



## Standard Operating Procedure for Textural Characterization of Boiled Yam by Ottawa Extrusion

#### Ibadan, Nigeria 17/10/2024

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<u>Ethics</u>: The activities, which led to the production of this document, were assessed and approved by the CIRAD Ethics Committee (H2020 ethics self-assessment procedure). When relevant, samples were prepared according to good hygiene and manufacturing practices. When external participants were involved in an activity, they were priorly informed about the objective of the activity and explained that their participation was entirely voluntary, that they could stop the interview at any point and that their responses would be anonymous and securely stored by the research team for research purposes.

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## RTBfoods / RTB Breeding



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The methodology for preparing boiled yam for instrumental texture analysis by Ottawa extrusion is outlined in this standard operating procedure (SOP). Fresh yam tubers are cleaned and about 16 % of its full length is trimmed off at the proximal and distal ends. A 2.5cm thick slice was cut from which a 62mm x 42mm x 20mm cuboid was obtained with the aid of a stainless-steel plunger. These cuboids are steamed for 23 minutes, ensuring even cooking. The extrusion test was conducted at 45<sup>o</sup> C using a Stable Microsystems TA-XT*plus* texture analyzer equipped with a 5-blade grid Ottawa cell and a 50 kg load cell. Key textural parameters obtained include End force, Max force, linear distance, Area, Initial gradient, and Distance at Max force. The extrusion texture parameters were all significant in discriminating among the yam genotypes with End force ( $p = 3.5 \times 10^{-19}$ ) and Max force ( $p = 4.7 \times 10^{-18}$ ) as the most discriminant textural parameters. Based on a hierarchical clustering analysis of the textural parameters, the genotypes were categorized into three groups: Oju iyawo (a *rotundata* check landrace) and TDr1437005 were identified as genotypes with highest values of texture. Oweigbo (a check *alata* landrace), TDa1511008, TDa11520050, and TDr1500042 were grouped as genotypes with intermediate values of texture. TDa1510119 and TDr1500100 were clustered together as genotypes with the least texture values for boiled yams.

Key Words: Boiled yam, Ottawa extrusion, Texture, Repeatability, Discriminant



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## **1** SCOPE AND APPLICATION

#### 1.1 Scope

This standard operating procedure describes the protocol for the preparation of boiled yam for instrumental texture analysis (extrusion method) using the Ottawa cell probe and the texturometer. The procedure is based on the average texture of the boiled yam when deformed by compression through blade grids which offer resistance to the deformation.

## **1.2 Definitions**

The textural parameters obtained from the Ottawa extrusion procedure include:

Max Force (N, or g): is the highest force registered during the extrusion. It represents the hardness of the sample or maximum resistance to deformation. Sometimes, it may be the same as or exceed the End force.

**Area (N.mm or N.s, or g.mm or g.s):** is the area under the curve of the extrusion process. It represents the work done or energy to extrude the sample. A large area represents a higher amount of energy dispensed to extrude the sample.

Additional textural parameters that can be obtained include:

**Distance at Max force (mm):** is the distance where the sample registers its maximum resistance to deformation by extrusion. Short distances represent harder texture at periphery of sample, and longer distances represent harder texture at core or end of sample height.

**Initial Gradient (N/mm or g/mm):** is the slope of the texture curve up to a specific distance during extrusion. It could represent the stress required for the material to yield to deformation by extrusion. It may also explain the surface nature of the sample, for example, a harder, crusty surface may register higher gradients, and consequently higher yield stress. In this study, the gradient was measured from the beginning of the curve up to 3mm where the majority of the curves depart from linearity.

**End force (N or g):** is the force registered at the end of the extrusion process. It represents hardness or resistance to deformation by extrusion at the end of extrusion (at the end of sample height) process. The End force may also be the Max force but never exceeds Max force.

**Linear distance, LD (N.mm or g.mm):** is the length of the whole texture curve. High linear distances may reveal that there are more peaks and troughs (jaggedness) or fracturing within the layers/matrices of the sample.

Other derived parameters that may be interesting to calculate are ratio LD/Area and LD to Max force.



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#### **1.3** Apparatus

- a. Texture analyzer equipped with an Ottawa texture-extrusion cell (the model used for this SOP is a TA-XTplus by Stable Micro System)
- b. Steam cooker to steam cook the yam samples
- c. Stopwatch (used to monitor the time)
- d. Digital balance to weigh yam samples before and after cooking for water absorption determination
- e. Aluminium foil
- f. Styrofoam box
- g. Knives
- h. Sample cutters
- i. Thermometer
- j. Meter rule

## 2 **PROCEDURE**

## 2.1 Sample preparation

Fresh yams tubers are cleaned from soil material and the full length is measured. Approximately 16% of the full length is cut off from the proximal and distal parts of the yam tuber each. Samples can be obtained from the tubers to represent at least two replications depending on the size of the circumference and length of the tubers and the number of tubers available. In this case, one big tuber was available per genotype. Therefore, after cutting off the proximal and distal parts, we divided the remaining central part into two latitudinal halves. One half was used for replicating 1 and the other half for replicate 2. From each replicate, measure and cut 2.5cm thick cross-section discs from the central part. Use a metal cutter to cut out 62mm x 42mm x 20mm cuboids from each cross-section cuts. The size of the yam sample ensures that it is compressed through the 5-blade extrusion grids of the Ottawa cell. Ideally, 3-5 cuboids are produced for each replicate. After making sure the steamer has produced steam, steam the samples for 23 mins, turning the cuboids to the other side after 12 min of steaming, in order to receive equal steaming periods on both sides of the sample. After steaming, wrap the cuboids in aluminium foil and place in insulated Styrofoam box.



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Figure 1 : Sample preparation workflow

## 2.2 Texture Analysis

The instrumental texture analysis was done using the Stable Microsystems TA-XT*plus* texture analyzer equipped with a 50kg load cell and an Ottawa cell with a 5-blade grid. Before the analysis, force calibration of the texture analyser was conducted using a 50kg load cell and a 5kg standard mass. The boiled yam samples (62mm x 42mm x 20mm) were wrapped in aluminum foil and kept in a Styrofoam box to prevent moisture and temperature loss. The instrumental texture analysis (extrusion test) was conducted by picking a sample, monitoring the surface temperature until it reaches 45°C, and measuring the texture using the Ottawa cell extruder. The test conditions are shown in the Table 1:

5 mm/s
2 mm/s
10 mm/s
0.981 N (or 100g)
95%
45°C
Cuboids, along/parallel to the fibre matrix
5-blade Ottawa grid
50kg
-

Table	1:	Instrumental	texture	test	parameters
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Figure 2 : An example of Ottawa Extrusion curve of boiled yam sample showing texture parameters

## **3 EXPRESSION OF RESULTS**

An exemplary test result shall indicate the method used and the statistical data including mean, standard deviation, standard error, and coefficient of variation. The repeatability of the procedure shall be assessed by non-significant difference (P > 0.05) between two or more replicates, each replicate consisting of 4-5 measurements.

The effect of sample mass or surface area on the value of the texture parameters of boiled yam may be normalized by dividing the value of the texture parameters by the mass or surface area, for example, Max force per g (N/g) or Max force per mm<sup>2</sup> (N/mm<sup>2</sup>).

## 4 CRITICAL POINTS OR NOTE ON THE PROCEDURE

- The texture analyzer must be switched on at least 15 mins before any calibration is done. Calibration must be performed before texture measurement.
- The sample size, shape, dimension, and temperature must be controlled.
- The texture analyzer must be placed on a rigid platform and vibration of the platform must be prevented.
- It must be ensured that there is no friction between the test probe and the Ottawa cell. To ensure this, a quick test without any sample in the Ottawa cell should be conducted and confirmed to register zero resistance prior to actual measurements with sample.
- The Ottawa cell blade grid and probe plate must be cleaned with damp cloth and wiped dry after each measurement.
- The sample orientation must be consistent for all measurements, i.e. along the yam fiber, or across the yam fiber alignment.



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## 5 TEST REPORT

The test report shall include all details necessary for the complete identification for the sample such as date of harvest, plot code, maturity of the crop, laboratory code name, sample name and replication.

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	Genotype	Ν	Mean	Std Dev	Std Err	CV (%)	CV, mean (%)
Gradient (N/mm)*	Oju iyawo	5 19.0		4.135	1.849	21.8	9.7
	Oweigbo	6	9.517	2.073	0.846	21.8	8.9
	TDa1510119	5	10.247	7.851	3.511	76.6	34.3
	TDa1511008	5	5.368	1.146	0.513	21.4	9.5
	TDa1520050	5	9.176	2.362	1.056	25.7	11.5
	TDr1437005	7	18.162	4.225	1.597	23.3	8.8
	TDr1500042	4	8.177	0.975	0.487	11.9	6.0
	TDr1500100	6	9.223	1.996	0.815	21.6	8.8
Max force (N)	Oju iyawo	5	132.825	13.088	5.853	9.9	4.4
	Oweigbo	/	/6.113	8.499	3.212	11.2	4.2
	TDa1510119	5	29.576	9.284	4.152	31.4	14.0
	TDa1511008	5	50.782	15.652	7.000	30.8	13.8
	TDa1520050	6	55.321	7.942	3.242	14.4	5.9
	TDr1437005	5	121.956	7.643	3.418	0.3	2.8
	TDr1500042	0	03.538	12.230	4.993	19.2	7.9
Distance at May famos (mm)		0	55.197	9.399	3.83/	17.0	7.0
Distance at Max force (mm)	Oju iyawo	0	17.230	3.541	1.440	20.0	8.4
		7	15.040	1.207	0.477	11.7	3.0
	TDa1510119	6	15.303	1.731	0.002	0 5	4.3
	TDa1511008	6	16 310	2.065	0.347	12.7	5.0
	TDa1320030	5	17 682	1 716	0.043	9.7	J.2 // 3
	TDr1437003	7	16 600	2 166	0.708	12.0	4.3
	TDr1500042	7	13 500	2.100	0.010	18.2	4.7
End force (N)		5	128 997	13 170	5 890	10.2	4.6
	Oweigho	7	65 730	7 080	2 676	10.2	4.0
	TDa1510119	5	24 254	3 231	1 445	13.3	6.0
	TDa1510117	5	46 138	12 174	5 444	26.4	11.8
	TDa1520050	6	49 246	8 351	3 409	17.0	6.9
	TDr1437005	5	111.356	9.216	4.121	8.3	3.7
	TDr1500042	6	58,719	10.551	4.308	18.0	7.3
	TDr1500100	6	54.000	10.187	4.159	18.9	7.7
Area (N.mm)	Oiu ivawo	5	1640.065	425.664	190.363	26.0	11.6
	Oweigbo	7	1220.088	149.652	56.563	12.3	4.6
	TDa1510119	5	500.968	136.491	61.041	27.2	12.2
	TDa1511008	5	739.629	203.880	91.178	27.6	12.3
	TDa1520050	5	862.478	57.299	25.625	6.6	3.0
	TDr1437005	5	1808.942	108.626	48.579	6.0	2.7
	TDr1500042	5	961.950	135.806	60.734	14.1	6.3
	TDr1500100	4	671.243	109.713	54.857	16.3	8.2
Linear Distance (N.mm)	Oju iyawo	5	153.349	6.928	3.098	4.5	2.0
	Oweigbo	7	103.927	12.969	4.902	12.5	4.7
	TDa1510119	6	59.855	18.290	7.467	30.6	12.5
	TDa1511008	5	70.438	16.911	7.563	24.0	10.7
	TDa1520050	6	77.049	7.516	3.068	9.8	4.0
	TDr1437005	5	145.640	8.637	3.863	5.9	2.7
	TDr1500042	7	76.252	17.273	6.529	22.7	8.6
	TDr1500100	6	65.584	9.686	3.954	14.8	6.0

Table 2 : Exemplary descriptive analysis of the textural parameters of boiled yam

\*Gradient at 3mm distance



## **APPENDICES**

# Annex 1: One-way analysis of variance (ANOVA) of extrusion texture by genotype

The extrusion texture parameters were all significant in discriminating among the boiled yam texture. The most discriminant textural parameters were the End force (which represents the resistance to deformation at 95% strain, at the end of the extrusion process.  $P = 3.5 \times 10^{-19}$ ) and Max force (which represents the hardness or maximum resistance to deformation of the boiled yam.  $P = 4.7 \times 10^{-18}$ ). The least discriminant textural parameters were the Distance at Max force (p = 0.045).

Generally, the genotypes Oju iyawo, TDr1437005 and Oweigbo recorded higher values of texture, while genotypes TDa1511008 and TDa1510119 recorded the least values of texture.

Source	DF	Sum of	Mean	F Ratio	Prob > F
		Squares	Square		
Gradient	7	913.8262	130.547	9.2504	1.957e-6
Max force	7	46133.831	6590.55	58.3121	4.66e-18
Distance at Max force	7	73.52629	10.5038	2.2925	4.457e-2
End force	7	43677.067	6239.58	68.0381	3.45e-19
Area	7	7587040.8	1083863	28.1603	3.6e-12
Linear distance	7	49989.025	7141.29	40.6123	5.66e-16

#### Analysis of Variance

#### Max force Connecting Letters Report

Level					Mean
Oju iyawo	Α				132.82480
TDr1437005	А				121.95600
Oweigbo		В			76.11286
TDr1500042			С		63.53750
TDa1520050			С		55.32083
TDr1500100			С		55.19733
TDa1511008			С		50.78220
TDa1510119				D	29.57600

Levels not connected by same letter are significantly different.

#### **End force Connecting Letters Report**

Level							Mean
Oju iyawo	Α						128.99740
TDr1437005		В					111.35640
Oweigbo			С				65.73014
TDr1500042			С	D			58.71867
TDr1500100				D	Е		54.00017
TDa1520050				D	E		49.24600
TDa1511008					E		46.13780
TDa1510119						F	24.25360

Levels not connected by same letter are significantly different.

## **Annex 2: Repeatability of texture analysis**

All the textural parameters were repeatable (P > 0.05) except Distance at Max force.

#### Exemplary Oneway Anova of texture parameters by Replicate

Source	DF	Sum of Squares	Mean Square	F Ratio	Prob > F
Gradient	1	31.8858	31.8858	0.9502	3.354e-1
Max force	1	587.369	587.37	0.5079	4.799e-1
Distance at Max force	1	34.51172	34.5117	7.1647	1.009e-2
End force	1	422.227	422.23	0.3892	5.36e-1
Area	1	274502.5	274502	1.2473	2.709e-1
Linear distance	1	573.762	573.76	0.4588	5.016e-1

#### **Analysis of Variance**

## **Annex 3: Hierarchical Clustering**

Based on a hierarchical clustering analysis of the textural parameters, the genotypes were categorized into three groups: Oju iyawo (a *rotundata* check landrace) and TDr1437005 were identified as genotypes with highest values of texture. Oweigbo (a check *alata* landrace), TDa1511008, TDa11520050, and TDr1500042 were grouped as genotypes with intermediate values of texture. TDa1510119 and TDr1500100 were clustered together as genotypes with the least texture values for boiled yams.



Hierarchical clustering of the boiled yam genotypes

