiencies in NAROCASS 1 coupled with low community phytosanitation measures, that we propose to release of candidate varieties (Mkumba, UG120156 and UG120193). These varieties will gradually replace NAROCASS 1 before it eventually becomes obsolete. The likely disapproval of NAROCASS 1 owing to its inferior processed products on the market is a phenomenon we want to diffuse timely!

#### 3.3 Acceptance of final-end cassava products

Two market segments predominate the cassava value-chain in Uganda i.e. boiled roots and flour-based meal products. These were the final-end products evaluated by farmers during the TRICOT evaluations. It was evident that most end-users preferred candidate variety UG120193 for making boiled cassava; this was followed by NAROCASS 1 (Table 7). Similarly, for 'flour-based meal' product, the candidate variety UG120193 was most preferred, with NAROCASS 1 ranking third (Table 7). Although candidate variety UG120156 was ranked seventh in making boiled cassava roots, it ranked fourth for flour-based meal product (Table 7). Importantly, all candidate varieties have acceptable levels of hydrogen cyanide (see Table 7; and <a href="https://cassavabase.org/">https://cassavabase.org/</a>). Thus, the social approvals by end-users for these varieties further justify their release to bolster the two predominant market segments.

Correlations were made between ranked attributes i.e. those: a) evaluated after harvest; b) evaluated after peeling and evaluation of boiled cassava roots; and c) evaluated after evaluation of the cassava flourbased meal. Accordingly, three findings were apparent. Firstly, most attributes at harvest had strong correlation with farmer's selection of overall best performing clone (Figure 7), and thus implying that production attributes are important drivers of adoption. Secondly, taste and how well cassava roots cooked, had the strongest positive correlation to the best performing boiled clone (Figure 8). This finding provides the insight that taste and softness of boiled roots, are major drivers of adoption in communities that primary consume boiled cassava (i.e. the boiled cassava market segment). Thirdly, taste of cassava flour-based meal had the strongest positive correlation to the best performing clone (Figure 9). This reinforcing the insight that taste of flour-based meal is a major driver adoption in communities that primarily consumer flour meal products (i.e. the flour-based meal market segment).

#### 3.4 Distinctiveness, uniqueness and stability

The candidate varieties were characterized by 24 morphological traits (Table 8). Exceptionally, Mkumba is characterized by pubescence on apical leaves, while other candidate varieties (UG120193 and UG120156), have no pubescence on apical leaves (Table 8). Secondly, Mkumba has red petiole colour, while UG120193 and UG120156, respectively have purple and purplish-green petioles (Table 8). The predominant variety to replace (NAROCASS 1) has exceptionally dark green leaf colour which is distinct from all the three candidate varieties. Exceptionally, UG120156 has a pink root cortex, while other candidate varieties and those in production have white-to-cream root cortex. The aforementioned characteristics highlight the distinctiveness and uniqueness of the candidate varieties. Stability analysis was performed using two traits i.e. fresh root yield and CBSD root necrosis (i.e. CBSD resistance). From the generated biplots, it was evident that the candidate variety UG120193 was the most stable while other candidate varieties exhibited modest location-specific adaptation (Figure 10). On the other hand, stability analysis based on CBSD root necrosis revealed that the candidate varieties were highly stable (Figure 11) and thus indicating their resilience to combat CBSD.

#### 4.0 Conclusions and recommendations

The cassava value chain will only survive provided that major actors i.e. producers, processors, traders and consumers, are each satisfied with products (e.g. varieties) they use. This philosophy guides cassava varietal improvement process at NARO. Thus, during the past 12 years, NARO has designed and systematically evaluated cassava clones to identify those fit-for-purpose for cassava end-users. In the end, three candidate varieties (Mkumba, UG12193 and UG12156) have been selected. Importantly, these candidate varieties have been designed to balance both "must-have" production attributes (i.e. resilience to major pests and disease notably CBSD and its whitefly vector), and quality attributes i.e. consumer preferred attributes of 'boiled roots' and 'flour-based meal' products. The participatory evaluation of these candidate varieties with 240 farmers offered valuable insights and hence basis for selection of these candidate varieties.

These candidate varieties once released will compliment previously released varieties notably NASE 14 and NAROCASS 1, which are increasing becoming susceptible to CBSD. This replacement is vital before both NASE 14 and NAROCASS 1 become obsolete. Specifically, we recommend Mkumba for "Flour-based food" product and industrial starch for distilling and/or generation of ethanol. Mkumba has up to 77.6% starch content as compared to NAROCASS 1 with only 73.7%. For both UG120156 and UG120193, we recommend them for both boiled roots' and 'flour-based meal' products. It is thus proposed that the candidate varieties (Mkumba, UG12156 and UG12193) be officially released as NAROCASS3, NAROCASS4 and NAROCASS5.

#### 5.0 Cultivar maintenance and quality seed access

Basic seed (virus-free indexed planting material) of the candidate varieties (Mkumba, UG12193 and UG12156) is maintained both in tissue culture and in screenhouse at NaCRRI, Namulonge. Foundation seed of these varieties (~1/2 an acre each clone) is available at National Semi Arid Resources Research Institute (NaSARRI), Serere. Additionally, at each of the TRICOT sites in Arua, Dokolo, Kaberamaido, Mityana, Luwero and Serere, each of these candidate varieties is established on ¼ an acre. Further, we plan to adopt the semi Semi-Autotrophic Hydroponics (SAH) technology, already established at NaCRRI, to enhance the quantities of virus-free clean seed of these candidate varieties.

Sources	Df	CBSD6s	CMD6s	CBSDRs	CBSDRi	FRWP	DMC
Clones	188	1.94***	0.47***	5.82***	5010***	9.43***	67.04***
Envts	9	6.78***	0.24***	3.76***	5243***	215.75***	2173.01***
Block/Envts	42	0.42***	0.133ns	0.88*	853**	4.23	42.24***
Clones x Envts	957	0.39***	0.09ns	0.62ns	629**	2.73	20.21**
Residual	226	0.26	0.11	0.58	452	4.09	14.36

Table 1: Analysis of variance for key agronomic traits across 10 locations in Uganda

D.f = degrees of freedom; CBSD6s = cassava brown streak disease foliar severity score at six MAP: CMD6s = cassava mosaic disease severity at six MAP; CBSDRs = cassava brown streak disease average root severity at 12MAP; CBSDRi = cassava brown streak disease root incidence at 12 MAP; DMC = root dry matter content; FRWP = fresh root weight per plant. \*\* and \*\*\* represents significance at P, 0.01, and 0.001, respectively. Envts = environments

able 2: Analysis of variance for key traits evaluated during uniform yield trial stage in Uganda
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Sources	Df	CBSDRs	CBSDRi	FRY	DMC
Clones	26	0.081***	403.6***	325.7***	23.2ns
Envts	3	0.024ns	99.5ns	269.9***	189.5***
Rep/Envts	4	0.006ns	56.1ns	133.3***	20.5ns
Clones x Envts	46	0.048***	216.5****	48.3*	10.7ns
Residual	62	0.012	55.9	30.1	17.2

D.f = degrees of freedom; CBSDRs = cassava brown streak disease average root severity at 12MAP; CBSDRi = cassava brown streak disease root incidence at 12 MAP; DMC = root dry matter content; FRY = fresh root weight per hectare \*, \*\* and \*\*\* represents significance at P, <0.05, 0.01, and 0.001, respectively. Envts = environments

Clones	CBSD6s	CBSD6s_Rank	CMD6s	CMD6s_Rank	CBSDRs	CBSDRs_Rank	CBSDRi.	CBSDRi_Rank	RWTP	RWTP_Rank	DMC	DMC_Rank
UG130022	1.10	13	1.00	10	1.15	26	7.41	26	2.80	45	30.52	59
UG110021	1.59	60	1.00	38	1.04	7	4.17	7	3.59	18	30.64	57
UG120156	1.00	4	1.00	4	1.09	19	7.33	19	1.81	105	31.27	48
UG130032	1.25	25	1.00	14	1.28	45	12.73	45	4.81	5	29.50	78
UG120186	1.33	33	1.00	18	1.00	1	0.00	1	0.45	173	39.58	1
UG120303	1.73	80	1.00	47	1.15	27	9.08	27	2.76	48	35.60	6
UG120115	1.33	35	1.00	20	1.34	52	24.84	52	3.94	13	30.09	68
UG110023	1.52	52	1.00	32	1.16	28	9.09	28	2.83	43	30.54	58
UG120182	1.00	5	1.00	5	1.00	1	0.00	1	0.58	167	30.34	62
UG120024	1.08	11	1.00	8	1.01	2	1.04	2	1.15	148	29.93	71
UG110026	1.28	29	1.00	16	1.09	18	6.82	18	1.88	98	30.22	65
UG120181	1.56	58	1.00	37	1.03	5	1.73	5	3.40	20	26.79	131
UG120193	1.21	23	1.51	168	1.17	30	10.53	30	2.97	33	35.25	8
UG120180	1.06	9	1.48	165	1.15	25	14.37	25	2.48	59	35.19	9
UG130105	1.67	72	1.00	43	1.00	1	0.00	1	1.60	120	30.21	66
UG110024	1.26	26	1.00	15	1.19	34	16.52	34	1.97	92	28.12	103
UG120044	2.00	96	1.00	54	1.00	1	0.00	1	1.37	133	33.10	22
UG120001	2.37	139	1.00	71	1.07	13	6.80	13	3.39	21	31.12	51
UG120006	2.18	118	1.00	61	1.23	40	15.00	40	4.33	7	31.42	45
UG120198	1.31	32	2.22	177	1.15	24	8.47	24	2.63	55	35.92	4
UG120037	1.95	94	1.00	52	1.01	3	1.39	3	1.07	153	34.57	12
UG110028	1.33	34	1.00	19	1.32	51	31.02	51	1.33	137	32.43	29
UG130004	2.44	149	1.00	77	1.02	4	1.58	4	2.57	57	32.30	32
UG120157	1.67	71	1.50	166	1.05	10	5.00	10	3.32	25	31.22	49
UG120138	1.00	3	1.00	3	1.47	73	22.44	73	1.65	117	30.09	67
UG120191	1.53	55	1.00	35	1.05	9	4.76	9	2.30	68	23.89	161
UG110025	1.68	74	1.17	144	1.00	1	0.00	1	1.75	109	35.04	10
UG120190	1.06	10	1.15	143	1.04	8	1.82	8	1.42	132	31.62	42
NAROCASS 1	1.19	22	1.12	134	1.35	55	18.33	55	5.48	3	29.75	74

Table 3: Performance of the top 30 Co clones across 10 locations in Uganda

CBSD6s = cassava brown streak disease foliar severity score at six MAP: CBSD6s\_Rank = rank of clone for CBSD foliar severity; CMD6s = cassava mosaic disease severity at six MAP; CMD6\_Rank = rank of clone for CMD severity; CBSDRs = cassava brown streak disease average root severity at 12MAP; CBSDRs\_Rank = rank of clone for CBSD root severity; CBSDRi = cassava brown streak disease average root severity at 12MAP; CBSDRs\_Rank = rank of clone for CBSD root severity; CBSDRi = cassava brown streak disease average root severity at 12MAP; CBSDRs\_Rank = rank of clone for CBSD root severity; CBSDRi = cassava brown streak disease average root severity at 12MAP; CBSDRs\_Rank = rank of clone for CBSD root severity; CBSDRi = cassava brown streak disease root incidence; RWTP = fresh root weight per plant; RWTP\_Rank = rank of clone for root weight per plant; DMC = root dry matter content; DMC\_Rank = rank of clone for DMC; Rank\_Sum = summation of clone ranking across the evaluated traits. Analysis based on 179 clones evaluated across 10 locations. Candidate clones (UG120156 and UG120193) outperformed the check variety (NAROCASS 1)

Clones	CBSDRs	CBSDRs_Sep	CBSDRi	CSBDRi_Sep	FRY	FRY_Sep	DMC	DMC_Sep
NAROCASS 1	1.12	cd	7.09	bc	31.76	а	35.45	а
UG110019	1.38	abc	28.96	ab	5.63	d	31.33	а
UG110021	1.15	abcd	14.27	abc	8.13	cd	35.13	а
UG110022	1.10	cd	7.61	bc	7.03	d	35.72	а
UG110023	1.12	cd	10.91	bc	7.37	d	37.22	а
UG110024	1.24	abcd	15.26	abc	11.25	cd	37.34	а
UG110026	1.14	bcd	14.15	bc	8.83	cd	34.34	а
UG110037	1.12	cd	9.87	bc	25.00	abc	36.82	а
UG120001	1.02	d	1.82	с	29.44	а	33.61	а
UG120020	1.00	d	0.00	с	25.00	abcd	41.27	а
UG120024	1.02	d	1.70	С	9.06	cd	33.42	а
UG120086	1.38	ab	33.42	а	6.03	d	37.48	а
UG120089	1.02	d	1.80	С	37.50	а	28.76	а
UG120124	1.03	cd	1.57	С	21.56	abcd	33.01	а
UG120136	1.20	abcd	11.47	bc	22.46	abcd	32.14	а
UG120156	1.02	d	1.69	С	12.59	cd	35.74	а
UG120180	1.02	d	1.84	С	14.17	bcd	40.13	а
UG120183	1.10	cd	7.09	bc	12.40	cd	36.86	а
UG120187	1.36	abc	24.42	ab	15.42	abcd	37.00	а
UG120190	1.04	cd	2.73	С	8.44	cd	33.02	а
UG120193	1.09	cd	7.63	bc	16.02	abcd	38.08	а
UG120198	1.05	cd	3.02	С	12.98	cd	36.93	а
UG120304	1.43	а	22.84	ab	22.81	abcd	32.93	а
UG130002	1.02	d	1.15	С	18.38	abcd	37.20	а
UG130007	1.06	cd	5.33	bc	27.19	ab	33.75	а
UG130014	1.13	cd	9.65	bc	13.59	cd	33.63	а
UG130016	1.03	d	1.96	С	20.55	abcd	35.02	а

Table 4: Performance of candidate clones at uniform yield trial stage across four sites in Uganda

CBSDRs = cassava brown streak disease average root severity at 12MAP; CBSDRs\_Sep = significance tests and thus clones, followed by same letter aren't significantly different; CBSDRi = cassava brown streak disease root incidence at 12 MAP; CBSDRi\_Sep = significance tests and thus clones followed by same letter aren't significantly different; FRY = fresh root yield in t/ha; FRYC\_Sep = significance tests and thus clones followed by same letter aren't significantly different; DMC = root dry matter content; DMC\_Sep = significance tests and thus clones followed by same letter aren't significantly different; same letter aren't significan

	Early-stage	whitefly res	istance		Early-stage whitefly resistance				
Variety	Kamuli	Luwero	Pallisa	Average	Kamuli	Luwero	Pallisa	Average	
MKUMBA	2.3	2.5	2.6	2.5	1.0	1.0	1.0	1.0	
NAROCASS2	2.8	3.5	4.1	3.5	1.6	2.6	2.6	2.3	
NAROCASS1	4.6	4.1	4.3	4.3	3.5	3	3.5	3.3	
ORERA	4.3	4.4	4.8	4.5	3.6	3.6	3	3.4	
NASE 12	5.3	4.2	4	4.5	5.5	3.3	2	3.6	
LOCAL	3.8	4.5	5.3	4.5	5	2.6	3.6	3.7	

Table 5. Field evaluation of whitefly resistance in selected clones

Whitefly resistance assessed on a scale of 1-6, where 1 = resistant and 6 = susceptible.

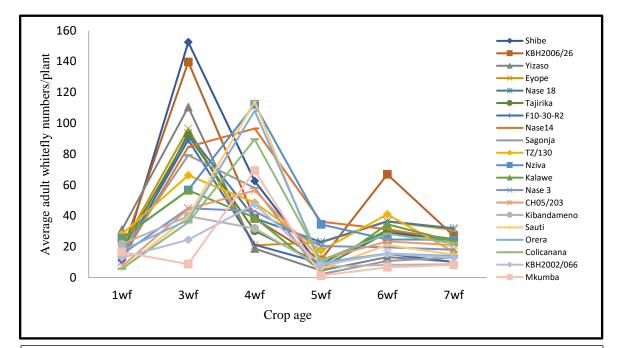


Figure 6: The trend of whitefly population dynamics and crop age. Abbreviations and acronyms: Wf = Mean Adult whitefly count from five most expanded leaves of a plant. Adult whitefly populations were computed as an average per plant by dividing the total population within a plot by the number plants per plot. The figures preceding wf on the horizontal axis represent crop age in months. Candidate clone "Mkumba" display high whitefly resistance levels as compared to the check clone NAROCASS 1 (TZ/130)

Clone	Rt.no	Frwt	CBSDRi	CBSDRs	DMC
Shibe	10.875a	0.579cde	28.4a	1.88bc	37.49abcde
CH05/203	10.8a	1.624bcd	21.73abcd	2.444abc	37.762abcde
NAROCASS1	10.765a	3.74a	8.21bcde	1.41bc	36.64bcde
Sauti	10.429a	1.938bc	17.4abcde	1.57bc	38.17abcd
NASE 18	10.33a	1.475bcd	19.98acd	2.333bc	37.27abcde
Eyope	9.0a	0.514de	27.38a	2.75ab	35.0cdef
Yizaso	8.923a	2.23b	19.44abcd	1.67bc	39.31abc
F10-30-R2	8.692ab	2.23ab	11.99abcde	1.46bc	39.0abcd
Sagonja	7.78ab	1.033bcde	27.24a	2.57abc	36.86bcde
Kalawe	7.45ab	0.656cde	12.97abcde	2.1bc	32.62ef
Nase 14	6.625ab	1.103bcde	8.39bcde	1.69bc	39.55abc
NASE 3	6.3ab	0.435de	12.45abcde	2.6abc	37.93abcde
Orera	6.23ab	1.979bc	10.78abcde	1.54bc	37.34abcde
Tajirika	5.8ab	1.333bcde	5.34de	1.64bc	37.05abcde
KBH2006/26	5.1ab	0.96bcde	9.79abcde	1.53bc	33.73def
Mkumba	4.55ab	1.736bcd	0.715e	1.0c	42.35a
KBH2002/066	4.27ab	0.379de	12.43abcde	2.22bc	35.08cdef
Nziva	3.8ab	0.63cde	6.06cde	1.3bc	41.05ab
Colicanana	3.67ab	0.403de	24.53abc	2.13bc	36.96bcde
Kibandameno	1.0b	0.029e	25ab	4.0a	30.01f
CV%	78.32%	74.88%	93.66%	60.74%	9.50%

Table 6. Agronomic performance of selected introductions in Uganda

CV% =coefficient of variation percentage, R.tno = Number of roots harvested per plot, Frwt = fresh root weight per plot, CBSDRi = cassava brown streak disease root incidence, CBSDRs = average CBSD root severity damage, DMC = dry matter content.

	Performance		Mean perform		Mean perfor		
	field at har	vest*	boiled cassava	n meal**	flour-based meal***		
Clone name	Mean	Rank	Mean	Rank	Mean	Rank	
NAROCASS 1	65.60a	1	54.16ab	2	44.21a	3	
UG130007	54.34ab	2	29.17abc	6	47.50a	2	
UG120193	43.03abc	3	58.72a	1	59.60a	1	
UG120124	33.84abcd	4	21.55abc	8	29.39a	6	
UG120198	29.33abcd	5	34.63abc	4	32.41a	5	
MM16/0707	28.94bcd	6	9.31c	11	17.78a	9	
UG120156	24.21bcd	7	22.96abc	7	39.42a	4	
MM06/123	14.48cd	8	35.34abc	3	24.44a	8	
UG130016	13.76cd	9	17.70bc	9	10.37a	12	
UG120180	13.70 cd	10	33.33abc	5	28.52a	7	
UG120024	2.78d	11	15.05c	10	16.20a	10	
MM16/1627	0.00d	12	3.52c	12	13.33a	11	

Table 7: Farmers' preference of clones at harvest and after evaluation of the boiled cassava roots and flour-based meal

\*Performance in the field was based on stem quality, root shape, root size, root yield, disease resistance and root cortex colour attributes. \*\*Performance of boiled cassava meal was based on ease of peeling, cooking time, mealiness, softness, taste and fibrousness attributes. \*\*\*Performance of flour-based meal was based on ease of drying of chips, ease of mingling, stickiness, texture, taste, and colour attributes. Means with different letter codes are significantly different from each other while those followed by same letter aren't significantly different. Across all evaluation trials the candidate clones are characterized by softness of boiled roots of: 2.5 KgF/cm-1 (Mkumba), 1.9 KgF/cm-1 (UG120193), 2.5 KgF/cm-1 (UG120156) and 2.2 KgF/cm-1 (NAROCASS 1). Similarly, for hydrogen cyanide (based on 1-9 scale), they are characterized by: 6.7 (Mkumba), (UG120193), 5.2 (UG120156) and 5.7 (NAROCASS 1). All data available on https://cassavabase.org/

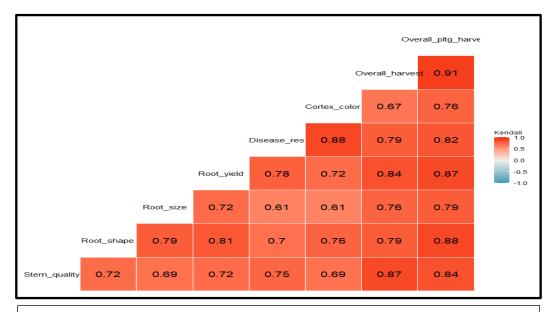


Figure 7: Correlation of attributes evaluated after harvest. Overall\_harvest means overall best performing clone at harvest. Overall\_pltg\_harve means overall best performing clone from planting to harvest.

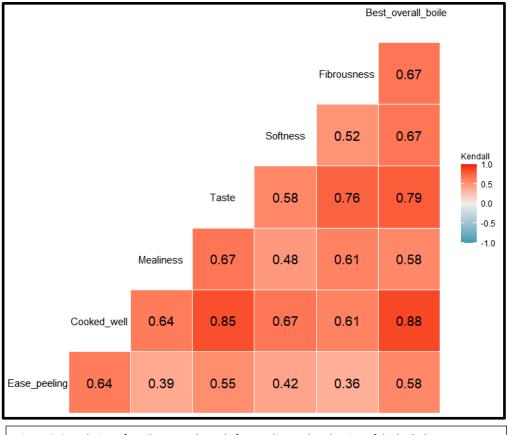


Figure 8: Correlation of attributes evaluated after peeling and evaluation of the boiled cassava roots. Best\_overall\_boile means overall best performance after evaluation of the boiled roots.

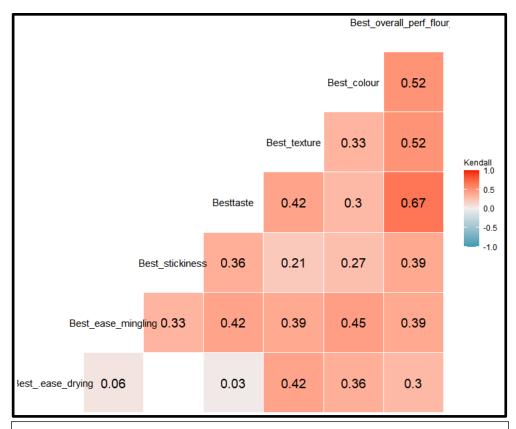


Figure 9: Correlation of attributes evaluated after evaluation of the cassava flour-based meal. Best\_overall\_perf\_flour means overall best performance after evaluation of cassava flour-based meal.

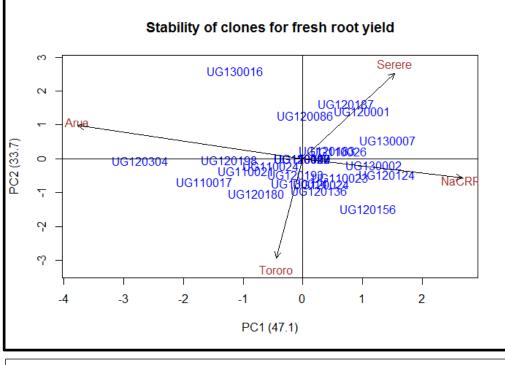


Figure 10: Stability of the clones for fresh root yield at four sites during National performance trial

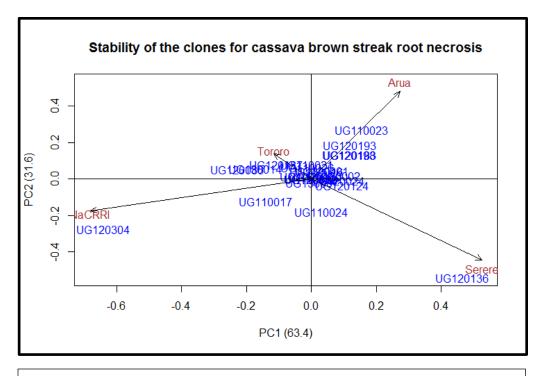


Figure 11: Stability of the clones for CBSD root necrosis resistance at four sites during National performance trial

Qualitative Description	МКИМВА	UG120156	UG120193
Color of apical leaves	Purplish green	Purplish green	Purplish green
Pubescence on apical leaves	Present	Absent	Absent
Petiole color	Red	Purplish green	Purple
Leaf color	Light green	Dark green	Light green
Number of leaf lobes	Seven	Seven	Seven
Color of leaf veins	Reddish-green in more than half of the lobe	Green	Reddish-green in less that half of the lobe
Flowering	Present	Present	Absent
Orientation of petiole	Horizontal	Inclined upwards	Inclined upwards
Color of stem exterior	Gray	Silver	Gray
Color of stem cortex	Dark green	Dark green	Dark green
Color of stem epidermis	Light brown	Light brown	Light brown
Growth habit of stem	Straight	Straight	Straight
Color of end branches of adult plant	Green-purple	Green-purple	Green-purple
Fruit	Present	Present	Absent
Seed	Present	Present	Absent
Branching habit	Dichotomous	Trichotomous	Trichotomous
Shape of plant	Open	Umbrella	Umbrella
Extent of root peduncle	Pedunculate	Pedunculate	Pedunculate
Root shape	Cylindrical	Conical-cylindrical	Conical-cylindrical
External color of storage root	Light brown	Light brown	Light brown
Color of root pulp (parenchyma)	White	White	White
Color of root cortex	White to cream	Pink	White to cream
Cortex: ease of peeling	Easy	Easy	Easy
Texture of root epidermis	Smooth	Intermediate	Intermediate

Table 8: Distinctiveness and uniqueness of the candidate clones

#### References

Kawano, K., 2003. Thirty years of cassava breeding for productivity-biological and social factors for success. Crop Science 43:1325-1335.

Nanyonjo AR, Kawuki RS, Kyazze F, Esuma W, Wembabazi E, Dufour D, Nuwamanya E, Tufan H. Assessment of end user traits and physicochemical qualities of cassava flour: a case of Zombo district, Uganda. Int J Food Sci Technol. 2021 Mar;56(3):1289-1297. doi: 10.1111/ijfs.14940.

Manze F, Rubaihayo P, Ozimati A, Gibson P, Esuma W, Bua A, Alicai T, Omongo C, Kawuki RS. Genetic Gains for Yield and Virus Disease Resistance of Cassava Varieties Developed Over the Last Eight Decades in Uganda. Front Plant Sci. 2021 Jun 21;12:651992. doi: 10.3389/fpls.2021.651992.

Iragaba P, Hamba S, Nuwamanya E, Kanaabi M, Nanyonjo RA, Mpamire D, Muhumuza N, Khakasa E, Tufan HA, Kawuki RS. Identification of cassava quality attributes preferred by Ugandan users along the food chain. Int J Food Sci Technol. 2021 Mar;56(3):1184-1192. doi: 10.1111/ijfs.14878.

Kawuki RS, Kaweesi T, Esuma W, Pariyo A, Kayondo IS, Ozimati A, Kyaligonza V, Abaca A, Orone J, Tumuhimbise R, Nuwamanya E, Abidrabo P, Amuge T, Ogwok E, Okao G, Wagaba H, Adiga G, Alicai T, Omongo C, Bua A, Ferguson M, Kanju E, Baguma Y. Eleven years of breeding efforts to combat cassava brown streak disease. Breed Sci. 2016 Sep;66(4):560-571. doi: 10.1270/jsbbs.16005.

Ozimati A, Kawuki R, Esuma W, Kayondo SI, Pariyo A, Wolfe M, Jannink JL. Genetic Variation and Trait Correlations in an East African Cassava Breeding Population for Genomic Selection. Crop Sci. 2019 Mar-Apr;59(2):460-473. doi: 10.2135/cropsci2018.01.0060.

Mukiibi DR, Alicai T, Kawuki R, Okao-Okuja G, Tairo F, Sseruwagi P, Ndunguru J, Ateka EM. Resistance of advanced cassava breeding clones to infection by major viruses in Uganda. Crop Prot. 2019 Jan;115:104-112. doi: 10.1016/j.cropro.2018.09.015.