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Cassava retting ability and textural attributes of *fufu* for demand-driven cassava breeding

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Abstract

BACKGROUND: Cassava retting ability and the textural qualities of cooked fufu are important quality traits. Cassava retting is a complex process in which soaking causes tissue breakdown, starch release, and softening. The rate at which various traits linked to it evolve varies greatly during fufu processing. According to the literature, there is no standard approach for determining retting ability. The retting indices and textural properties of fufu were measured using both manual and instrumental approaches.

RESULTS: Different protocols were developed to classify 64 and 11 cassava genotypes into various groups based on retting ability and textural qualities, respectively. The retting protocols revealed considerable genetic dissimilarities in genotype classification: foaming ability and water clarity should be measured at 24 h, while penetrometer, hardness, turbidity, pH, and total titratable acidity data are best collected after 36 h. The stepwise regression model revealed that pH, foaming ability, and dry matter content are the best multivariates (with the highest R^2) for predicting cassava retting. These predictors were used to develop an index for assessing the retting ability of cassava genotypes. The retting index developed showed a significant relationship with dry matter content and fufu yield. The study also showed significant correlations between instrumental cohesiveness and sensory smoothness (r = -0.75), moldability (r = -0.62), and stretchability (r = 0.78). Instrumental cohesiveness can correctly estimate *fufu* smoothness ($R^2 = 0.56$, P = 0.008) and stretchability ($R^2 = 0.60$, P = 0.005).

CONCLUSION: pH, foaming ability, and dry matter content are the best traits for predicting cassava retting ability, while instrumental cohesiveness can effectively estimate *fufu* smoothness and stretchability.

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Supporting information may be found in the online version of this article.

Keywords: high-throughput protocols; consumer preferences; varietal adoption; heritability; genetic progress

INTRODUCTION

In Nigeria, as in many tropical developing countries, especially in Africa, cassava (*Manihot esculenta* Crantz) is one of the most important food crops and a major source of carbohydrates.^{1,2} Cassava is a versatile crop and can be processed into a wide range of products such as *gari, fufu,* starch (*tapioca*), flour, and chips for industrial use, human consumption, and animal feed.

Fufu, a fermented cassava mash, in wet or dry form, is a popular product from cassava consumed by most people in southeast Nigeria.^{3,4} The unit operations for fufu production are root peeling, washing, cutting into pieces, steeping in water for 3–4 days to ret or soften (i.e., ferment), sieving of retted root pieces to remove fiber and, finally, extracting starch by pressing through a muslin cloth.

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Retting is a process in which cassava roots are immersed in water; they become softened, and the pectic materials that bind the fibers are broken down, thereby liberating soluble constituents such as starch. This has been attributed to an increase in pectinolytic and cellulolytic enzymatic activities.⁵⁻⁷ It has also been reported that varietal differences have a significant influence on the extent of retting of cassava roots.⁸ The degree of root softening determines the easy separation of the water-insoluble material, such as fiber strands, during the sieving operation. This determines the yield of wet fufu mash, which is the primary interest of *fufu* processors.⁹ Retting has also been linked to the textural properties of cooked *fufu*. The textural properties of *fufu* refer to the adhesiveness or stickiness, cohesiveness, and hardness of the fufu product. A study¹⁰ reported a higher and better index for drawability, moldability, easy-to-form dough, smoothness, and color in *fufu* processed using the retting method compared to alternative methods of processing fufu.

The quality and acceptance of fufu by consumers and the adoption of cassava varieties by processors are influenced by the retting ability of the roots and the textural characteristics of the *fufu*.¹¹

However, there is no standard protocol to assess the retting ability of cassava root and the textural properties of *fufu*. Lack of appropriate high- or mid-throughput methods for selecting and integrating these end-user preferred traits into the breeding pipeline at the early stages of breeding has led to an unintended loss of genetic variation for root quality traits over time.^{12,13} Furthermore, the lack of these appropriate methods for screening quality traits at the early breeding stage has led to the breeding of genotypes that do not meet the end-users needs, resulting in a low adoption rate of released varieties.

This study, therefore, aims at profiling the retting ability using high- and mid-throughput methods developed and standardized within the RTBfoods project (https://rtbfoods.cirad.fr), and consumer-preferred textural properties of *fufu* processed from selected cassava genotypes. The study also identifies key traits that influence variation in the retting ability of cassava genotypes and the textural attributes that drive preference among fufu consumers. This will hopefully accelerate the breeding of cassava varieties with key quality traits that drive the adoption, processing, and consumption of *fufu* by end-users.

MATERIALS AND METHODS

Genotypes

Studies were carried out on cassava roots from 64 genotypes obtained from the NRCRI-NextGen population (43 and 21 clones from preliminary (PYT) and advanced (AYT) yield trials, respectively). Trials with the 64 genotypes were established over 2-year cropping seasons (2020 and 2021) and were laid out in the field using a randomized complete block design of two replications in two locations in Nigeria (PYT: Umudike; AYT: Umudike and Otobi). Umudike is located at 5° 28' 0" N latitude and 7° 330' 0" E longitude, and Otobi at 7° 47' 0 " N latitude and 10° 0' 0" E longitude.

Profiling for retting of fresh cassava roots

Fresh cassava roots were processed to *fufu* using the validated RTBfoods standard operating procedure (SOP) for sensory characterization of *fufu*, and retting ability was evaluated using SOPs developed within the RTBfoods project (https://doi.org/10. 18167/agritrop/00595). The instrumental firmness of 64 cassava

genotypes was measured with a handheld penetrometer of 3.5 mm diameter cylindrical probe (FHP-802 model), through conventional fermentation times of 0, 24, 48, and 72 h. In order to predict early and quick selection for the breeders, an unconventional average time of 36 h (average between 24 and 48 h) was added. A total of ten repeated sample measurements were taken on each genotype by measuring the proximal and distal parts of five randomly selected cassava roots per genotype through fermentation time. The maximum resistance force (newtons) per penetration was recorded by pushing the tip of the handheld penetrometer to a depth of 1 cm of the selected roots. The average of these ten penetrations per genotype is referred to as *firmness*.

Sensory (hand) firmness of cassava roots (five randomly selected roots per genotype), and foaming and water clarity (i.e., water visibility) of the fermenting liquor through days of fermentation were also visually evaluated by five champion processors at NRCRI, Umudike. Manual or visual assessment by the processors was rated on a 5-point scale ranging from extremely soft to extremely hard for firmness and from extremely clear/absence of foam to extremely turbid/extremely foamy for clarity and formation of bubbles on the fermenting liquor. The turbidity of the fermented liquor was also measured in the lab using an ultraviolet-visible spectrophotometer (752 N model, Bosch searchtech Instrument, England). An aliquot of the fermenting liquor per fermenting vessel in two replicates was poured into a cuvette, and the transmittance was measured at 650 nm. Material losses and fermented mash recovered during fufu processing were also estimated. Fufu yield was calculated as the weight of dewatered *fufu* mash divided by the weight of peeled cassava roots used.

Biophysical analysis of fresh cassava roots

The dry matter content (DMC) of fresh cassava roots was determined using the oven drying method as described by AOAC.¹⁴ Total titratable acidity (TTA) and pH were determined for the fermenting liquor per fermenting vessel through fermentation time. TTA was determined following the method described by Lees.¹⁵ A sample of 10 mL was titrated with 0.1 mol L⁻¹ NaOH using phenolphthalein as an indicator. pH was determined using a pH meter (PHS-25 model, England). The pH meter was calibrated using buffers of pH 7 and 10. These biophysical analyses were carried out in two replicates per genotype.

Sensory and textural properties of *fufu* from 11 cassava genotypes

The validated RTBfoods SOP for *fufu* textural analysis¹⁶ (https://doi.org/10.18167/DVN1/RFZMLC) and sensory texture profile analysis (STPA) were used to assess the texture properties of *fufu* produced from 11 AYT cassava genotypes planted in Otibi.

Instrumental texture profile analysis (ITPA) by double compression was used to determine *fufu* hardness, adhesiveness, cohesiveness, springiness, gumminess, chewiness, and resilience using a texture analyzer (TA.XT2, Stable Micro Systems Ltd, Godalming, UK). Two cooking replicates per genotype were considered, and six measurements per cooking replicate were collected. STPA was conducted on the same samples in duplicate sessions by 12 trained panelists.¹⁷ Smoothness, moldability, stickiness, hardness, and stretchability of *fufu* were scored on a scale of 0 (lowest attribute score) to 10 (highest attribute score).

Consumer testing of *fufu* from five cassava genotypes

Following Madu *et al.*,¹⁸ the five *fufu* products processed from five varieties (*Ichenke*, *NR-68*, *NR-9*, *TMS* 13–1, and *TMEB419*) by the

Table 1. Statistical parameters and broad sense heritability of the retting ability of 64 cassava genotypes evaluated across two repetitions in each of

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processors with varying quality characteristics were tested by 300 fufu consumers in Nigeria's South-East (Abia State) and North-Central (Benue State) regions. The five varieties used were selected from the AYT planted at Otobi. Two of the five genotypes (TMEB419 and the local check Ichenke) had also been included among the 11 genotypes involved in the study of the sensory and textural properties of *fufu* described above.

Statistical analyses

Variables analyzed included sensory and instrumental texture attributes of *fufu*, across the different clones used for the study of retting traits. Statistical analyses carried out included analyses of variance (ANOVA), bivariate correlations, discriminant analysis using ranking coefficient, and ranking indices with assigned weights depending on the correlation coefficient values, using JMP Pro 15 software (SAS Institute Inc., Cary, NC, USA). Pearson correlation analysis and hierarchical classification were conducted to show the association among the different retting traits and other factors influencing retting ability. An alpha level ≤ 0.05 was chosen to indicate significance in the correlations. The broad sense heritability was estimated by calculating the ratio between genetic (σ^2_G) and phenotypic (σ^2_P) variances; σ^2_P is the sum of σ^2_G and the error (σ_{e}^{2}) .

ANOVA was used to test for significant differences in the overall acceptability of *fufu* products among the varieties tested in consumer testing. Supporting Information Table S3 explains the important triangulation-ranking traits (retting ability and texture) that were among the sensory traits chosen by consumers. Triangulation is a synthesis of four steps following Forsythe et al.¹⁹ in their study on interdisciplinary and participatory methods to improve the acceptability of root, tuber, and banana varieties. Step 1 was the scope of the study and the gaps in research (demand characteristics and gaps in knowledge were identified). Step 2 showed the set of ranked quality characteristics from users playing different roles in the food chain (characteristics/trait preferences identified and knowledge produced on the broader food system and the role of gender and gender relations). Step 3 described the qualitative characteristics of the cassava varieties under study for fufu (processing diagnostics/demonstration of the fufu product profile). Step 4 provided robust data on preferences regarding the final product among a diverse set of consumers. The development of the questionnaires used in the implementation of

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Step 4 benefited from the results and observations of Step 3. The preferred trait highlighted in S3 (retting ability) was the same in Steps 2 and 3, and therefore selected.

RESULTS

Roots: retting ability and processing parameters

Table 1 presents the estimation of broad sense heritabilit and variance components of selected retting parameters using instrumental approaches (handheld penetrometer for firmness, spectrophotometer for water turbidity and pH meter) and sensory procedures (hardness, foaming ability and water visibility). The results showed that the instrumental techniques (firmness by penetrometer and turbidity by spectrophotometer) have higher broad sense heritability estimates (65.3% and 57.2%, respectively), than the sensory methods: hardness (by hand), foaming ability and water visibility (visual), with broad sense heritability estimates of 27.9%, 39.0%, and 32.8%, respectively. Heritability for pH was the lowest (24.9%).

Profiling of the retting ability of cassava genotypes using different protocols, shown in Fig. 1, illustrates the kinetic patterns for the different variables measured. After only 24-48 h of retting, clear changes are already obvious. Penetrometer data show a drastic reduction in the firmness of the root from 24 to 48 h. Hardness shows a similar pattern, although differences between 24, 48 and 72 h are more gradual. Foaming ability and water clarity show similar trends, with drastic changes from 0 to 24 h, which then slow down considerably, accompanied by a large dispersion of data. Turbidity and pH are gradually and consistently reduced through time, with noticeable changes early on (between 0 and 24 h). TTA, on the other hand, increases consistently through time with a larger dispersion of data, late in the retting process.

The correlation between biophysical properties of cassava genotypes, processing parameters and retting indices shows that there is a significant relationship between retting indices, biophysical parameters, and product yields of different genotypes. Since most changes take place between 24 and 48 h of retting, an average of the two measurements was added (this could be considered the response after 36 h of retting). As expected, there were positive, significant, and strong correlations for measurements of the same parameter through consecutive times, except for hand hardness (36 vs. 72 h); foaming ability (36 and 48 vs.

Statistic	Instrumental			Manual	Visual	
Statistic	Penetrometer (N)	Turbidity (NTU)	pН	Hardness	Foaming ability	Water visibility
Heritability	0.65	0.57	0.25	0.28	0.39	0.33
Genotype variance	0.51	15.42	0.00	0.03	0.04	0.03
Genotype × Environment variance	0.00	3.87	0.01	0.00	0.01	0.021
Residual variance	1.01	37.48	0.03	0.29	0.35	0.17
Grand mean	3.42	42.93	4.62	2.01	3.13	2.66
LSD	0.86	4.17	0.08	0.29	0.31	0.27
% CV	30.03	14.26	3.47	26.75	18.96	15.34
Genotype significance	0.00	0.00	0.28	0.03	0.05	0.02
GenxEnv significance	1.00	0.27	0.04	1.00	0.81	0.39

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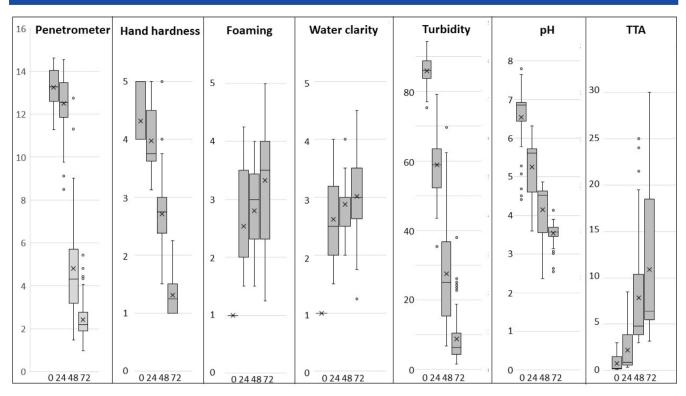


Figure 1. Kinetics of different traits at 0, 24, 48, and 72 h of retting (horizontal axis). The units of measurement were newtons (N) for penetrometer and a 1–5 scale for the sensory assessment through hand hardness, foaming ability, and water clarity. Turbidity and total titratable acidity (TTA) were measured in nephelometric turbidity units (NTU) and as a percentage, respectively.

72 h); and water clarity (24 vs. 72 h), which failed to reach statistical significance. Moreover, penetrometer, turbidity, pH, and TTA measurements taken at different times always showed positive and significant correlations (Supporting Information, Table S1). However, an interesting evolution from positive to negative correlations through time could be observed in some traits: hand hardness (24 vs. 48 and 72 h); and foaming ability (24 vs. 72 h).

Table 2 summarizes the most relevant correlations between different traits measured at 24, 36 (average between 24 and 48 h), 48, and 72 h of retting. Penetrometer data showed significant correlations with hand hardness (except when the latter was measured at 24 h). Hardness at 24 h showed significant positive correlations with foaming ability; however, at longer fermenting times the correlations evolved to be significantly negative. Negative and significant correlations between hardness (24 and 36 h) versus water clarity and pH were observed. A similar (but positive) correlation was observed with turbidity and TTA. Correlations between foaming ability versus water clarity or pH were generally negative and significant (except for the associations between foaming ability and pH, when either variable was measured at 72 h). On the other hand, correlations of foaming ability versus turbidity and TTA were significantly positive (except when foaming ability was measured at 72 h).

As expected, water clarity and turbidity were always negatively correlated. In most instances, these correlations were significant (Table 2). An interesting change in the sign of correlations took place between water clarity *versus* pH and TTA. The coefficients between water clarity (measured at 24 and 36 h) *versus* pH or TTA were significantly negative and positive, respectively. However, the sign of the correlations changed when water clarity was measured at 48 or 72 h. Turbidity showed positive and

(in most cases) significant correlations with pH and TTA. Finally, pH and TTA were negatively correlated.

In general, correlations tended to be highest when measured at 36 h, except in the case of hardness, which seemed to provide the most reliable information when taken at 24 h. This suggests that the critical changes during retting take place in the first 48 h. Correlations involving the penetrometer were not significant (except for the correlation with hand hardness shown in Table 2). The weakest correlations among different traits were observed for measurements taken at 72 h. The changes in the sign of correlations for the same traits taken at different times illustrate the strong sensitivity of the timing of evaluation on retting ability.

Correlations between different variables (measured at different times during the retting process) and fufu yield are presented in Table 3. There was no association between penetrometer data and fufu yield. Hand hardness, on the other hand, showed significant and positive correlations with fufu yield when measured after 24 or 36 h of retting. Foaming ability at 24–36 h of retting showed significant and positive correlations. The correlation between water clarity and turbidity correlations and fufu yield did not reach statistical significance. Strong correlations were consistently found for pH and TTA (negative and positive, respectively), except for pH at 72 h.

The result of regression modeling for classifying genotypes according to their retting ability (retting index)

Up until now, there has been no clear and objective way to assess retting ability in cassava. No single correlation provided a satisfactory explanation for the variation observed in *fufu* yield and quality, which is a function of the retting ability (Table 3). Therefore, a weighted retting index, integrating all the variables measured at

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Table 2. 72 h of rett		ations among tr	aits (Pearson's c	orrelations) me	asured in cassa	va roots at 24, 3	36 (average betw	ween 24 and 48	3 h), 48, and
P/H	H-24	H-36	H-48	H-72	H/FA	FA-24	FA-36	FA-48	FA-72
P-24	0.07	0.27*	0.28	0.45**	H-24	0.44**	0.54**	0.44**	0.04
P-36	0.03	0.44**	0.55**	0.64**	H-36	0.23	0.29*	0.24	-0.25*
P-48	0.01	0.44**	0.58**	0.62**	H-48	-0.12	-0.14	-0.11	-0.36*
P-72	-0.16	0.25*	0.49**	0.66**	H-72	0.20	0.00	-0.13	-0.37**
H/WC	WC-24	WC-36	WC-48	WC-72	H/T	T-24	T-36	T-48	T-72
H-24	-0.43**	-0.61**	-0.58**	-0.39**	H-24	0.41**	0.65**	0.62**	0.39**
H-36	-0.37**	-0.46**	-0.39**	-0.12	H-36	0.29*	0.54**	0.55**	0.41*
H-48	-0.07	-0.01	0.05	0.22	H-48	-0.01	0.08	0.12	0.16
H-72	-0.17	0.00	0.17	0.29*	H-72	-0.20	-0.05	0.04	0.01
H/pH	pH-24	pH-36	pH-48	pH-72	H/TTA	TTA-24	TTA-36	TTA-48	TTA-72
H-24	-0.79**	-0.80**	-0.80**	-0.03	H-24	0.83**	0.82**	0.79**	0.83**
H-36	-0.55**	-0.58**	-0.61**	-0.15	H-36	0.46**	0.46**	0.44**	0.55*
H-48	0.04	0.01	-0.03	-0.17	H-48	-0.20	-0.19	-0.18	-0.09
H-72	-0.17	-0.19	-0.20	-0.26*	H-72	0.08	0.05	0.03	0.10
FA/WC	WC-24	WC-36	WC-48	WC-72	FA/T	T-24	T-36	T-48	T-72
FA-24	-0.32*	-0.34**	-0.24	0.06	FA-24	0.12	0.31*	0.34**	0.07
FA-26	-0.27*	-0.56**	-0.66**	-0.19	FA-26	0.40**	0.33**	0.21	0.28*
FA-48	-0.15	-0.53**	-0.72**	-0.28*	FA-48	0.46**	0.24	0.06	0.33**
FA-72	0.04	-0.08	-0.16	-0.64**	FA-72	0.15	-0.01	-0.10	-0.08
FA/pH	pH-24	pH-36	pH-48	pH-72	FA/TTA	TTA-24	TTA-36	TTA-48	TTA-72
FA-24	-0.62**	-0.61**	-0.58**	-0.25*	FA-24	0.62**	0.61**	0.59**	0.61**
FA-26	-0.57**	-0.57**	-0.55**	-0.01	FA-26	0.63**	0.64**	0.62**	0.65**
FA-48	-0.37**	-0.36**	-0.35**	0.14	FA-48	0.44**	0.46**	0.45**	0.48**
FA-72	0.20	0.18	0.16	0.32*	FA-72	-0.05	-0.08	-0.10	-0.08
WC/T	T-24	T-36	T-48	T-72	WC/pH	pH-24	pH-36	pH-48	pH-72
WC-24	-0.04	-0.27*	-0.34**	-0.09	WC-24	-0.74**	-0.77**	-0.78**	-0.74**
WC-36	-0.27*	-0.42**	-0.40**	-0.23	WC-36	-0.36**	-0.39**	-0.42**	-0.36*
WC-48	-0.41**	-0.42**	-0.32*	-0.29*	WC-48	0.61**	0.61**	0.60**	0.61**
WC-72	-0.31*	-0.31*	-0.23	-0.27*	WC-72	0.67**	0.61**	0.62**	0.67**
WC/TTA	TTA-24	TTA-36	TTA-48	TTA-72	T/pH	pH-24	pH-36	pH-48	pH-72
WC-24	0.73**	0.73**	0.71**	0.80**		0.41**	0.43**	0.43**	0.27
WC-36	0.35**	0.37**	0.36**	0.45**		0.52**	0.53**	0.53**	0.12
WC-48	-0.61**	-0.59**	-0.57**	-0.59**		0.45**	0.45**	0.44**	-0.06
WC-72	-0.72**	-0.58**	-0.67**	-0.67**	T-72	0.12	0.15	0.18	-0.31
T/TTA	TTA-24	TTA-36	TTA-48	TTA-72	pH/TTA	TTA-24	TTA-36	TTA-48	TTA-72
T-24	0.38**	0.44**	0.45**	0.40**	pH-24	-0.86**	-0.86**	-0.84**	-0.88*
T-36	0.60**	0.63**	0.63**	0.60**	рН-36	-0.86**	-0.87**	-0.85**	-0.90*
T-48	0.58**	0.59**	0.57**	0.56**	pH-48	-0.84**	-0.86**	-0.84**	-0.90**
T-72	0.32*	0.25*	0.21	0.26*	pH-72	-0.16	-0.28*	-0.32*	-0.29*

Note: P, penetrometer; H, hand hardness; FA, foaming ability; WC, water clarity; T, turbidity; TTA, total titratable acidity; shade with the same colour are associated; * and ** means significance at 5% and 1%.

36 h, was created to assess retting ability. The weights for each parameter were the respective correlation coefficients with fufu yield. Figure 2 illustrates the variation of the retting index among the 64 genotypes evaluated, and Table 4 shows the correlations between the index and each individual trait (measured at the optimal timing).

A stepwise regression analysis involving all measured variables was carried out to determine the sequence of relevant parameters

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Parameter	24 h	36 h	48 h	72 h
Penetrometer	0.19	0.19	0.13	0.04
Hand hardness	0.32*	0.31*	0.10	0.19
Foaming ability	0.36**	0.34**	0.22	-0.08
Water clarity	0.34**	0.15	-0.31*	-0.29
Turbidity	0.26*	0.37**	0.34**	0.13
pH	-0.41**	-0.41**	-0.41**	-0.09
TTA	0.34**	0.29*	0.27*	0.30*

Note: Shade with the same colour are associated; * and ** means significance at 5% and 1%.

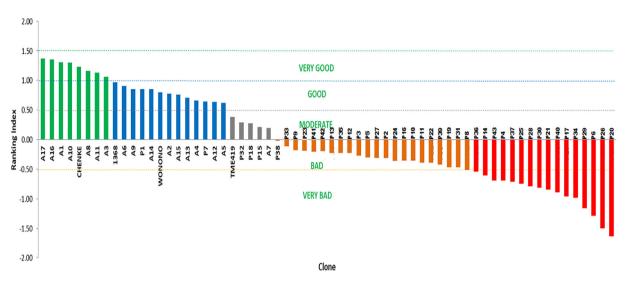


Figure 2. Variation of retting index among the 64 genotypes evaluated.

Table 4.	Relationships between the biophysical properties of the
cassava g	enotypes, processing parameters (product yield and mate-
rial losses)) and retting index

Trait	Retting index			
Penetrometer	0.17			
Hardness	0.63***			
Foaming ability	0.54***			
Water clarity	0.08			
Turbidity	0.55***			
рН	-0.73***			
ТТА	0.72***			
Chaff loss	0.34*			
DMC	0.49**			
<i>Fufu</i> yield	0.68***			
<i>Note</i> : Asterisks indicate significance level at *5%, **1%, ***0.1%.				

in the retting index model The most efficient model (based on the lack-of-fit test) included pH, DMC, and foaming ability, with an R^2 of 73.9. Adding water clarity to the model would increase R^2 to 77.7 (Supporting Information, Table S2 and Fig. S1). A model with additional parameters would increase (as expected) the R^2 value but would also become unnecessarily complex. The proposed model with three parameters offers an acceptable balance between the

amount of variability explained on the one hand and relative simplicity on the other.

The textural profile of fufu

Study of the sensory and instrumental textural profile of *fufu* shows that, apart from retting ability, the textural qualities of cooked *fufu* have been identified as traits of interest to consumers. The repeatability of the ITPA measurements was adequate. All instrumental texture attributes varied significantly among the cassava varieties evaluated. However, variety–cooking replicate interaction significantly affected some attributes (hardness, cohesiveness, and resilience).

Significant correlations were found between some sensory and instrumental textural properties of *fufu* (Table 5). Although instrumental and sensory assessments of hardness were not significantly associated, instrumental hardness was significantly correlated with stickiness (-0.51) and stretchability (-0.44). The strongest association was observed between sensory hardness and gumminess (0.82). The highest correlations involving sensory smoothness were found with cohesiveness (-0.75), gumminess (-0.70), and chewiness (-0.70). In general, sensory stickiness showed a weak correlations with springiness (-0.72), cohesiveness (-0.62), and chewiness (0-0.62). Finally, the most important correlations involving sensory stretchability were with cohesiveness (0.78), resilience (0.75), chewiness (0.68), and

	Sensory						
Instrument	Smoothness	Stickiness	Moldability	S-Hardness	Stretchability		
I-Hardness	0.12	-0.51*	0.37	0.27	-0.44*		
Adhesiveness	0.40*	-0.51*	0.23	-0.42*	-0.21		
Springiness	-0.62**	0.55*	-0.72**	0.43*	0.67**		
Cohesiveness	-0.75**	0.48*	-0.62**	0.46*	0.78**		
Gumminess	-0.70**	-0.06	-0.24	0.82***	0.42*		
Chewiness	-0.70**	0.39	-0.62**	0.61**	0.68**		
Resilience	-0.64*	0.03	-0.41*	0.38	0.75**		

Note: Asterisks indicate significance level at *5%, **1%, ***0.1%; -SHardness means Sensory hardness, -IHardness means Instrumental Hardness; Shade with the same colour are associated.

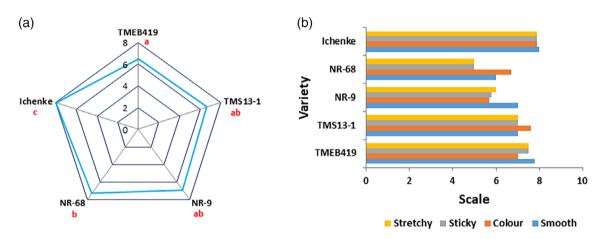


Figure 3. (a) Overall acceptability (ANOVA) and (b) preferred traits of fufu from five cassava varieties.

springiness (0.67). Linear models were developed to estimate key mean sensory scores of *fufu* from instrumental texture measurements. Smoothness and stretchability of *fufu* can be estimated (Supporting Information, Fig. S3) from instrumental cohesiveness using the regression equations shown below:

Smoothness = $11.6 - 10.1 \times \text{Cohesiveness} (R^2 = 0.559, P = 0.008)$ Stretchability = $-1.1 + 13.5 \times \text{Cohesiveness} (R^2 = 0.602, P = 0.005)$

The consumer study results (Fig. 3) show the ANOVA for acceptability of five different cassava varieties, including a local check. ANOVA results indicated that the overall acceptability of *Ichenke* (7.94) was significantly higher than that of the four remaining varieties. *NR-68* was the second-best variety, with an acceptability of 7.27, followed by *NR-9* (6.89) and *TMS13-1* (6.64). The texture (smoothness) of *fufu* quality was highest for the local check, implying this was the consumers' most crucial trait of interest (Fig. 3(b)). The closely related genotypes with similar textural properties are shown in Supporting Information, Fig. S2.

DISCUSSION

Roots: retting ability and processing parameters

The findings in this study revealed that the retting of cassava is a complex process, and the speed at which different traits associated with retting varies considerably with time. There was no single variable that could satisfactorily assess retting ability; hence the need to rely on the retting index as a proxy variable.

The various methods/protocols used in this study include the instrumental methods (penetrometer, turbidity, TTA, and pH) and the manual/visual methods (hand hardness, foaming ability, and water clarity).

The penetrometer method, which gave the highest heritability value, also showed a high ability to discriminate the firmness of cassava root. However, it exhibited a poor relationship with fufu yield – an important trait that drives fufu end-user preference. The ability of the handheld penetrometer to effectively discriminate the retting ability of cassava genotypes based on firmness, as shown in this study, supports earlier findings²¹ indicating strong repeatability during evaluation of sugar beet firmness using the penetrometer. Another study²² also supported the effectiveness of handheld penetrometers in distinguishing textural and phenotypic differences in cooked cassava roots. This current study agrees with previous studies^{2,22-24} Assessing water clarity as fermentation progresses was also applied as a means of characterizing cassava genotypes into distinct clusters with respect to their retting ability. The result revealed that the clarity/visibility of the fermenting liquor decreased with an increase in days of fermentation. An alternative instrumental approach for evaluating water clarity (turbidity) involving transmittance measurements, using the spectrophotometer produced a similar result. A decrease in the transmittance of the retting liquor was

observed as the days of retting increased; this decrease in the transmittance (increase in turbidity or decrease in water clarity) could be attributed to the release of water-soluble materials (starch) into fermenting liquor during fermentation.^{25–27} An increase in turbidity/decrease in water clarity may also be explained by the rise in microbial flora as a result of the production of metabolites, such as glucose, that support bacterial growth during fermentation.²⁸

Instrumental turbidity had higher heritability than the visual assessment of water clarity. Correlations between water clarity (24 h) and turbidity (36 h) with *fufu* yield, as shown in Table 3, were positive but not statistically significant (0.34 and 0.37, respectively).

This study showed that, with the exception of pH, instrumental methods for measuring retting indicators had higher precision compared to manual methods. The development of high- and mid-throughput methods for screening could serve as a milestone for the inclusion of these processor/consumer-priority quality traits at the early stages of breeding.

The study also showed that biophysical properties such as DMC influence fermentation periods, which means that fresh cassava roots with high DMC tend to require a longer time to soften during *fufu* processing but will likely result in higher *fufu* yield production. According to Hahn,²⁹ variability in *fufu* quality could be attributed to varietal differences in the DMC of cassava roots.

There was a clear variation in the sensitivity of the different methods with respect to time. Penetrometer and hardness data should be taken at 36 h of retting. Hand hardness, foaming ability and water clarity, on the other hand, should be measured after 24 h. Differences in turbidity, pH, and TTA are obvious after only 36 h of retting; however, waiting for one or two more days provides a better distinction between genotypes. Conventionally, retting at 36 h has not been considered, but this study has proven that the phenomena that determine retting ability have better predictive capacity when measured at 36 h, which will help breeders make a quick selection of materials with good retting ability. It remains unclear, however, whether a single measurement taken at 36 h provides similar results to the average of two measurements taken at 24 and 48 h.

The inconsistencies in these different methods to assess retting ability indicated the need to develop a retting index as the most powerful and reliable approach to quantifying differences in the retting ability of different genotypes. Although this index is practical for selection purposes, further studies for a better understanding of the accuracy of each individual instrumental or manual method should be conducted. Moreover, these studies could result in an improvement of the model for selection purposes.

Retting is best predicted by combining and using a multivariate approach, where the best (three-parameter) index includes pH, foaming ability, and dry matter content. The use of models to predict variables has been employed in previous studies to explain the thermal softening of cassava roots.²⁰

The textural profile of fufu

Sensory and instrumental textural attributes of fufu

Principal component analysis (PCA) was used to classify cassava genotypes with similar textural quality into clusters. The first two-component PCA showed that *lchenke* (good landrace), *Wonono* (intermediate landrace) and *TMS98* (a good elite variety) were linked with smoothness, moldability, and adhesiveness.

NR-24 was associated with stickiness and *NR-3* was associated with hardness. *TMS13-3*, *TMS13-2*, and *TMEB419* were cohesive, resilient, springy, stretchable, and chewy. It is clear, therefore, that cassava varieties with unknown *fufu* quality may be predicted to belong to a class of *fufu* quality by associating them with key sensory and textural attributes. PCA also showed that sensory and instrumental hardness are unrelated; adhesiveness and smoothness are closely related; while stretchability is closely associated with springiness, resilience, and cohesiveness. The genotypes were clustered in three classes (hierarchies) representing *good*, *intermediate*, and *poor* textural quality for making *fufu*.

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There were promising associations between instrumental and sensory texture measurements. The development of high- and mid-throughput methods for screening key *fufu* quality traits is a critical step towards the inclusion of processor/consumer priority traits at the early selection stages of breeding. This will lead more quickly to the release of varieties that meet end-user needs.

Texture, smoothness, color, and stretchability are among the most liked characteristics of *fufu*.^{11,18} The texture of cooked root and tuber crops and their products has often been cited as a primary determinant of the acceptability of improved and local cassava varieties.³⁰ Current consumer studies confirm these earlier reports.²⁴ There were significant differences in the overall acceptability of the varieties among the consumers, with the local check (*lchenke*) having the best overall textural acceptability for *fufu* quality, consistent with consumers' most important trait of interest. *lchenke* distinguished itself (along with *Wonono* and *TMS98*) for its sensory smoothness and instrumental cohesiveness, which are strongly correlated with each other.

The current study demonstrated the complexities associated with assessing retting ability in cassava roots, including the influence of timing of measurements and the fact that no single trait can reliably predict retting. Considering the single instrumental parameter to predict retting ability pH (alternatively, the closely correlated TTA) ranked the best. This was followed by turbidity and penetrometer, which also correlated acceptably well with sensory hardness; among manual methods, hardness and foaming ability showed the best correlations with the retting index. However, the low to moderate heritability, poor correlations with *fufu* yield, and high dissimilarities in the classification of cassava genotypes by these individual methods led to the development of a multivariate approach.

From regression analysis, pH, foaming ability, and turbidity were the best multivariates that reliably predicted the retting ability. The index could be considered a mid-throughput method for selection purposes for breeders. In addition, the correlations between instrumental and sensory fufu textural attributes demonstrated the feasibility of developing high- or mid-throughput protocols for screening fufu textural attributes. The implementation of these protocols would allow the selection of genotypes with end-users' preferred *fufu* quality traits at the early stages of the breeding process.

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DATA AVAILABILITY STATEMENT

The data that supports the findings of this study are available in the supplementary material of this article.

SUPPORTING INFORMATION

Supporting information may be found in the online version of this article.

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