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Pitout JDD, Church DL, Gregson DB, Chow BL, McCracken M, Mulvey M, Laupland KB (2007). Molecular epidemiology of CTXM-producing *Escherichia coli* in the Calgary Health Region: emergence of CTX-M-15-producing isolates. *Antimicrob. Agents Chemother.* 51: 1281-1286.

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## Full Length Research Paper

# Beverage quality and biochemical attributes of arabusta coffee (*C. arabica* L. x *C. canephora* Pierre) and their parental genotypes

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Arabica coffee (*Coffea arabica* L.) is known for the production of high quality beverage while Robusta coffee (*Coffea canephora* Pierre) has been characterized as a neutral, weak flavored and occasionally with strong acid and pronounced bitterness. Viable and reasonably fertile interspecific hybrids can easily be obtained from crosses between the allotetraploid *C. arabica* L. and induced autotetraploid forms of *C. canephora* P. This study was carried out to determine beverage quality characteristics and biochemical components of 15 coffee genotypes, nine of them being interspecific Arabusta F<sub>1</sub> hybrids. Beverage quality was determined by a panel of six judges using the prescribed sensory evaluation procedures, while caffeine, oil, trigonelline, total chlorogenic acids (CGA) and sucrose were analyzed in green coffee samples using recommended methodologies. The results indicated significant ( $p < 0.05$ ) variations among the genotypes for all the sensory attributes. The total score, which is a reflection of the broad coffee quality performance showed that SL34 and SL28 (which served as reference in sensory quality), were not significantly different from Arabusta hybrids SL34 x UT8, SL28 x UT8, N39 x UT8, SL34 x UT6, CaturraxUT6 and SL28 x UT6. The quality of some Arabusta hybrids was found to be similar to that of pure Arabica genotypes. Similarly, biochemical variables revealed significant ( $p < 0.05$ ) variations for caffeine, oil and sucrose, among genotypes except for CGA and trigonelline which were not significantly different. There were positive significant correlations between all the sensory characteristics. Sucrose showed significant ( $P < 0.05$ ) correlations with fragrance flavour, aftertaste and overall. Trigonelline showed a significant negative correlation with body and caffeine. All the Arabusta hybrids scored specialty grade (80 points and above for total score) and therefore future studies on their performance in many locations with more variable climatic conditions is recommended.

**Key words:** Coffee, Arabusta, F<sub>1</sub> interspecific hybrids, sensory variables, biochemical components.

## INTRODUCTION

Coffee belongs to the genus *Coffea* in the Rubiaceae family that contains 640 genera and 1000 species

(Charrier and Berthaud, 1985). *Coffea arabica* L. commonly referred to as Arabica coffee, is tetraploid

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( $2n = 4x = 44$ ) and is known for the production of high quality beverage (Gichuru et al., 2008; Kathurima et al., 2009; Gichimu and Omondi, 2010). *Coffea canephora* Pierre, also referred to as Robusta coffee, is diploid ( $2n = 2x = 22$ ) and generally self-incompatible (Combes et al., 2000; Lashermes et al., 2011). Robusta coffee has been characterized as a neutral, weak flavored and occasionally with strong acid and pronounced bitterness (Bertrand et al., 2003). Robusta coffee is less susceptible to pests than Arabica coffee (Tshilenge et al., 2009)

The transfer of desirable genes particularly for disease resistance from diploid species like *C. canephora* and *C. liberica* into tetraploid *C. arabica* cultivars without affecting quality traits has been a major objective of Arabica coffee breeding. However, the ploidy level differences between the tetraploid *C. arabica* and other diploid coffee species has been a major bottleneck for interspecific gene transfer (Ky et al., 2001a).

Viable and reasonably fertile interspecific hybrids between *C. arabica* and various diploid species including *C. canephora* have been successfully produced (Lashermes et al., 2011). Such hybrids have been produced through crosses between the allotetraploid *C. arabica* with induced autotetraploid forms of *C. canephora* obtained through doubling of the chromosome number by colchicine treatment (Owuor and Van Der Vossen, 1981). The first successful interspecific hybrids between induced tetraploid *C. canephora* and *C. arabica* were made in Brazil in 1950. Those hybrids have been used in coffee breeding programs to introgress genes for resistance to coffee Coffee Leaf Rust (*Hemileia vastatrix*) and Coffee Berry Disease (*Colletotrichum kahawae*) from *C. canephora* into *C. arabica* or to improve the quality of Robusta coffee by direct use of the  $F_1$  Arabusta hybrids (Owuor and Van der Vossen, 1981). New Arabica coffee cultivars with better quality, higher yield potential and resistance to diseases have started to replace the traditional varieties on a large scale in several countries (Gichimu and Omondi, 2010). Arabica coffee plants have a narrow genetic base attributed to the few seeds/plants used for dissemination, successive genetic reduction due to human impacts and reproduction nature of Arabica coffee which is autogamous (Teressa et al., 2010). Reduced genetic diversity is reported to compromise the ability of populations to evolve so that they can cope up with environmental changes and thus reducing their chances of long-term persistence (Frankham et al., 2002). As for many other crops, evaluation of the genetic diversity and available resources within the genus *Coffea* is an important step in coffee breeding (Cubry et al., 2008).

Coffee is one of the most popular beverages consumed all over the world with a total annual consumption of coffee exceeding 400 billion cups (Nebesny and Budryn, 2006). Sensory assessment is one of the methods used in identifying the market acceptability especially in food or drink based products. It is also used for product develop-

ment and improvement as most important factors for a particular market can be identified and improved (Lazim and Suriani, 2009). For coffee, quality of liquor also referred to as beverage quality, determines the desirability of coffee for consumption purposes and acts as a yardstick for price determination (Agwanda et al., 2003; Kathurima et al., 2009; Gichimu et al., 2012). Production and supply of coffee with excellent quality is important for coffee exports and success of a new coffee variety depends to a great extent on its bean and beverage quality (Gichimu et al., 2012). Different levels of biochemical components in coffee contribute variously to the final quality of the cup (Buffo and Freire, 2004). The presence of those biochemical components could have a favorable effect on the coffee beverage quality, as for trigonelline and sugars, or an unfavorable one, as for chlorogenic acids and caffeine (Clifford, 1985; Macrae, 1985). Trigonelline is considered to be important for both taste and nutrition (Ky et al., 2001a). Coffee oil carries most of the coffee aroma and contributes to brew viscosity (Buffo and Freire, 2004). This study was carried out with an aim of assessing the variation of cup quality traits and biochemical components of 15 coffee genotypes (including 9 interspecific Arabusta  $F_1$  hybrids). The study also aimed at determining the suitability of the interspecific  $F_1$  Arabusta in the improvement of the beverage quality of Robusta coffee or for further selection and use as coffee varieties.

## MATERIALS AND METHODS

### Description of the study site

The study was conducted at the Coffee Research Station (CRS), Ruiru, about 33 km North of Nairobi. CRS lies within the upper midland (UM2) at latitude  $1^\circ 06'S$  and longitude  $36^\circ 45'E$  and is approximately 1620 m above sea level. The area receives a bimodal rainfall of 1063 mm annually with mean temperature of  $19^\circ C$  (min.  $12.8^\circ C$ , max.  $25.2^\circ C$ ). The soils are classified as complex humic nitisols and plinthic ferrasol (Jaetzold and Schmidt, 1983). They are well drained, deep, reddish brown, slightly friable clays with murram sections occasionally interrupting. The soil pH ranges from 5 to 6 (Jaetzold and Schmidt, 1983).

### Test materials

The coffee genotypes in this study comprised of nine interspecific Arabusta  $F_1$  hybrids, two commercial arabica coffee varieties (namely SL 28 and SL 34), two museum accessions (N39 and Caturra), Hibrido De Timor (HDT), a natural Arabusta accession and a diploid Robusta variety (Table 1). SL 28 and SL 34 served as reference in quality evaluation. The  $F_1$  hybrids are interspecific crosses between four Arabica and four induced tetraploid Robusta accession. Induced tetraploid Robusta accessions (ex Fr.) were introduced from Uganda. These genotypes are conserved by Coffee Research Foundation (CRF) at the main station, Coffee Research Station (CRS) and Oakland Estate. Cherry samples for analysis were collected during the peak harvesting period of October to December, 2012. Ripe healthy berries were harvested from each of the genotypes and processed using wet processing

**Table 1.** List of coffee genotypes evaluated for sensory and biochemical components.

Genotype	Status	Introduced from
SL28 x UT3	Arabusta F1 Hybrid	Kenya
SL28 x UT6	Arabusta F1 Hybrid	Kenya
SL34 x UT6	Arabusta F1 Hybrid	Kenya
N39 x UT6	Arabusta F1 Hybrid	Kenya
Caturra x UT6	Arabusta F1 Hybrid	Kenya
SL28 x UT8	Arabusta F1 Hybrid	Kenya
SL34 x UT8	Arabusta F1 Hybrid	Kenya
N39 x UT8	Arabusta F1 Hybrid	Kenya
SL28 x UT10	Arabusta F1 Hybrid	Kenya
SL34	Commercial variety	Kenya
SL28	Commercial variety	Kenya
N39	Museum accession	Lyamungu Tanzania
Caturra	Museum accession	Brazil
Hibrido De Timor	Natural Arabusta	Portugal
Robusta	Diploid Robusta	Uganda

**Table 2.** Descriptors used by the sensory panel to describe the sensory properties of the coffee samples.

Scale	Attribute	Word anchor
1 - 10	Fragrance/Aroma	Very poor - Outstanding
1 - 10	Flavour	Very poor - Outstanding
1 - 10	Aftertaste	Very poor - Outstanding
1 - 10	Balance	Very poor - Outstanding
1 - 10	Preference	Very poor - Outstanding
1 - 10	Acidity	Very flat - Very bright
1 - 10	Body	Very thin - Very heavy

procedures (Mburu, 2004). The cherry samples were pulped, fermented, washed, wet parchment dried to final moisture content of 10.5-11% and then hulled to produce green coffee beans for analysis (Kathurima et al., 2010). The laboratory experiment was carried out in a complete randomized design in six replicates each representing a judge.

#### Roasting and sensory analysis

The green coffee beans were roasted within 24 h of evaluation, in order to ensure a fresh brew, using a laboratory roaster (Probat BRZ 4, Rhein, Germany). The beans were roasted to a medium level roast and allowed to rest for at least eight hours. The roasted samples were ground individually (five cups per sample), using a sample grinder (Probat vtv-633T, Rhein Germany), not more than 15 min before infusion with water. The samples were weighed out to the predetermined ratio of 8.25 g per 150 ml of water. Sensory evaluation was conducted using the procedures described by Lingle (2001). Seven sensory variables namely; fragrance/aroma, flavour, aftertaste, acidity, body, balance and overall were assessed and scored together with three process control variables (uniformity, clean cup and sweetness) by a panel of six trained cuppers on a 10-point scale whose descriptors are as Table 2.

Fragrance is the smell of the ground coffee when still dry and aroma is the smell of the coffee when infused with hot water while

aftertaste are vapors remaining after the coffee is swallowed (Lingle, 2001). Balance is the assessment of how well the flavour, aftertaste, acidity and body fit together in a synergistic combination (Kathurima, 2013). The attribute overall, is a reflection of the panelists personal appraisal based on the holistically integrated rating of the sample as perceived by the individual panelist (Kathurima, 2013). All the sensory parameters (including the three process control parameters) were added together to constitute the total score which was a reflection of the broad coffee quality performance. On the basis of scores obtained, the coffee was classified into either specialty grade (80-100 points) or commercial grade (79 points and below) (Specialty Coffee Association of America, SCAA).

#### Determination of coffee biochemical components

The genotypes were analyzed for five attributes namely, caffeine, trigonelline, oils, sucrose and chlorogenic acids (CGA). Portions of the green coffee samples were placed in small plastic bottles and stored under -80°C. After 24 h of freezing, the samples were ground in liquid nitrogen using an analytical mill (Model A10, IKA work inc. Wilmington, NC, USA). Caffeine, trigonelline and CGA were extracted from green coffee powder by refluxing in distilled water. Caffeine, trigonelline and CGA were analysed using a HPLC system (KNEUR) equipped with a Supel Co. discovery diode array detector at three wavelengths, 278 nm for caffeine, 266 nm for trigonelline and 324 nm for CGA. Sucrose was extracted from green coffee powder using the method of Osborne and Voogt (1978). Sucrose was analysed using a HPLC system (KNEUR) equipped with a Eurospher 100-5 NH<sub>2</sub> column and a refractive index detector. Caffeine, trigonelline CGA and sucrose were identified by comparing the retention times of standards and their concentrations calculated from peak areas using calibration equations. Coffee oil was analysed as outlined in the AOAC (1995). The laboratory experiment was carried out in a complete randomized design (CRD) in four replicates each representing different extraction time.

#### Data analysis

Sensory and biochemical data were subjected to analysis of

**Table 3.** Mean sensory characteristics and specialty classification of fifteen coffee genotypes evaluated in this study.

Genotype	Sensory variables							Mean total score
	Fragrance	Flavour	Aftertaste	Acidity	Body	Balance	Overall	
SL28	7.8 <sup>a</sup>	8.04 <sup>a</sup>	7.88 <sup>ab</sup>	8.17 <sup>a</sup>	7.79 <sup>a</sup>	7.79 <sup>a</sup>	8.04 <sup>a</sup>	85.54
SL34	7.8 <sup>a</sup>	8.08 <sup>a</sup>	7.96 <sup>a</sup>	8.13 <sup>a</sup>	7.75 <sup>a</sup>	7.79 <sup>a</sup>	7.92 <sup>ab</sup>	85.46
CaturraUT8	7.7 <sup>ab</sup>	7.58 <sup>ab</sup>	7.54 <sup>abc</sup>	7.46 <sup>abc</sup>	7.46 <sup>ab</sup>	7.50 <sup>ab</sup>	7.38 <sup>bcdef</sup>	83.83
N39 x UT8	7.7 <sup>ab</sup>	7.63 <sup>ab</sup>	7.63 <sup>abc</sup>	7.63 <sup>abc</sup>	7.75 <sup>a</sup>	7.58 <sup>ab</sup>	7.67 <sup>abcd</sup>	83.67
SL34 x UT6	7.7 <sup>ab</sup>	7.67 <sup>ab</sup>	7.63 <sup>abc</sup>	7.63 <sup>abc</sup>	7.54 <sup>ab</sup>	7.63 <sup>ab</sup>	7.54 <sup>abcde</sup>	83.54
CaturraUT6	7.6 <sup>ab</sup>	7.71 <sup>ab</sup>	7.63 <sup>abc</sup>	7.58 <sup>abc</sup>	7.58 <sup>ab</sup>	7.63 <sup>ab</sup>	7.63 <sup>abcd</sup>	83.38
SL28 x UT8	7.6 <sup>ab</sup>	7.67 <sup>ab</sup>	7.79 <sup>abc</sup>	7.79 <sup>abc</sup>	7.50 <sup>ab</sup>	7.63 <sup>ab</sup>	7.67 <sup>abcd</sup>	83.29
SL28 x UT6	7.5 <sup>abc</sup>	7.54 <sup>abc</sup>	7.54 <sup>abc</sup>	7.58 <sup>abc</sup>	7.71 <sup>a</sup>	7.50 <sup>abc</sup>	7.50 <sup>abcde</sup>	82.92
SL34 x UT8	7.5 <sup>abc</sup>	7.67 <sup>bc</sup>	7.79 <sup>abc</sup>	7.83 <sup>ab</sup>	7.63 <sup>a</sup>	7.63 <sup>ab</sup>	7.75 <sup>abc</sup>	82.17
N39 x UT6	7.5 <sup>abcd</sup>	7.46 <sup>abc</sup>	7.29 <sup>bcd</sup>	7.33 <sup>bcd</sup>	7.46 <sup>ab</sup>	7.42 <sup>abcd</sup>	7.33 <sup>cdef</sup>	81.75
SL28 x UT3	7.4 <sup>abcd</sup>	7.17 <sup>bc</sup>	7.38 <sup>abcd</sup>	7.17 <sup>bcd</sup>	7.38 <sup>ab</sup>	7.04 <sup>d</sup>	7.17 <sup>def</sup>	81.33
N 39	7.3 <sup>abcd</sup>	7.50 <sup>abc</sup>	7.46 <sup>abc</sup>	7.58 <sup>abc</sup>	7.50 <sup>ab</sup>	7.42 <sup>abcd</sup>	7.38 <sup>bcdef</sup>	80.67
Caturra	7.1 <sup>bcd</sup>	6.58 <sup>d</sup>	6.88 <sup>d</sup>	6.88 <sup>de</sup>	7.00 <sup>b</sup>	7.08 <sup>cd</sup>	6.96 <sup>f</sup>	79.96
Robusta	7.0 <sup>cd</sup>	6.54 <sup>d</sup>	6.88 <sup>d</sup>	6.58 <sup>e</sup>	7.42 <sup>ab</sup>	6.71 <sup>e</sup>	6.54 <sup>g</sup>	78.50
HDT	6.96 <sup>d</sup>	7.00 <sup>c</sup>	7.17 <sup>cd</sup>	7.08 <sup>cde</sup>	7.46 <sup>ab</sup>	7.25 <sup>bcd</sup>	7.04 <sup>ef</sup>	77.04
LSD(p<0.05)	0.260	0.337	0.335	0.392	0.295	0.259	0.316	
P-value	< 0.0001	< 0.0001	< 0.0001	< 0.0001	0.012	< 0.0001	< 0.0001	

Means along a column not sharing the same letter are significantly different (P<0.05) using Student-Newman-Keuls test. HDT- Hibrido De Timor. The means for total score were not separated as they were derived from additions.

**Table 4.** The first two principle components (PC) of the seven sensory variables.

Variable	PC1	PC2
Fragrance/aroma	0.371	-0.181
Flavour	0.395	-0.019
Aftertaste	0.392	0.001
Acidity	0.388	-0.175
Body	0.318	0.926
Balance	0.383	-0.223
Overall	0.393	-0.17
Eigen value	6.309	0.384
Variability (%)	90.133	5.489
Cumulative (%)	90.133	95.623

variance (ANOVA) using COSTAT statistical software. Student-Newman-Keuls (SNK5%) test was used to separate the means at 5% level of significance. Multivariate analyses was done using principle component analysis (PCA) for both sensory quantitative variables and biochemical components the characteristics plotted for important principle components using XLSTAT statistical software, version 2012.

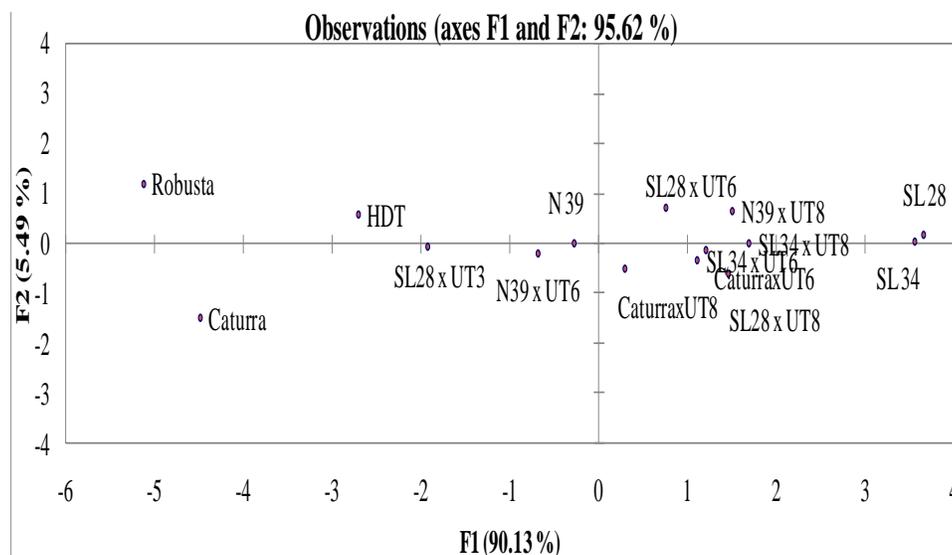
Sweetness refers to a pleasing fullness of flavor as well as any obvious sweetness and its perception is the result of the presence of certain carbohydrates. Clean cup refers to a lack of interfering negative impressions from first ingestion to final aftertaste. Any non-coffee like tastes or aromas will disqualify an individual cup. Two (2) points are awarded for each cup displaying the attribute of clean Cup. Uniformity refers to consistency of flavor of the different cups of the sample tasted. If the cups taste different, the rating of this aspect would not be as high. Two (2) points are awarded for each

cup displaying this attribute, with a maximum of 10 points if all 5 cups are the same. To get the total score, the scores of sweetness, clean cup and uniformity were added to the scores of the other attributes.

## RESULTS

### Diversity of coffee genotypes as determined by sensory variables

A total of 15 genotypes were evaluated for sensorial attributes. The results indicated highly significant (p<0.05) variation among the genotypes for all the sensory attributes studied (Table 4). The commercial varieties SL28 and SL34 got the highest scores in all the sensory variables studied, though they were not significantly different from the Arabusta hybrids for several traits. All the sensory attributes attained scores above 6.5 meaning they were all rated as good by the panel of assessors. Robusta and Caturra were significantly (p<0.05) different from the other genotypes in flavour and acidity. The overall scoring aspect is meant to reflect the holistically integrated rating of the sample as perceived by the individual panelist. Robusta showed the lowest score in terms of balance, acidity, flavor and overall. All interspecific F<sub>1</sub> Arabusta hybrids genotypes were characterized with bitterness as was Robusta and HTD. All the genotypes scored a maximum of 10 points for each of the variables clean cup, sweetness and uniformity, which were added to the scores of the other sensory variables to classify the coffee as specialty grade (80 to 100 points) or commercial grade (79 and below).



**Figure 1.** Principle component (PC) analysis plot of first two principle components, illustrating relationship among the coffee genotypes assessed for sensory attributes.

The mean total scores of the coffee genotypes in this study is shown in Table 3. SL 28 and SL34 scored the highest but similar to all the interspecific Arabusta  $F_1$  hybrids (specialty quality). Robusta, Caturra and HDT scored the lowest (77.04, 78.50 and 79.96 respectively), scores that were below specialty quality.

Sensory data was subjected to principle component analysis (PCA). PCA results indicated that the first two principle components explained 95.62% (PC1 90.13% and PC2 5.49%) of the total variation (Figure 1). All the sensory attributes contributed almost equally to PC1 while body contributed the most to the variations observed in PC2 (Table 4). The genotypes Robusta, Caturra, HDT, SL28xUT3, N39xUT6 and N39 were placed in the negative side of PC1 while all the other genotypes were placed in the positive side of PC1. The genotypes placed in the negative side of PC1 were characterized by having lower beverage quality (Figure 1).

#### Diversity of coffee genotypes as determined by biochemical components

The coffee genotypes showed significant ( $p < 0.05$ ) differences in the levels of caffeine, oil and sucrose while CGA and trigonelline did not show significant differences among the genotypes (Table 5). Robusta recorded the highest percentage of caffeine content (2.39%). All the interspecific  $F_1$  Arabusta hybrids recorded average to high caffeine content ranging from 1.98 to 2.25% except SL28 x UT8 and SL28 x UT6 (which recorded caffeine content of 1.53 and 1.77% respectively). Arabica genotypes recorded low caffeine content among all the genotypes with SL34 recording the lowest at 1.09% and

Caturra with the highest at 1.65%. All the Arabica genotypes recorded significantly ( $p < 0.05$ ) higher oil contents than Robusta and the interspecific  $F_1$  Arabusta hybrids except SL28 x UT8 whose oil content was similar to Arabica. Robusta recorded the lowest oil content of 13.39%. Similarly, Robusta showed the lowest content of sucrose (5.75%) while UT6 x SL34 accession recorded the highest amount (9.99%).

The data of the five biochemical components analyzed for the 15 coffee genotypes was subjected to principle component analysis (PCA). The first three principle components explained 84.870% (38.86, 25.59 and 20.43%) of the total variation respectively (Table 6). Six coffee genotypes, (SL34, SL28, Caturra, N39, UT8 x SL28 and HDT) were placed in the positive side of PCA graph while the other nine genotypes Robusta, SL28 x UT3, SL34 x UT6, Caturra x UT6, SL34 x UT8, SL28 x UT6, Caturra x UT8, N39 x UT6, and N39 x UT8 were placed in the negative side of the PCA graph. The genotypes in the negative side of the PC plot were characterized by high caffeine content while those in the positive were characterized by high oil contents (Figure 2). The biochemical components contributed differently to the variations observed in PC1, PC2 and PC3 (Table 6).

#### Correlation coefficients between sensory and biochemical variables of the coffee genotypes

The results indicated significant ( $p < 0.001$ ) positive correlations among all the cup quality traits (Table 7). Sucrose showed significant ( $P < 0.05$ ) correlations with fragrance flavour, aftertaste and overall. Trigonelline showed a significant negative correlation with body and

**Table 5.** Mean caffeine, trigonelline, oil, sucrose and total chlorogenic acids (CGA) % dry weight basis (DWB) for 15 coffee genotypes in this study.

Genotype	Biochemical components				
	Caffeine	Trigonelline	Oil	CGA	Sucrose
Robusta	2.4 <sup>a</sup>	1.2 <sup>a</sup>	13.4 <sup>b</sup>	4.7 <sup>a</sup>	5.8 <sup>e</sup>
SL34x UT8	2.2 <sup>b</sup>	1.2 <sup>a</sup>	15.2 <sup>b</sup>	5.47 <sup>a</sup>	7.95 <sup>cd</sup>
SL28 xUT3	2.2 <sup>b</sup>	1.6 <sup>a</sup>	14.5 <sup>b</sup>	5.55 <sup>a</sup>	9.91 <sup>ab</sup>
Caturra x UT8	2.1 <sup>b</sup>	1.05 <sup>a</sup>	13.9 <sup>b</sup>	5.52 <sup>a</sup>	8.96 <sup>abc</sup>
Caturra x UT6	2.0 <sup>b</sup>	1.15 <sup>a</sup>	14.7 <sup>b</sup>	5.36 <sup>a</sup>	6.92 <sup>d</sup>
UT6 x N39	2.0 <sup>b</sup>	1.38 <sup>a</sup>	14.56 <sup>b</sup>	5.25 <sup>a</sup>	7.47 <sup>d</sup>
UT6 x SL34	2.0 <sup>b</sup>	1.16 <sup>a</sup>	14.17 <sup>b</sup>	4.96 <sup>a</sup>	9.99 <sup>a</sup>
UT8 x N39	2.0 <sup>b</sup>	1.18 <sup>a</sup>	15.25 <sup>b</sup>	5.90 <sup>a</sup>	8.94 <sup>abc</sup>
UT6 x SL28	1.8 <sup>c</sup>	1.14 <sup>a</sup>	14.32 <sup>b</sup>	5.74 <sup>a</sup>	8.67 <sup>bc</sup>
Caturra	1.6 <sup>cd</sup>	1.46 <sup>a</sup>	19.43 <sup>a</sup>	5.13 <sup>a</sup>	7.61 <sup>d</sup>
UT8 x SL28	1.5 <sup>de</sup>	1.44 <sup>a</sup>	18.26 <sup>a</sup>	5.01 <sup>a</sup>	8.71 <sup>bc</sup>
N39	1.4 <sup>ef</sup>	1.32 <sup>a</sup>	18.13 <sup>a</sup>	5.65 <sup>a</sup>	7.44 <sup>d</sup>
SL28	1.2 <sup>fg</sup>	1.19 <sup>a</sup>	18.53 <sup>a</sup>	5.22 <sup>a</sup>	8.67 <sup>bc</sup>
HDT	1.1 <sup>g</sup>	1.19 <sup>a</sup>	18.49 <sup>a</sup>	5.10 <sup>a</sup>	7.16 <sup>d</sup>
SL34	1.1 <sup>g</sup>	1.13 <sup>a</sup>	18.27 <sup>a</sup>	4.96 <sup>a</sup>	9.18 <sup>abc</sup>
LSD (P<0.05)	0.054	0.422	0.502	0.805	0.746
P-value	< 0.0001	0.400NS	< 0.0001	0.200NS	< 0.0001

Means within a column not sharing a letter are significantly different at P<0.05.

**Table 6.** The first three principle components (PC) of the five biochemical variables.

Variable	PC1	PC2	PC3
Caffeine	-0.660	-0.132	0.305
Trigonelline	0.152	0.200	0.936
Oil	0.693	0.067	0.026
CGA	-0.246	0.626	-0.163
Sucrose	-0.014	0.739	-0.063
Eigenvalue	1.943	1.279	1.021
Variability (%)	38.857	25.586	20.428
Cumulative (%)	38.857	64.442	84.870

CGA: Total chlorogenic acids.

caffeine.

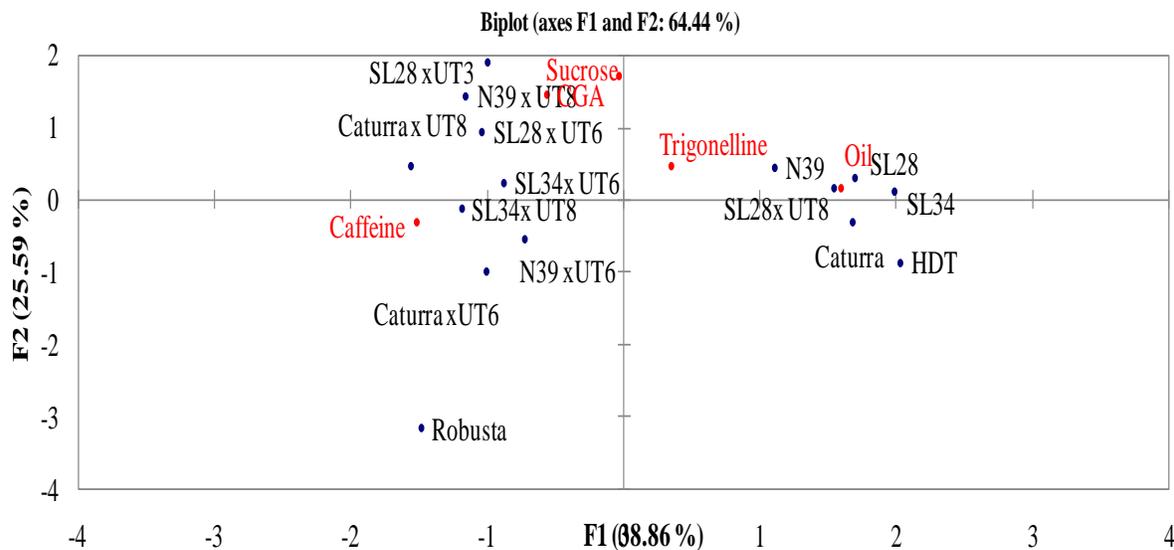
## DISCUSSION

The results indicated significant ( $p<0.05$ ) variation among the 15 coffee genotypes in this study for sensory attributes. This is an indication of high genetic variation among the genotypes for all sensory traits. This was in agreement with previous findings reported by Dessalegn et al. (2008), Kathurima et al. (2009), Kathurima et al. (2010) and Tessema et al. (2011). This result partly agrees with Gichimu et al. (2012) who reported significant ( $p<0.05$ ) variations in all the sensory traits except body in

34 Ruiru 11 sibs. Coffees graded according to the Specialty Coffee Association (SCAA) grading system should, receive more than 80 points in total score to qualify as specialty (Lingle, 2001). All the Arabusta hybrids, SL28, SL34 and N39 attained an overall sensory score of above 80 and were therefore of specialty grade. This was in agreement with Owuor (1988), who reported that introgressed lines were found to produce good beverage quality similar to non-introgressed standards. Robusta, HDT and Caturra attained 77.71, 79.96 and 78.50 points respectively that were below the specialty grade. Gichimu et al. (2012) reported a total mean of 82 points for all the sensory traits for Ruiru 11 sibs. Although the introgressed genes in Arabusta hybrids did affect the beverage quality, undesirable effect (bitterness) often associated with introgressed segments (Bertand et al., 2003) from Robusta genome, was picked out by the judges among the hybrids.

Van der Vossen (1985) recommended overall standard as the best cup quality selection trait due to its high heritability. On the other hand, based on correlation, repeatability and sensitivity analyses, Agwanda (1999) recommended flavour rating as the best selection criterion for genetic improvement of cup quality in Arabica coffee. However, this study showed that all the sensory variables analyzed in this study using trained panel of tasters were important in determining the overall quality of a coffee.

The 15 coffee genotypes recorded highly significant ( $p<0.05$ ) differences for caffeine, oils and sucrose while



**Figure 2.** Principle component (PC) analysis plot of first two principle components, illustrating relationship among the coffee genotypes assessed for biochemical components.

**Table 7.** Correlation coefficients between sensory and biochemical variables of the coffee genotypes.

Variable	Fragrance	Flavour	Aftertaste	Acidity	Body	Balance	Overall	Caffeine	Trigonelline	Oil	CGA
Flavour	0.92**										
Aftertaste	0.90**	0.97**									
Acidity	0.88**	0.97**	0.97**								
Body	0.68**	0.80**	0.79**	0.76**							
Balance	0.86**	0.95**	0.91**	0.95**	0.69**						
Overall	0.90**	0.96**	0.96**	0.98**	0.74**	0.96**					
Caffeine	-0.08	-0.3	-0.28	-0.42	-0.19	-0.39	-0.36				
Trigonelline	-0.34	-0.39	-0.34	-0.33	-0.58*	-0.43	-0.31	0.05			
Oil	-0.09	0.05	0.09	0.25	0.14	0.2	0.21	-0.86**	0.25		
CGA	0.24	0.27	0.23	0.23	0.27	0.24	0.29	0.14	-0.03	-0.19	
Sucrose	0.64*	0.54*	0.58*	0.53*	0.29	0.48	0.54*	0.1	0.08	-0.03	0.29

\*\*Correlation significant at the 0.01 level; \*Correlation significant at the 0.05 level.

trigonelline and CGA did not show significant differences. This result was in agreement with Tessema et al. (2011) and in partial agreement with Anthony et al. (1993) and Kathurima et al. (2010). Anthony et al. (1993), studied biochemical diversity on genus *Coffea* L. using HPLC analyses to determine the contents of caffeine and chlorogenic acids (CGA) and reported highly significant differences for caffeine content within species variation. This study indicated significant variations within Arabica and Arabusta hybrids for caffeine content.

Kathurima et al. (2010) reported genotype effect factors on the levels of total chlorogenic acids (CGA) and caffeine but no significant ( $p < 0.05$ ) differences were observed in the levels of trigonelline and oils among the composite Ruiru 11 hybrids. This partly agrees with the

result from this study as there was significant variation among the interspecific  $F_1$  Arabusta hybrids for caffeine, oils and sucrose. This result is also in agreement with Tessema et al. (2011) who studied variability and association of biochemical attributes in *C. arabica* germplasm collection and reported that the performance of all the study genotypes were highly significant ( $p < 0.01$ ) for caffeine and oils.

Correlations between coffee cup quality and some chemical attributes may be used as an additional tool for coffee quality evaluation (Farah et al., 2006). There were positive significant correlations between all the sensory characteristics. Sucrose showed significant ( $P < 0.05$ ) correlations with fragrance, flavour, aftertaste and overall. Trigonelline showed a significant negative correlation with

body and caffeine. The content and nature of sugars in the green coffee beans is important in the development of flavour and pigmentation during roasting. Sucrose is the main contributor of reducing sugars which are implicated in Maillard reactions occurring during the roasting process (Grosch, 2001). As the most abundant, sucrose acts as aroma precursors that affect both taste and aroma of the beverage (Maria et al., 1994). Higher sucrose contents in Arabica green beans have been shown to partially explain its better cup quality (Ky et al., 2001b). Trigonelline is a pyridine alkaloids that has been associated with flavor formation in coffee during roasting. Trigonelline negatively correlated with caffeine, that is, high caffeine values were accompanied by low trigonelline values and vice versa, indicating a close but competing linkage of the two pathways (Baumann, 2006).

## Conclusion

The study demonstrated the existence of a high diversity in cup quality among all the genotypes studied. The interspecific  $F_1$  Arabusta hybrids demonstrated variation for all the sensory attributes with a total mean score of >80 points, a Specialty Quality as per the SCAA green coffee classification chart. All the interspecific  $F_1$  Arabusta hybrids produced results comparable to commercial varieties SL28 and SL34. The study also indicated significant diversity in biochemical traits among the genotypes for caffeine, oils and sucrose and no variation for CGA and trigonelline.

## Recommendation

Future studies should be done to test the performance of the Arabusta hybrids in many locations with more variable climatic conditions and seasons.

## Conflict of Interests

The authors did not declare any conflict of interests.

## ACKNOWLEDGEMENTS

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*Full Length Research Paper*

# Homogenization of milk and its effect on sensory and physico-chemical properties of yoghurt

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**This study was designed to evaluate the chemical composition, sensory properties and microbial load of differently homogenized milk for yoghurt-making. Milk was homogenized with a hand whisker (HW), pressure sprayer (PS) and high-speed mixer (HM) while the control was not homogenized (NH) prior to yoghurt-making. Samples were stored in a refrigerator for 10 days at 4°C and thereafter examined for microbial counts using pour plate technique. Results show that homogenization had no significant ( $p > 0.05$ ) influence on taste and flavor of yoghurt. The chemical composition of the yoghurt samples in all the treatment groups were improved as the days in storage progressed. The highest total viable count (TVC), coliform and fungal counts were obtained with NH while PS recorded the least counts.**

**Key words:** Chemical composition, sensory properties, microbial load, homogenizer and yoghurt.

## INTRODUCTION

Yoghurt is one of the most popular fermented dairy products widely consumed all over the world and its consumption has increased considerably since the 1970s to the present decade (Deeth and Tamime, 1981; Hassan and Amjad, 2010) due to its perceived health benefits. Ayebo and Shahani (1980) reported that fermented dairy products are more nutritious than the milk from which they are made. Furthermore, the higher nutritional value of these products has been attributed to the increased production of certain nutrients and to the pre-hydrolysis of the major milk components by lactic starter cultures, rendering them more digestible (Hewitt and Bancroft, 1985; Bystron and Molenda, 2004). In general, the overall properties of yoghurt, such as acidity level, free fatty acid production, production of aroma compounds (diacetylene, acetaldehyde and acetoin) as well as the sensory profile

and nutritional value, are important traits of the product (Lee and Lucey, 2010). The production of yoghurt entails many processes including standardization and homogenization of milk. This study was designed to evaluate the effect of different homogenization methods on the microbial, physico-chemical and sensory properties of yoghurt.

## MATERIALS AND METHODS

### Milk sampling and transportation

Fresh cow milk was collected from lactating White Fulani cows. The cows were milked between 06.00 and 07.00 h by hand milking procedure in hygienic conditions. The milk was thoroughly mixed with a ladle spoon for about three (3) minutes. The milk collected in

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plastic containers was ice-packed in a cooler and immediately transported to the laboratory for pH determination, yoghurt making and chemical analysis.

### Preparation of yoghurt samples

The fresh milk obtained was clarified to remove dirt, debris and udder tissues using a clean cheese cloth. Thereafter, the milk was pre-heated to 50-60°C and 10% skimmed milk was added. The whole milk was then divided into four equal portions (treatments). Thus, not homogenized (NH) served as the control and hand whisker (HW), pressure sprayer (PS) and high-speed mixer (HM) were homogenized with a whisker (Rudong Jiahua Food Machinery Co., China), pressure sprayer (Hymatic Agro, New Delhi, India) and a high-speed mixer (10,000 to 13,000 rpm; Qlink<sup>R</sup> Shanghai, China), respectively. The different milk portions were then pasteurized at 85°C for 20 min and sucrose was added at 6% inclusion level. The milk was allowed to cool to 40 to 45°C and then inoculated with a mixed culture of *Lactobacillus bulgaricus* and *Streptococcus thermophilus* (yoghurt starters) and then allowed to ferment in an incubator set at 42°C till a firm coagulum was formed. After incubation, the yoghurt so formed was cooled to 4-5°C, stirred and then filled into small transparent cups (covered) with labels. The labeled cups were then placed into larger plastic containers and kept under refrigerated conditions at 4°C until further analyses.

### pH determination

The pH of fresh filtered milk and laboratory prepared yoghurt were obtained using a digital pH meter (PHS – 3C, TBT, Jiangsu, China). The pH meter was calibrated with buffer standards of pH 4 and pH 10 prior to use. 50mL each of the samples was placed in a beaker, the probe of the pH meter was inserted and pH value was recorded. The probe was rinsed thoroughly with distilled water before use on other samples.

### Determination of titratable acidity (TTA)

The titratable acidity (TTA) of the fresh milk and freshly prepared yoghurt were determined using 0.1 M NaOH and phenolphthalein indicator according to the procedures of AOAC (2005).

### Ash content determination

The ash content of milk and yoghurt samples was determined at 550°C according to AOAC (2005). The ash content was expressed as the inorganic residue left as a percentage of the total weight of milk and yoghurt incinerated.

### Total solids

The weight of the residue obtained from moisture content analysis was expressed as percentage total solids using the formula below:

$$\text{Total solids (\%)} = \frac{(\text{Weight of dish + Dry yoghurt}) - (\text{Weight of dish})}{\text{Weight of the sample}} \times 100$$

### Chemical analysis

The fresh milk and the yoghurt samples prepared were studied for dry matter, fat, protein, lactose while ascorbic acid, calcium, iron and phosphate using atomic absorption spectrophotometer. All measurements were in triplicates.

### Microbiological analysis

The pour plate technique (Adegoke, 2000) was used for the microbiological examination of fresh milk and the various yoghurt samples as described below.

### Preparation of media

#### Nutrient agar (NA)

28 g of powdered commercially prepared nutrient agar was accurately weighed into clean, dry 1L flask and 1000 ml of distilled water was added and placed inside water bath (Classic Equipment, Mumbai, India) set at about 90°C to allow the agar to dissolve. The dissolved agar was then distributed into MacCartney bottles and placed inside autoclave (Systec GmBh, Germany) set at 121°C for 15 min.

#### MacConkey agar (MCCA)

55 g of MacConkey Agar (Sigma-Aldrich) was accurately weighed and 1000 mL of distilled water added and boiled to dissolve the agar. The dissolved agar was then distributed into MacCartney bottles and autoclaved as for nutrient agar.

#### Potato dextrose agar (PDA)

39 g of PDA (BD Worldwide) was accurately weighed and 1000mL of distilled water added and bring to boil to dissolve the agar. The dissolved agar was then distributed into MacCartney bottles and autoclaved as for Nutrient Agar.

#### Serial dilution/pouring of plates

9 mL of distilled water was pipette into clean test tubes and plugged with cotton wool and wrapped with aluminum foil. This was then sterilized in autoclave at 121°C for 15 min.

1 mL each of the samples (milk/yoghurt) was measured into a clean test tube containing 9 mL of sterile distilled water and serially diluted until a dilution factor of 10<sup>-5</sup> was achieved and 1 ml of the last dilution factor plate out into sterile plates. The media was poured individually; that is, NA, MCCA and PDA into separate plates and each was duplicated.

The plate for total viable count (NA) and coliform counts (MCCA) were allowed to cool and set and incubated invertedly at 37°C for 48 h. However, the plates for fungal counts (PDA) were inverted and incubated at 28 - 30°C for 72 h.

### Sensory evaluation

The fresh yoghurt produced in the different treatment groups was subjected to evaluation. It was assessed for colour, taste, texture, flavour and overall acceptability. A total of sixty (60) respondents who were familiar with the taste of yoghurt were provided with the score cards comprising a 9-point hedonic scale (Larmond, 1977). The hedonic scale ranged from 1 (dislike extremely) to 9 (like extremely).

### Statistical analysis

The data collected were subjected to statistical analysis in a completely randomized design using ANOVA procedure of SAS (1999)

**Table 1.** Chemical composition of White Fulani cow milk used for the study.

Parameter	Mean
Total solids (%)	15.00
Protein (%)	3.90
Fat (%)	4.70
Ash (%)	1.20
Lactose (%)	4.80
pH	6.60
Titratable acidity (%)	0.18
Ca <sup>2+</sup> (mg/100 g)	130.00
Fe <sup>2+</sup> (mg/100 g)	0.60
PO <sub>4</sub> <sup>3-</sup> (mg/100 g)	80.00
Ascorbic acid (mg/100 g)	1.10

\*Each value is a mean of 3 determinations.

**Table 2.** Chemical composition of yoghurt produced with different homogenizers at Day 0.

Parameter	Homogeniser type				±SEM
	NH	HW	PS	HM	
TS (%)	17.97 <sup>a</sup>	14.77 <sup>b</sup>	18.87 <sup>a</sup>	14.97 <sup>b</sup>	0.59
Fat (%)	3.87 <sup>a</sup>	3.51 <sup>c</sup>	3.59 <sup>b</sup>	3.59 <sup>b</sup>	0.04
Protein (%)	3.69	3.90	3.68	3.67	0.07
Lactose (%)	4.29 <sup>c</sup>	4.50 <sup>a</sup>	4.49 <sup>a</sup>	4.39 <sup>b</sup>	0.03
Ash (%)	1.63 <sup>b</sup>	2.01 <sup>a</sup>	1.03 <sup>c</sup>	2.04 <sup>a</sup>	0.13
TTA (%)	0.92 <sup>a</sup>	0.96 <sup>a</sup>	0.96 <sup>a</sup>	0.88 <sup>b</sup>	0.01
pH	5.10 <sup>a</sup>	4.44 <sup>b</sup>	4.61 <sup>b</sup>	5.07 <sup>a</sup>	0.09

\*Each value is a mean of 3 determinations; <sup>abc</sup> means with different superscripts along the same row are significant ( $p < 0.05$ ); SEM=standard error of mean; TS = total solids; TTA = titratable acidity; NH = not homogenized; HW = hand whisker; PS = pressure sprayer; HM = high-speed mixer.

**Table 3.** Chemical composition of yoghurt produced with different homogenizers at Day 5 of storage.

Parameter	Homogenizer type				±SEM
	NH	HW	PS	HM	
TS (%)	19.08 <sup>b</sup>	19.89 <sup>b</sup>	21.30 <sup>a</sup>	16.83 <sup>c</sup>	0.51
Fat (%)	3.93 <sup>a</sup>	3.57 <sup>c</sup>	3.66 <sup>b</sup>	3.64 <sup>b</sup>	0.04
Protein (%)	3.83 <sup>ab</sup>	3.88 <sup>a</sup>	3.92 <sup>a</sup>	3.77 <sup>b</sup>	0.02
Lactose (%)	4.46 <sup>ab</sup>	4.52 <sup>b</sup>	4.59 <sup>a</sup>	4.43 <sup>c</sup>	0.02
Ash (%)	2.09 <sup>b</sup>	2.13 <sup>b</sup>	2.20 <sup>a</sup>	2.09 <sup>b</sup>	0.01
TTA (%)	0.97 <sup>b</sup>	1.00 <sup>b</sup>	1.10 <sup>a</sup>	0.93 <sup>b</sup>	0.02
pH	5.07 <sup>a</sup>	4.92 <sup>b</sup>	4.99 <sup>b</sup>	5.33 <sup>a</sup>	0.05

\*Each value is a mean of 3 determinations; <sup>abc</sup> means with different superscripts along the same row are significant ( $p < 0.05$ ); SEM = standard error of mean; TS = total solids; TTA = titratable acidity; NH = not homogenized; HW = hand whisker; PS = pressure sprayer; HM = high-speed mixer.

**Table 4.** Chemical composition of yoghurt produced with different homogenizers at Day 10 of storage.

Parameter	Homogenizer type				±SEM
	NH	HW	PS	HM	
TS (%)	17.32 <sup>c</sup>	18.63 <sup>b</sup>	19.32 <sup>a</sup>	16.82 <sup>c</sup>	0.30
Fat (%)	3.96 <sup>a</sup>	3.63 <sup>c</sup>	3.73 <sup>b</sup>	3.73 <sup>b</sup>	0.04
Protein (%)	3.89 <sup>b</sup>	3.97 <sup>a</sup>	3.82 <sup>b</sup>	3.84 <sup>b</sup>	0.02
Lactose (%)	4.55 <sup>b</sup>	4.64 <sup>b</sup>	4.76 <sup>a</sup>	4.51 <sup>b</sup>	0.03
Ash (%)	2.21 <sup>b</sup>	2.31 <sup>a</sup>	2.32 <sup>a</sup>	2.23 <sup>b</sup>	0.02
TTA (%)	1.05 <sup>bc</sup>	1.12 <sup>ab</sup>	1.17 <sup>a</sup>	0.99 <sup>c</sup>	0.08
pH	5.59 <sup>a</sup>	5.36 <sup>b</sup>	5.36 <sup>c</sup>	5.62 <sup>a</sup>	0.53

\*Each value is a mean of 3 determinations; <sup>abc</sup> means with different superscripts along the same row are significant ( $p < 0.05$ ); SEM = standard error of mean; TS = total solids; TTA = titratable acidity; NH = not homogenized; HW = hand whisker; PS = pressure sprayer; HM = high-speed mixer.

version 8. Treatment means were separated by Duncan multiple range test of the same package.

## RESULTS

### Chemical composition of milk

Chemical composition of milk samples are presented in Table 1. Table 2 shows chemical composition of yoghurt produced with different homogenizers at day 0. Table 3 shows chemical composition of yoghurt produced with different homogenizers at day 5 of storage. Chemical composition of yoghurt produced with different homogenizers at day 10 of storage is presented in Table 4. Table 5 shows mean scores of sensory properties of yoghurt samples produced from different homogenizers and stored at refrigeration temperature of 4°C.

### Day 0

The colour, taste, flavour, texture and overall acceptability (OA) mean scores for yoghurt produced using different homogenizers at day 0 are as shown in Table 5. There was no significant ( $p > 0.05$ ) difference recorded for all the parameters measured. However, NH (control) had the best mean scores for taste, flavour and overall acceptability. Furthermore, best colour.

### Day 5

The type of homogenizer used had significant ( $p < 0.05$ ) influence on the colour, texture and overall acceptability of yoghurt stored for 5 days under refrigeration temperature. Yoghurt homogenized with HM had the highest scores for all the parameters measured except colour. There were significant differences ( $p < 0.05$ ) in colour scores across the treatments, with PS giving the

**Table 5.** Mean scores of sensory properties of yoghurt samples produced with different homogenizers and stored at refrigeration temperature of 4°C.

Storage (day)	Homogenizer (type)	Colour	Taste	Flavour	Texture	OA
0	NH	6.85	7.50	7.70	6.85	7.23
	HW	6.90	6.45	6.95	6.90	6.80
	PS	7.15	6.60	6.45	6.45	6.66
	HM	7.65	7.00	6.90	7.00	7.14
	SEM	0.15	0.18	0.15	0.17	0.16
5	NH	7.25 <sup>ab</sup>	6.75	6.75	6.90 <sup>b</sup>	6.91 <sup>b</sup>
	HW	7.10 <sup>b</sup>	6.75	6.85	6.65 <sup>b</sup>	6.84 <sup>b</sup>
	PS	7.75 <sup>a</sup>	7.10	6.90	7.40 <sup>ab</sup>	7.29 <sup>ab</sup>
	HM	7.70 <sup>ab</sup>	7.45	7.30	7.80 <sup>a</sup>	7.56 <sup>a</sup>
	SEM	0.11	0.15	0.14	0.15	0.12
10	NH	7.60	7.60	7.60	7.10 <sup>b</sup>	7.48
	HW	7.80	7.60	7.20	7.00 <sup>b</sup>	7.40
	PS	8.10	7.20	7.10	7.70 <sup>ab</sup>	7.53
	HM	8.20	7.90	7.30	8.00 <sup>a</sup>	7.85
	SEM	0.12	0.13	0.13	0.15	0.10

\*Each value is a mean of 10 determinations; <sup>ab</sup> means with different superscripts within a column on storage are significant ( $p < 0.05$ ); OA = overall acceptability; NH = not homogenized; HW = hand whisker; PS = pressure sprayer; HM = high-speed mixer.

highest score and HW the least. Overall acceptability also varied significantly ( $p < 0.05$ ) across the treatments with HM having the highest acceptability score and HW the least.

### Day 10

Except for texture, there were no significant differences ( $p > 0.05$ ) in sensory properties of yoghurt when milk was not homogenized or homogenized with HW, PS or HM. At day 10 of storage, texture of yoghurt was significantly ( $p < 0.05$ ) affected by type of homogenizer. The highest texture score was recorded for PS and least for HW. Although overall acceptability of yoghurts with differently homogenized milk did not differ significantly, the highest overall acceptability score (7.85) was recorded for HM.

Generally, taste and flavour were not significantly ( $p > 0.05$ ) influenced from day 0-10 in all the treatment groups. Furthermore, all the sensory parameters evaluated were improved as the days in storage progressed (Table 5).

Table 6 shows the microbial load of fresh milk and yoghurt produced with different homogenizers and stored at refrigeration temperature.

## DISCUSSION

The total solids content of milk fell within the range 11.1

to 16.8% as reported by Adeneye et al. (1970) and Onatola (2004). The fat content (4.7%) also agreed with the range of 3.0-8.2% (Adeneye et al., 1970; Ogunsiiji, 1974; Onatola, 2004). The White Fulani cow is traditionally a low milk yielder (Adeneye et al., 1970; Olaloku et al., 1971; Olaloku, 1972). The fat content of its milk is supposed to be high because of the inverse relationship between milk yield and the butterfat (Schmidt, 1971; Bath et al., 1978; Belewu, 2006). The low fat content observed for White Fulani cow's milk in the present study did not follow such expectation. This may be attributed to the season at which the milk was collected (during rainy season). There is increase in milk yield during this time. It is a well known fact that milk yield is inversely proportional to the fat content of milk. The crude protein (CP) content (3.9%) was within the range of 3.3 to 4.8% (Adeneye et al., 1970), 3.4 to 4.2 (Ogunsiiji, 1974) and 2.9 to 5.0 (O'Mahony, 1988). The CP content (3.9%) was lower than that of fat (4.7%) as earlier reported by Laben (1963) and Williamson and Payne (1978).

The ash value (1.2%) obtained in the present study is above the range (0.5-0.8%) reported by Basu et al. (1962), Olaloku (1972), Ogunsiiji (1974) and Adebowale (1976). The average pH of milk used for this study was 6.6. This suggested that the milk was normal. A range of 6.5-6.7 was considered to be normal pH for cow milk (O'Mahony, 1988). pH 6.3 and 6.5 had been reported for

**Table 6.** Microbial load of fresh milk and yoghurt produced with different homogenizers and stored at refrigeration temperature of 4°C.

Storage (day)	Homogenizer (type)	TVC (cfu/g)	Coliform count (cfu/g)	Fungal Count (cfu/g)
	Fresh milk	$3.2 \times 10^5$	$7.5 \times 10^5$	$6.5 \times 10^5$
0	NH	$8.0 \times 10^3$	$1.0 \times 10^3$	Nil
	HW	$5.0 \times 10^3$	$1.0 \times 10^3$	Nil
	PS	$3.0 \times 10^3$	Nil	Nil
	HM	$4.0 \times 10^3$	Nil	Nil
5	NH	$8.0 \times 10^3$	$1.0 \times 10^3$	Nil
	HW	$6.0 \times 10^3$	$3.0 \times 10^3$	Nil
	PS	$3.0 \times 10^3$	Nil	Nil
	HM	$6.0 \times 10^3$	Nil	Nil
10	NH	$1.4 \times 10^4$	$4.0 \times 10^3$	$1.0 \times 10^3$
	HW	$9.0 \times 10^3$	$5.0 \times 10^3$	$1.0 \times 10^3$
	PS	$5.0 \times 10^3$	$3.0 \times 10^3$	$1.0 \times 10^3$
	HM	$8.0 \times 10^3$	$2.0 \times 10^3$	$1.0 \times 10^3$

NH = Not homogenized; HW = hand whisker; PS = pressure sprayer; HM = high-speed mixer; TVC = total viable count.

White Fulani cow's milk (Afolabi, 1991; Onatola, 2004). These lower values indicated that the milk used by these authors was acidic probably as a result of activity of lactic acid bacteria or presence of colostrum. pH values higher than 6.7 are always associated with mastitic milk. The titratable acidity (TTA) of 0.18% obtained in this study was higher than 0.14% reported by Aworh and Akinniyi (1989). TTA is normally expressed as percentage lactic acid (Lee, 1985). The milk used for this experiment was ice-packed in a gallon to prevent lactose breakdown to lactic acid and to prevent multiplication of lactic acid producing bacteria.

Homogenization is known to break fat globules down increasing surface area for ingredients and fermentative bacteria to act. Homogenized milk is whiter in colour with better taste, flavour and texture. This suggests the high nutritive and best sensory scores obtained with HM and PS. The least nutritive and sensory scores obtained with HW as compared with HM and PS is due to the lower force or pressure applied with the whisker and probably due to inadequate timing of homogenizing.

Fresh milk had the highest microbial load (Table 6). This suggests that homogenization reduced the TVC, coliform and fungal counts of the yoghurt produced. Milk homogenization has been reported to reduce the microbial load of milk since the microbes tends to adhere to fat globules and are lost in the homogenizer (Eckles et al., 1951). The nil coliform and fungal counts in HM and PS on the day of manufacture may probably be due to the shear force applied by the high-speed mixing and the high pressure of the pressure sprayer. The appearance of coliforms and fungi in the latter days of storage might possibly be as a result of contamination which provided conducive environment for the growth of the microbes.

## Conflict of Interests

The authors did not declare any conflict of interests.

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## Full Length Research Paper

# Nutraceutical and health benefits of some vegetables eaten in Enugu State Nigeria

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Little or nothing is known about the nutritional and medicinal values of some of the vegetables eaten in Enugu state of Nigeria. This study aimed to quantify the total antioxidants as phenols, total flavonoids, vitamin C content and reducing properties of six vegetables [*Teltaria occidentals* (ugu), *Gnetum africanum* (ukase), *Piper quineense* (uziza), *Gongronema latifolium* (utazi), *Achyranthes spendens* (awa), *Occimum grattissimum* (nchegnwu)]. Colorimetric method was performed using Gallic acid and rutin standard reagents for total antioxidants (phenols) and total flavonoids, respectively. The vitamin C and reducing properties contents were also determined using 2, 6-dichlorophenol indophenols (titrimetric) and trichloroacetic acid colorimetric methods, respectively. The result reveals that *G. africanum* (ukazi) ranked the best of all the vegetables assayed for total flavonoids (0.58 mg/g), total antioxidants (49 mg/g) as phenols, vitamin C (0.91 mg/100 mL) and reducing property (64 mg/g) followed by *A. spendens* (awa) (total flavonoids, 1.57 mg/g; total antioxidants; 50 mg/g; vitamin C; 0.43 mg/100 mL and reducing property. 81 mg/g), *T. occidentalis* (ugu), (total flavonoids, 0.58 mg/g; total antioxidant; 49 mg/g; vitamin, C 0.69 and total reducing property 64 mg/g), *O. grattissimum* (nchanwu) (total flavonoids, 0.36 mg/g; total antioxidant, 45 mg/g; vitamin C, 0.60 mg/100 mL; and total reducing property 80 mg/g) while *P. guineense* (uziza) (total flavonoids, 0.71 mg/g; total antioxidants, 36 mg/g; vitamin C, 0.40 mg/100 MI and total reducing property, 42 mg/g) and *G. latifolium* (utazi) (total flavonoids, 0.36 mg/g; total antioxidant, 8 mg/g; vitamin C, 0.33 mg/100 MI and total reducing property, 81 mg/g) are the least. However, the result of the study have highlighted the nutritional and medicinal richness of these vegetables and encourages the increased consumption of them to compensate nutrient deficiency and therefore could be a useful source to prevent or attenuate damages such as lipid peroxidation, glycation of proteins and inactivation of enzymes caused by free radicals.

**Key words:** Vegetables, total antioxidants, flavonoids, vitamin C, reducing properties.

## INTRODUCTION

Vegetables are low in fat but contain good amounts of vitamins and minerals. All the green- yellow - orange vegetables are rich sources of calcium, magnesium, potassium, iron, beta- carotene, vitamin B - complex,

vitamin C, vitamin A, and vitamin K. Vegetables are home for many antioxidants that help protect the human body from oxidant stress, diseases and cancer, and also help the body develop the capacity to fight against these by

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boosting its immunity (Umesh, 2014)

Today awareness is increasing among consumers regarding foods and the therapeutic properties of nutrients. Some people assert that it is misguided to assume that rapid technological and pharmaceutical developments are good for our health (Higdon, 2005).

In order to improve immune system, vitamins, minerals and antioxidants are essential. Most vegetables are very rich in phytonutrients (vitamins A, B, C, K, beta carotene and a host of anti-oxidants which assure a normal progression of the metabolic processes of the body. The nutrient composition of different types of vegetables varies considerably and contains vitamins, essential amino acids as well as minerals and antioxidants that play several roles in the body (Fasuyi, 2006; Mnzava, 1997; Segal et al., 1983; Watson, 2001).

Anti-oxidant are the nutritional equivalent of man's best friend. They are loyal protectors and nurturers of our cells, repelling disease, and promoting good health (Rani et al., 2004; Karadeniz et al., 2005). Antioxidants can be derived from healthy foods or in the form of supplements and they include a family of naturally occurring compounds like vitamins A, C and E, beta-carotene, lycopene, flavonoids, and more (Enemo et al., 2010; Hamzah et al., 2013; Oyewole et al., 2013; Venter et al., 2014). These antioxidants are believed to protect cells from free radicals, harmful oxygen molecules which are thought to cause the cell damage that contributes to the development of cancer, atherosclerosis, Alzheimer's disease, and rheumatoid arthritis. Free radicals may be the underlying reason for aging (Cherubini et al., 2005; Christen, 2000; Di Matteo and Esposito, 2003; Hitchon and El-Gabalawy, 2004; Kham et al., 2010; Larsen, 1993; Nakabeppu et al., 2006; Nunomura et al., 2006; Rhee, 2006; Sohal, 2002; Stadtman, 1992; Valko et al., 2007; Wood-Kaczmar et al., 2006)

Free radicals are formed in the body, but their production is increased by factors such as smoking, alcohol consumption, air pollution, infection, stress, excessive sunlight and toxin like radiation, heavy metals and asbestos (Bannister and Rottliog, 1987; Beck et al., 2000; Ignatowicz et al., 2013; Johnson and Giulivi, 2005; Stohs and Bagchi, 1995; Valko et al., 2005; Wozniak et al., 2012).

The role of antioxidants in vegetables and other foods is to mop up excessive free radicals in the human body that lead to oxidative stress and thereby slowing or preventing diseases and aging. The study aims to quantify the total flavonoids, total antioxidants (phenols), vitamin C and total reducing properties of the six vegetables under study.

## MATERIALS AND METHODS

### Sample collection and preparation

Fresh leaves of six popularly eaten vegetable in Enugu metropolis

namely: *T. occidentals* (ugu), *O. grattismum* (ewa), *G. latifolium* (utazi) *P. guineense* (uzuza), *G. Africanum* (ukazi) and *A. splendens* (nchanwu) were bought from a commissioned farmer in Ogbete Main Market on July 18, 2013 and authenticated by Prof. J. C Okafor of Applied Biology and Biotechnology Department, Enugu State University of Science and Technology. The leaves were washed with distilled water and 10 g ground with an electric blender and stored in well labeled polypropylene films placed in a clean well-dried black container.

### Determination of total anti-oxidant content

The Gulcin et al. (2003) method was employed to assay the total antioxidant content of the six vegetables. About 100 mL of 70% (v/v) ethanol was added into the ground samples in the ratio of 1:5, stirred and left for 24 h at room temperature. The extract was separated from the residue by filtration through Whatman No. 1 filter paper. The remaining residue was re-extracted and then the extracts were combined. The extract was concentrated to 100 mL under reduced pressure at 40°C using a rotary evaporator. One milliliter of the extract and standard Gallic acid solution (10, 20, 30, 40, 50 and 100 mg/L) was added to 9 mL of water. Then 1 mL of Folin Ciocalteus reagent was added to the mixture and vortexed twice. After 5 min 10 mL of 7% sodium carbonate was added to the mixture and incubated for 90 min at 25°C. The absorbance against a reagent blank was determined using UV/visible spectrometer at 750 nm.

### Determination of total flavonoids

Aluminum chloride colorimetric method was used for total flavonoid determination (Zhishen et al., 1999; Ghasemi et al., 2009). Rutin was used as a standard for the calibration curve. Hundred milligram of rutin was dissolved with 84 mL of 60% ethanol (v/v) and the volume made up to 100 mL with 30% ethanol (v/v). The standard curve was constructed by diluting 0.01, 0.2 0.3, 0.4 and 0.5 mL rutin to 1 mL with water to obtain 0,10, 20, 30, 40 and 50 mg/mL of rutin. To this, 0.5 mL of 5% NaNO<sub>2</sub> was added alongside with 1 mL of the sample extract (extracted with 5 ml of 95% ethanol in the ratio of 1:10 for 30 min with constant stirring and shaken for 5 min). Then, 0.5 mL 10% AlCl<sub>3</sub> was also added and the test tube shaken for 5 min. Four milliliter of 4% NaOH was then added and the test tube shaken for 15 min. The absorbance was determined at 510 nm using test tube 0 to zero the UV/visible spectrometer.

### Determination of total reducing properties

The ground vegetable was soaked in methanol in the ration of 1:5 for 48 h. Twenty five milliliters of methanol extract of the vegetable sample was mixed with 2.5 mL of 0.2 M sodium phosphate buffer and 2.5 mL of 1% potassium ferricyanide, and incubated at 50°C for 20 min then 2.5 mL of 10% trichloroacetic acid was added and the mixture centrifuged at 1000rpm for 10 min. The upper layer (2.5 mL) was mixed with 2.5 mL of glass distilled water and 0.5 mL of 0.1% ferric chloride and its absorbance measured spectrophotometrically at 700 nm against a blank. Ascorbic acid was used as the reference standard (Sarah et al., 2013).

### Extraction and determination of vitamin C

Ten grams of vegetable sample was ground using mortar and pestle with the aid of 2 g of 2 M hydrochloric acid washed sand to facilitate the breakdown of the sample until a fine paste was obtained. The vitamin C content of the sample was extracted with 5

**Table 1.** Total anti-oxidant, Flavonoid, reducing property and Vitamin C content in vegetable sample.

Sample	Total anti-oxidant (mg/g)	Total flavonoids (mg/g)	Total reducing properties (mg/g)	Vitamin C (mg/100 mL)
<i>Telfaria occidentalis</i> (ugu)	49	0.58	64	0.69
<i>Gnetum africanum</i> (ukazi)	51	0.82	64	0.91
<i>Piper Guineense</i> (uziza)	36	0.71	42	0.40
<i>Gongronema latifolium</i> (utazi)	8	0.36	81	0.33
<i>Achyranthes spondens</i> (awa)	50	1.57	81	0.43
<i>Occimum grattissimum</i> (Ncheanwu)	45	0.36	80	0.60

mL of 2% HCl into a 100 mL measuring cylinder through a pad of cotton wool. The extraction was repeated three times and the extract combined and the volume made up to 100 mL with distilled water.

Ten milliliter of the extract was titrated with 0.001 M of 2, 6-dichlorophenol indophenols solution until a pink coloration persist for 30 s (Amadi et al., 2004).

## RESULTS AND DISCUSSION

The assay of total anti-oxidants, flavonoids reducing properties and vitamin C content of the vegetables *T. occidentalis* (ugu), *G. africanum* (utkazi), *P. guineense* (uziza), *G. latifolium* (utazi), *A. spondens* (awa), *O. grattissimum* (ncheanwu), popularly eaten in eastern Nigeria were analyzed using UV/visible spectrometer and titrimetric method for vitamin C.

The results revealed that *G. afranium* (ukazi), *A. spondens* (awa) and *T. occidetalis* (ugu) are very rich in anti-oxidants (51, 50 and 49 mg/g), respectively. This may be the underlying reason why people who eat natural and whole foods are healthier and live longer than those who mostly eat processed foods (Nakabeppu et al., 2000). *G. latifolium* (utazi) is not very rich in anti-oxidants. Anti-oxidants mostly come from the fresh vegetables and fruits we eat. They prohibit and sometimes prevent the oxidation of other molecules in the body. The benefits of antioxidants are very important to good health because if free radicals are left unchallenged they can cause a wide range of illnesses and chronic diseases (Rui, 2003; Joao, 2012).

The result of the total flavonoids content determination (Table 1) showed that *A. spendens* (awa) has twice as much flavonoids (1.57 mg/g) as *G. africanum* (Ukazi, 0.82 mg/g) and *P. guineense* (uziza, 0.71 mg/g). This is followed by *Telfaria occidentalis* (ugu 0.58 mg/g) while *O. grattissimum* (Ncheanwu, 0.36 mg/g) and *G. latifolium* (utazi, 0.36 mg/g) had the least total flavonoids content. This result supports the claim that diets rich in fruits and vegetables are beneficial to health and that dietary flavonoids are key drivers in anti-allergic, anti-cancer, anti-oxidants, anti-inflammatory, anti-viral, anti bacterial, anti-leukemic and vasodilator activity (Beenu and Rajni,

2012; Lisa, 2013; Shirma, 2006; Yao et al., 2004).

The reducing properties of a vegetable are due to phytochemicals such as steroidal saponins, insulin-like peptides and alkaloids (Khanna et al., 1981; Sandra, 2011; Singh et al., 2004). These chemicals probably may be responsible for the bitter taste and high reducing properties of *G. latifolium* (utazi, 81 mg/g), *A. spondens* (awa, 81 mg/g) and *O. grattissimum* (ncheanwu, 80 mg/g), (Khanna et al., 1981; Sandra, 2011; Singh et al., 2004)

Results of the vitamin C content of the six vegetables showed that *G. Africanum* (ukazi) has the highest content of vitamin C (0.91 mg/mL) followed by *T. occidentalis* (ugu, 0.69 mg/mL), *O. grattissimum* (Ncheanwu, 0.60 mg/mL), while *A. spondens* (awa, 0.43 mg/mL), *P. guineense* (uziza, 0.40 mg/mL) and *Gongronema latifolium* (utazi, 0.33 mg/mL) have the least vitamin C content. Vitamin C is the single most important vitamin that has an anti-oxidant property, an important factor in collagen production and useful in wound healing of all types. Vitamin C may protect the skin from free radical damage after exposure to ultraviolet (UV) rays. It is a popular remedy for common cold. It helps to prevent cataracts and lower one's risk of heart disease and strokes. Vitamin C can help regulate blood sugar levels in people with diabetes and it also acts as an oxidative modifier of low density lipoprotein (LDL) (Levin, 1986; Douglas et al., 2000; Shukla, 1969; Steinbrecher et al., 1990; Neu et al., 1997). The increased consumption of these vegetables rich in Vitamin C will be of great health benefit.

## Conclusion

Vegetables are low in fat but contain good amount of vitamins such as Vitamin C, antioxidants, flavonoids and reducing agents. Vegetable nutrition has widely drawn the attention of fitness-conscious individuals and food scientists because of their proven health benefits. The increased consumption of *A. spondens* (Awa), *G. Africanum* (ukazi), *T. occidentalis* (ugu), *O. grattissimum* (ncheanwu) and *P. guineense* (Uziza) has beneficial

effects such as anti-allergic, anti-cancer, anti-oxidants, anti-inflammatory, anti-viral, anti-bacterial, and anti-leukemic and vasodilator activities. It can be concluded that *G. africanum* (ukazi) ranked the best of all the vegetables assayed followed by *A. spondens* (awa) and *T. occidentalis* (ugu).

## Conflict of Interests

The authors did not declare any conflict of interests.

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Full Length Research Paper

# Effect of processing on the quality of flour, *abacha* slices and its flour derived from cassava (*Manihot esculenta Crantz*) TMS 97/4779

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This study was undertaken to evaluate the effect of processing methods on the quality of *abacha* slices. *Abacha* slices were produced from cassava (TMS 97/4779) roots by four different methods. The *abacha* processing methods investigated were of different peeling, boiling, slicing and soaking methods. Also, raw cassava flour, which was peeled, sliced and dried (PSD) was produced. The sensory properties and yield of *abacha*, the chemical and physical properties of the flour from the dried *abacha* and raw cassava flour were determined. The sensory properties of the fresh *abacha* were evaluated which showed that boiled, peeled and sliced sample (BPS) was most preferred. The yield of the *abacha* ranged from 29.17-39.13%. Significant differences ( $p < 0.05$ ) were observed in the proximate composition of the samples. The processing methods significantly affected the swelling index and water absorption capacity of the *abacha* flour. PSD sample showed the highest value in swelling index and water absorption capacity. The cyanide content, which ranged from 4.20-7.20 mg/kg, was below the recommended safety level of 10 mg/kg HCN. The processing methods significantly ( $p < 0.05$ ) affected the pasting properties of the *abacha* flour except peak viscosity and peak time.

**Key words:** Cassava, peeling, boiling, slicing, soaking, hydrogen cyanide.

## INTRODUCTION

*Abacha* is a snacks or main meal in the eastern states of Nigeria. It is a shredded or sliced dry or wet product, obtained by shredding or slicing boiled cassava roots, soaking the shreds for 8-24 h in cold water and washing to remove the scum that causes sliminess. It may then be dried. Preliminary work carried out on the effect of variety on consumer acceptability of *abacha* slices, showed that slices made from TMS 97/4779 were most preferred. And

also, ascertained that texture is the main attribute in determining the acceptability of sliced *abacha*. The most common method (traditional) employed in the processing of *abacha* is peeling, size reducing into chunks, boiling, slicing and soaking. However, alternative methods include boiling the root before peeling, boiling of the peeled root before slicing and different peeling method. There is need to understand the effect of the different

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processing methods on the quality of *abacha* product. The effects of these alternative processing methods on the chemical, physical (functional and rheological) and sensory properties of the product is the focus of this investigation.

## MATERIALS AND METHODS

### Source of sample

Roots of bitter cassava variety (*Manihot esculenta* Crantz), 97/4779, were harvested 14 months after planting, from a farm at the Ebonyi State Agricultural Development Programme, Abakaliki.

### Preparation of *abacha* slices samples

Four kilograms each of the cassava roots was used to produce *abacha* slices, using different sequence of treatments as follows:

#### **Traditional method: Peeled, sliced and boiled *abacha* slices (PSB)**

Four kilograms (4 kg) of the root was washed with cold tap water and manually peeled with stainless steel knives. The peeled root was thinly sliced (0.5 mm thick), boiled (PSB) in 3 L tap water for 45 min using stainless cooking pot, cooled and washed once to reduce the scum which causes sliminess before steeping in 5 L cold tap water at room temperature (28±2°C) for 16 h. The steep water was changed at 4 h intervals for 16 h before final washing.

#### **Alternative method 1: Peeled, boiled, sliced and soaked *abacha* (PBS)**

Another 4.0 kg of the roots was peeled manually, washed with cold tap water, boiled in 3 L tap water for 45 min, cooled and sliced (0.5 mm). The sliced sample (PBS) was steeped in 5 L tap water for 16 h and washed as in PSB.

#### **Alternative method 2: Peeled, boiled, sliced and soaked *abacha* (CBS)**

Four kilograms of the root was washed and the papery brown peel (cork) removed. It was boiled in 3 L tap water for 45 min, cooled, sliced (CBS), steeped in 5 L cold tap water for 16 h and washed as above.

#### **Alternative method 3: Boiled, peeled and sliced *abacha* slices BPS**

Another portion (4.0 kg) of the cassava roots was washed and boiled without peeling in 3 L tap water for 45 min, cooled and peeled with a stainless steel knife. It was then sliced (BPS) steeped in 5 L tap water for 45 min and washed in tap water as in PSB above.

#### **Peeled, sliced and dried chips PSD**

The roots were peeled, washed, sliced and sun dried (35-40°C). All

the *abacha* samples were sun dried for 72 h to get dried *abacha* slices which was packaged in a polyethylene bag and put in an air tight plastic container for analysis.

## Analyses

### *Yield of abacha*

This was determined by using the expression.

$$\text{Yield (\%)} = \frac{\text{Weight of } abacha \text{ after washing}}{\text{Weight of cassava used}} \times 100$$

Where, weight after washing was determined after draining.

### **Determination of chemical properties of wet or dried *abacha***

The moisture, ash and crude fibre, contents were determined by standard methods of the AOAC (1990). Fat content was determined by the continuous solvent extraction method using the Soxhlet apparatus. Crude protein was determined by the Kjeldahl method (Kirk and Sawyer, 1998) in which the N<sub>2</sub> content was determined and multiplied by the factor, 6.25. Carbohydrate content was calculated by difference.

The energy content of the samples was determined from the result of the proximate analysis by multiplying with the relevant factors- 17, 17 and 37 which are the conversion factor for carbohydrate, protein and fat, respectively.

The hydrogen cyanide (HCN) content was determined by the alkaline picrate method of Bradbury et al. (1999), while pH was determined by the method of Matil (1971). The method of Kirk and Sawyer (1998) was used to determine the titratable acidity.

### **Determination of physical properties**

The method described by Okaka et al. (1997) was used to determine bulk density. Swelling capacity was determined following the procedure of Ukpabi and Ndimele (1990), while the water absorption capacity was determined using the method of Lin et al. (1994). A rapid visco analyzer (RVA) (Model RVA 3d+ Network Scientific, Australia) was used to determine the pasting characteristics according to the procedure adopted by Omodamiro et al. (2007).

### **Statistical design and analysis**

All data obtained were subjected to analysis of variance (ANOVA) using completely randomized design (CRD). Means were separated using Fisher's least significant design (FLSD) at p<0.05 (Steel and Torrie, 1980).

## RESULTS AND DISCUSSIONS

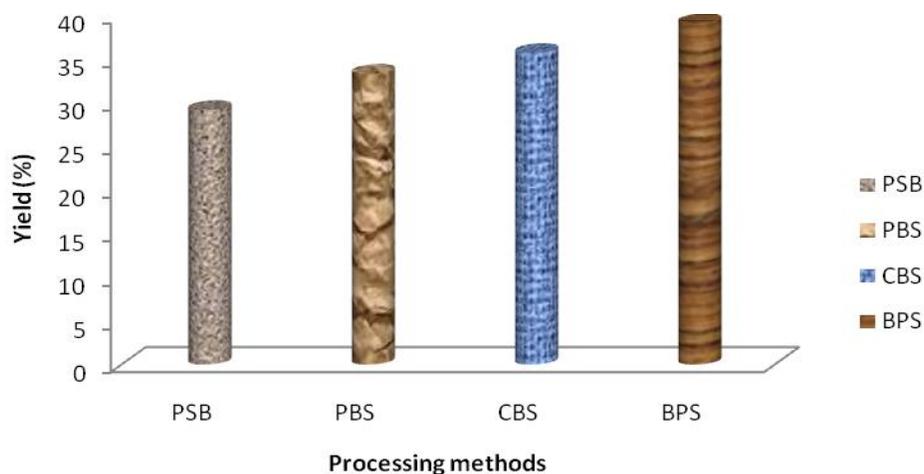
### **Sensory properties**

The sensory properties of *abacha* samples (Table 1) showed that consumers preferred the boiled, peeled and sliced (BPS) followed by peeled, sliced and boiled (PSB)

**Table 1.** The sensory properties of the *abacha* samples.

Sample	Sensory properties			
	Appearance	Texture	Taste	General acceptability
CBS	2.90 <sup>b</sup>	4.80 <sup>c</sup>	4.15 <sup>b</sup>	3.80 <sup>c</sup>
PBS	7.00 <sup>a</sup>	6.10 <sup>b</sup>	6.70 <sup>c</sup>	7.00 <sup>b</sup>
PSB	7.05 <sup>a</sup>	6.60 <sup>a</sup>	7.35 <sup>a</sup>	7.05 <sup>b</sup>
BPS	7.55 <sup>a</sup>	7.15 <sup>a</sup>	7.43 <sup>a</sup>	7.85 <sup>a</sup>
LSD	0.718	0.746	0.612	0.796

Values in the same column with different superscript are significantly ( $p < 0.05$ ) different. PSB = Peeled, sliced and boiled; PBS = peeled, boiled and sliced sample; CBS = only papery brown layer peeled, boiled and sliced sample; BPS = boiled, peeled and sliced sample.

**Figure 1.** Effect of processing on the yield of *abacha* slices.

sample in terms of texture. The texture of BPS and PSB differed significantly ( $p < 0.05$ ) from the others.

The appearance of BPS was rated highest (7.55), while CBS sample was the least (2.90). The processing method drastically affected the appearance of the *abacha* slices from CBS. It was significantly ( $p < 0.05$ ) different in appearance from other samples. The poor appearance of CBS sample may be due to dark colour imparted by the corky tissue (cortex) of the cassava roots. This could lead to low economic value for the *abacha*.

Sensory scores for taste of the samples ranged from 4.15 to 7.45. The BPS had the highest score of 7.45, while CBS had the lowest score of 4.15. There were significant differences in the taste of the four different *abacha* samples.

The score of the general acceptability of the *abacha* ranged from 3.80 - 7.85. BPS had the highest in general acceptability, while CBS had the least. There was no significant difference ( $p > 0.05$ ) between PSB and BPS samples but they differed significantly ( $p < 0.05$ ) from others in general acceptability.

The appearance, texture and the taste of the *abacha* influenced the general acceptability of the *abacha*. This is so because BPS that had the highest score for appearance (7.55), texture (7.15) and taste (7.43) scored the highest for general acceptability, while CBS that had the least score for appearance (2.90), texture (4.80) and taste (4.15), scored least.

### Yield

The yield of the *abacha* samples ranged from 29.17 to 39.13% (Figure 1). Boiled, peeled and sliced (BPS) sample had the highest yield of 39.13%, while the peeled, sliced and boiled (PSB) sample had the least value (29.17%). This is probably due to high loss of materials during peeling and slicing in addition to losses sustained from disintegration of *abacha* slices during boiling. The BPS sample had the highest yield of *abacha* slices because during boiling, the cementing material between the peel and the edible fleshy portion breaks down,

**Table 2.** Effect of processing on the proximate composition of the *abacha* slices.

Sample	Proximate composition (%)					
	Moisture	Fibre	Ash	Protein	Fat	Carbohydrate
PSB	9.86 <sup>a</sup>	2.68 <sup>a</sup>	2.35 <sup>c</sup>	2.53 <sup>a</sup>	1.55 <sup>a</sup>	81.03 <sup>a</sup>
PBS	8.82 <sup>b</sup>	2.77 <sup>b</sup>	2.47 <sup>a</sup>	1.29 <sup>b</sup>	1.67 <sup>b</sup>	82.98 <sup>b</sup>
CBS	9.23 <sup>c</sup>	2.86 <sup>c</sup>	2.28 <sup>e</sup>	2.48 <sup>a</sup>	1.48 <sup>c</sup>	81.68 <sup>c</sup>
BPS	9.62 <sup>d</sup>	2.64 <sup>d</sup>	2.33 <sup>a</sup>	1.59 <sup>b</sup>	1.33 <sup>d</sup>	82.48 <sup>d</sup>
PSD	9.43 <sup>e</sup>	2.56 <sup>e</sup>	2.38 <sup>a</sup>	2.33 <sup>a</sup>	1.71 <sup>b</sup>	81.60 <sup>e</sup>
LSD	0.075	0.0158	0.322	0.051	0.0775	0.016

Data are means of 3 replications. Values in the same column with different superscripts are significantly different ( $p < 0.05$ ). PSB = Peeled, sliced and boiled; PBS = peeled, boiled and sliced '*abacha*'; CBS = only papery brown layer peeled, boiled and sliced '*abacha*'; BPS = boiled, peeled and sliced '*abacha*'; PSD = peeled, sliced and dried '*abacha*'.

thereby making peeling easy with minimum loss of material. Yield range of 29.17-39.13% recorded is lower than 86.60-87.40% yield for *abacha* shreds reported by Ekwu et al. (2009) and lower than the 51% flour recovery for cooked and steeped cassava reported by Njoku and Banigo (2006).

### Proximate composition

The proximate composition of the *abacha* samples are presented in Table 2. The moisture content of the dried *abacha* slices ranged from 8.82 to 9.86 g/100 g. The moisture content of the *abacha* processed from alternative methods was significantly lower than the moisture content of *abacha* processed from traditional method. The lower moisture content is advantageous as it will help in the storing of the *abacha* flour. PBS sample had the least value (8.82 g/100 g). Okechukwu and Okoye (2010) reported a similar moisture range (7.72-10.83 g/100 g) for *abacha* samples. With this range of moisture content, the *abacha* slices will be stored well provided it is kept in an airtight container. There was significant ( $p < 0.05$ ) differences between the moisture content of samples, which may be attributed to differences in processing methods such as peeling, slicing and boiling, boiling, peeling and slicing etc, applied.

The protein (Nx6.25) content ranged between 1.29 and 2.53 g/100 g. The protein content of PSB (traditional method) was higher than the protein content of *abacha* processed using the alternative methods. The low protein range is similar to the low range usually quoted in literature (Oke, 1968; Oyenuga, 1968) for cassava. Statistical results showed significant differences ( $p < 0.05$ ) in the protein content of some of the samples.

The fat content of the samples ranged from 1.33 to 1.77g/100 g. PSD had the highest value of 1.77 g/100 g,

indicating that boiling and soaking reduces the fat content of cassava. The lower fat content would enhance the keeping quality of the *abacha* since oxidative rancidity would be reduced. The fat content of the samples obtained in this study is higher than the 1.15% for cassava products (Oluwole et al., 2004). The differences could probably be attributed to the processing method employed during sample preparation or to differences in the variety of the cassava used. There were significant differences ( $p < 0.05$ ) in the fat content of the samples except for PSD and peeled, boiled and sliced (PBS) samples, which had similar values.

The crude fibre content ranged from 2.56 to 2.86 g/100. CBS that had only the brown papery peel removed had the highest crude fibre content of 2.86 g/100 g. This could imply that the cortex (corky layer which was not removed) has higher fibre content than the inner tissue. PSD sample had the least (2.56 g/100 g). The crude fibre of the *abacha* (2.56-2.86 g/100 g) was within the range of 2.0 to 5.0 g/100 g as reported by the NIS (1998) for cassava products. It is significantly lower than the value of fibre in raw cassava flour (3.52 g/100 g) and may be attributed to the different processing method, age and varietal differences (Njoku and Banigo, 2006). With the high fibre content, consumers of *abacha* processed in these forms may not suffer from gastro and circulatory disease epidemiologically linked to low dietary fibre intake (Okaka et al., 2002). The samples differed significantly ( $p < 0.05$ ) in their fibre content.

The ash content ranged from 2.28 to 2.47 g/100 g. The PBS sample had the highest of 2.47 g/100 g, indicating that leaching of organic matter during boiling of the root probably occurred to a greater extent than the mineral content, leading to higher concentration of minerals in the sample. This was followed by PSD sample (2.35 g/100 g), while sample CBS had the lowest ash content (2.24 g/100 g). The ash content of samples differed significantly ( $p < 0.05$ ) from each other and may be due to

**Table 3.** The TTA and pH of *abacha* slices from 97/4779.

Sample	TTA (%)	pH
CBS	0.22 <sup>ab</sup>	6.78 <sup>a</sup>
PBS	0.18 <sup>a</sup>	6.38 <sup>c</sup>
PSB	0.21 <sup>ab</sup>	6.55 <sup>b</sup>
BPS	0.26 <sup>b</sup>	6.25 <sup>d</sup>
PSD	0.20 <sup>a</sup>	6.74 <sup>a</sup>
LSD	0.051	0.051

ND = Not determined. Data are means of 3 replications. Values in the same column with different superscripts are significantly different ( $p < 0.05$ ). PSB = Peeled, sliced and boiled; PBS = peeled, boiled and sliced sample; CBS = only papery brown layer peeled, boiled and sliced sample; BPS = boiled, peeled and sliced sample; PSD = peeled, sliced and dried sample.

**Table 4.** The hydrogen cyanide content of the samples.

Samples	Fresh cassava (mg/kg)	Wet samples (mg/kg)	Reduction (%)	Dried samples (mg/kg)	Total reduction (%)
CBS	6.78	84.04	4.60	89.18	42.50
PBS	6.17	85.48	4.50	89.41	42.50
PSB	42.50	6.72	84.19	4.50	89.41
BPS	42.50	6.25	85.29	4.20	90.11
PSD	42.50	ND	ND	7.20	83.06
		0.135		0.0256	

ND = Not determined. Data are means of 3 replications. PSB = Peeled, sliced and boiled; PBS = Peeled, boiled and sliced sample; PPBS (CBS) = only papery brown layer peeled, boiled and sliced sample; BPS = boiled, peeled and sliced sample; PSD = peeled, sliced and dried sample.

differences in the processing methods employed.

The carbohydrate content of the samples ranged from 81.03 to 82.98 g/100 g. The carbohydrate content of the *abacha* from the alternative methods (PBS- 82.98 g/100 g, CBS - 81.69 g/100 g and BPS- 82.48 g/100 g) was higher than the carbohydrate content of *abacha* processed from traditional method (81.03 g/100g). The range of 81.03-82.98 g/100 g is slightly lower than the value of 84.22-86.99 g/100 g obtained for pre-gelled and raw cassava flour (Njoku and Banigo, 2006). The values were significantly different ( $p < 0.05$ ) from each other and may be attributed to differences in the processing methods.

#### pH, total titratable acidity (TTA) and hydrogen cyanide (HCN) content

The pH and TTA content of the *abacha* samples are

presented in Table 3. The TTA of the samples ranged between 0.18 and 0.26%. The sample from boiled, peeled and sliced cassava had the highest value of 0.26%, while that from peeled, boiled and sliced cassava had the least (0.18%). There were significant differences ( $p < 0.05$ ) in the total titratable acidity of samples BPS, PSD and PBS, while BPS did not differ from PSB and CBS. High TTA is expected to lower the pH of a cassava product due to increase in acidity.

The pH of the *abacha* samples ranged from 6.25 to 6.78. The high pH may be due to the fact that the samples were not fermented. The sample from CBS had the highest pH, while the sample from BPS cassava root had the least pH of 6.25. The pH of the *abacha* samples differed significantly ( $p < 0.05$ ) from each other except samples from CBS and PSD, which were statistically the same.

The hydrogen cyanide content of the samples are presented in Table 4. The hydrogen cyanide content of

**Table 5.** Functional properties.

Sample	Energy (J)	Swelling index	Water absorption capacity (%)	Starch (%)	
				Dry	Fresh
PSB	753.93 <sup>a</sup>	8.12 <sup>a</sup>	76.33 <sup>a</sup>	70.46 <sup>a</sup>	9.74 <sup>a</sup>
PBS	767.09 <sup>a</sup>	7.48 <sup>b</sup>	73.55 <sup>b</sup>	69.25 <sup>b</sup>	8.24 <sup>b</sup>
CBS	759.35 <sup>c</sup>	8.45 <sup>c</sup>	74.17 <sup>c</sup>	69.15 <sup>c</sup>	8.74 <sup>c</sup>
BPS	762.41 <sup>d</sup>	7.24 <sup>d</sup>	68.50 <sup>d</sup>	69.05 <sup>d</sup>	8.73 <sup>c</sup>
PSD	758.98 <sup>e</sup>	9.05 <sup>e</sup>	95.44 <sup>e</sup>	69.84 <sup>e</sup>	ND
LSD	0.544	0.051	0.0485	0.051	0.0336

ND = Not determined. Data are means of 3 replications. Values in the same column with different superscripts are significantly different ( $p < 0.05$ ). PSB = Peeled, sliced and boiled; PBS = Peeled, boiled and sliced sample; CBS = only papery brown layer peeled, boiled and sliced sample; BPS = boiled, peeled and sliced sample; PSD = peeled, sliced and dried sample.

the *wet abacha* slices ranged from 6.17 to 6.87 mg HCN/kg, while the dry samples ranged between 4.20 and 7.2 mg HCN/kg. The wet samples had a higher HCN than the dried samples. The lower HCN value of dry *abacha* slices was due to the volatile nature of the compound, which was easily lost during drying. For the dried samples, the peeled, sliced and dried sample had the highest HCN content (7.20 mg/kg) with the least percentage reduction of HCN, 83.06%. This is because only drying was employed during processing which is one of the least effective method in detoxification of cassava. Statistically significant differences ( $p < 0.05$ ) were observed in the HCN content of the fresh sliced *abacha* samples. For the *abacha* samples, CBS had the least percentage reduction of HCN for both wet and dry *abacha* slices. This could be because of the cortex that was not removed, which contains higher proportions of HCN (Ihekoronye and Ngoddy, 1985).

The range (4.20-7.20 mg/kg) of HCN for both the fresh and dry samples of the product (Table 4) were below the recommended safety level of 10 mg/kg HCN reported by World Health Organization (Adindu et al., 2003). Therefore the methods of processing of *abacha* used in this study adequately detoxified the cassava.

### Functional properties

The functional properties of flour derived from sliced *abacha* samples are presented in Table 5. The peeled sliced and dried samples had the highest swelling index and water absorption capacity of 9.05 and 95.44%, respectively, while the *abacha* from boiled, peeled and sliced cassava had the least values of 7.24 and 68.50%, respectively. The high water binding capacity of the PSD samples may be attributed to a high proportion of undegraded starch granules (Soni et al., 1985; IITA, 2003) or to a lack of association of the starch polymers in the native granules. Increase in water absorption capacity

in food systems enables bakers to manipulate the functional properties of dough in baked products (Omodamiro et al., 2007). This implies that PSD sample can confer good functionality when used as composite flour in confectionery products. Processing significantly ( $p < 0.05$ ) affected the swelling indices and the water absorption capacities of the samples.

Starch in the peeled, sliced and dried (PSD) samples ranged from 69.09 to 70.46% for dry, while the range was 8.24 to 9.74% for the fresh samples. For both fresh and dried samples, the *abacha* from peeled, sliced and boiled cassava roots had the highest values of 70.46 and 9.74%, respectively. The starch content of dried *abacha* was higher than the fresh ones in all the samples, due to removal of moisture by drying (Canovas and Mercado, 1996). Values obtained in this study are close to the reported value of 70% (Asiedu, 1998). Significant differences ( $p < 0.05$ ) were observed in starch contents of the samples.

The energy content of the samples ranged from 753.93 to 767.09 Joules. The PBS samples had the highest (767.09 J) energy content, while the PSB sample had the lowest energy content (753.93 J). There were significant ( $p < 0.05$ ) differences in the energy content of the samples except for sample PSB and PBS.

### Pasting properties

The pasting properties of the flour derived from samples are presented in Table 6. Peak viscosity of the *abacha* samples ranged from 100.83 to 109.00 RVU (rapid visco-analyser unit). The peeled, sliced and dried (PSD) sample had the highest peak viscosity (181.00 RVU) at a temperature of 64°C in 8.99 min. The peak viscosity of the *abacha* samples was significantly lower than the PSD sample, with the peeled, boiled and sliced (PBS) sample having the least peak viscosity (100.00 RVU) at 63.90°C in 9 min. The higher peak viscosity of PSD indicates that

**Table 6.** Effect of processing on the pasting properties of flour derived from *abacha* slices from 97/4779.

Sample	Pasting properties						Pasting	
	Peak viscosity	Trough	Break down	Setback viscosity	Final viscosity	Time	Temperature	
	RVU	RVU	RVU	RVU	RVU	(min)	(°C)	
CBS	117.92 <sup>a</sup>	81.02 <sup>c</sup>	36.90 <sup>c</sup>	94.44 <sup>c</sup>	175.40 <sup>c</sup>	9.01 <sup>a</sup>	64.20 <sup>c</sup>	
PBS	100.83 <sup>a</sup>	54.58 <sup>b</sup>	46.25 <sup>b</sup>	107.00 <sup>b</sup>	161.58 <sup>b</sup>	9.00 <sup>a</sup>	63.90 <sup>b</sup>	
PSB	110.52 <sup>a</sup>	87.08 <sup>a</sup>	22.44 <sup>a</sup>	81.50 <sup>a</sup>	168.52 <sup>a</sup>	8.99 <sup>a</sup>	64.00 <sup>a</sup>	
BPS	109.33 <sup>a</sup>	74.92 <sup>d</sup>	34.41 <sup>d</sup>	96.56 <sup>d</sup>	171.00 <sup>d</sup>	8.98 <sup>a</sup>	64.80 <sup>d</sup>	
PSD	181.00 <sup>b</sup>	93.73 <sup>e</sup>	87.27 <sup>e</sup>	53.43 <sup>e</sup>	152.13 <sup>e</sup>	4.17 <sup>b</sup>	64.01 <sup>e</sup>	
LSD	26.763	0.05	0.055	0.05	0.0613	0.055	0.0613	

RVU = Rapid visco analyser Units. Values are means of 3 replications. Values in the same column with different superscripts are significantly different ( $p < 0.05$ ). PSB = Peeled, sliced and boiled; PBS = Peeled, boiled and sliced sample; CBS = only papery brown layer peeled, boiled and sliced sample; BPS = boiled, peeled and sliced sample; PSD = peeled, sliced and dried sample.

boiling and soaking reduced the peak viscosity of cassava flour and also shows that PSD should have the best starch content (Osungbaro, 1990). The relatively high peak viscosity exhibited by flours from PSD samples is an indication that the flour may be suitable for products requiring high gel strength and elasticity. The peak viscosity of PSD was significantly ( $p < 0.05$ ) different from the peak viscosity of all the *abacha* samples, which did not differ ( $p > 0.05$ ) from each other.

The setback viscosity of the PBS sample was the highest (107.00 RVU), while PSD had the lowest value of 53.43 RVU. The high setback viscosity of PBS indicates higher potential for retrogradation in food products (Olapade et al., 2004; Otegbayo et al., 2006) as it is a measure of gel stability. The setback viscosity of samples differed significantly ( $p < 0.05$ ) from each other.

The pasting temperature of the *abacha* flour samples ranged from 63.90 to 64.80°C. Flour from BPS had the highest value of 64.80°C, while that from PBS had the lowest (63.90°C) indicating low temperature at which the sample formed paste during heating. Increase in viscosity would therefore be detected first in BPS. Pasting temperatures obtained in this study (for *abacha* flour) is similar to the value reported by Sanni et al. (2005) for gari, but Omodamiro et al. (2007) reported slightly higher pasting temperatures (69-80°C) for *lafun* and starch processed from different cassava genotypes. Differences in the reported values may be due to the processing methods used, varietal differences and the nature of the starch involved. A higher pasting temperature implies higher water binding capacity, higher gelatinization time and lower swelling property of starch due to a high degree of association between starch granules (Numfor et al., 1996). Flour from *abacha* of BPS would therefore have lower swelling properties as observed (Table 4). There were significant differences ( $p < 0.05$ ) in pasting temperature of the samples.

The trough (holding strength) ranged from 54.58 to 93.73 RVU. The peeled, sliced and dried (PSD) sample had the highest value (93.73 RVU) indicating that the granules would be less prone to rupture during pasting and its cooked paste would be more stable than the paste of other samples (Adebowale et al., 2008). The peeled, boiled and sliced (PBS) sample had the lowest value (54.58 RVU). There were significant differences ( $p < 0.05$ ) in the holding strength of the samples.

Breakdown viscosity ranged from 22.44 to 87.72 RVU. The lowest breakdown viscosity of PSB implies that it probably had very weak cross-linking within the granules (Oduro et al., 2005), which may result from much rupture of the starch granules. The *abacha* processing method significantly ( $p < 0.05$ ) affected the value of breakdown viscosity of the samples.

The peak time of samples ranged from 4.17 to 9.01 min. The PSD sample had the lowest value (4.17 min), while the flour from CBS had the highest (9.01). This shows that the peeled, sliced and dried (PSD) sample attained peak viscosity in the shortest time and will gel faster than others. This implies that less energy would be used during its thermal processing. Processing methods did not significantly ( $p > 0.05$ ) affect the peak time of the *abacha* samples but PSD sample significantly ( $p > 0.05$ ) differed from the *abacha* samples

## Conclusions

From the result in this study, the following conclusions are made. Boiling, peeling before slicing cassava roots (BPS) produced *abacha* with highest (39.13%) yield. The high fibre content of the samples (2.56-2.86 g/100 g) would make consumers of *abacha* processed following the methods adopted in this study not to suffer gastro and circulatory disease epidemiologically linked to low fibre

intake. HCN (6.17-7.20 mg HCN/kg) of the samples were within the safe range for human consumption. The PSD sample, which had high peak viscosity would be suitable for products that require high gel strength and elasticity. Flour made from *abacha* processed from peeled, boiled and sliced cassava roots would be stable in foods, while higher retrogradation would be experienced when flour from PBS is used in food manufacture.

Consumers preferred *abacha* slices that were boiled, peeled and sliced in all sensory attributes, while they did not like the one made by peeling only the brown papery peel of cassava root. Therefore, *abacha* slices from TMS 97/4779 should be processed by boiling, peeling and slicing before soaking and washing.

### Conflict of Interests

The authors did not declare any conflict of interests.

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