EFFECTS OF PACKAGING AND STORAGE CONDITION ON FUNCTIONAL PROPERTIES AND QUALITY ATTRIBUTES OF CASSAVA FLOUR (CVS. ‘TME 419’ AND ‘UMUCASS 36’)

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DECLARATION

By submitting this thesis/dissertation, I declare that the entirety of the work contained therein is my own, original work, that I am the sole author thereof (save to the extent explicitly otherwise stated), that reproduction and publication thereof by Stellenbosch University will not infringe any third party rights and that I have not previously in its entirety or in part submitted it for obtaining any qualification.

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SUMMARY

Cassava flour is recommended for substitution with wheat flour in composite flour for baking. The potential use of cassava flour in the food and pharmaceutical industries is attributed to its gluten-free nature and excellent functional properties. However, optimum packaging solution and storage conditions for cassava flour is critical in maintaining the quality attributes and shelf-life stability during storage. Therefore, this study focused on investigating the effects of package types (plastic buckets, low density polyethylene (LDPE) bags and brown paper bags) and storage conditions (cool condition (15 °C, 90% RH); ambient condition (23 °C, 60% RH); and higher condition (38 °C, 60% RH)) on the functional properties, quality attributes and shelf-life stability of cassava flour (cvs. 'TME 419' and 'UMUCASS 36') developed at the National Root Crops Research Institute, Umudike, Nigeria. Proximate composition, physicochemical attributes, functional properties, and microbial safety of flour were analysed every 4 weeks for 12 weeks storage duration.

Flour stored under cool condition with paper bags became moist and sticky with appearance of mould growth before 4 weeks of storage. However, at the end of 12 weeks storage, a decline in moisture content of 11.00 ± 0.02 and 7.05 ± 0.01% flour of 'TME 419' was observed at ambient and higher conditions, respectively. Rate of moisture decline was similar in flour of 'UMUCASS 36'. A slight decrease in protein content of flour was observed during the 12 weeks storage from 1.9 ± 0.07 to 1.30 ± 0.001% for cv. 'TME 419' and 3.0 ± 0.05 to 2.27 ± 0.001% for cv. 'UMUCASS 36'; however, no significant difference was observed under ambient and higher conditions. Cassava flour packed in paper bags and stored under higher condition (38 °C, 60%) had the highest loss (50%) of carotenoid content from 1.84 ± 0.10% to 0.91 ± 0.08%, while a minimal loss (24%) of carotenoid was observed in flour packed in plastic buckets under ambient condition. The concentration of hydrogen cyanide (HCN) decreased across all treatments and was below the safe cyanide level of 50 µg/ mL for food products. After the 12 weeks of storage, flour packed in plastic buckets had the highest aerobic mesophilic bacterial counts (3.43 ± 0.04 log cfu/ g) followed by flour in LDPE bags (3.37 ± 0.03 log cfu/ g) and paper bags (3.35 ± 0.01 log cfu/ g). No significant difference was observed in the package types; however the counts observed were within the acceptable microbial limit

Swelling power (SP), solubility and peak viscosity were used to characterise the changes in functional and pasting properties of cassava flour relevant in food industries. Flour packed in plastic buckets under ambient condition had the lowest swelling power (8.48 ± 0.55%) and peak viscosity (260 ± 0.51 RVU) compared to flour packed in LDPE and paper bags with

(9.10 ± 0.13 and 9.32 ± 0.41%) SP and (263.67 ± 4.04 RVU and 302 ± 9.52 RVU) peak viscosity, respectively. The essential minerals (sodium, potassium, copper, and iron) were significantly higher in flour of ‘TME 419’ compared to ‘UMUCASS 36’.

In summary, for the production of high grade foods such as bread where higher swelling power and viscosities are required, flour from ‘TME 419’ packed with paper bags under higher condition could be desirable. In addition, for infant formulation, flour from ‘UMUCASS 36’ packed in plastic buckets and stored under ambient condition which best maintained nutritional contents (protein and fat) and had the lowest peak viscosity would be more suitable. Flour from both cassava cultivars could be stored up to 12 weeks duration under ambient and hot tropical conditions using all package types evaluated. However, storage with paper bag under higher condition offers the chances of better shelf -life stability of cassava flour.

OPSOMMING

Daar word aanbeveel dat kassavameel in plaas van koringmeel in saamgestelde meel by gebak gebruik word. Die potensiële gebruik van kassavameel in die kos- en farmaseutiese industrieë word toegeskryf aan die glutenvrye aard en funksionele kenmerke daarvan. Optimale verpakking en stoortoestande is egter belangrik vir die instandhouding van die gehalte kenmerke en raklewe stabiliteit tydens stoor. Daarom is die fokus van hierdie studie op die effek van verskillende tipes verpakking (plastiekemmers, lae densiteits politelien (LDPE) sakke en bruin papiersakke) en stoortoestande (koel toestande (15 °C, 90% RH); omringende temperature (23 °C, 60% RH); en hoër temperature (38 °C, 60% RH) op die funksionele kenmerke, gehalte kenmerke en raklewe stabiliteit van kassavameel (kultivare. 'TME 419' en 'UMUCASS 36') wat by die Nasionale Wortelgewasse Navorsingsinstituut, Umudike, Nigerië ontwikkel is. Die komposisie, fisiochemiese kenmerke, funksionele kenmerke en mikrobiale veiligheid van meel is elke vier weke tydens die 12-weke stoortydperk ontleed.

Meel wat onder koeltoestande in papiersakke gestoor word, word klam en taai en swamme maak by vier weke van stoor 'n verskyning. Teen die einde van 12 weke stoortydperk is daar 'n afname in klammigheid van 11.00 ± 0.02 en 7.05 ± 0.01% in 'TME 419' meel by onderskeidelik omgewings- en hoër temperature. Die afname in klammigheid is soortgelyk by 'UMUCASS 36' meel. 'n Effense afname in die proteïen inhoud van die meel is tydens die 12-weke stoortydperk vanaf 1.9 ± 0.07 tot 1.30 ± 0.001% by die kultivaar . 'TME 419' en 3.0 ± 0.05 tot 2.27 ± 0.001% vir kultivaar 'UMUCASS 36' opgemerk. Geen noemenswaardige verskil is egter onder omgewings- en hoër temperature opgemerk nie. Kassavameel wat in papiersakke en onder hoër temperature (38 °C, 60%) gestoor is het die hoogste verlies (50%) aan karotien inhoud vanaf 1.84 ± 0.10% tot 0.91 ± 0.08% getoon , terwyl 'n minimale verlies (24%) by meel wat in plastiekemmers onder omgewingstemperature verpak is, opgemerk is. Die konsentrasie van waterstof hidrosianied (HCN) het tydens alle behandelinge afgeneem en was onder die veilige vlak van 50 µg/ mL vir kosprodukte. Na 'n 12-weke stoortydperk het die meel wat in plastiekemmers verpak is, die hoogste mesofiliese bakterië telling getoon \ (3.43 ± 0.04 log cfu/ g) gevolg deur die meel in die LDPE sakke (3.37 ± 0.03 log cfu/ g) en papiersakke (3.35 ± 0.01 log cfu/ g). Daar was geen merkbare verskil ten opsigte van verpakkingstipes nie; die tellings wat geneem is, was almal binne die aanvaarbare mikrobiale perk.

Swelkrag (SP), oplosbaarheid en piek viskositeit is gebruik om die veranderinge in funksionele kenmerke van kassavameel wat betrekking het op die kosindustrie, te ondersoek. Meel wat onder omgewingstemperature in plastiekemmers verpak is, het die laagste swelkrag

(8.48 ± 0.55%) en piekviskositeit getoon (260 ± 0.51 RVU) getoon vergeleke met meel wat in LDPE- en papiersakke (9.10 ± 0.13 en 9.32 ± 0.41%) swelkrag en (263.67 ± 4.04 RVU en 302 ± 9.52 RVU) piekviskositeit, onderskeidelik toon. Die belangrike minerale (natrium, kalium, koper en yster) was noemenswaardig hoër in die 'TME 419' meel vergeleke met 'UMUCASS 36'.

Ten slotte, vir die produksie van hoëgraad kossoorte soos brood waar hoë swelkrag en viskositeit belangrik is, is In 'TME 419' meel onder hoër toestande verpak in papiersakke, die beste keuse. In die geval egter van babakosse is 'UMUCASS 36'meel wat in plastiekemmers verpak en onder omgewingstemperature gestoor is, en wat dus koswaardes (proteïen en vette) behou en wat die laagste piek viskositeit het, meer geskik. Meel van albei kultivaars kan vir tot twaalf weke onder omgewings- en hoë, tropiese temperature in al die verpakkingstipes wat evalueer is, gestoor word. Stoor in papiersakke onder hoër temperature verbeter egter die kanse op beter raklewe stabiliteit.

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Language and style used in this thesis are in accordance with the requirements of the International Journal of Food Science and Technology. This thesis represents a compilation of manuscripts where each chapter is an individual entity and some repetition between chapters has, therefore, been unavoidable.

CHAPTER 1

INTRODUCTION

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INTRODUCTION

Cassava (*Manihot esculenta* Crantz*)* is a tuberous root crop grown in the tropics between latitudes 30°N and 30°S, with low cost vegetative propagation. It belongs to the family of *Euphorbiaceae* and originated from South America (Nhassico *et al.*, 2008). The root is drought resistant and capable of growing in different types of soil and seasons (Taiwo, 2006). As one major staple food in the tropical and subtropical region, cassava provides food for a population of more than 500 million across Africa, Latin America and Asia (Opara, 1999; Montagnac *et al*., 2009). The root, which is the major edible part of the crop, is rich in carbohydrates, and the starch content (86.49 ± 2.68%) is higher compared to other root and tuber crops such as yam (10.7 ± 1.1%), sweet potato (69.15 ± 5.85%), and taro (11.2 ± 1.26%) (Lebot *et al.*, 2009). However, it is low in protein, fat, fibre as well as some vitamins and minerals (Charles *et al.*, 2005). Utilisation of cassava root as a food source and as industrial raw material is limited because of the rapid postharvest deterioration which starts within two days after harvest (Sánchez *et al.*, 2006; Opara, 2009; Iyer *et al.*, 2010). This situation shortens the shelf-life of the root, leading to postharvest loss, low products yield and poor market quality of fresh root and minimally processed cassava food products such as gari and flour (Van Oirschot *et al.*, 2000).

Fresh cassava root contains a toxic compound (hydrogen cyanide), which is harmful for human consumption and apparently detrimental for the use of cassava in food industries (Iglesias *et al.*, 2002). However, research have shown that processing techniques such as peeling, fermentation, soaking and drying can detoxify and reduce the cyanide content, improve palatability and add value to the root (Cardoso *et al.*, 2005; Burns *et al.*, 2012). Converting cassava root into food forms and raw materials such as fufu, garri, tapioca, flour, chips and pellets can extend the shelf-life, facilitate trade and promote industrial use (Taiwo, 2006; Fadeyibi, 2012).

Globally, there is a notable increase in demand and price of cereals, especially wheat, which consequently influences the price of cereal-based products (FAO, 2013). The rising cost of importing wheat flour for food in many developing countries such as Nigeria has spurred the need for research to develop suitable flour from local agricultural materials such as cassava, which are cheaper but also possesses suitable quality attributes and functional properties. Furthermore, wheat flour contains gluten which causes celiac disease especially to gluten intolerant persons (Briani *et al.*, 2008). Studies have recommended gluten-free diet as a suitable treatment for patients with celiac disease, gluten intolerance and wheat allergic reactions (Gaesser & Angadi, 2012; Alvarez & Boye, 2014). Therefore, non-wheat gluten-free flour developed from root and tuber crops such as sweet potato (*Ipomoea batatas*), cassava (*Manihot esculenta*), potato (*Solamum tuberosum*), yam (*Dioscorea spp*) and cocoyam (*Xanthosoma sagitifolium*) offer the potential to alleviate the double burden of rising cereal prices and gluten intolerance (Aryee *et al.*, 2006; Ammar *et al.*, 2009; Sanful & Darko, 2010). In particular, the use of gluten-free flour from high quality cassava root as composite flour in highly sought after foods such as bread has gained popularity in many developing countries (Eddy *et al.*, 2007).

High quality cassava flour is white or creamy, unfermented and gluten-free flour obtained from cassava root and it is used in the food industry for the production of pasta and confectionery (Taiwo, 2006; Shittu *et al.*, 2008). When wheat was substituted by up to 20% in bread, Eddy *et al.* (2007) found that cassava flour added no foreign odour or taste to the product formed and no significant changes were observed in other bread characteristics. The physicochemical properties of cassava flour offer the benefit of good functionality as raw material for the manufacturing of various food products. For instance, the high starch content of cassava flour contributes to crispy texture of processed products (Falade & Akingbala, 2010), while its low fat content is an excellent attribute for controlling rancidity and enhancing shelf-life stability of the product (Charles *et al.*, 2005; Eleazu *et al.*, 2011). Flour and other materials used in manufacturing food products need to be packaged and stored properly prior to utilisation to ensure the quality, safety and storage stability. To realise the full potential of cassava flour in food processing, either alone or in combination with other raw materials such as wheat flour, knowledge of the effects of package types and storage conditions on quality and shelf-life stability of cassava flour is important.

Packaging materials used for the storage of flour products include plastic containers, polymeric and paper bags (Aryee *et al.*, 2006; Opara & Mditshwa, 2013). The packaging type and storage conditions applied affect the quality, shelf-life and safety of food products through their influences on moisture content, water activity and nutrient compositions of the food product (Opara & Mditshwa, 2013). Previous studies have shown that both moisture and package types contribute to influence the microbial load, the shelf-life and other quality attributes of flour products (Butt *et al.*, 2004; Mridula *et al.*, 2010; Robertson, 2012). When packaging films are used, the permeability of the film to water vapour and gases is particularly important, especially with regard to the shelf-life of dry products (Siracusa, 2012).

Several studies have investigated the effects of different processing techniques on cyanide content retention, the proximate composition, physicochemical, pasting and functional properties of cassava flour and other cassava products (Aryee *et al.*, 2006; Cumbana *et al.*, 2007; Iwe & Agiriga, 2014). Aryee *et al*. (2006) reported that cassava flour vary in its functionality because of the differences in its inherent physicochemical properties. The authors also noted that some cultivars could be useful for the production of starch, glucose syrup, ethanol, baking and other industrial purposes. Other researchers evaluated the acceptability of cassava flour-based products in different proportions with wheat flour (Eddy *et al.*, 2007; Nwabueze & Anoruoh, 2011). Iwe and Agiriga (2014) observed that the use of mechanical shredder will facilitate commercial production of ighu (cassava flakes) thereby promoting economy in developing countries. Several extensive studies have reported the effects of packaging and storage condition on quality of a wide range of whole and minimally processed food products such as fruit and vegetables, meat and dairy (Caleb *et al*., 2012; Opara & Mditshwa, 2013; O'Grady *et al.*, 2014). However, there is limited knowledge on the potential impacts of packaging and storage conditions on quality attributes of stored cassava flour, particularly the cultivars bred for wheat substitution in baking. The hypothesis of the current study is that cassava flour packaged and stored at different storage conditions will show variations in the quality attributes and shelf-life stability.

In order to test the hypothesis, this study focused on investigating the effects of storage conditions 15 °C, 90% RH (cool condition); 23 ± 2 °C, 60% RH (ambient conditions); and 38 ± 2 °C, 60% RH (higher condition) and three selected packaging materials (brown paper bags, low density polyethylene bags (LDPE) and plastic buckets) on the quality, functional properties and shelf-life of two cassava flour (cvs. 'TME 419' and 'UMUCASS 36') developed at the National Root Crops Research Institute, Umudike, Abia state, Nigeria. This was accomplished by the following specific objectives:

1. Investigating the effects of storage conditions and duration on physicochemical properties and microbial quality of the flour of two cassava cultivars (cvs. 'TME 419' and 'UMUCASS 36';
2. Evaluating the impact of selected packaging materials on the quality attributes and microbial stability of cassava flour; and
3. Characterising the functional, pasting properties and mineral content of cassava flour under different storage conditions.

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CHAPTER 2 LITERATURE REVIEW

LITERATURE REVIEW ON POSTHARVEST HANDLING AND STORAGE OF FRESH CASSAVA ROOT AND PRODUCTS

CHAPTER 2

LITERATURE REVIEW ON POSTHARVEST HANDLING AND STORAGE OF FRESH CASSAVA ROOT AND PRODUCTS

1 Introduction

Cassava (*Manihot esculenta* Crantz), also referred to as *yucca* in Spanish, *mandioca* in Portuguese and *tapioca* in French, belongs to the *Euphorbiaceae* family (Opara,1999; Burrell, 2003). It has been reported that the crop originated from South America and was domesticated between 5,000 and 7,000 years B.C. (Olsen & Schaal, 2001). The first import of cassava to Africa was by the Portuguese from Brazil in the 18th century, but now cassava is cultivated and consumed in many countries across Africa, Asia and South America (Nhassico *et al.*, 2008; FAOSTAT, 2013). The crop has drought resistant root which offers low cost vegetative propagation with flexibility in harvesting time and seasons (Haggblade *et al.*, 2012). Cassava can be cultivated throughout the year between latitude 30º N and 30º S, in different soil types except hydromorphic soil with excess water (Iyer *et al.*, 2010). The stem grows to about 5 m long with each plant producing between 5 to 8 long tubers with firm, homogenous fibrous flesh covered with rough and brownish outer layer of about 1 mm thick (Fig. 1). The root can be stored in the ground for over 2 years, and this serves as a means of food security to the farmer in West African countries such as Nigeria (Nhassico *et al.*, 2008; Falade & Akingbala, 2010).

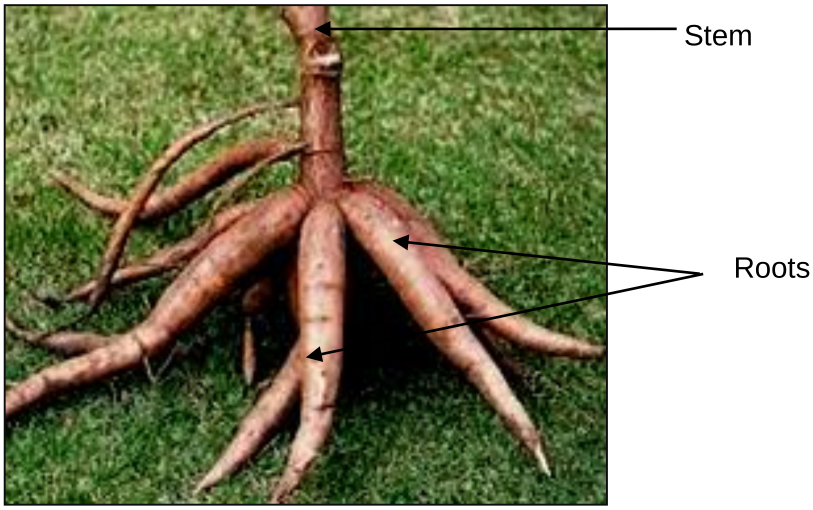

Figure 1 Picture of cassava root (http://www.greenharvest.com.au)

Cassava is a subsistence crop in Africa, and supplies about 200 - 500 calories per day (836.8 – 2092J) for households in the developing countries (Sánchez *et al.*, 2006; Omodamiro *et al.*, 2007). In the early years, cassava was neglected as food crops because of its low protein content (< 2%) and high cyanide content (120-1945 mg HCN equivalent/ kg) (Iglesias *et al*., 2002; Charles *et al*., 2005), but it is considered the fourth most energy rich food source due to the high (>70 %) carbohydrate content (Falade & Akingbala, 2010). The leaf of cassava plant is higher in protein (3 - 5%) and some macro nutrients, and therefore consumed as vegetable in some countries (Salcedo *et al.*, 2010; Burns *et al*., 2012). However, the tuberous root is the major edible part of the crop. The root serves as a source of food security against famine because of its long storage ability in the ground prior to harvest (El-Sharkawy, 2004). The root can be processed into different food forms for human consumption, animal feed and as industrial raw material for paper, textiles and alcoholic drinks (Falade & Akingbala, 2010; Haggblade *et al.*, 2012). In Thailand, cassava dry chips and pellets are the major export commodity (Falade & Akingbala, 2010), while in Nigeria, it is processed mainly into gari and fufu.

Utilisation of cassava root in food is numerous, however, the potential in food and other industrial applications is limited by the rapid postharvest physiological deterioration, which reduces the shelf-life and degrades quality attributes (Sánchez *et al.*, 2006). This physiological deterioration is attributed to its high moisture level (60 to 75%), and respiration rate which continues even after harvest (Salcedo *et al.*, 2010), resulting in softening and decay of the root and thus rendering it unwholesome for human consumption. Other factors that can cause deterioration of cassava root include pests, disease, and mechanical damage such as cuts and bruises which occur during postharvest handling and processing (Falade & Akingbala, 2010; Iyer *et al.*, 2010). The cut area exposes the root to vascular streaking and microbial attack, thereby accelerating deterioration and decay (Opara, 1999; Opara, 2009; Buschmann *et al.*, 2000). Studies have shown that physiological changes start within 24 h after harvest with a blue black discolouration commonly appearing on the root after 72 h (Iyer *et al.*, 2010; Zidenga *et al.*, 2012). The colour change of the root is accompanied by fermentation and thereafter an offensive odour indicating complete rotting (Reilly *et al.*, 2004). This rapid degradation of quality in fresh cassava roots is a major reason for the poor utilisation, poor market quality, short root storage life and low processing yield (Reilly *et al.*, 2004; Sánchez *et al.*, 2006).

Converting cassava root to other food forms creates products with longer shelf-life, adds value to the root, and reduce postharvest loses (Falade & Akingbala, 2010). Furthermore, the application of novel postharvest handling, processing, packaging and storage techniques is of critical importance for successful large scale production and utilisation of cassava roots and products. Successful application of these postharvest technologies will contribute towards maintaining product quality and safety as well as reducing incidence of postharvest losses, and thereby, improve food security (Opara, 2013). An overview of some key peer review articles on aspects of postharvest handling, processing and utilisation of cassava root is presented in (Table 1). However, information on the postharvest handling, processing and storage of cassava roots and products are limited in comparison with other globally important food crops such as wheat (Butt *et* al., 2004; Kolmanič *et al*., 2010) and rice (Falade *et al.,* 2014). Therefore, this study reviews the postharvest handling and spoilage mechanisms of cassava root, and the role of packaging and storage on quality of fresh cassava root and products.

Table 1 Overview of selected peer reviewed articles on cassava with emphasis on postharvest handling and processing.

Scope of review	Recommendations/ findings	References
A. Postharvest handling and processing		
The influence of texture modifiers on the quality attributes of dried fufu flour	Increase in starch stability and on the cooked fufu	Adebowale *et al*. (2005)
Utilisation of cassava for food; challenges, processing and raw material improvement	The use of appropriate techniques such as fermentation enhances value addition	Falade and Akingbala (2010)
Fermentation activities of the latic acid bacteria in garri production	Traditional fermentation of cassava is dominated by a lactic acid bacteria (LAB) population	Kostinek *et al*. (2005)
Developments in processing of cassava for value addition through biotechnological means	Different products from cassava	Pandey *et al*. (2000)
The domestic and industrial uses of cassava roots and products	Need to improve the cultivation of cassava because of the many uses	Taiwo (2006)
Traditional cassava foods and processing	Different processing techniques for cassava products	Aloys and Hui Ming (2006)
Comparison of cyanogen content and chemical composition of cassava products	Wet fermentation decreases nutritional value of cassava but reduces the cyanogenic level	Muzanila *et al*. (2000)
Composition, structure and physicochemical attributes of some root and tuber starches	The utilisation of the starches in the industries	Hoover (2001)
Identification of gluten substitutes with low cost to improve cassava bread volume and structure	Cassava bread was accepted and egg white improved its nutritional composition	Eggleston *et al*. (1992)
Proposed models for the regulation of cyanogenesis in cassava	Biochemical and transgenic plant approaches to reducing the cyanogen content of cassava	McMahon *et al*. (1995)

Table 1 continued

Scope of review	Recommendations/ findings	References
Effect of high production cost, starch loss and environment impact	Starch processing technology for industrial uses	Sriroth *et al*. (2000)
Cyanide reduction in cassava flour	Wetting and drying reduces cyanide content in flour.	Cumbana *et al*. (2007)
Nutritional value of cassava as food and recent advances for advance	Bio fortification key to alleviating some aspect of food insecurity	Montagnac *et al*. (2009)
Effects of cyanogenic glucoside and glucosidases in cassava roots	Effect of processing and detoxification in cassava roots	Jansz and Uluwaduge (2012)
B. Others		
Spread of cassava brown streak disease in the eastern part of Africa and its control.	Creating awareness of the disease will help to control its spread	Hillocks and Jennings (2003)
Development of transgenic technology in cassava.	Application of technology to improve cassava	Taylor *et al.* (2004)
The use of improved cassava cultivar to alleviate economy of the less privileged	Breeding, one major aspect in cassava productivity.	Kawano and Cock (2005)
The use of industrial fatty waste such as cassava flour as biosurfactant production.	Biosurfactants help to lower chemical toxicity.	Gautam and Tyagi (2006)
Disorders associated with cassava diet	Tropical ataxic neuropathy (TAN) and kenzo	Adamolekun (2011)
BioCassava Plus (BC+) program intended to improve the health of Africans through modern biotechnologies.	Efficacy of using transgenic strategies for the biofortification of cassava	Sayre *et al*. (2011)

2 Economic importance of cassava

Annual global production of cassava is estimated to be over 238,000 tonnes, with Africa contributing about 54 %, followed by Asia and South America (Table 2). Cassava root produces excellent flour quality and therefore has been promoted as composite flour for use in the food industries (Shittu *et al.*, 2008). Cassava flour is also highly recommended in the diet of celiac patients with strict adherence to gluten-free food products (Briani *et al.*, 2008; Niewinski, 2008). Celiac disease is an autoimmune complex that affects the bowel after the ingestion of gluten-containing grains or cereals such as wheat and rye (Briani *et al.*, 2008).

Table 2 World leading cassava producers (tonnes)

Countries/ year	**2008**	**2009**	**2010**	**2011**	**2012**
World	233,083,324	237,985,098	243,489,480	262,753,309	262,585,741
Africa	122,246,224	123,080,801	134,406,803	147,597,851	149,479,840
Angola	10,057,375	12,827,580	13,858,681	14,333,509	10,636,400
Benin	3,144,551	3,787,918	3,444,950	3,645,924	3,295,785
Cameroon	2,882,734	3,340,562	3,808,239	4,082,903	4,200,000
Congo	1,196,300	1,231,000	1,148,500	1,150,000	1,200,000
Côte d Ivoire	2,531,241	2,262,170	2,306,839	2,359,015	2,412,371
Ghana	11,351,100	12,230,600	13,504,086	14,240,867	14,547,279
Malawi	3,491,183	3,823,236	4,000,986	4,259,301	4,692,202
Mozambique	4,054,590	5,670,000	9,738,066	10,093,619	10,051,364
Nigeria	44,582,000	36,822,250	42,533,180	52,403,455	54,000,000
Sierra Leone	1,988,561	2,814,576	3,250,044	3,412,546	3,520,000
Uganda	5,072,000	5,179,000	5,282,000	4,757,800	4,924,560
Tanzania	5,392,358	5,916,440	4,547,940	4,646,523	5,462,454
Zambia	1,185,600	1,160,853	1,151,700	1,266,295	1,300,000
Asia	76,046,076	81,345,012	74,951,223	80,477,236	80,744,003
China, mainland	4,400,000	4,500,000	4,550,000	4,500,000	4,560,000
Cambodia	3,676,232	3,497,306	4,247,419	8,033,843	7,613,697
Democratic Republic	15,013,490	15,054,450	15,013,710	15,024,172	16,000,000
India	9,056,000	9,623,000	8,059,800	8,076,000	8,120,000
Indonesia	21,593,052	22,039,148	23,918,118	24,009,624	23,922,075
Philippines	1,941,580	2,043,719	2,101,454	2,209,684	2,223,144
Thailand	25,155,797	30,088,024	22,005,740	21,912,416	22,500,000,
Viet Nam	9,309,900	8,530,500	8,595,600	9,897,913	9,745,546
South America	33,041,504	31,448,411	31,936,808	32,097,924	30,057,840
Brazil	26,703,039	24,403,981	24,967,052	25,349,088	23,044,557
Colombia	1,803,911	2,250,233	2,082,440	2,164,850	2,274,358
Paraguay	2,218,530	2,610,000	2,624,084	2,453,837	2,560,000
Peru	1,171,818	1,166,017	1,240,121	1,115,593	1,119,560

FAO (2013) http://faostat3.fao.org/faostat-gateway/go/to/download/Q/QC/E

In view of enhancing cassava productivity to promote economic development, the global mandate on cassava research was given to the International Centre for Tropical Agriculture (CIAT) in Colombia while the International Institute for Tropical Agriculture (IITA) in Nigeria obtained the regional mandate on cassava research (El-Sharkawy, 2007). However, due to widespread consumer preference for maize, cassava cultivation in South Africa is low compared to other African countries like Nigeria, Ghana, Angola,Tanzania, Uganda and Malawi. Cassava production in South Africa is limited to small scale farmers close to Mozambican border, with annual production between 8 and 15 t/ha (Mabasa, 2007) compared to 54,000 tonnes productions in Nigeria (FAO,2013).

Cassava is one of the major tropical staple foods alongside yam, plantain, and sweet potato, and is considered as a good source of carbohydrate and the fourth most energy-giving diet (Mudombi, 2010). Some cultivars are produced for human consumption while some are for animal feed (Falade & Akingbala, 2010), however, studies have shown that cultivars such as TMS 94/0330, 91/02324, 92/0035, 001/0355, TME 1, UMUCASS 36, and 92/0057 are suitable for food as well as feed (Aryee *et al.,* 2006; Eleazu *et al.,* 2011). The starch obtained from the root of most cultivars is used for making traditional desserts, salad dressing, soup thickener, binding agent in sausages, high fructose syrup, and in textile industries (Montagnac *et al.*, 2009). In countries such as Brazil, cassava is basically cultivated for local industrial purposes, while in Thailand it is an export commodity. In parts of sub-Saharan Africa it is grown mainly by subsistence farmers for consumption as staple food and as a source of income (Falade & Akingbala, 2010). Cassava is being explored as a potential bio-fuel crop in countries like China and Thailand (Zidenga *et al.*, 2012). In Brazil, the bio-fuel from cassava is used by flex-fuel light vehicles while in the United States it is used as gasoline (Adelekan, 2010; Adelekan, 2012).

Demand for cassava has increased most especially in developing countries where low supplies of cereals are experienced. This is because of its significant uses in food and beverage industries as composite flour (Eddy *et al.*, 2012). Over the years, there have been cases of geographical scarcity and low supply of wheat thus leading to high demand for wheat, high cost of wheat flour, and wheat based food products (Olaoye *et al.*, 2006; Olaoye *et al.*, 2011). This situation led to the production of different flour products such as plantain flour, cocoyam flour, taro flour as well as cassava flour. These are substitutes to wheat flour in varying proportions ranging from 5 to 30% (Giami *et al.*, 2004; Eddy *et al.*, 2007). Based on sensory evaluation studies, 20% wheat/cassava composite flour was recommended for bread recipe because the product quality attributes showed no distinct variation when compared with 100% wheat flour

(Eddy *et al.*, 2007; Sanful & Darko, 2010). In addition to the food uses of cassava root, it can also be used in the production of paper, textiles, plywood, glue and alcohol (Raemakers *et al.*, 2005; Adelekan, 2012). Cassava leaves are rich in protein (3 - 5%) and some essential minerals such as calcium, nitrogen and potassium; as a result of this, leaves serve as vegetable in soups to supplement the low protein content of the root (Odii, 2012). The root, which is the major source of food, can be boiled or roasted and eaten as fresh root with sauce or soup especially the low cyanide or sweet type of cassava roots (Lebot *et al.*, 2009). Cassava roots could also be minimally processed into various primary and secondary products (Fig. 2) (Falade & Akingbala, 2010).

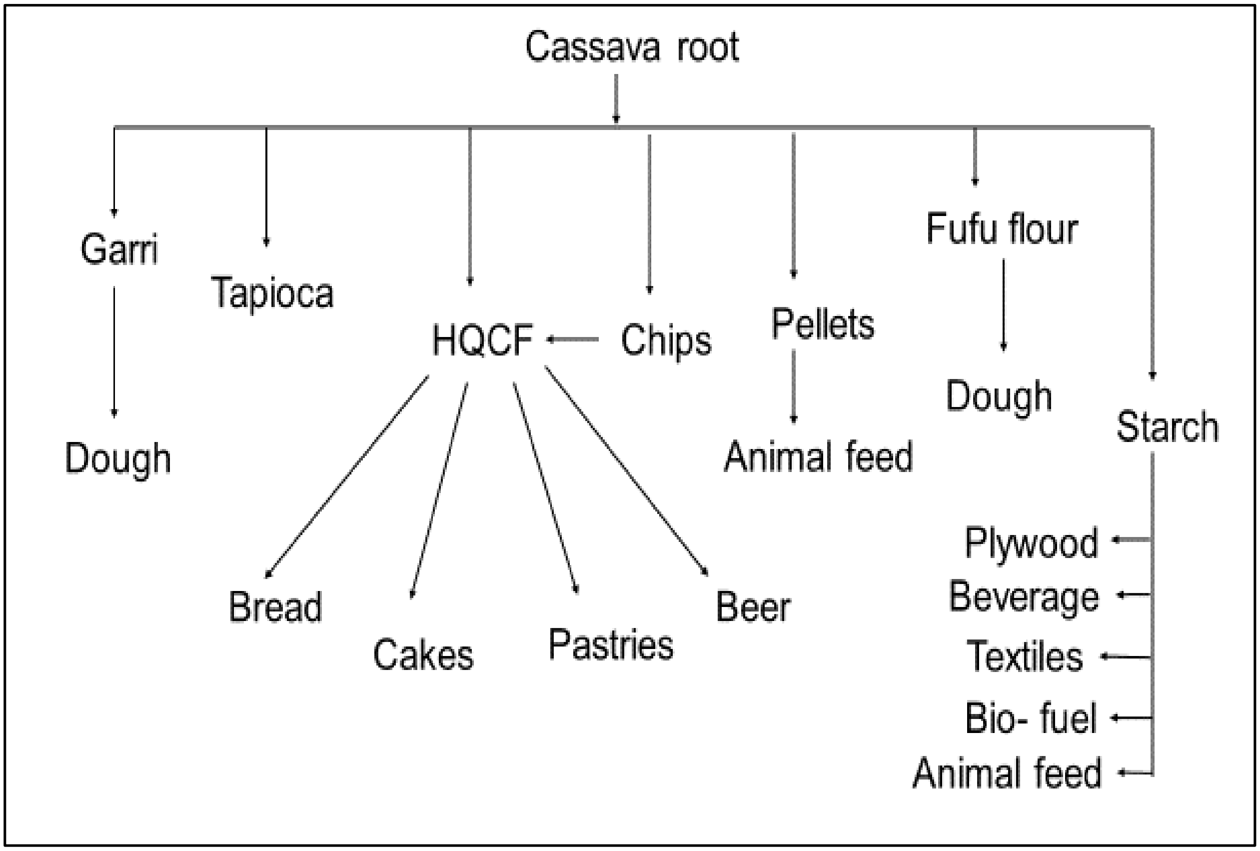

Figure 2 Different products derived from minimal processing of cassava root (Montagnac *et al.*, 2009; Falade & Akingbala, 2010).

3 Classification of cassava root

Cassava roots may be classified into sweet and bitter based on the level of cyanogenic glucoside in the tissue. The major cyanogenic glucosides found in cassava are linamarin and lotaustralin (Fig. 3), which can be hydrolysed into hydrogen cyanide (HCN) (Iglesias *et al.*, 2002). Hydrogen cyanide is a toxic compound harmful to human health and could lead to death if consumed in excess (Nhassico *et al.*, 2008; Burns *et al.*, 2012). Bitter cultivars of cassava root have higher level of cyanide content (28 mg HCN/ kg) than the sweet type (8 mg HCN/ kg) dry weight bases (Chiwona-Karltun *et al.*, 2004; Charles *et al.*, 2005). Sweet cassava root cultivars with lower cyanide content can be eaten fresh or boiled (Nhassico *et al.*, 2008) while the bitter type with higher cyanide concentration require further processing to eliminate the toxins before consumption (McKey *et al.*, 2010).

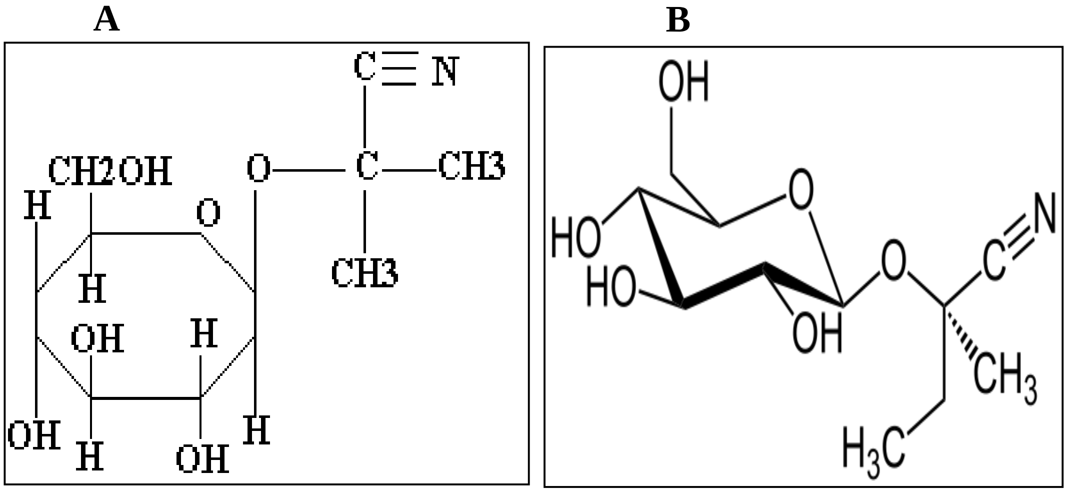

Figure 3 Molecular structures of (a) linamarin and (b) lotaustralin, the major cyanogenic glucosides found in cassava (Kannangara *et al.*, 2011)

Symptoms of cyanide consumption include fast breathing, restlessness, dizziness, headache, nausea and vomiting. In chronic cases, symptoms could result in convulsion, low blood pressure, and loss of consciousness, lung injury and respiratory failure which could lead to death (Burns *et al.*, 2012). It has also been reported that consumption of these cyanogens causes irreversible paralysis of the legs and stunted growth in children (Ernesto *et al.*, 2002; Nhassico *et al.*, 2008). Greater quantity of these glucosides are biosynthesised in the leaves and are absorbed in the root but predominantly on the peels (Siritunga & Sayre, 2004; Cumbana *et al.*, 2007). Total cyanide found in the fresh unpeeled root and the leaves range from 900 – 2000 ppm and 20 – 1860 ppm, respectively, depending on cultivar (Cardoso *et al.*, 2005). However, during processing about 90% of the HCN is lost due to the linamarin breakdown and the residual cyanogen levels should be below the safe limit (10 ppm) recommended by the World Health Organisation (WHO) for cassava flour (FAO/WHO, 1995; FAO/WHO, 2005). Removal of cyanogenic compound from the root during processing for production of cassava-based foods is one major approach to promoting safety in cassava consumption (Iglesias *et al.*, 2002). Processing techniques such as peeling, soaking/wetting, grating, dewatering, and sun drying are employed as they enhance the detoxification of cassava roots for safe consumption and prevent the occurrence of diseases (Chiwona-Karltun *et al.*, 2004; Cumbana *et al.*, 2007). Furthermore, the application of innovative/technologies such as modified processing techniques and the use of breeding of cultivars with low cyanogenic compound have been recommended for the reduction of the cyanide level in cassava root (Iglesias *et al.*, 2002; Nhassico *et al.*, 2008). The level of hydrogen cyanide is also influenced by root age, varietal and environmental factors (Charles *et al.*, 2005). Another significant factor that influences cyanogenic level in the root is seasonal variation as cyanide content in cassava flour was observed to be high when roots were harvested during the period of low rainfall, which was attributed to root dehydration during dry seasons (Cumbana *et al.*, 2007).

Various cultivars of cassava are grown worldwide most of which have been bred by the Centro International Agricultural Tropical (CIAT) in Colombia, International Institute for Tropical Agriculture (IITA) and National Root Crops Research Institute, Umudike, Nigeria (NRCRI) (Eleazu *et al.*, 2011; Sayre *et al.*, 2011). Presently, improved cultivars with desirable character traits such as high carotenoid content have been released by researchers from these institutes (Eleazu *et al.*, 2012). Carotenoids are among the most valuable food constituents because of the health benefit they offer in fighting against diseases such as cancer and cardiovascular diseases and these health-promoting properties have been attributed to their antioxidant activity (Rodriguez-Amaya *et al.*, 2011). As vitamin A precursor, they also prevent cataracts (Krinsky & Johnson, 2005). In addition, cassava with high beta carotene shows longer shelf-life of the flour and can also reduce the onset of postharvest physiological deterioration of root (Sánchez *et al.*, 2006; Eleazu *et al.*, 2011).

Cassava cultivars with novel starch content, also known as waxy cassava (Sanchez *et al*., 2010), with amylose-free or low amylose starch, have been developed using genetic mutation techniques (Zhao *et al.*, 2011). High amylose starch is associated with paste retrogradation, which is undesirable for many applications of starch paste as well as composite flour for baking purposes (Koehorst-van Putten *et al.*, 2012). In addition, paste from high amylose starch shows low viscosity and low gel clarity unlike the waxy starch (Raemakers *et al.*, 2005). The gels from waxy cassava cultivar show little or no syneresis (liquid separation in gel) during storage even as low as -20 °C and this justifies the use of flour from waxy cassava cultivars in the formulation of refrigerated or frozen foods (Sanchez *et al*., 2010). Similarly, waxy cultivars need no modification with chemicals such as alkenyl succinic anhydride and phenyl isocyanate because they form stable gels (Shimelis *et al*., 2006). The chemicals could contribute in degrading the essential nutrients in the starch, they are unfavourable to the environment and are expensive to use (Raemakers *et al.*, 2005; Sánchez *et al.*, 2010).

4 Nutritional composition of cassava root and products

Cassava root (and products) is a major staple food in many African countries, especially in West Africa. Nutritional composition could be influenced by the type of cultivar as well as the geographical location, age of the plant, environmental conditions, processing and cooking methods (Tewe & Lutaladio, 2004). In comparison with other staple foods, cassava root proved to be the third highest energy and carbohydrate source (Table 3).

Table 3 Nutritional composition of cassava root compared with other major staple foods (per 100 g).

Compositions	**Cassava**	**Maize**	**Rice**	**Wheat**	**Potato**	**Sweet potato**	**Yam**
Water (g/ 100g)	60	76	12	11	79	77	70
Energy (kJ)	670	360	1528	1419	322	360	494
Protein(g/ 100g)	1.4	3.2	7.1	13.7	2	1.6	1.5
Fat (g/ 100g)	0.28	1.18	0.66	2.47	0.09	0.05	0.17
CHO (g/ 100g)	38	19	80	71	17	20	28
Fibre (g/ 100g)	1.8	2.7	1.3	10.7	2.2	3	4.1
Sugar (g/ 100g)	1.7	3.22	0.12	0	0.78	4.18	0.5
Calcium (mg/ 100g)	16	2	28	34	12	30	17
Magnesium (mg/ 100g)	21	37	25	144	23	25	21
Phosphorus (mg/ 100g)	27	89	115	508	57	47	55

Source: http://www.nal.usda.gov/fnic/foodcomp/Data/SR18/sr18.html.

4.1 Macro- and micro-nutrient contents

Cassava is a starchy fibrous root crop, with low contents of protein, fat and fibre. However, it is rich in carbohydrate contents, ranging from 32 to 35% in fresh weight and about 80 to 90% in dry matter making it a good source of energy (Montagnac *et al.*, 2009). Carbohydrate content of the fresh root is more than that of potatoes but less when compared with rice and wheat from the table above (Montagnac *et al.*, 2009). The starch formed has about 80% amylopectin and 17 to 20% as amylose and this ratio gives cassava a functional quality for use in making confectioneries (Rawel & Kroll, 2003). It contains monosaccharide level of about 17% sucrose and little amount of fructose and dextrose and therefore could serve as a valuable raw material in high fructose syrup, beverages and pastries (Charles *et al.*, 2005). Fibre content ranges from

1.5% to 4% in processed products such as flour; however the content varies in different cultivars (Gil & Buitrago, 2002). The lipid content is relatively low (0.3) when compared with other staple foods with the exception of potato and rice.

Protein content in cassava root is very low (1 to 2%), therefore, excessive consumption of cassava for a prolonged period of time could lead to protein energy malnutrition. About 50% of the protein in cassava is whole protein while the remaining 50% is of the amino acids such as glutamic and aspartic acids and some non-proteins component (Montagnac *et al.*, 2009). Most of the macronutrients such as fat, protein and carbohydrates are higher in the un-peeled root than in peeled as shown in Table 4.

Table 4 Composition of cassava peeled and unpeeled root (Gil & Buitrago, 2002)

Constituent	**Peeled root**	**Unpeeled root**
Water (%)	71.50	68.06
Carbohydrate (%)	26.82	29.06
Crude fibre (%)	0.12	0.99
Crude protein (%)	0.74	0.87
Fat (%)	0.13	0.17
Ash (%)	0.69	0.85

Micronutrients are required by the body in smaller quantities. Most of these micronutrients are found in the cassava leaves and they include iron, zinc, manganese, magnesium, and calcium while the root contains minimal amount of the following micronutrients: iron, potassium, magnesium; copper; zinc; and manganese (Charles *et al.*, 2005). However, the calcium content is relatively high (16 mg/ 100 g) compared to maize (2 mg/ 100 g) (Montagnac *et al.*, 2009). The lipid content of cassava roots in fresh wet bases have been reported lower compared to maize and rice but higher than yam and potato, it ranges from 0.1% to 0.3% and the glycolipids are mainly galactose-diglyceride (Gil & Buitrago, 2002). The high water content of the root (> 65%) spurs the early postharvest physiological deterioration and thus limiting its utilisation and production yield. Therefore further processing will help to expand the utilisation of the root, improve the yield, stabilise shelf-life and increase palatability.

4.2 Anti-nutrients in cassava root

Cassava contains some anti-nutrients and toxic substances which inhibit the digestibility and intake of major nutrients, although these compounds can still be healthy to human health depending on the amount consumed (Montagnac *et al.*, 2009). For example, HCN is the most toxic compound found in higher level in the bitter type which makes the consumption of fresh cassava root to be restricted. It is obtained from the hydrolysis of a nitrogenous plant metabolite from amino acid known as cyanogenic glucoside (Falade & Akingbala, 2010). This compound is predominant both in the roots and the leaves although more abundant in the leaves, with contents above the FAO/WHO (1995) recommendation of < 10 ppm (Siritunga & Sayre, 2004). Studies have shown that consumption of cassava products with cyanide level within the recommended is not harmful to health but prolonged intake could lead to glucose intolerance, spastic paralysis of the legs (kenzo) (Ernesto *et al.*, 2002). Furthermore, cyanide intake, in combination with iodine deficiency, could cause goitre, cretinism and stunted growth in children (Nhassico *et al.*, 2008). Monotonous consumption of cassava diet has been associated with a chronic disease known as tropical ataxic neuropathy observed mainly in adults (Oluwole *et al.*, 2002), which results in weakness of the joints, hardness to hearing, poor vision and even blindness. In addition, cassava leaves contain higher content of cyanide and nitrate, making consumers prone to stomach cancer (Wobeto *et al.*, 2007).

Another anti-nutrient is the phytate which is a non-toxic nutrient (Fig. 4). Phytate provides storage for phosphate and insitol and is normally contained in the seed of plant (Kumar *et al.*, 2010). Phytate is formed during plant maturation and represents between 60 to 90% of the total phosphate found in the whole plant (Loewus, 2002). Irrespective of the action of this anti-nutrient against different terminal diseases like cancer, the negative effect of phytate in the body includes formation of insoluble phytate-mineral complexes leading to a decrease in mineral availability and deficiency of iron, zinc, calcium and magnesium in the body (Konietzny & Greiner, 2004). It also forms non-phytate protein complex and inhibit amlyase activities thereby degrading carbohydrate utilisation (Selle *et al.*, 2000). Processing techniques such as soaking, malting and fermentation have been observed to reduce phytate content by increasing activity of naturally occurring phytate properties (Hambidge *et al.*, 2008).

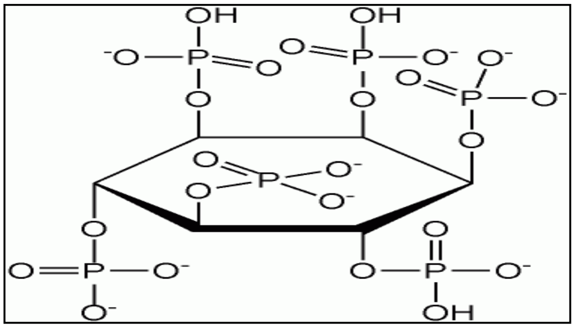

Figure 4 The molecular structure of phytate

4.3 Proximate composition

Protein, fat, and carbohydrate contents contribute to the total energy content of cassava root and products while water and ash only contribute the total mass of the product and influences shelf-life stability (Etudaiye *et al.*, 2009). Ash also indicates the availability of inorganic minerals in the product (Eleazu *et al.*, 2011).

Table 5 shows the proximate composition of fresh cassava root and some processed cassava products. While moisture content is higher in fresh cassava root, studies have shown that the composition of protein, fat, ash and carbohydrates are higher in the products formed from cassava root (Charles *et al.*, 2005; Falade & Akingbala, 2010; Falade *et al.*, 2014). This suggests that the products will have a longer shelf-life than the fresh roots, because low moisture level inhibits microbial growth while moisture level above 12% results in poor shelf stability (Aryee *et al.*, 2006). Therefore, processing is a key factor to reduce loss and maintain the quality of products and promotes adequate supply of the crop in all seasons (Akingbala *et al.*, 2005; Falade & Akingbala, 2010).

Table 5 Proximate composition of roots and some cassava products (% dry weight base) (Aryee *et al.*, 2006)

Constituent (%)	Root	Flour	Fufu	Garri
Moisture	68.1	9.9	11.9	5.8
Protein	1.1	4.4	10.9	1.0
Crude fat	0.4	3.6	4.5	0.2
Crude fibre	1.1	3.8	3.2	1.9
Ash	0.5	2.1	3.5	1.0
Carbohydrate	29.1	9.9	77.9	90.9

4.4 Functional properties of cassava products

The size and morphology of the starch granules influence the functional behaviour of cassava flour as well as the value-added products (Shittu *et al.*, 2008). For example, swelling power is the measure of starch ability to imbibe and expand in volume at a particular temperature. Low swelling power suggests that starch granules have strong binding force and low amylose content (Ikegwu *et al.*, 2009). Low amylose starch has an excellent functionality of easy digestibility when compared with the high amylose and this property is desirable for use in the food industries (Kaur *et al.*, 2013). In addition, Shimelis *et al*. (2006) reported that low swelling power in cassava flour is a clear indication of a restricted starch which shows a high resistance to breaking during cooking and the gel formed is stable at cooling phase. Swelling capacity and solubility of flour and starch are proportional to each other, such that an increase in swelling power results to increased solubility pattern. However, solubility is also influenced by the starch granular size and gelatinisation, which is a reflection of the breaking phase of the intermolecular hydrogen bond (Shimelis *et al.*, 2006).

Another unique functionality of flour is the water binding capacity (WBC). This is the ability of food products to take up and retain water either by adsorption or absorption. It contributes to the easy handling of dough during preparation of high grade food like noodles and bread (Doporto *et al.*, 2012). However, WBC of flour is influenced by the extent of starch disintegration and this implies that the rate at which starch granules break loose is proportional to the water binding capacity of the products (Falade & Okafor, 2013). Low WBC could be attributed to the high protein content in a product because high protein content has shown to limit the ability of water uptake in the food material (Wani *et al.*, 2013).

Paste stability during product development is greatly influenced by the gelatinisation and pasting properties. This pasting behaviour of starch is important in flour characterisation and use in the food industries (Shimelis *et al.*, 2006). Variation in the viscosity of gels depends on the amylose content of the starch hence gelatinised food could be used for different purposes based on the level of viscosity (Iwe & Agiriga, 2014). High viscous cassava starch is useful in the production of jelly, food thickener and binder while starch with low viscosity is suitable for weaning food production (Tsakama *et al.*, 2010).

5 Physiology of cassava root

Cassava plant requires a warm climate of greater than 20 ºC mean day temperature for optimum growth, photosynthesis and for production of roots (Olaleye *et al*., 2013). The physiological resistance to drought is attributed to the rapid enclosure of its stomata during stress from water (Allem, 2002). However, cultivation on a poor soil during severe drought results in low transpiration rate as well as reduction in the leaf canopy, which in turn leads to reduced photosynthesis rate (El-Sharkawy, 2004). Irrespective of the scattered nature of the root, it can tolerate a depth of more than 2 m in the soil, thus allowing the crop to utilise available water and nutrient. This condition makes in-ground storage for up to two years period possible (El-Sharkawy, 2007). The in-ground storage method is vital for preventing the physiological streaking of the root which leads to further decay and has also been observed to preserve the root and maintain freshness until they are needed especially during food shortage periods (Olaleye *et al*., 2013). Cassava roots as well as other root and tuber crops continue to respire after harvest and as a result, low shelf-life and high incidence of postharvest losses (Fadeyibi, 2012). The physiological conditions that occur shortly after harvest is associated to root respiration, water loss and attack by pest and diseases on any cut surface (Fadeyibi, 2012). Physical changes on the root associated with quality degradation commence 2 - 3 days after harvest, and this poses a challenge to optimal storage of freshly harvested root, marketing of the root and/or consumption.

5.1 Deterioration and spoilage of cassava root

Rapid deterioration is a major challenge limiting commercial production of cassava. As soon as the root is uprooted from the ground, it begins a process of postharvest physiological deterioration within the next 24 h. This situation proposes that fresh root cannot be stored longer than 4 days because of its high moisture content. The effective utilisation of cassava root is greatly constrained not only by low protein level and the high cyanide content but also by the rapid rate of deterioration normally characterised by a blue black discolouration of the root and the streaking of the xylem tissue (Iyer *et al.*, 2010). Therefore, the shelf-life of the root is shortened leading to wastage, poor products yield, economic losses, reduction in market quality and poor commercialisation (Van Oirschot *et al.*, 2000). However, the early signs leading to this postharvest deterioration are not fully understood, but it has been attributed to the increase in cellular respiration and the biochemical changes observed from 3 to 4 h after harvest (Iyer *et al.*, 2010). The accumulation of secondary metabolite from the phenyl-propanoid pathway (Bayoumi *et al*., 2010) as well as the increase in enzyme activities such as phenylalanine ammonia lyase (PAL) glucanase, proteinase, peroxidase and polyphenol oxidase have also been seen as factors contributing to the rate of this deterioration (Iyer *et al.*, 2010). The onset of the enzyme PAL begins within 2 - 3 min of cuts or wounds on the root and follows the progress of postharvest physiological deterioration (PPD) on the root on the second day (Akingbala *et al.*, 2005). Additionally, the point of cuts, wounds or abrasion have been observed to be the breeding place for the microorganism thus enhancing the postharvest physiological deterioration (Reilly *et al.*, 2004).

Over the years, postharvest deterioration was not a problem but with the increasing rate of urbanisation, distance between fields and processing site coupled with the unstable transport scheme in the developing countries, deterioration of root has become a serious problem.

Similarly, because of the rapid deterioration rate of the root, storage of the root becomes like an impossible task. The marketing of 2 to 3 day harvested cassava root becomes challenging as the roots are regarded unwholesome for consumption and have poor processing quality. Production of reactive oxygen species (ROS) is another early sign leading to the onset of PPD. This process is an unavoidable situation caused by the aerobic respiration, damage during harvest under stress, or when attacked by pathogens (Zidenga *et al.*, 2012). However, this early sign was induced by the level of cyanogenesis in the root, therefore to prevent the rapid case of PPD, reduction in cyanide-induced accumulation of ROS is recommended (Bayoumi *et al.*, 2010).

The postharvest deterioration which generally causes spoilage of the root can be classified into 3 different factors namely: physiological, microbial and mechanical. Spoilage of cassava root under storage has been observed to be instigated from the activities of polyphenol oxidases as they were observed in the discoloured root (Buschmann *et al.*, 2000).

Physiological deterioration

The postharvest physiological deterioration (PPD) often known as primary deterioration has been assumed to be triggered by the breaks and cuts created on the roots during harvest or processing leading to the colour change on the roots. Often there are cuts and bruises when the roots are pulled out of the ground and such areas form the onset of deterioration (Reilly *et al.*, 2003). This type of deterioration is not caused by microorganisms, but as a result of the mechanical damage and stress induced by water loss from wounds, which therefore encourages the growth of microbes (Iyer *et al.*, 2010). PPD is associated with colour change and the streaking of the xylem parenchyma tissue. These signs begin from the second day of harvest and have been likened to the normal biochemical and oxidative changes that occur as plants respond to wounds. Increased respiration of about 20 to 30 ºC and low relative humidity between 65 to 80% encourages deterioration (Sánchez *et al.*, 2006). This implies that cassava root will still undergo deterioration and spoil even without any mechanical damage because of the aerobic respiration process which continues in the root even after harvest. In addition, oxidation is observed within 15 min from the part of the injured root leading to alteration in the genes and accumulation of the secondary metabolite (Reilly *et al.*, 2004). Various techniques to reduce this postharvest deterioration have been investigated such as use of paraffin wax to coat each root but this method could only extend and maintain quality of root for few weeks (Reilly *et al.*, 2004). The exclusion of oxygen or submerging roots in water or storing in an anaerobic environment can inhibit the streaking of the xylem tissue. In addition, Van Oirschot *et al*. (2000)

observed that pruning cassava plant 2 weeks before harvesting is another technique that reduces the susceptibility of this physiological deteriorative response of the root. Similarly, Sánchez *et al*. (2006) in their study on the effect of the total carotenoid content in cassava root on the reduction and delay of postharvest deterioration concluded that roots kept at 10 ºC and 80% relative humidity can remain fresh till after two weeks. Also, total amount of carotenoids in the roots is proportional to the rate of postharvest physiological deterioration in the root; this means that higher carotenoid can reduce or delay the onsets of PPD and extend the shelf-life of the root thereby broadening the industrial uses of cassava (Chávez *et al.*, 2007).

Microbial deterioration

Physical damage such as wounds, cuts and bruises (especially during harvesting, handling operations and transportation) can serve as focal points for microorganisms and lead to the second stage of cassava root spoilage known as the microbial deterioration or the secondary deterioration (Falade & Akingbala, 2010). Microbial deterioration is induced by microorganisms that cause rotting, and two types of rot have been identified. These include: *Aspergillus niger*, *A. flavus*, *A. furngatus*, *Penicillium citrinum* and *Rhizopus* spp. (Onyimonyi, 2002). Under aerobic conditions, these organisms cause a dry rot, which results in discoloration and a slight increase in acidity of the cassava root. Also fungi attack on cassava product like chips has an influence on colour change, off flavor and taste attributes (Gnonlonfin *et al.*, 2008). Also under anaerobic conditions, deterioration is initiated by the activities of bacteria such as the *Bacillus* spp., this result in rapid development of acid in the root (Onyimonyi, 2002). The microbial deterioration of cassava root is characterised by fermentation and softening of the root tissue, this ccommences 4 - 5 days shortly after the primary deterioration from the wounded point (Buschmann *et al.*, 2000; Reilly *et al.*, 2004). Therefore this highlights on the importance of optimum postharvest handling practices, in order to minimise mechanical damage of the cassava root.

Mechanical damage

Mechanical damage occurs as a result of careless handling when harvesting and transporting from the field to the processing site and during processing and peeling of the root (Iyer *et al.*, 2010). Unfortunately, the effects of the injuries on the root are overlooked but it has been attributed as the major factor constituting the physiological deterioration of the root (Fadeyibi, 2012). In most cases damages occur during harvest with the use of farm tools and machinery or in pulling of the root from the ground. The damage on the root during transportation could be caused by the vibration or compression in the packaging materials used.

However, mechanical damage can be avoided through careful handling of root after harvest. Therefore, to minimize the effect of mechanical damage, once the root is harvested with cuts, the fresh root with cuts are subjected to a curing process for about four days under storage temperature between 30 – 40 ºC and 90 - 100% relative humidity (RH) (Fadeyibi, 2012). The time for complete curing cannot be estimated with certainty because it is determined by factors such as extent of wound, season, condition of the crop at harvest and the cultivars. The process has been seen as a means of reducing the onset of microorganism and disease as well as PPD and extending the shelf-life of the fresh root.

6 Postharvest handling and storage of fresh root

The quality, sustainability and the safety of the plant lies not just on the pre-harvest factors but most importantly on the postharvest management especially for crop like cassava with rapid deterioration rate (Iyer *et al.*, 2010). Reduction of postharvest losses can help to improve the quality of fresh cassava root. Some proven measures to prevent loss include: the use of improved cultivars with longer shelf-life; application of proper agricultural practices during cultivation; proper handling during and after harvest; and the use of the appropriate processing techniques (Kader & Rolle, 2004).

Postharvest handling of cassava roots include: storage of the fresh root, processing, packaging and storage of the processed product (Opara & Mditshwa, 2013). In addition packaging and storage are the most major factors in postharvest handling that ensures food security and safety of the final product (Daramola *et al.*, 2010). Packaging guarantees the quality of the root by protecting it from bruises and injuries and also prevents excessive moisture loss. Also the stored root influences the quality of the product formed as well as its yield (Akingbala *et al.*, 2005).

6.1 Storage of fresh cassava root

Storage of agricultural raw materials is a vital aspect in postharvest handling. It guarantees that produce remains available and adequate even when they are out of season. Root and tuber are crops with high moisture content and they are usually bulky to carry, therefore storing and transportation is often challenging. During storage, most tropical root crops such as yam and cassava transpire and lose moisture; this leads to reduction in acidity level of the crops and thereby degrading the cooking quality as well as the market value. In the rural areas, storage of cassava root was not an issue because the farmers harvest and process root immediately for consumption and this practice reduces the epidemic of deterioration (Reilly *et al.*, 2004). However, with the quest of future need of cassava, several methods were developed to extend the shelf-life of fresh cassava root (Westby, 2002; Akingbala *et al.*, 2005; Fadeyibi, 2012).

Traditional methods

In-ground storage is the simplest and easiest traditional way of extending the shelf-life of cassava root and to improve food security. Cassava root has an optimum harvesting age and flexibility of harvest which offers the advantage of longer in-ground storage. However, the stored root can be lignified due to long storage and some characteristic features are degraded in the process (Westby, 2002). Also, root could be infested by some pathogens (*Fusarium solani, Phaeolus manihotis*) or even by rodents. Some methods such as burying the root in the soil and piling the root in heaps with constant watering were also exploited (Westby, 2002). The methods were not reliable as deterioration could set in from the root kept under. Another traditional method is the coating of root with loamy soil. Nevertheless, the traditional methods were only successful in extending the shelf-life of the root for few days. It has been shown that during storage the cyanide, moisture and starch contents in the root decrease while the ash, sugar, crude fibre as well as the acidic contents increase with the length of storage (Table 6) (Akingbala *et al.*, 2005).

Table 6 Chemical compositions (g/ kg) of cassava root under 21 day storage at different storage conditions (Akingbala *et al.*, 2005)

Storage	**Moisture**		**Ash**		**Crude fiber**		**Sugar**		**Starch**	
days	**P**	**T**	**P**	**T**	**P**	**T**	**P**	**T**	**P**	**T**
0	670	670	6	6	13	13	51	51	764	764
7	610	646	8	8	14	14	73	88	673	694
14	620	648	13	14	21	19	111	100	627	599
21	571	641	18	15	26	24	151	132	558	534

*P = Polyethylene bags; *T = Trench

Improved method

Several improved methods to prevent deterioration and extend the shelf-life of root were introduced such as breeding cassava cultivars with resistance or tolerance to physiological deterioration (Morante *et al.*, 2010). Pruning of cassava plants before harvest (Iyer *et al.*, 2010), and advanced processing technology, which is the most reliable means of preventing the occurrence of PPD (Buschmann *et al.*, 2000). These technologies are encouraging as they further improve the market situation of the crop and guarantee the quality of the fresh root for longer days, but that notwithstanding, very few of the technology is being harnessed in the developing countries. The methods include:

- *Field clamp*

This method has been considered as a traditional method of storage of cassava roots and roots can be stored for up to 8 weeks as it was also confirmed with the storage of potato. Clamps are constructed by laying straws on the floor and arranging the roots followed by another layer of straws and then with soil making a heap. There is always an opening for ventilation but this method is usually affected by the wet season as the ground should be kept dry during the storage period (Fadeyibi, 2012). It is also labour intensive and requires skilled labourers (Westby, 2002).

- *Storage in a box with moist sawdust*

In this method sawdust and cassava root are placed alternatively in the box with the sawdust being on the top and at the bottom. The sawdust is kept moist as deterioration could occur if the sawdust becomes dry and if too damp it would favour the growth of spoilage microorganism causing root decay, so the box should always be monitored and water applied when necessary (Fadeyibi, 2012). Olaleye *et al*. (2013) investigated the effectiveness of trench and sawdust storage in maintaining moisture content of cassava root. They observed that the root stored at ambient temperature using both methods showed high level of water retention up to the 6th week of storage. This is a clear indication of the good keeping quality of and postharvest of freshly harvested roots (Karim *et al.*, 2009). However, the limitation of these techniques is that the high relative humidity in the storage environment could inhibit the growth of microorganisms (Olaleye *et al.*, 2013).

- *Storage in plastic bags*

The use of plastics like polyethylene and low density or high density polyethylene bags are the most useful means of transportation to the urban sectors (Fadeyibi, 2012). Studies have proven storage of cassava root up to three weeks in plastic airtight films. Also treating the root with fungicides like thiabendazole for few seconds before packaging in the polymeric film could extend the shelf-life of the root. Hence the technology could favour commercial exportation of cassava root. However, due to limited mechanical protection of the plastic bags, the root could be damaged during transportation (Fadeyibi, 2012). Akingbala *et al*. (2005) investigated the effect of packaging cassava root in polyethylene bags and trench for 21 days. Their study showed low water retention in polyethylene bag after the period of storage indicating deterioration of root and poor keeping quality. Another improved method is digging pit with soil of 15% moisture content. This method was observed to extend the root to about 8 weeks but nutrient loss in the root was observed after the storage period.

Advanced technology

Some advanced methods have been developed by the Centro Internacional de Agricultura Tropical (CIAT) in conjunction with National Resources Institute (NRI) for the storage of cassava (Fadeyibi, 2012). These methods include: refrigeration at lower temperature range of values 0 - 5 ºC; freeze drying; and waxing with paraffin wax (Reilly *et al.*, 2004). These methods are used for export purposes but it is expensive to maintain the equipment used for the purpose, and also handling requires skilled personnel (Fadeyibi, 2012). It can improve storage of fresh cassava root up to 2 weeks but refrigeration is not a most practised method in the developing countries (Reilly *et al.*, 2004). The use of paraffin wax also creates the possibility to market the crop and further increase the margin of holding stock of fresh root only for few weeks. Storage conditions of high temperature and low RH favours the curing process, but prior to storage it requires proper handling and selection of root. However, none of these methods can extend the shelf-life of root while maintaining the quality attributes. Therefore, processing into different food forms is a better option for extending the shelf-life, eliminate some toxic compounds like cyanide and also retain the quality of the product for a longer period of time (Cardoso *et al.*, 2005; Kolawole *et al.*, 2009).

6.2 Processing of cassava roots

Processing cassava roots into different food forms helps to stabilise shelf-life, improve quality and detoxify the roots (Inyang *et al.*, 2006; Kolawole *et al.*, 2009). Additionally, processing can also increase or decrease the quality attributes of the processed products. Studies have shown that during traditional processing over 40% of the produce is lost on drying. This is because the products are usually dried on bare floor where they are exposed to various contaminants such as dust and birds (Inyang *et al.*, 2006). However, these contaminations can be avoided by modifying the production and drying process, adherence to food sanitary and hygienic practices (Tsav-Wua *et al.*, 2004).

The products from cassava root are either processed into unfermented or fermented foods and drinks, but their processing methods such as boiling, steaming, roasting as well as the form (solid, semi-solid or liquid) in which they are consumed differ (Falade & Akingbala, 2010). The processed products from the root can be used for industrial purposes or for consumer foods (Table 7). Some of the unfermented products are common in some African countries while others are available in several regions of the world and they include the following.

Unfermented cassava products

- *Tapioca*

Tapioca grit is a partially gelatinised flake commonly consumed in many countries in West Africa as a convenience food (Adebowale *et al*., 2007). Tapioca processing is varietal sensitive and can be processed using rotary dryer or traditionally by roasting method but the former is widely used as it is applicable for all varieties (Adebowale *et al.*, 2007). Moreover, with the rotary drying method, it has been found that the time of drying, the changes in moisture content, as well as shelf-life stability of tapioca and other products can be predicted (Falade & Akingbala, 2010).

- *Cassava starch*

Cassava starch is a very good raw material in the food industry. It can be processed by peeling and washing of the roots, grating, and sieving to remove the fibre (Inyang *et al.*, 2006). The mash is allowed to sediment then followed by decanting to collect the starch (Raji *et al.*, 2008). The starch has a low gelatinisation temperature, high water binding capacity (thus a good stabiliser of food) and it has high viscosity and does not retrograde easily. The lipid, protein ash and phosphorus contents are generally low, but its carbohydrate content ranges between 73.5 to 84.9 %. However, the quality of cassava starch can be altered during drying and therefore renders it unacceptable (Jekayinfa & Olajide, 2007). Both the modified and unmodified starch are used as raw materials in food industries, either directly as starch food in form of custard, or as a thickener in baby foods and gravies and as a binder for products during cooking to prevent drying out (Taiwo, 2006).

Table 7 Common names used to describe some cassava products

Common names	Description	Location	Use	References
Unfermented products				
Tapioca	Partially gelatinised dried cassava starch	West Africa/Asia	Consumer food	Adebowole *et al*. (2007)
Chips and pellets	Dried irregular slices of root	Nigeria	Industrial purposes	Adamade and Azogu (2013)
Cassava starch	Dried mash from which the fibre is removed	South India, Asia & Nigeria	Food industry	Inyang *et al*. (2006)
Wafer	fried cassava starch in oil	India	Consumer food	Falade and Akingbala (2010)
Cassava flour	Milled cassava chip or reconstituted mash	Nigeria	Industrial and domestic use	Taiwo, (2006)
Unfermented fufu	Boiled and pounded dough	West Africa	Consumer food	Falade and Akingbala (2010)
Ampesi	Mashed boiled root	Brazil	Consumer food	Falade and Akingbala (2010)
Cassava puddings	Grated cassava root and mixed with banana	Indonesia	Consumer food	Falade and Akingbala (2010)
Fermented products				
Cassava beer	Flour mixed with water and yeast and made to ferment	Uganda	Consumer drink	
Banu or Uala	Distilled liquor from crushed cassava root	Uganda	Consumer drink	Falade and Akingbala (2010)
Gatot	Dried cassava cut into pieces	Cameroun	Consumer food	
Cassava bread	Made from wheat and cassava flour	West African/Asia	Consumer food	Shittu *et al*. (2008)
Fermented cassava starch	Fermented root used for baking	Brazil	Consumer food and industrial use	Srinivas (2007)
Garri	Pregelatinized granulated mash	West Africa	Consumer food	Fadeyibi (2012)
Lafun	Cassava porridge	Nigeria	Consumer food	Falade and Akingbala (2010)
Agbelima	Fermented root	West Africa	Consumer food	Obilie *et al*. (2003)
Akyeke/Attieke	Steamed sour granulated product	Cote d'Iviore and Ghana	Consumer food	Obilie *et al*. (2003)

- *Cassava chips and pellets*

Exporting of cassava products in a portable form is becoming a common practice and cassava chips and pellets are very efficient means for this purpose. They readily satisfy the market demand (Adamade & Azogu, 2013). They are simply dried slices of cassava roots of varying sizes between 2 cm to 5 cm usually packed in a jute bag or paper bag for exporting (Adebowale *et al*., 2007). Chips can be used for animal feed but its use is being constrained by many factors such as growth of mould as a result of environmental conditions in the package during longer distance of shipping. In addition, chips production and demand is inconsistent and there is also the problem of market competition in supply (Westby, 2002). However, chips and pellet have reduced moisture content and therefore prevent both quality and quantity postharvest loss (Adamade & Azogu, 2013).

Pellet is similar to chips but has lower moisture content of about 9% compared to chips. Hence, pellets have a longer storage life compared to chips (Falade & Akingbala, 2010). It is cylindrical in shape, dry and hard with length of about 2 - 3 cm and diameter about 0.4 - 0.8 cm (Adebowale *et al*., 2007). Pellets can be processed either from the root and leaves. It can also be processed from the peels of the root and it involves milling and extrusion, resulting in gelatinised products which become hardened on cooling. Pellet is recommended for shipment purposes because it has less storage weight and the ability to retain the quality after long distance of travel due to the low moisture content (Raji *et al.*, 2008).

- *Unfermented cassava flour*

Unfermented cassava flour is generally referred to as high quality cassava flour (HQCF). It is white, smooth and odourless flour and can be used as composite flour. The introduction of HQCF to the developing countries will encourage the use of cassava-based products and thereby reduce the dependency on imported cereals and grains (Taiwo, 2006). Traditionally, cassava flour was processed by sorting and peeling, washing and grating. The grated pulp is then dewatered (using rock to compress the sack bags) and pounded with pestle and mortar; this process contributes to soften the tuber. The mash is then sun-dried and pounded again and then sieved (sieving the flour gives a good quality product) and finally packaged for further use in the food industries (Eddy *et al.*, 2007; Fadeyibi, 2012). This method however could lead to fermentation as the dewatering and drying may take longer time thereby making the flour lose the functionality for composite flour (Falade & Akingbala, 2010). As a result, a new technique of processing was developed which is widely used to improve the system of agriculture and encourage the local farmers (Olaoye *et al.*, 2011). It is a fast method which involves: harvesting and sorting of good roots, peeling and washing manually, grating (usually done with a motorised cassava grater), dewatering (with screw or hydraulics press), pulverise, drying (solar or oven drying), fine milling, sifting the milled flour with a motorised flour sifter 250 µm and then packaging (Jekayinfa & Olajide, 2007). Products from unfermented cassava include: cassava macaroni and wafer (India), cassava puddings (Indonesia), and cassava cakes or wayano (India, Thailand and Malaysia) (Falade & Akingbala, 2010).

Fermented cassava products

Fermentation is one method of processing cassava into another food form which not only improve the flavour and taste of the product but extends the shelf-life (Falade & Akingbala, 2010). Fermentation is one major method employed during processing, which enhances the reduction of the cyanide level and detoxification of the root (Kostinek *et al.*, 2005). Some notable products from fermented cassava include:

- *Cassava bread*

Cassava bread is a fermented product prepared from the combination of wheat flour and cassava flour in the ratio of 5:1 (Shittu *et al.*, 2008). This proportion has been observed to give acceptable fresh loaf. Cassava flour is processed into dry flour by drying at temperature of about 50 ºC to ensure that flour retains its creamy colour after drying. This process has the ability of improving the use of cassava flour as composite flour in baking industries.

- *Fermented cassava starch*

This is a modified starch from fermentation of cassava root. It can be used for frying and baking of cheese bread in some countries such as Brazil (Srinivas, 2007). The process involves steeping already peeled and grated cassava roots in a tank of water for a period of 20 to 70 days to allow fermentation. This steeping process in adequate water helps in separating the starch granules from the fibre and other soluble compound. After fermentation, the obtained starch is dried to produce a powdered product. Although, soaking process is essential, it could cause deterioration of starch and thus reduce its usefulness in the food and pharmaceutical industries (Taiwo, 2006). The quality and the physicochemical properties of the fermented starch obtained are greatly affected by the varietal and environmental factors such as the temperature during fermentation.

- *Cassava fufu*

Fufu is an acid-fermented cassava product that is processed through the submerged fermentation of peeled roots in water. Fufu is a common traditional food for the West African countries (Oyewole & Sanni, 1995). The softened root is then pounded into wet fufu and the following processes are adopted: steeping the root in water for 2 – 3 days to soften the pulp and there after it is screened, allowed to sediment, dewatered with cloth bags, cooked and finally pounded into *fufu*. The quality of fufu is determined by the texture, aroma and colour (creamy-white or yellowish) depending on the variety used. The quality of fufu is greatly affected by season, the processors and also most especially the variety (Obadina *et al.*, 2009). Deterioration rate is high because it is processed as a wet paste with moisture content of about 50%, therefore the shelf-life is short and will not be useful or be suitable for large-scale and commercial purposes. However, a modern technology has been developed to extend the shelf-life and market quality of fufu. This is obtained by drying in high temperature for about 60 ºC to produce flour which can be further reconstituted with hot water (Dipeolu *et al.*, 2001).

- *Garri*

Garri is the most commercial and useful product from cassava processing. It is creamy-white, pregelatinised granular and high calorie food with a slightly sour taste (Falade & Akingbala, 2010). It is processed from fresh cassava roots following very tedious operation of peeling and grating into mash (Fadeyibi, 2012). The grated pulp is put in sacks (Jute or polypropylene) and the sacks are placed under heavy stones or pressed with a hydraulic lack between wooden platforms for 3 - 4 days to dewater the pulp and allow fermentation to take place (Falade & Akingbala, 2010). This traditional way of processing cassava root into garri is monotonous, time-consuming, requires more labour and hazardous to health because processors are usually exposed to smoke and heat during frying (Taiwo, 2006).

Thereafter the pulp is sieved with traditional woven splinters of cane and finally fried over a heated metallic surface (garrification) to dextrinise and dry the grits (Akingbala *et al.*, 2005). During this time constant stirring with a wooden paddle is required until low moisture content usually between 8 - 10% is achieved (Falade & Akingbala, 2010). Garri is regarded as pre-cooked convenient food which can be eaten as a snack and the long period of frying contributes greatly to its longer shelf-life (Fadeyibi, 2012).

- *Lafun and Agbelima*

Lafun, is fermented cassava flour common in southwest Nigeria, it is prepared like porridge and eaten with soup (Falade & Akingbala, 2010). The process involves manual peeling and chipping to enhance fermentation and detoxification of the root, the tubers are soaked in large quantity of water for 2 to 3 days for fermentation to take place and the mash is dewatered and dried for maximum of 3 days before packaging for house hold consumption or marketing (Falade & Akingbala, 2010). Agbelima is a traditional food of the West African especially in Ghana, Togo and Benin. It involves grating and fermentation of the cassava tuber with inocula, although these inocula enhance the fermentation to about 2 days, it also degrades the taste and texture. Fermentation process in agbelima promotes detoxification of cassava root and gives a peculiar organoleptic quality such as a souring taste and a softened texture (Obilie *et al.*, 2003).

- *Akyeke or Attieké*

Akyeke or attieké is an indigenous food product from cassava popular in Cote d'Iviore and Ghana. It is a steamed sour granulated product, fermented to modify the rubbery texture of the peeled cassava roots. Peeled cassava roots are washed and grated together with traditional inocula (peeled cassava chunks soaked in water for 3 days). The mash is packed into polyethene sacks and allowed to ferment for 5 - 7 days depending on the amount of inoculum added. The mash is further dewatered with screw press and the granules are then sundried and steamed to obtain the akyeke. The inoculum is prepared by soaking peeled cassava chunks in water for about 3 days to soften and ferment (Obilie *et al.*, 2003).

Bioenergy

- *Ethanol from cassava*

Another significant use of cassava which has become of scientific interest globally is its use as a renewable energy source (Adelekan, 2010). This was necessitated because of the rate of deforestation in the developing countries to produce firewood for domestic as well as industrial purposes (Fadeyibi, 2012). Cassava being an energy crop offers a cheaper alternative to conventional energy sources and provides favourable impression for the utilisation of natural resources (Ubalua, 2007). Similarly, development of ethanol from cassava will further enhance value addition and reduce postharvest losses since cassava waste could be used for the production (Ubalua, 2007). Also this technology will propel agricultural productivity especially in the developing countries and reduce the reliance on fossil fuel for source of energy (Adelekan, 2012; Fadeyibi, 2012). Ethanol fuel (ethyl alcohol) is similar to the alcohol found in alcoholic beverages and can be used for biofuel, an alternative to gasoline, and as oxygenate to gasoline in the United States (Adelekan, 2010).

7 Packaging and storage of cassava products

Packaging is essential in food systems because it helps to reduce losses, add value, extend shelf-life, maintain quality and wholesomeness of products, improve market standard and food safety (Inyang *et al.*, 2006; Opara & Mditshwa, 2013). It is the easiest and cheapest way of preventing food contamination with undesirable organisms and foreign matter (Opara & Mditshwa, 2013). Postharvest losses often occurr due to poor or inadequate packaging, which results in shelf-life instability (Ogiehor & Ikenebomeh, 2006)

The shelf-life of packaged cassava products is influenced by the storage conditions, moisture content and the water activity (a_w) of the product. Water activity influences the microbial load and facilitates physicochemical deterioration due to high water activity level often associated with increase in moisture content and high relative humidity (Ikhu-Omoregbe, 2006). Optimum packaging keeps the product readily available for the consumers in a wholesome quality and prevents deterioration caused by microorganisms. Ogiehor and Ikenebomeh (2006) observed garri stored in different packaging materials, at the end of the storage period each of the stored samples in the materials showed an increase in the fungal and viable bacterial count which varied between each material. Also, studies have shown that during storage in the different packaging materials, changes occurred in the proximate composition and this was suggested to be due to the different relative permeabilities of the packaging materials either to atmospheric conditions, gases and environmental changes (Butt *et al.*, 2004). Similarly, during long periods of storage the pH of the sample is decreased due to the microbial activities and the metabolites are reactivated leading to increase in titratable acid in the cassava products (Ogiehor & Ikenebomeh, 2006). However, the use of the appropriate packaging materials could inhibit the growth of microorganism and extend the shelf-life of the product (Ogugbue & Gloria, 2011).

Most products from cassava are hygroscopic in nature and if exposed to the atmosphere can absorb moisture and gases from the environment and will in turn encourage the growth of microbes (Ogugbue & Gloria, 2011). On exposure to high humidity and moisture content, flour products tend to glue to the body of the packaging film due to increase in the wall friction (Iqbal & Fitzpatrick, 2006). This situation therefore reduces the crispiness of the final product processed from such flour sample (Kulchan *et al.*, 2010). Various packaging materials have been recommended in literature for the packaging of cassava processed products. The product quality and shelf-life is a function of the characteristics of the packaging film used, therefore each product has a particular packaging material suitable for it (Robertson, 2012). The different packaging materials used for storage of cassava products are mainly paper, plastic and sack bags or a combination of one or two of the materials (Table 8).

Paper materials can be coated or laminated with aluminium and they are superior over other materials because they can be recycled at low cost (Opara & Mditshwa, 2013). Plastic films which include low density polyethylene (LDPE), high density polyethylene (HDPE), laminated aluminium foil (LAF), polyethylene (PE), polypropylene (PP) and plastic containers are widely used for packaging. However, some of these materials are faced with a major setback of the inability to resist light and moisture, which can further contribute to degrading the physicochemical properties and quality of products such as the colour and carotenoids which are heat and light sensitive (Chávez *et al.*, 2007).

Table 8 Packaging materials and storage conditions for handling different cassava products

Products	Package materials	Storage conditions	Duration (Weeks)	Comments	Reference
Cassava-Flour baked products	LDPE,OPP, plastic bag	30±1 °C	24	Shelf-life stability in LDPE,OPP	Kulchan *et al*. (2010)
Garri	Gunny sack, HDPE, LDPE, Plastic container	Ambient	9	Microbial stability in HDPE	Ogugbue and Gloria (2011)
Garri	LDPE, HDPE, plastic bucket & hessian bags.	30 ± 1 °C	24	Proximate best in HDPE	Ogiehor and Ikenebomeh (2006)
Fufu flour	Air tight dishes	28 ± 2 °C, 33 and 76% RH	8	Flour was best at 33% RH	Obadina *et al.* (2007)
Garri	Unpackaged, PP, PE, Hessian bag	16 & 36 °C	24	Best in PP & PE	Adejumo and Raji (2012)

8 Summary and future prospects

Harvested cassava root is highly susceptible to postharvest deterioration because of its high moisture content which reduces the market quality. Latest developments in postharvest handling and storage of freshly harvested cassava root and processed products have been discussed in this review. Various processing techniques intended to facilitate commercial production of cassava root into more convenient food forms such as garri, flour, fufu, and tapioca and the different packaging methods for root and processed product were highlighted. The review presented above showed that traditional storage methods of burying cassava root in the soil or piling the root in heaps with constant watering could contribute to the drastic reduction in the produce quality and percentage production yield, due to increased incidence of physiological disorders and decay.

Recent advancements in postharvest technology offer the possibility for cassava root to be stored for about 8 weeks under optimum conditions of ambient temperature (≥ 23 °C) and (< 60%) relative humidity (Zidenga *et al.*, 2012). However, in-ground storage is the most preferred way of root storage; however, long-term storage under this condition could result in nutrient losses and degradation of some functional properties of fresh root, in addition to the possibility of root lignification. Unless effective control measures are taken, postharvest losses could also arise due to microbial decay (rot), pests and rodents attack.

In addition to applying optimum storage techniques, processing is another effective way to reduce postharvest losses and add value to fresh cassava root. However, processing must be done within the first two days after harvest before deterioration sets in. Proper hygienic practices such as thorough washing of roots with potable water should be adopted and machines should also be washed to avoid cross-contamination during processing. Packaging of processed products is also vital in postharvest handling as it determines the longevity and availability of the processed products. This means that degradation of quality attributes could still occur during storage of the produce depending on the type of packaging materials used, thereby resulting in physicochemical, functional as well as the nutritional losses of the processed products.

Therefore, to explore the utilisation of fresh cassava produce, research should focus more on evaluating the optimum packaging and storage conditions for the shelf-life stability of fresh cassava products especially high quality cassava flour. This is because cassava flour offers more technical uses compared to the other minimally processed produce such as fufu, garri and lafun. Furthermore, the quality attributes of cassava flour during storage are essential as they will guarantee its use as composite flour in the industries. In addition, the influence of packaging materials and storage conditions on the functional properties of cassava flours should be investigated during storage. This will enable the food processors to make proper selection of composite flour during product development. Most importantly, more research on technology applicable in reducing the onset of postharvest physiological deterioration and extending the shelf life of freshly harvested cassava root for future commercial and domestic purposes should be emphasied.

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CHAPTER 3

INVESTIGATING THE EFFECTS OF STORAGE CONDITIONS AND DURATION ON PHYSICOCHEMICAL PROPERTIES AND MICROBIAL QUALITY OF CASSAVA FLOUR (CVS. ‘TME 419’ AND ‘UMUCASS 36’)

CHAPTER 3

INVESTIGATING THE EFFECTS OF STORAGE CONDITIONS AND DURATION ON PHYSICOCHEMICAL PROPERTIES AND MICROBIAL QUALITY OF CASSAVA FLOUR (CVS. ‘TME 419’ AND ‘UMUCASS 36’)

Summary

An optimum storage condition is essential for extending shelf-life and maintaining quality of cassava flour and other minimally processed food products. This study investigated the effects of storage conditions (15 °C, 90% relative humidity (RH)) cool condition, (23 ± 2 °C, 60% RH) ambient condition, and (38 ± 2 °C, 60% RH) higher condition on changes in physicochemical quality attributes of cassava flour (cvs. ‘TME 419’ and ‘UMUCASS 36’). Cassava flours from each cultivar were packaged in paper bags and stored for 12 weeks during which changes in physicochemical properties were studied at periodic intervals. Physicochemical quality attributes of both cultivars differed significantly due to their genetic makeup. The ‘UMUCASS 36’ flour had higher protein, carbohydrate and dry matter contents compared to the ‘TME 419’ flour. Proximate composition of cassava flour was significantly influenced by the storage conditions and duration. Moisture content decreased from 12.0% to 7.1% and 9.8 to 6.8% in cultivars ‘TME 419’ and ‘UMUCASS 36’, respectively. Water activity (a_w) level of cassava flour decreased significantly ($p < 0.05$) over storage duration from 0.57 to 0.54 for cv. ‘TME 419’ and from 0.54 to 0.51 for cv. ‘UMUCASS 36’ under higher condition, while a slight decrease in a_w level was observed for flour stored under ambient (23 °C, 60% RH) condition. A decrease in microbial load from 5.4 to 4.8 log cfu/ g was observed in both cultivars, with increase in storage temperature from 15 °C to 38 °C. However, cassava flour stored under cool condition (15 °C, 90% RH) absorbed moisture and visual observation of decay was seen on the packages after three weeks of storage. The carotenoid content after processing was higher in cv. ‘UMUCASS 36’ (2.5 ± 0.10 mg/ g) compared to cv. ‘TME 419’ (1.8 ± 0.11 mg/ g). Yellowness index of the flour from yellow cassava (cv. ‘UMUCASS 36’) was significantly influenced by storage duration with about 31% decrease in yellowness observed at the end of 12 weeks of storage. The ambient condition (23 °C, 60% RH) best maintained nutritional and physicochemical quality.

Introduction

High quality cassava flour is one of the major processed by-products from cassava root. It is gluten-free flour and beneficial in the treatment of celiac patients (Briani *et al.*, 2008; Biagi *et al.*, 2009). Cassava flour has high carbohydrate content and a source of high caloric food. Research has shown that consumption of cassava-based food can help to support the nervous system, alleviate stress, anxiety and irritable bowel syndrome (Baffour, 2009). This is accredited to the high energy value and carbohydrate content. Flour obtained from improved cassava varieties (bio fortified with high carotenoid pigment crops) are rich in carotenoids, which serves as a good and cheap source of vitamin A (Liu *et al.*, 2011; Nassar & Ortiz, 2009). The nutritive importance of carotenoid plants is attributed to its conversion to vitamin A, as in the case of β-carotene, and to its antioxidant property (Sánchez *et al.*, 2006). Therefore, optimum postharvest handling process is critical in maintaining the bioactive components of cassava flour.

Furthermore, cassava flour is a useful supplement in the production of baby food, pastas and glucose syrup (Nwabueze & Anoruoh, 2011). As composite flour, studies have shown that cassava flour is a better supplement to wheat flour compared to other root and tuber crops (Eddy *et al.*, 2007; Olaoye *et al.*, 2011). Hence, it is used in the production of confectionery in the food industries. The availability of cassava flour also helps to reduce dependency on wheat flour in developing countries (Shittu *et al.*, 2008; Kulchan *et al.*, 2010; Gyedu-Akoto & Laryea, 2013).

In recent years, consumers and food processors have shown considerable interest in the nutritional quality and safety of food (free of any environmental, chemical or microbial contamination) rather than the quantity of the product (Shobha *et al.*, 2012). Thus, optimum packaging and storage conditions are essential in maintaining desired quality attributes in food. Food powders (wheat flour, tea powder and whey permeate) exposed to 20 °C and 66% RH in a previous study were observed to have an increase in moisture content, which resulted in increase in wall friction and caking of the flour (Iqbal & Fitzpatrick, 2006). This situation was ascribed to the hygroscopic nature of food powders. However, storage of soybean flour at 40 °C and 90% RH had a significant influence on the flour quality, as the decrease in fat content was higher at the accelerated storage temperature than the ambient (25 - 35 °C) during the 75 d storage duration (Agrahar-Murugkar & Jha, 2011). The authors attributed this observation to be the effect of high temperature on the lypolytic enzymes thus causing a decrease in fat content of the beans flour. Similar studies have been carried out on cassava roots and its by-products (Chávez *et al.*, 2007; Rodríguez-Sandoval *et al.*, 2008; Sánchez *et al.*, 2013), but there is limited information with regard to the influence of storage conditions and duration on the physicochemical properties of cassava flour. Sanchez *et al*. (2013) evaluated the changes in roots of two cultivars of cassava (susceptible and tolerant to postharvest physiological deterioration (PPD)) stored for 14 days in ambient tropical temperature (19.0 - 29.6 ± 2 °C) and 61.3 - 94.7% RH and observed a progressive decrease in starch content. This could limit the extent at which cassava root would be stored. The observation with different storage conditions for cassava root therefore gives an indication of possible variations in the physicochemical properties and shelf-life stability of the flour products during storage. Therefore, considering the effect of storage on the physicochemical properties of cassava flour, it is paramount for the food scientist and industries to establish suitable storage conditions for packaged cassava flour. This research was therefore designed to evaluate the effects of storage conditions and duration on the proximate composition, physicochemical properties and microbial stability (total plate count and fungi) of cassava flour using two newly bred cultivars (‘TME 419’ and ‘UMUCASS 36’) in Nigeria.

Materials and methods

Plant materials

Cassava cultivars ‘TME 419’ and ‘UMUCASS 36’, white and yellow roots, respectively (Fig.1), were harvested at commercial maturity 12 months after planting from National Root Crops Research Institute (NRCRI) Umudike (5º28′33"N 7º32′56"E), Abia state, Nigeria. The fresh cassava roots were harvested from a randomised complete block design field plot of three replications. These cultivars were selected based on their functional properties (paste stability), which are desirable for use in the food industries (Nwabueze & Anoruoh, 2011; Eleazu & Eleazu, 2012).

Figure 1 Cross section of cassava root (A) TME 419 and (B) UMUCASS 36

Cassava processing, packaging and storage

Clean cassava root (without breaks/cuts) were sorted, washed, peeled and re-washed with running tap water. The roots were sliced into chips of about 2 - 3 cm in length using electric stainless steel chips making machines (YS QS400, Shandong, China) and sun-dried (40 ± 2 °C) for three days to a constant moisture content of about 13% (Falade & Akingbala, 2010). Dried cassava chips obtained from the two cultivars were packaged separately in sterile polyethylene bags and transported under ambient conditions to the Department of Food Science, Stellenbosch University, South Africa. The chips were milled into flour with a Cyclone Laboratory milling machine (Model 3100, Perten Instruments, Hagersten, Sweden) fitted with a sieve size of 0.5 mm. Baseline/initial (week 0) analysis of cassava flour quality attributes was carried out separately for both cultivars prior to packaging and storage.

The processed cassava flour from both cultivars was packaged (200 g) in separate brown paper bags (150 x 250 mm) (Fig. 2) under aseptic conditions. The packed flour were stored at selected environmental conditions using a test chamber (MLR-351H, Sanyo, Japan) as follows: 15 ± 1 °C, 90% RH (cool condition); 23 ± 2 °C, 60% RH (ambient condition); and 38 ± 2 °C, 60% RH (higher condition). Separate bags of cassava flour were taken for analysis at 4 weeks interval for 12 weeks duration. Storage was stopped at 12 weeks due to time factor and most of the quality attributes such as carotenoid had declined rapidly. All analysis for physicochemical properties, proximate composition and selected bioactive components (carotenoid and HCN) were performed in triplicates on each sampling day.

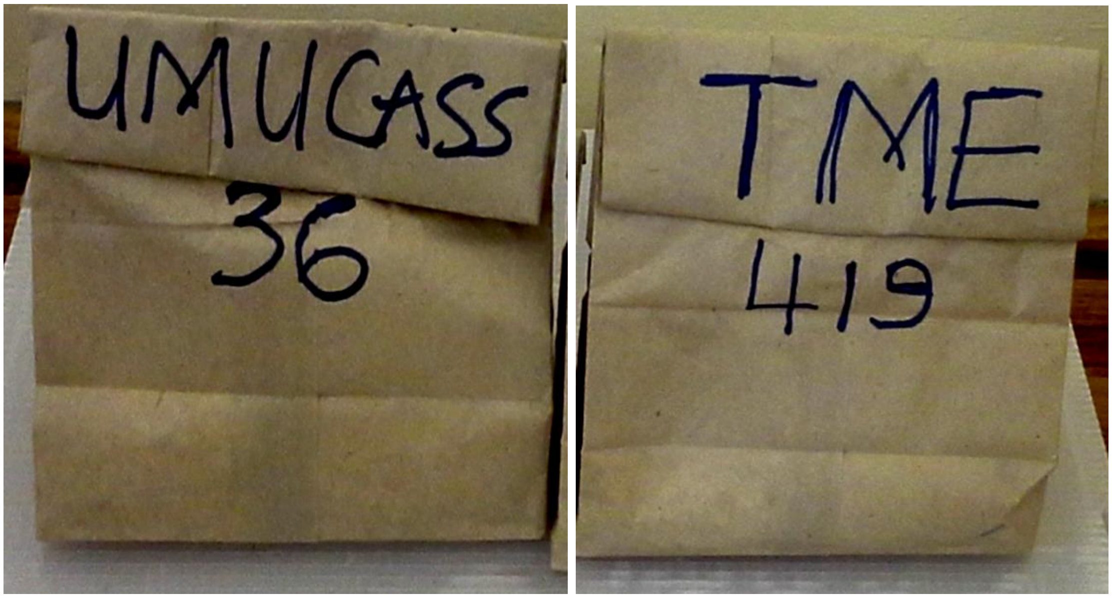

Figure 2 Paper bags for cassava flour packaging at different storage conditions

Proximate compositions analysis

Moisture and ash contents were carried out according to the relevant Association of Official Analytical Chemist (AOAC, 2012) methods, (925.09) for moisture and (923.03) for ash. Protein content was determined following the Kjeldahl method; 2 g of cassava flour were digested for 90 min at 400 °C in sulphuric acid with Kjeldahl tablet (Merck, South Africa) as catalyst. The digest containing ammonium sulphate and carbon dioxide was diluted with 40 mL distilled water before neutralising with 35% sodium hydroxide through a distillation unit (UDK129, Italy) for 4 min. The digest was distilled into boric acid solution containing methyl red and bromocrescyl green indicator (Merck, South Africa). Finally, the greenish boric acid solution was titrated against 0.1 M HCl until a permanent light pink colour was observed. Protein content was calculated from % N using a conversion factor 6.25 (N × 6.25) methods 920.152 (AOAC, 2006).

Fat content was determined through the soxhlet extraction method (AOAC, 2006) using 70 mL petroleum ether as the extraction solvent. Total fat extraction was done in 3 h and pre weighed beakers were weighed again after extraction and evaporation of ether. Percentage carbohydrate content was calculated by difference equation 1 (Nwabueze & Anoruoh, 2011). Energy value was calculated using Atwater factor equation 2 (Akubor *et al*., 2003). The dry matter was determined with equation 3 (Etudaiye *et al.*, 2009).

$$\mathrm{CHO}(\%) = 100 - \%\ (\text{protein} + \text{fat} + \text{Moisture} + \text{Ash}) \qquad (1)$$

$$\text{Energy value (KJ/kg)} = (\text{protein} \times 4 + \text{Fat} \times 9 + \mathrm{CHO} \times 4) \qquad (2)$$

$$\text{Dry matter } (\%) = (100 - \text{moisture content}) \qquad (3)$$

Physicochemical analysis

Colour

The 3-dimensional colour values which are expressed as *L** (lightness), *a** (green) and *b** (yellowness/blueness) were taken using a chromameter (CR-400/410 Konica Minolta Sensing Inc., Japan). The instrument was calibrated against a white plate prior to use. The cassava flour was poured into dry petri dishes and colour snapshots were taken three times from each flour treatment by placing the lens of instrument on the flour (Rhim & Hong, 2011). The mean of the nine measurements per treatment was calculated. Total colour difference (ΔE), which indicates the magnitude of change in colour parameters between the initial and final colour values during storage was calculated using equation 4 (Pathare *et al*., 2013).

$$\Delta E *= \sqrt{\Delta a *^2 + \Delta b *^2 + \Delta L *^2} \qquad (4)$$

Furthermore, whiteness indices (WI) which describes the total whiteness of food products and may indicate the degree of discolouration or deterioration in some products during storage was calculated (Hsu *et al.*, 2003; Lin *et al.*, 2009). Whiteness is one of the major attributes the end users of flour desire either for domestic or industrial uses and it is often used to express the quality of the flour during storage (Rodriguez-Aguilera *et al.*, 2011). Whiteness is based on the scale of 0 -100, with higher values describing brighter colour appearance (Hsu *et al.*, 2003). The WI of cassava flour was calculated using equation 5 (Pathare *et al*., 2013):

$$\mathrm{WI} = 100 - \sqrt{(100 - L *^2) + a *^2 + b *^2} \qquad (5)$$

Yellow index (YI) could be used to express the level of yellow colouration especially in products rich in carotenoid (Rodriguez-Amaya *et al.*, 2011). The yellowness of fresh or minimally processed produce can degrade during processing and exposure to light during storage (Pathare *et al*., 2013). The extent of yellow degradation can be determined with the YI equation 6 (Pathare *et al*., 2013).

$$\mathrm{YI} = \frac{142.86 \times b*}{L*} \qquad (6)$$

Water activity

The water activity of cassava flour was determined using Pawkit water activity (a_w) meter (Decagon Devices, Inc., Pullman, Washington, USA) at a constant temperature of 21 °C. The standard procedure for the instrument was followed. The sample cups were half-filled with 5g cassava flour to ensure accuracy of result, and to avoid flour covering the instrument sensor. The Pawkit a_w meter was positioned over the cups and the water activity level was measured within 5 min.

Total carotenoids

Spectrophotometric method as described by Opiyo and Ying (2005) with modifications was used to determine the total carotenoid of the cassava flour. According to Rodrigez Amaya and Kimura (2004) the determination of total carotenoid gives a more accurate estimate for trans-β-carotene; this is because about 90% of the total carotenoid present in cassava root is β-carotene. One gram of flour was measured into centrifuge tube and homogenised with 14 mL solution of n-hexane: acetone (3:2 v/v), and then agitated for 1 min before centrifuging with Eppendorf centrifuge (Mark Chemicals, (Pty) Ltd., South Africa) at 6000 rpm for 10 min at 4 ºC. Absorbance was read immediately at 450 nm using a spectrophotometer (Helios Omega UV-Vis Thermo Scientific, USA). Sample preparation and readings were done under less intense (reduced brightness) light in the laboratory because of the sensitivity of carotenoid to light. Total carotenoids content were calculated and expressed in (mg/ 100g) dry weight (DW) basis following the equation 7 (Opiyo & Ying, 2005; Opara & Al-Ani, 2010).

$$\text{Total carotenoid (mg/ 100g)} = \frac{\text{Ab}^{450} \times 4}{\text{Sample weight}} \times 1000 \qquad (7)$$

Hydrogen cyanide determination

The alkaline picrate method was used with minor modification to quantify the total cyanide content in the cassava flour samples (Onwuka, 2005). Cassava flour (5 g) was measured into a conical flask and dissolved with 50 mL distilled water. The mixture was kept at room temperature overnight. Alkaline picrate was prepared by dissolving 5 g Na_2CO_3 and 1 mL of picric acid with warm water in 200 mL volumetric flask. Flour solution left overnight was then filtered using Whatman filter paper No. 1 and 1 mL of the filtrate was mixed with 4 mL alkaline picrate solution. The mixture of solution containing 1 mL flour filtrate and 4 mL alkaline picrate was incubated in a water bath (TMK 14R20, FMH instruments, India) at 50 °C for 5 min. A light reddish brown colour was observed and the absorbance was read using a spectrophotometer (Helios Omega UV-Vis Thermo Scientific, USA) against a reagent blank at 490 nm. Standard curve prepared by diluting potassium cyanide (KCN) in water acidified with HCL at different concentrations ranging from (0.01 to 0.05 µg/.mL) (Fig. 3) was used to extrapolate the concentration of HCN from the equation y = (2.4x+ 0.216)*10, with R^2 = 0.92. Where y = concentration, 2.4 = slope of curve, X = absorbance, 0.216 = intercept and 10 is the dilution factor. Results were expressed as mean of triplicate values of each treatment in µg/ mg.

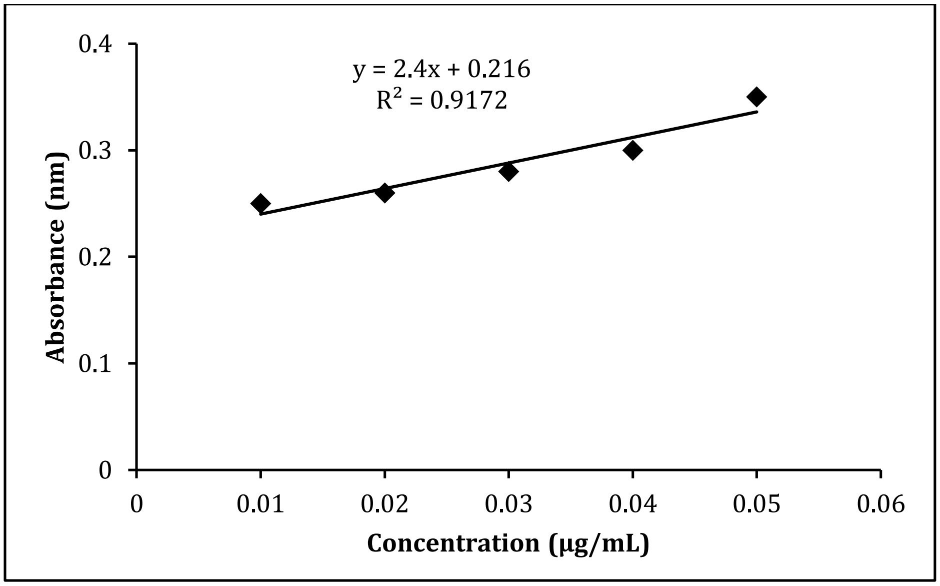

Figure 3 Standard curve for determining hydrogen cyanide content prepared by diluting potassium cyanide (KCN) in water acidified with HCL

Determination of pH

The pH levels of cassava flour were determined with a reference glass electrode pH meter basic 20^+ (Crison, 52-01 PI1000, Spain). Cassava flour (10 ± 0.02 g) was dissolved in 10 mL of deionized water (Ogiehor & Ikenebomeh, 2006). The pH meter was first calibrated with buffer pH 4 and 7 by placing the electrode in each buffer and rinsing before placing in test samples.

Microbial analysis

Microbial analysis was done to determine the microbial stability of cassava flour over time. Physiological solution (PS) was prepared by dissolving 9 g of NaCl in 1000 mL distilled water. A weight of 10 g of cassava flour was aseptically taken from each bag and homogenised in 90 mL of the sterilised PS. Dilution was done 5 times by taking 1 mL from first dilution and transferred into 9 mL tubes of PS. Each dilution was plated out in triplicate on a plate count agar (PCA) for aerobic mesophilic bacteria and potato dextrose agar (PDA) for mould and yeast using pour plate method. Sterile plates of both agars were open for 10 min around the work area and in the laboratory to ascertain the microbial state of the environment. The plates were allowed to cool, kept at inverted position to avoid condensation of moisture in the plate and then incubated at 37 °C for 48 h and 26 °C for 72 h for bacteria and yeast and mould count, respectively (Akhtar *et* *al.*, 2008). Visible colonies were counted after incubation and the results were reported as log cfu/ g

Statistical analysis

Factorial analysis of variance (ANOVA) was performed using Statistica software (Statistica 12.0, Statsoft Inc., Tulsa, OK, USA). The significant differences between the mean were determined with Duncan's multiple range tests at 95% confidence interval. All results obtained for each cultivar and storage conditions were expressed as mean of triplicate values ± standard deviations.

Results and discussion

Proximate composition of fresh cassava flour

The initial compositions of cassava flour cvs. 'TME 419' and 'UMUCASS 36' showed significant variations in the cassava flour cultivars (Table 1). Cultivar differences had a significant effect on the proximate composition ($p < 0.05$). Carbohydrate content of flour was 85.5 ± 0.17% for 'UMUCASS 36' and 83.6 ± 0.09% for 'TME 419'. Protein was generally low for both cultivars and significantly higher in 'UMUCASS 36' (3.0 ± 0.05%) than in 'TME 419' flour (2.0 ± 0.07%). Dry matter content was higher in 'UMUCASS 36' flour (90.2 ± 0.15%) than in the flour of 'TME 419' (88.8 ± 0.11%). The average moisture content for both cultivars was 10.9%, cv. 'UMUCASS 36' was lower (9.78 ± 0.15) compared to 'TME 419'. However, moisture content in both cultivars was within the recommended moisture range (12%) for flour products and within the range reported in literature for cassava flour (Charles *et al.*, 2005; Shobha *et al.*, 2012; Falade *et al.*, 2014). Although similar processing and drying techniques were used, moisture content in flour of cv. 'TME 419' was higher than in cv. 'UMUCASS 36' in this study. The observed variations in moisture content could be attributed to the inherent attributes such as dry matter content of the different cultivars. Dry matter is determined by taking the weight of the flour before and after drying, the weight loss is usually as result of moisture. Low moisture content in flour is essential during storage because it favours shelf-life stability of flour (Eleazu *et al.*, 2012). Higher moisture content > 12% has been reported to accelerate or enhance microbial growth (Aryee *et al.*, 2006). Therefore, both cultivars used for this study possess the potential of good storage quality although cv. 'UMUCASS 36' has a better prospect for longer shelf stability because of the lower moisture and higher dry matter contents.

Table 1 Comparison of the proximate compositions of cassava flour (cvs. 'TME 419' and 'UMUCASS 36') (dry weight basis)

Compositions	**TME 419**	**UMUCASS 36**
Moisture content (%)	11.96 ± 0.11^a	9.78 ± 0.15^b
Ash (%)	1.19 ± 0.03^b	1.59 ± 0.03^a
Protein (%)	1.97 ± 0.07^b	3.03 ± 0.05^a
Fat (%)	1.00 ± 0.07^b	1.21 ± 0.05^a
CHO (%)	83.63 ± 0.09^b	85.45 ± 0.17^a
Dry matter content (%)	88.84 ± 0.11^b	90.22 ± 0.15^a
Energy value (KJ/kg)	1487.83 ± 0.05^b	1508.84 ± 0.34^a

Mean ± SD, similar letters in rows are not significantly different ($p < 0.05$)

Dry matter content (88.84 ± 0.11%) of flour of cv. 'TME 419' was significantly lower than 'UMUCASS.36' flour. This result was consistent with research done on cassava flour and other cassava products (87.73 - 71.60%) from other cultivars (Etudaiye *et al.*, 2009; Eleazu & Eleazu, 2012). Eleazu and Eleazu (2012) in their study on proximate composition of cassava flour also observed that flour of cv. 'UMUCASS 36' (the yellow root) had higher (90.30 ± 1.13) dry matter content compared to 'TMS98/0505' white root (87.73 ± 0.95) suggesting a better functional and cooking quality of the flour during food processing. Therefore, knowledge of dry matter content of flour is essential for the food industries as it could affect cooking time, quality of cooked food and shelf-life stability of processed products.

Protein, fat, and ash contents were lower in flour of cv. 'TME 419' (2.0 ± 0.07, 1.0 ± 0.07, and 1.2 ± 0.03%), respectively compared to the 'UMUCASS 36' cultivar (3.0 ± 0.05, 1.2 ± 0.05 and 1.6 ± 0.03%), respectively. This result was expected because cassava root is generally low in protein, fat and ash components. Previous studies on cassava flour by other researchers also observed low values for protein, fat and ash. Charles *et al*. (2005) reported mean values of protein (1.5 ± 0.2%), fat (0.2 ±0.1%) and ash (1.8 ± 0.6%) of the flour of five cassava cultivars from north Thailand. Similarly, Nwabueze and Anoruoh (2011) reported low protein (2.6 ± 0.12%), fat (0.62 ± 0.1%) and ash (1.48 ± 0.07%) contents for the flour of cassava cv. 'TME 419'. In addition, the disparity observed in these components was due to the differences in cassava cultivars. Aryee *et al*. (2006) reported cultivar differences in proximate composition of the flour of 31 different cassava cultivars; this in turn influenced the utilisation of the flour cultivars for food and industrial purposes.

The energy values for both cassava flour cultivars were 1487.8 ± 0.05 KJ/ kg (355.6 kcal/ kg) and 1508.8 ± 0.34 KJ/ kg (360.5 kcal/ kg) for cultivars 'TME 419' and 'UMUCASS 36', respectively. Cassava flour from cv. 'UMUCASS 36' had higher caloric value consistent with the higher carbohydrate content compared to the other cassava flour cultivars with substitution potential and cassava-wheat composite blend previously studied in literature (Jisha *et al*., 2010). These findings highlight the need for the critical evaluation of proximate compositions of different cultivars of cassava flour to assess their suitability in the food industries.

Physicochemical analysis

The pH range of flour from both cultivars was from 5.5 to 6.2, with cv. 'TME 419' having higher pH values (5.76 ± 0.57 – 6.21 ± 0.13) than cv. 'UMUCASS 36' (5.50 ± 0.03 – 6.03 ± 0.01). Similar ranges of pH (5.3 to 6.5) were reported in literature for cassava flour (Aryee *et al*., 2006), and for other flour products from other root crops (Falade & Okafor, 2013). Knowledge of the pH value of flour is essential, as it provides a guide in determining the ratio of cassava flour that could be substituted when mixing composite flour for baking (Aryee *et al.*, 2006).

Colour attributes of both cultivars before storage were as follows: L^* (lightness) value for cv. 'TME 419' was 90.2 ± 0.73 while cv. 'UMUCASS 36' was 87.7 ± 0.60. Values were significantly different in both cultivars because of their genetic makeup, 'TME 419' (white cultivar) and 'UMUCASS 36' (yellow cultivar). Falade *et al*. (2014) reported similar range in L^* value (86.54 - 92.13) for Africa rice flour thus confirming considerable lightness of flour products for the food industries. The colour parameters of two traditionally processed Burundian cassava flour showed similar range of L^* value (97 - 74), which was attributed to the long fermentation period (Aloys & Hui Ming, 2006). The authors reported that fermentation just like the other processing method (drying) could have a profound influence on the colour attributes of cassava flour.

The initial whiteness index of both cassava flours were 85.1 ± 0.72 for cv. 'TME 419' and 78.8 ± 1.86 for cv. 'UMUCASS 36'. The higher whiteness index of cultivar 'TME 419' flour is not surprising given its normal white colour compared with the whiteness index of cv. 'UMUCASS 36'. Similarly, the yellowness index of cv. 'UMUCASS 36' was higher (28.19 ± 2.31) due to the yellow pigmentation compared to the cv. 'TME 419' (17.80 ± 0.86) with white colour. The values obtained in this present study were consistent with those reported in literature for white yam flour processed under different drying methods (45.8 ± 0.15 to 85.7 ± 0.07) (Hsu *et al.*, 2003).

Additionally, the authors established that colour and carotenoid retention can be influenced by the drying method employed during processing. Thus, the distinction in whiteness and yellowness indices observed in this study could be attributed to the differences in cultivars, carotenoid content and the effect of sun-drying during chips processing. Carotenoid degradation of the flour of both cassava cultivars resulted to reductions in the yellowness indices because carotenoid pigments can be denoted with yellow colour in food.

Effects of storage conditions on the proximate composition of cassava flour

Table 2 summarises the effects of storage conditions and durations on proximate composition of the flour of two cassava cultivars (‘TME 419’ and ‘UMUNCASS 36’). Moisture content (MC) in both flour cultivars decreased significantly with increase in storage time from 4 to 12 weeks (Table 2). The lowest value in MC (6.83 ± 0.13%) was observed in cv. ‘UMUCASS 36’ under higher condition (38 °C, 60%) because of its high dry matter content. Further analysis showed that storage conditions (temperature and relative humidity) had a significant impact on the moisture content of cassava flour of both cultivars. The reduced trend followed for moisture content during the storage period in both cultivars were consistent with literature findings for wheat flour and other food powders (Iqbal & Fitzpatrick, 2006). Iqbal and Fitzpatrick (2006) evaluated the effect of storage conditions (5, 15, and 30 °C) and (22, 44, and 66% RH) on the moisture content of three food powders and observed the lowest value of MC (12.5%) at 30 °C, 22% RH. The authors also noticed an increase in MC under lower temperature and higher relative humidity (5 °C, 66% RH) which was similar to the observation in the current study under cool condition (15 °C, 90% RH). According to Charles *et al*. (2005) the observed moisture range in this study after 12 weeks storage (6.8 ± 0.13 to 11.9 ± 0.08) shows that flour from both cassava cultivars are moderately hygroscopic. The low moisture level reported after the storage of both cassava flour cultivars (Table 2) suggests longer shelf-life stability, however processing methods, storage conditions and type of packaging materials should be considered during storage for better quality.

Table 2 Changes in proximate compositions of cassava flour under different storage conditions (dry weight basis)

Cultivars	Storage conditions		Proximate composition of cassava flour during storage							
	Time (weeks)	**Temp (°C)**	**Moisture (%)**	**Ash (%)**	**Protein (%)**	**Fat (%)**	**CHO (%)**	**DM (%)**	**Energy value (KJ/kg)**	**pH**
TME 419	0	_	11.96 ± 0.11^{a}	1.19 ± 0.03^{d}	1.97 ± 0.07^{e}	1.00 ± 0.07^{e}	83.63 ± 0.09^{f}	88.84 ± 0.11^{f}	1487.87 ± 0.05^{e}	5.76 ± 0.02^{b}
	4	15	DO	DO	DO	DO	DO	DO	DO	
	4	23	11.79 ± 0.08^{a}	1.79 ± 0.2 ^{b}	1.76 ± 0.02^{ef}	0.94 ± 0.03^{e}	82.65± 0.28^{g}	88.20± 0.08^{g}	1448.17± 0.85^{f}	5.68 ± 0.33^{bef}
		38	8.20 ± 0.58^{e}	1.71 ± 0.19^{b}	1.79± 0.04^{e}	0.80 ± 0.00^{g}	86.53 ± 0.48^{d}	91.79± 0.57^{c}	1508.29±2.06^{d}	5.50 ± 0.57^{e}
	8	23	11.41 ±0.35^{ab}	1.58 ± 0.06^{bc}	1.70± 0.07^{g}	0.76 ± 0.00^{h}	83.53 ± 0.25^{f}	88.58± 0.35^{fg}	1455.15 ± 1.18^{f}	6.12 ± 0.10^{ab}
		38	7.48 ±0.25^{f}	1.71 ± 0.08^{bc}	1.73 ± 0.03^{gf}	0.82 ± 0.00^{f}	87.20 ± 0.30^{c}	92.51± 0.24^{b}	1519.33 ± 1.09^{bc}	5.95 ± 0.56^{ab}
	12	23	11.00± 0.02^{b}	1.60 ± 0.03^{c}	1.31 ± 0.00^{h}	0.72 ± 0.00^{i}	83.93 ± 0.05^{f}	88.99 ± 0.02^{f}	1453.61 ± 0.21^{f}	6.21 ± 0.13^{a}
		38	7.05± 0.01^{fg}	1.61 ± 0.00^{c}	1.30± 0.00^{h}	0.45 ± 0.00^{j}	88.17 ± 0.02^{b}	92.95 ± 0.01^{ab}	1514.39 ± 0.09^{cd}	5.85 ± 0.02^{c}
UMUCASS 36	0	_	9.78 ± 0.15^{c}	1.59 ± 0.03^{c}	3.03± 0.05^{a}	1.21± 0.05^{b}	85.45 ± 0.17^{e}	90.20 ± 0.15^{e}	1508.49 ± 0.34^{d}	5.54 ± 0.05^{d}
	4	15	DO	DO	DO	DO	DO	DO	DO	
	4	23	9.58± 0.30^{cd}	1.49 ± 0.10^{bc}	2.82 ± 0.05^{b}	1.15 ± 0.00^{c}	84.95 ± 0.34^{e}	90.42 ± 0.30^{de}	1512.35± 1.20^{cd}	5.68± 0.03^{ce}
		38	8.30± 0.52^{e}	1.73 ± 0.06^{bc}	2.75 ± 0.04^{b}	1.08 ± 0.00^{d}	86.13 ± 0.51^{d}	91.69 ± 0.51^{c}	1528.16± 1.93^{b}	5.74 ± 0.16^{cd}
	8	23	9.97± 0.03^{c}	2.04 ± 0.08^{bc}	2.88 ± 0.00^{a}	1.21 ± 0.00^{b}	83.53 ± 0.15^{f}	90.03 ± 0.03^{e}	1498.03± 0.23^{e}	5.71± 0.14^{cd}
		38	7.32± 0.09^{fg}	1.76± 0.19^{bc}	2.88±0.00^{a}	1.45 ± 0.00^{a}	86.59 ± 0.12^{d}	92.68 ± 0.09^{ab}	1552.36 ± 0.49^{a}	5.78± 0.10^{cd}
	12	23	9.12± 0.14^{d}	1.37 ± 0.34^{cd}	2.27 ± 0.00^{c}	0.83 ± 0.00^{f}	86.41 ± 0.73^{d}	90.87 ± 0.14^{d}	1515.36± 2.92^{cd}	6.03 ± 0.01^{b}
		38	6.83± 0.13^{g}	1.35 ± 0.09^{d}	2.27 ± 0.00^{c}	0.79 ± 0.00^{g}	88.76 ± 0.15^{a}	93.17 ± 0.13^{a}	1553.10± 0.58^{a}	5.50 ± 0.03^{f}

The values are given as means of triplicate determinations ± standard deviation.
Similar superscript letters in columns are not significantly different ($p < 0.05$).
DO = Decay observed

The highest moisture value (11.8 ± 0.08%) was observed in cv. 'TME 419' flour at week four of storage at ambient conditions (23 °C, 60%RH) , which later declined with storage duration (Fig. 4). However, the reduction in MC observed for both cvs. 'TME 419' (12.0 ± 0.11 to 7.1 ± 0.01%) and 'UMUCASS 36' (9.8 ± 9.15 to 6.8 ± 0.13%) was consistent under higher condition. This suggests that storage under higher condition (38 °C, 60% RH) could help maintain the MC of cassava flour below critical level (12%) during storage. However, storage under high temperature (38 to 45 °C) has been shown to negatively impact nutritional contents in flour products during long storage (Agrahar-Murugkar & Jha, 2011). Ambient storage condition (23 ± 2 °C, 60% RH) would best retain quality attributes and maintain shelf-life stability of cassava flour for more than 12 weeks of storage. Cool storage condition (15 °C, 90% RH) was not effective, as flour absorbed moisture from the environment which led to increase in MC and mould growth on the paper bag before the fourth week of storage.

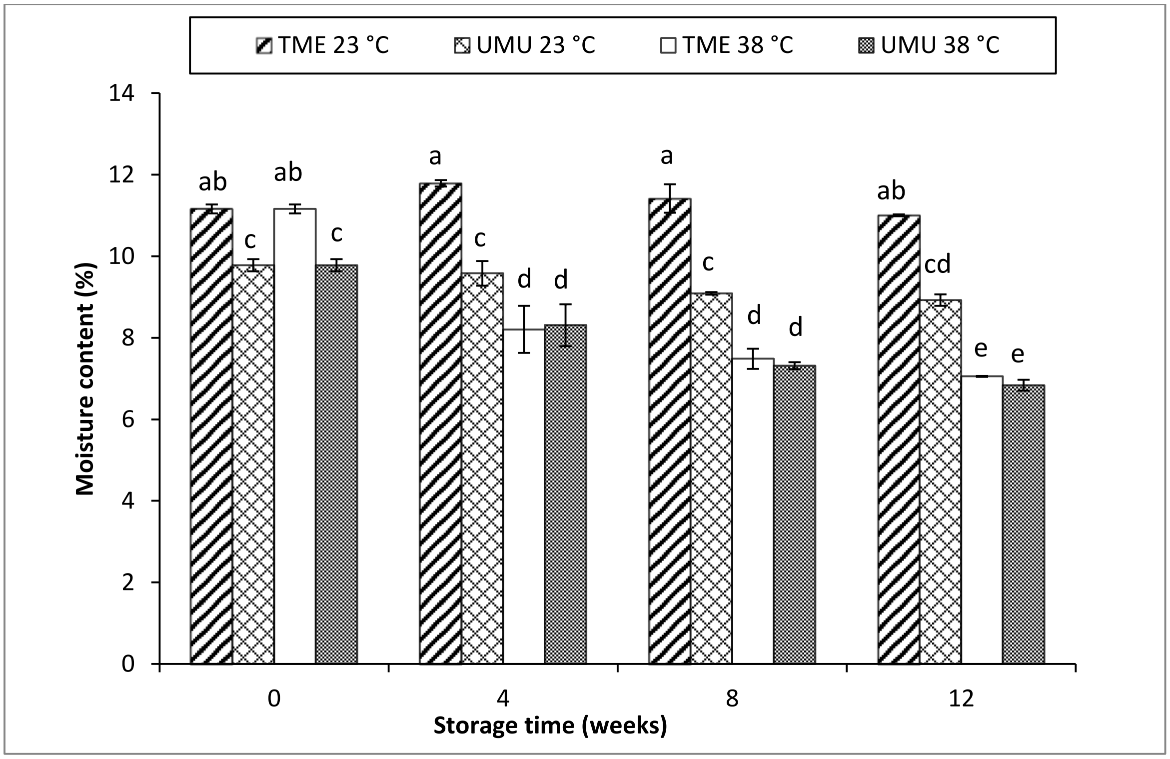

Figure 4 Changes in moisture content in cassava flour during storage at 23 and 38 °C, 60% RH. Similar superscript letters are not significantly different between data points ($p > 0.05$).

Ash content gives a quantitative estimation of the minerals available in a given food product (Eleazu *et al.*, 2012). Knowledge of the ash content in flour is essential because it allows the milling industries to estimate the expected flour yield as well as identify the milling functionality of flour

(Park & Henneberry, 2010). Ash content of 'TME 419' flour (1.60 ± 0.03%) was not significantly higher than 'UMUCASS 36' (1.37 ± 0.34%) at the end of the 12 weeks storage. This result falls within the literature range of 1.3 to 2.8% ash content of flour from different cassava cultivars (bitter and sweet) (Charles *et al.*, 2005). From the current study we can deduce that high ash content in white flour could influence the brightness and quality appearance of the final product such as white bread. However, for products like brown bread and whole wheat snack, high ash content is recommended.

At the end of the 12 weeks storage, the protein content of flour decreased by 34.0% for 'TME 419' and 25.1% for cv. 'UMUCASS 36'. The observed decrease in protein could be attributed to the rate of proteolytic activities on change in MC. Although protein content is expected to increase at lower MC because of less proteolytic activities, reverse was the case in this study. Therefore, the decline in protein could also be as a result of the decrease in the activities of the microorganism as moisture content reduced (Butt *et al*., 2004). This trend concurs with literature on the influence of moisture content on the protein content of soybean flour as increase in moisture led to increase in protein concentration (Agrahar-Murugkar & Jha, 2011). In addition, The fat content decreased with storage duration across treatments. The decrease in fat could be attributed to the lipolytic activities of the enzyme lipase and lipoxidase (Agrahar-Murugkar & Jha, 2011), which resulted in the decline in fat content. Carbohydrate content ranged from 83.6 to 88.0% in cv. 'TME 419' and from 85.45 to 88.76% in cv. 'UMUCASS 36'. Variation in carbohydrate content could be attributed to the cultivar differences, as well as the level of moisture content of flour (Charles *et al.*, 2005). It is expected that cultivar with lower moisture content will have higher carbohydrate, because of the differences in the percentage of other proximate compositions (Nwabueze & Anoruoh, 2011). The CHO contents obtained at the end of the 12 weeks storage (88.17 and 88.76%) for cvs. 'TME 419 'and 'UMUCASS 36', respectively were higher than the base line (Table 1). This outcome was consistent with reports in literature on carbohydrate content of five different cultivars of cassava flour (Charles *et al.*, 2005). In addition, the interaction between cultivars, storage conditions and duration had significant effect on percentage CHO ($p = 0.004$) (Fig. 5).

The energy contents of cassava flour during the storage period were generally high in the range of 1514.2 KJ/ kg to 1553.3 KJ/ kg (361.9 to 371.2 Kcal/ kg) (Table 2). Etudaiye *et al*. (2009) also reported high energy value of cassava fufu flour; therefore cassava root and product are good source of high caloric food. Storage conditions and duration had no significant effect on the energy values of cassava flour.

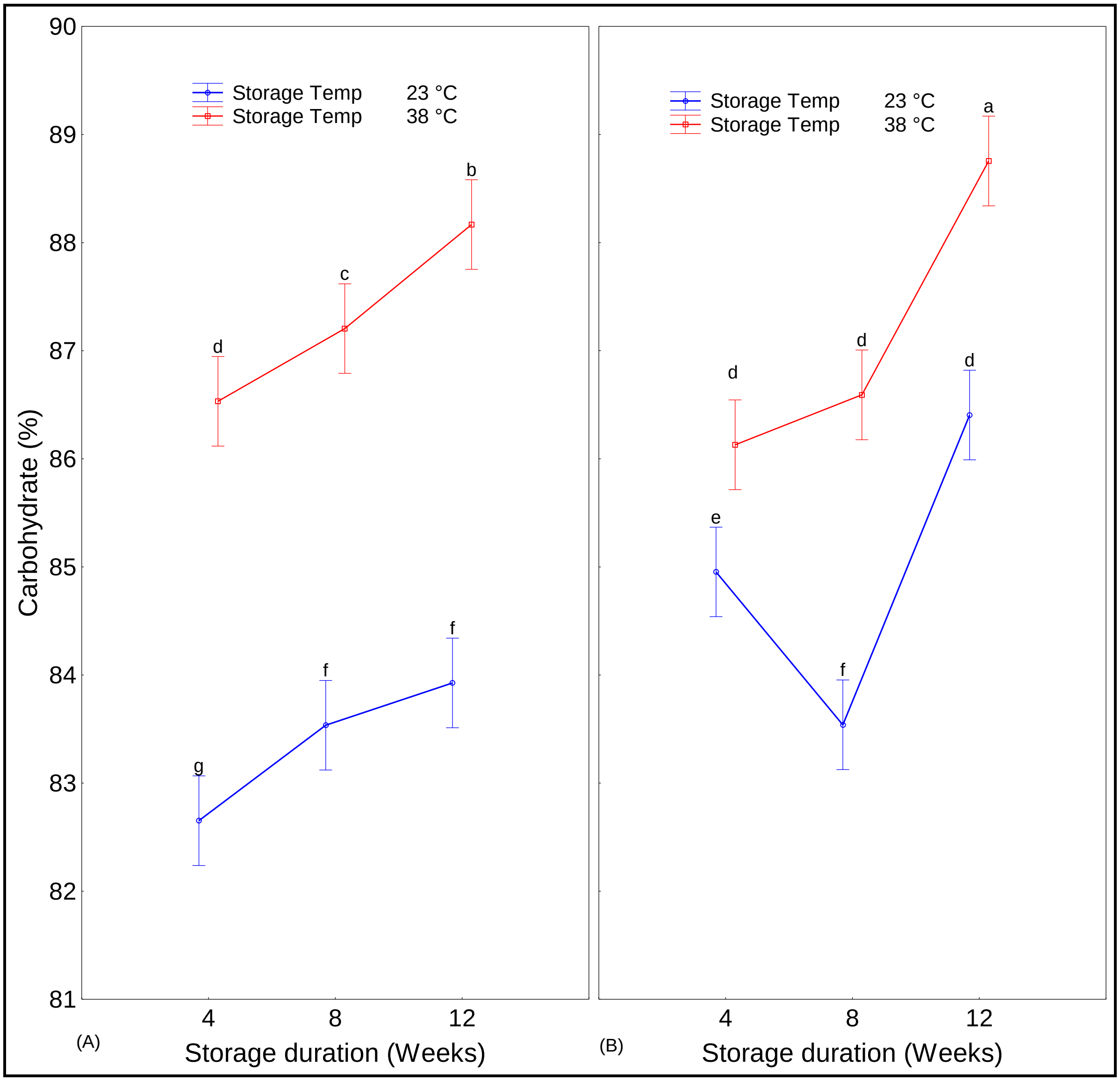

Figure 5 Interactions between cultivar, storage conditions and duration on changes in percentage carbohydrate (CHO). (A) ‘TME 419’, (B) ‘UMUCASS 36’. Data points with similar letters are not significantly different according to Duncan LSD test ($p > 0.05$).

Effects of storage on the physicochemical properties of cassava flour

Colour parameters

The measured CIE *L** *a** *b** colour parameters changed over the duration of the storage, and was significantly different in both cultivars after the storage period. Table 3 gives a summary on the colour parameter. The *L** and *b** values for both cultivars after storage ranged from 84.6 ± 0.28 to 91.2 ± 2.30. The flour of both cassava cultivars had appreciable lightness in flour after the 12 weeks storage. However, cv. 'TME 419' had the lowest *L** value (84.6 ± 0.28) under higher condition which could be attributed to the degradation in whiteness enhanced at higher temperature. Similar result range of *L** (84.64 to 90.64) and *b** (10.74 to 14.29) was reported in literature for cocoyam, taro, corn, and potato flour (Falade & Okafor, 2013; Kaur *et al.*, 2013). The *L** value was highest in cultivar 'UMUCASS 36' (91.20 ± 0.30) under higher conditions after storage because of the level of the loss in yellow pigmentation and consequently a lighter colour compared to the ambient condition (89.64 ± 0.13). Yellowness denoted by *b** was observed to be higher in cv. 'UMUCASS 36' (12.55 ± 1.02) than in cv. 'TME 419' (10.74 ± 0.53). Variation in *b** value of flour was reported to be a function of the level of protein and the carbohydrate values in the different flour cultivars (Kaur *et al.*, 2013). Thus the decrease observed in *b** value could possibly be due to degradation of yellow pigment, cultivar differences and the significant variations in carbohydrate values.

The total colour difference (ΔE) of cassava flour during the storage period ranged from 2.3 ± 0.47 to 5.1 ± 0.26 in cv. 'TME 419' and 5.6 ± 0.34 to 6.4 ± 0.32 in cv. 'UMUCASS 36'. Colour change increased across the storage duration, and the highest change in colour was observed on flours stored under higher condition (38 °C, 60 %). This highlights on the impact of high storage conditions on the colour quality of cassava flour. The percentage colour difference after the 12 weeks storage period was about 59.5% for cv. 'TME 419' and 21.9% for cv. 'UMUCASS 36'. This observation showed that colour degradation was faster in the white cv. ('TME 419') than yellow cv. ('UMUCASS 36'), which implies that 'UMUCASS 36' flour cultivar best maintained cassava flour colour. Degradation of colour during storage could also indicate loss of nutritional values due to the auto-oxidation reaction of the anthocyanin and β-carotene (Kaur *et al.*, 2013).

The whiteness index of the cassava flour varied from 81.3 ± 0.47 to 85.1 ± 0.02 in cv. 'TME 419' while cv. 'UMUCASS 36' ranged from 78.7± 0.47 to 84.5 ±.0.53 (Table 3). The whiteness index for cv. 'TME 419' decreased during storage, while about 6% increase in whiteness was observed in the flour of cv. 'UMUCASS 36' after the 12 weeks storage period. The decrease in whiteness in cv. 'TME 419' would be attributed to the oxidation reactions during storage while the increase in whiteness in cv. 'UMUCASS 36' was as a result of oxidation and degradation of yellow pigment in the flour. The lowest decrease in whiteness was observed in ambient condition at the last storage week compared to the higher condition and the baseline value prior to storage. Hence, ambient storage condition (23ºC, 60% RH) could be suggested for better stability for whiteness of stored cassava flour. In addition, the variation in whiteness in both cultivars of cassava flour during the storage period could be accredited to the genotypic difference of the cultivar. However, change in yellowness of flour of cv. 'UMUCASS 36' could be attributed to the degradation of the yellow pigment (Lin *et al.*, 2009). Storage duration had an impact on the whiteness of cassava flours in this study and there was significant difference between the two cultivars. This finding agrees with the observation reported by Hsu *et al.* (2003). The authors observed that drying had an impact on the WI of white yam flour. This highlights the need of critical evaluation of flour appearance during storage because higher WI of the flour influences consumers' acceptability.

The yellowness index of cassava flour (Table 3) ranged from 17.8 to 18.1 in cv. 'TME 419' and 28.2 to 19.3 in cv. 'UMUCASS 36'. Decrease in yellowness over time was significant in cv. 'UMUCASS 36' at 23 °C (Fig. 6). This decrease resulted in the corresponding increase in the whiteness observed in cv. 'UMUCASS 36' which infers corresponding degradation of carotenoid in the flour. Therefore, from this study, yellow colour degradation could be as a result of the intensity of exposure to light during processing or storage duration thus the need to reduce light intensity during processing is essential.

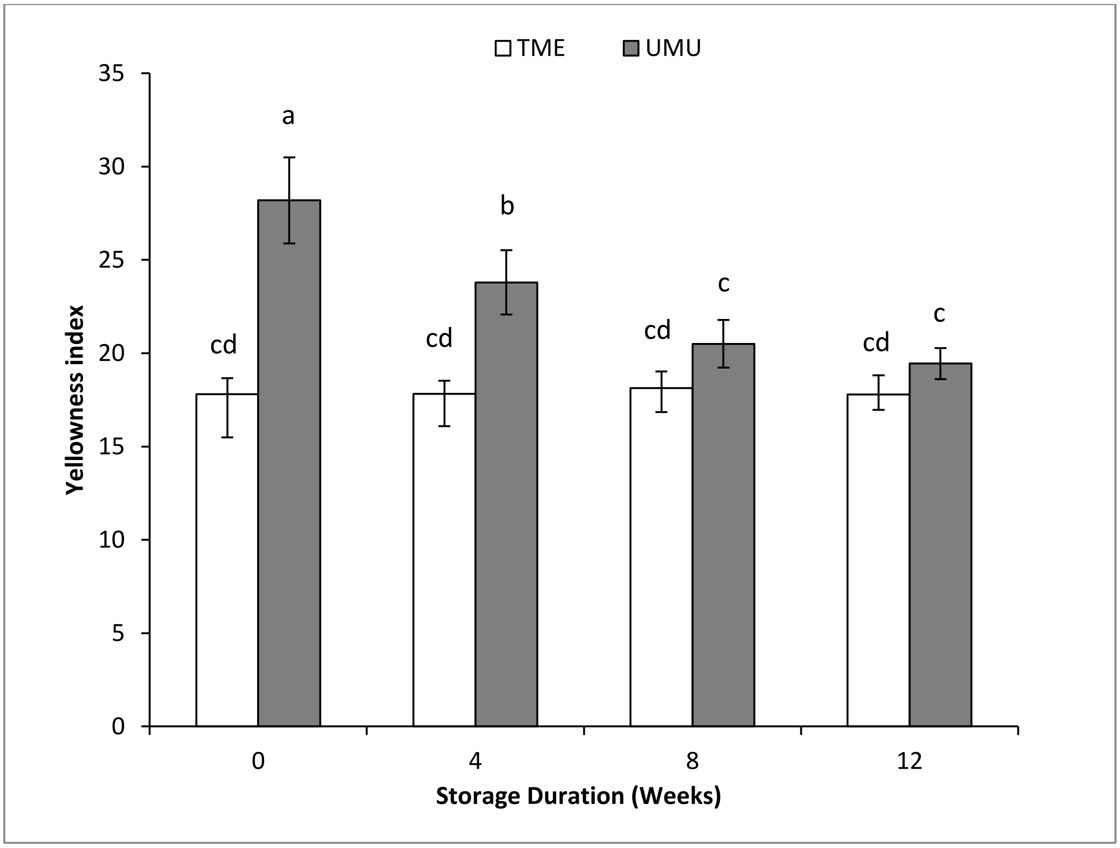

Figure 6 Changes in yellowness index of cassava flour cv. 'TME 419' and 'UMUCASS 36' stored under ambient (23 °C, 60% RH). Data points with similar letters are not significantly different ($p >$ 0.05).

Table 3 Colour analysis of cassava flour stored at different storage conditions (23 - 25 °C & 30-40 °C, 60% RH)

Cultivar	**Storage conditions**		**Colour parameters**					
	Time (weeks)	**Temp (°C)**	**L***	**a***	**b***	**ΔE***	**WI**	**YI**
TME	0	**	90.21± 0.73^{a}	-0.30± 0.00^{a}	11.24± 0.63^{d}	0.00 ± 0.00	85.09 ± 0.02^{a}	17.80 ±0.86^{fg}
	4	23	88.07 ± 0.34^{b}	-1.03 ± 0.14^{c}	10.88 ± 0.27^{d}	2.27 ± 0.47^{f}	84.3 4 ± 1.22^{abc}	17.65 ± 0.49^{fg}
		38	87.59 ± 0.25^{b}	-1.24 ±0.33^{cd}	11.02 ± 0.57^{d}	2.89 ± 0.26^{ef}	83.17 ± 0.50^{ec}	17.98 ± 0.92^{f}
	8	23	87.37 ± 0.89^{b}	-1.07 ± 0.08^{c}	10.98 ± 0.60^{d}	2.93 ± 0.93^{ef}	82.20 ± 0.36^{ef}	17.64 ± 0.97^{fg}
		38	87.68 ± 0.80^{b}	-1.09 ± 0.12^{c}	11.23 ± 0.54^{cd}	3.35 ± 0.18^{e}	82.20 ±0.25^{bc}	19.14 ± 1.52^{e}
	12	23	85.61 ± 0.94^{c}	-0.60 ± 0.17^{b}	10.45 ± 0.60^{e}	4.72 ± 0.83^{d}	82.20 ± 1.01^{ef}	17.45 ± 1.13^{fg}
		38	84.64 ± 0.28^{cd}	-0.55 ± 0.13^{b}	10.74 ± 0.53^{de}	5.61 ± 0.26^{bc}	81.25 ±0.47^{f}	18.13 ± 0.93^{f}
UMU	0	**	87.66 ± 1.68^{b}	-0.60± 0.00^{b}	17.29± 1.08^{a}	0.00 ± 0.00	78.75 ± 1.86^{fg}	28.19 ± 2.31^{a}
	4	23	84.74 ± 1.12^{cd}	-1.31 ± 0.22^{d}	13.80 ± 0.93^{b}	4.99 ± 0.34^{cd}	79.20 ± 1.67^{g}	23.35 ± 1.97^{c}
		38	83.99 ± 0.80^{d}	-1.28 ± 0.28^{d}	14.29 ± 0.86^{b}	5.03 ± 0.22^{cd}	78.38 ± 1.21^{g}	24.48 ± 1.73^{b}
	8	23	88.50 ± 0.41^{b}	-1.12 ± 0.08^{d}	12.11 ± 0.59^{bc}	5.59 ± 0.25^{cb}	83.07 ± 0.50^{de}	19.54 ± 0.93^{a}
		38	91.05 ± 0.46^{a}	-1.40 ± 0.11^{e}	13.68 ± 0.46^{b}	5.04 ± 0.45^{cd}	83.59 ± 0.44^{abcd}	21.47 ± 0.71^{d}
	12	23	89.64 ± 0.13^{a}	-1.35 ± 0.06^{de}	12.22 ± 0.36^{c}	5.99 ± 0.47^{ab}	84.52 ± 0.53^{a}	19.26 ± 0.51^{e}
		38	91.20 ± 0.30^{a}	-1.46 ± 0.11^{e}	12.55 ± 1.02^{c}	6.36 ± 0.32^{a}	84.46 ± 0.64^{ab}	19.63 ± 1.12^{e}

Data represent mean of triplicate values and standard deviation
Similar superscript letters in columns are not significantly different ($p > 0.05$).

Total carotenoids content

Carotenoid rich plants contain antioxidant components which offer various health benefits, such as reducing the risk of cardiovascular diseases, cancer and other degenerating diseases (Eleazu *et al.*, 2012). Prior to flour packaging and storage, the total carotenoid content of both cassava flour cultivars was 1.8 ± 0.11 mg/ 100 g and 2.5 ± 0.10 mg/ 100 g, for cv. 'TME 419 and 'UMUCASS 36', respectively. The flour of the yellow cv. 'UMUCASS 36' had higher carotenoid content than the white cv. 'TME 419'. The levels of carotenoid in both cassava flour cultivars were lower when compared to the cassava root (Chávez *et al.*, 2007). The ranges were from 1.8 ± 0.11 to 0.84 ± 0.01 mg/ 100g for cv. 'TME 419' and from 2.5 ± 0.10 to 1.1 ± 0.04 mg/ 100g for cv. 'UMUCASS 36'. The highest retention was observed under the ambient condition in both cultivars (Fig. 7).

The lower carotenoid content of cassava flour could be attributed to degradation during processing and exposure to light and oxygen during postharvest handling and storage (Chavasit *et al.*, 2002; Chávez *et al.*, 2007; Opara & Al-Ani, 2010; Oliveira *et al.*, 2010). Other researchers reported that carotenoid pigments especially β-carotene which is predominant in cassava are sensitive to light and can be affected by processing methods like sun-drying (Bechoff *et al.*, 2009). A study investigating the loss of carotenoid during drying of orange-fleshed sweet potato and cassava chips also reported highest percentage loss of carotenoid in sun-drying in comparison to other drying methods (Rodriguez-Amaya *et al.*, 2011). Furthermore, an evaluation study on the degradation of carotenoid during storage of Einkorn and bread wheat flours, deduced that the retention of carotenoid in flour is a function of storage temperature and duration (Hidalgo *et al.*, 2009). The authors also observed degradation of carotenoid with increase in storage time.

Therefore, we can attribute the decrease in concentration of carotenoid in the flour especially on the yellow cultivar to be degradation caused through the processing methods employed (chipping, sun-drying and milling). In addition, the higher condition (38 °C, 60% RH) had the highest drop in carotenoid concentration than the ambient condition (23 °C, 60% RH). From this result, we can conclude that carotenoid pigment would best be maintained at ambient condition.

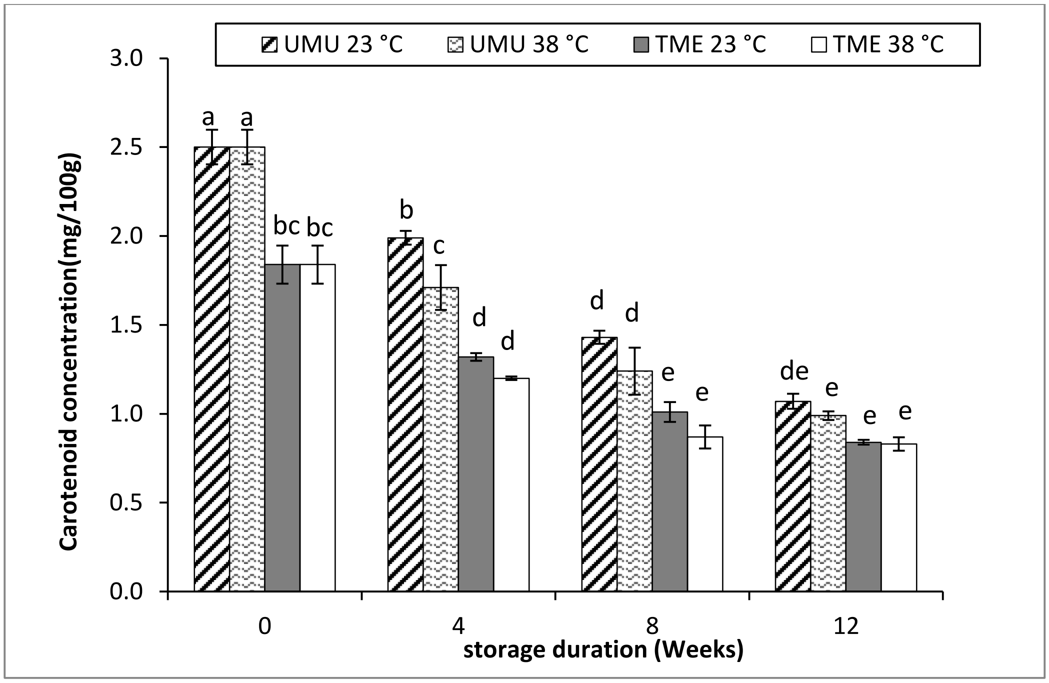

Figure 7 Effects of storage temperature and duration on carotenoid contents of the flour of two cassava cultivars ('TME 419' and 'UMUCASS 36'). Data points with similar letters are not significantly different (*p* > 0.05).

Hydrogen cyanide (HCN) *content*

After processing the cassava flour, the amount of cyanide retained in the flour was about 3.9 ± 0.06µg/ mL in cv. 'TME 419' and 4.9 ± 0.21 µg/ mL in cv. 'UMUCASS 36'. The yellow cultivar 'UMUCASS 36' was slightly higher in hydrogen cyanide retention than the white cultivar 'TME 419'. The ranges of total cyanide retained in both cultivars were lower than the reported concentration for fresh cassava root parenchyma (1 – 1550 mg/ kg) and (900 – 2000 mg/ kg) in the root cortex (Cardoso *et al.*, 2005). Both cultivars were significantly lower in cyanide concentration compared to the reported lethal dose for cyanide in human (50 - 300mg/ kg) body weight (Akiyama *et al.*, 2006). The reduction in HCN could be credited to the processing methods which include chipping and sun-drying as both methods, had been reported in literature to quicken the rate of linamarine breakdown and cyanogen reduction (Eleazu & Eleazu, 2012).

The observed differences in HCN levels could also be attributed to the difference in pH values of both cultivars. For instance, Cumbana *et al*. (2007) evaluated the effect of pH on the HCN retention in cassava flour and concluded that near neutral pH value (6.2 - 6.7) of flour will lead to corresponding decrease in HCN concentration. Thus the HCN concentration was lower in cv. ‘TME 419’ which had the highest pH value after 12 weeks of storage. The reduction in cyanide level of flour confirms previous reports that processing cassava root helps to reduce the level of cyanide in the product and improve palatability of cassava products (Cumbana *et al.*, 2007; Nwabueze & Anoruoh, 2011). The milling process of flour from cassava chips created more surface area thus enhancing the breakdown of the cyanognic compounds (linamarin and lotaustralin) in the flour.

Total cyanide retained in both cultivars after processing falls within the safe level (0 - 50mg/ kg) for human consumption as justified in literature (Akiyama *et al.*, 2006). This comparison of both cultivars in this study showed that cultivar differences as well as pH had an influence on the cyanide level of cassava flour. A slight decrease in HCN was observed during the storage duration and the rate of decrease was higher in flours stored under the higher condition than at ambient condition. This observation correlates with reports in literature on the effect of storage condition and duration on HCN level in cassava flour and other flour products (Burns *et al.*, 2012; Eleazu & Eleazu, 2012). The total HCN content retained in both cassava flour cultivars at the end of the 12 weeks storage period ranged from 3.3 ± 0.14 - 4.4 ± 0.05 µg/ mL, with the highest values observed under ambient condition in cv. ‘UMUCASS 36’. Therefore, from the result of this study, the concentration of cyanide retained in the flour before and after 12 weeks of storage may not confer any toxic effect to the consumers as it falls below the safe limit for cyanide in human foods.

Microbial stability of cassava flour during storage

Effects of storage condition (temperature and RH) and duration on the microbial stability of cassava flour was monitored for 12 weeks. To determine the microbial stability of cassava flour stored in the cool storage conditions (15 °C, 90% RH) a weekly evaluation was conducted on week 0, 1 and 2, in order to monitor the accelerated microbial growth prior to spoilage and visible decay observed at the week. The observed colonies at the first week were higher than the microbial level on the initial week (fresh/unpacked) samples (5.4 ± 0.0 - 5.6 ± 0.04 log cfu/ g) for total aerobic mesophilic bacteria and (5.0 ± 0.04 - 5.2 ± 0.02 log cfu/ g) for yeast and mould. Similar trends were also observed in the cultivar ‘UMUCASS 36’. By week 2, significant increase in microbial load was observed in both cultivars (6.3 ± 0.05 log cfu/ g) for total aerobic mesophilic bacterial and (5.6 ± 0.03 log cfu/ g) for yeast and mould with a slight discolouration of the flour colour. The values were already higher than the acceptable microbial limits (250 g)

2.4 log cfu/ g for South Africa (DOH, 2001, HPA, 2009). Visual observation of mould growth and decay was first noticed on the packages stored at 15 °C, 90% RH after the third week of storage. The observed decay could be attributed to the effect of high RH, which contributed to increase in water activity (a_w) and percentage moisture content of the flour (Table 1). This condition consequently enhanced the growth of microorganisms. Therefore, paper bag storage under cool condition will result to shelf-life instability and unwholesome flour because of the high microbial growth. Hence, this situation clearly indicates loss of flour quality and generally unacceptable for domestic and industrial uses.

The total aerobic mesophilic bacteria load in cassava flours stored at 23 °C and 38 °C were significantly influenced by a_w level of the flour (Fig. 8). At week four of storage the colony forming unit of cv. 'TME 419' increased significantly from 5.4 ± 0.01 to 6.1 ± 0.07 log cfu/ g as the water activity level increased from 0.57 ± 0.01 to 0.68 ± 0.00. Subsequently, the colony forming unit decreased with the decrease in a_w at the end of the 12 weeks storage time in both storage conditions (5.1 ± 0.04 and 4.8 ± 0.05 log cfu/ g) ambient and higher conditions, respectively.

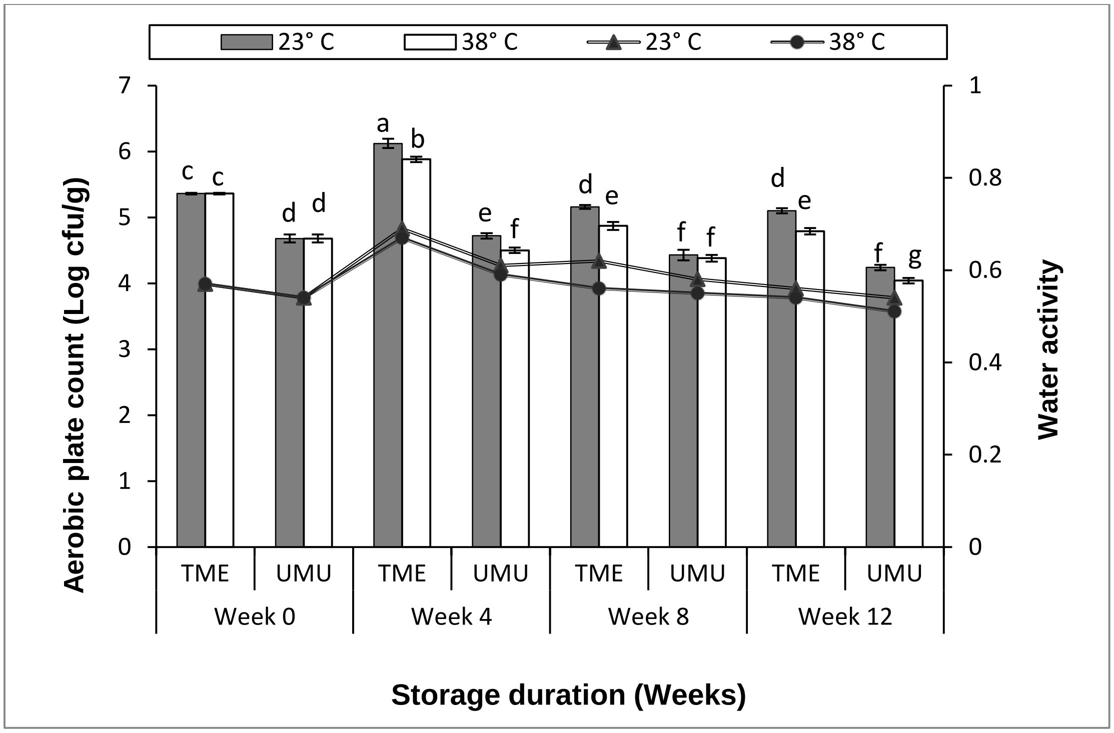

Figure 8 Effects of storage conditions and duration on the growth of aerobic mesophilic bacteria of the flour of two cassava cultivars 'TME419' and 'UMUCASS 36'. Data points with similar letters are not significantly different ($p > 0.05$). Line graphs indicate the water activity, while the bars the aerobic plate counts.

The presence of yeast and mould was also observed to decrease with the increase in storage duration at hot storage condition. Similar trend was observed in both cultivars, but cv. 'TME 419' had higher yeast and mould counts at the end of 12 weeks storage period (4.1 ± 0.04 log cfu/ g) under ambient condition and (3.8 ± 0.04 log cfu/ g) under higher condition (Fig. 9). Yeast and mould count observed at 23 °C agrees with the evaluation of flour stored at ambient conditions (Berghofer *et al.*, 2003). In addition, the yeast and mould count at the end of 12 weeks storage was within the acceptable limit for flour products (10,000 g) 4.0 log cfu/ g of (HPA, 2009). Microbial load decreased during weeks 8 and 12 of storage. Storage conditions and cultivar differences had significant effect on the microbial load. This result was consistent with observation in literature and reduced moisture level and low a_w has been reported to retard microbial growth (Padonou *et al.*, 2009).

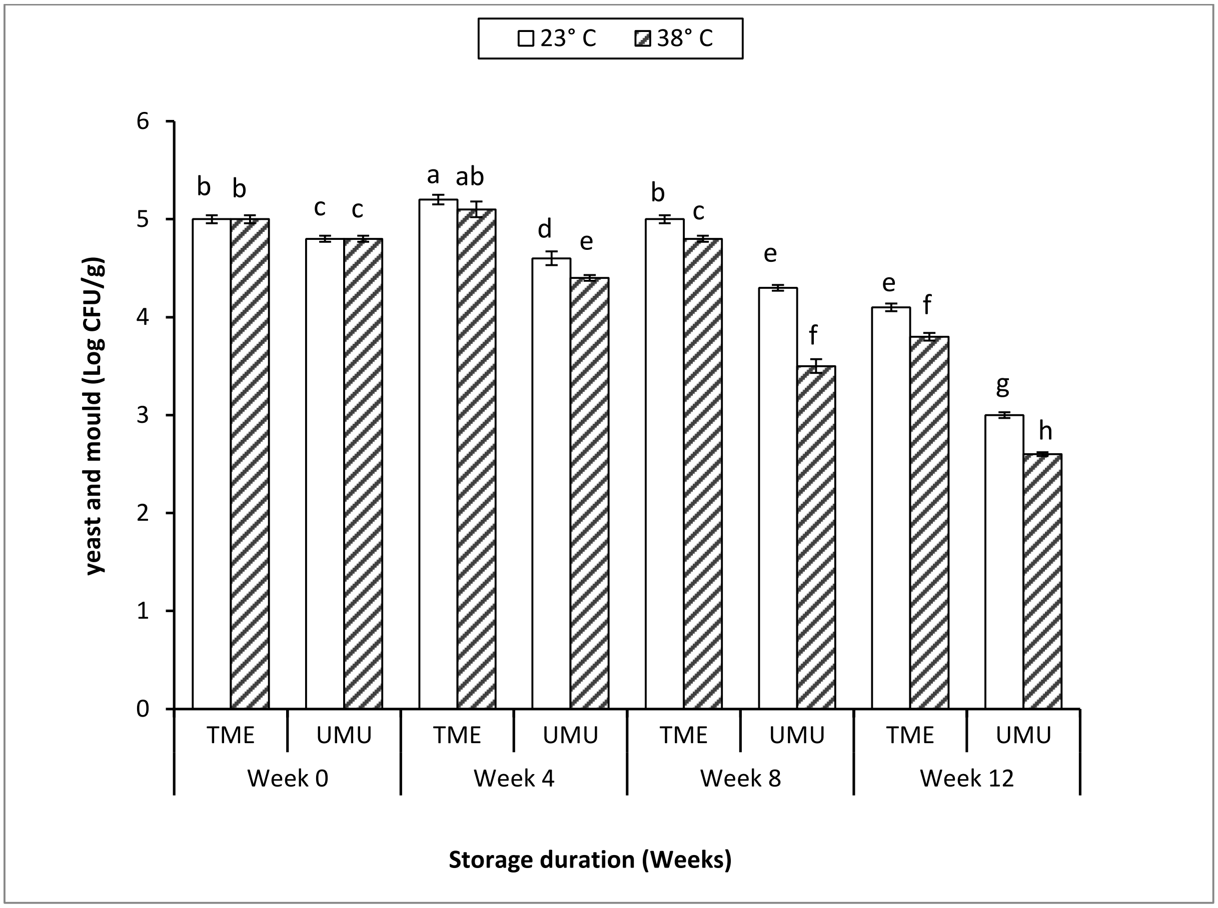

Figure 9 Effects of storage conditions and duration on yeast and mould counts of cassava flour cultivars 'TME419' and 'UMUCASS 36'. Data points with similar letters are not significantly different (*p* > 0.05).

Conclusion

Based on the outcome of this study, quality attributes of cassava flour can be significantly influenced by proper cultivar selection, storage conditions and duration. Storage conditions (temperature and relative humidity) affected the physicochemical properties of cassava flour during the 12 weeks storage period. The hot storage condition (38 °C, 60% RH) gave the lowest moisture content in both cultivars; this result gives a clear indication of longer shelf-life stability for cassava flour in such condition. However, higher condition was not favourable in maintaining the nutritional quality of cassava flour because it facilitated the proteolytic and lypolytic enzymatic reactions thus leading to significant loss of protein and fat. Conversely, the ambient condition (23 °C, 60% RH) best stabilised protein and fat contents at the end of 12 weeks storage. In addition, the change in colour attributes were reported to be lower under ambient condition, therefore cassava flour quality could be best maintained during storage under ambient condition. However, the decay observed on package at cool condition (15 °C, 90% RH) after three weeks confirms that such condition is not appropriate for flour storage with paper bags.

Total carotenoid and HCN concentration significantly decreased with the length of storage. This reduction in carotenoid concentration during storage is a concern for enjoying the health benefits of carotenoid as vitamin A precursor. This is attributed to the sensitivity of carotenoid to light and oxygen. Therefore, food processors and industries should adopt appropriate measures during processing and storage to ensure adequate retention of carotenoid in cassava flour. Reduction in HCN content in the cassava flour at the end of 12 weeks storage corroborate with other research findings that processing cassava root into flour and other products helps to reduce the HCN content. However, processing of flour should be properly done as it gives the highest percentage loss of the cyanogenic compounds.

Furthermore, the influence of storage conditions (temperature and relative humidity) on microbial stability, moisture content and water activity level of cassava flour were significant. The decrease observed in microbial load during the storage duration is favourable for shelf-life stability of cassava flour. Highest decrease in moisture and water activity level was noticed under hot storage condition (38 °C, 60% RH) which also resulted in low microbial count in cassava flour at the end of 12 weeks storage. This observation confirms that hot storage condition will be best to stabilise moisture content and extend shelf-life stability except that it causes significant loss of essential nutrients. Based on this study, it can be concluded that ambient storage (23 °C, 60% RH) could best maintain flour quality of the cassava cultivars investigated.

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CHAPTER 4

EVALUATING THE IMPACT OF SELECTED PACKAGING MATERIALS ON THE QUALITY ATTRIBUTES OF CASSAVA FLOUR (CVS. ‘TME 419’ AND ‘UMUCASS 36’)

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EVALUATING THE IMPACTS OF SELECTED PACKAGING MATERIALS ON THE QUALITY ATTRIBUTES OF CASSAVA FLOUR (CVS. TME 419 AND UMUCASS 36)

Summary

The influence of packaging materials (plastic buckets, low density polyethylene (LDPE) bags, and paper bags) on quality attributes of the flour of two cassava cultivars (‘TME 419’ and ‘UMUCASS 36’) stored at ambient condition (23 ± 2.0 °C, 60% RH) were investigated for 12 weeks. Cassava flour from each package was evaluated for proximate, physicochemical properties and microbial growth at 4 weeks interval. The result indicated that moisture content (MC), protein and fat contents decreased with storage duration across all the treatments. Flour packed in paper bags had the lowest MC (11.0 ± 0.02 and 9.1 ± 0.01%), while flour in plastic buckets had the highest MC (11.4 ± 0.02 and 9.2 ± 0.13%) for ‘TME 419’ and ‘UMUCASS 36’, respectively, at the end of 12 weeks storage period. Total colour difference (∆E) of both cassava flour cultivars increased with storage duration. Flour packed in plastic buckets had the lowest change in colour (3.2 ± 0.42) for cv. ‘TME 419’ and (4.1 ± 0.87) for cv. ‘UMUCASS 36’. The concentration of total caroteniod decreased across all treatments. After the 12 weeks storage, the highest total carotenoid retention (1.7 ± 0.02 and 2.0 ± 0.05 µg/ mL) was observed in flour packed in plastic buckets. The highest microbial load at the end of 12 weeks storage period (3.8 ± 0.1 log cfu/ g and 3.6 ± 0.07 log cfu/ g) for total aerobic mesophilic bacteria and fungi respectively was observed in flour packed in plastic bucket while flour in paper bag had the lowest microbial count (3.4 ± 0.03 log cfu/ g and 3.4 ± 0.08 log cfu/ g) for total aerobic mesophilic bacteria and fungi, respectively. Similar trend was followed in ‘TME 419’ cassava flour cultivars. Cassava flour packed in paper bag and stored under ambient condition, which had the lowest MC and lowest microbial load, could be recommended as the best packaging type for both cassava flour cultivars at the end of 12 weeks storage duration. However, for carotenoid retention and reduction in colour change, flour packaging with plastic buckets would be ideal.

Introduction

The global annual production of cassava was estimated to be over 238,000 tonnes (FAO, 2013), about 85% of the roots are processed into different food forms such as garri, lafun, fufu and composite flour for domestic and industrial purposes (Shittu *et al.*, 2008). Most of these minimally processed by-products of cassava roots are affected by poor postharvest handling such as packaging and storage, which consequently result in losses and shelf-life instability (Ogiehor & Ikenebomeh, 2006). This is because of the continuous respiration rate of the root even in the processed forms. However, products like cassava flour give the benefit of longer shelf-life stability as well as reducing the cyanogenic compound in the root. Having the root reconstituted in flour form adds value and also helps to maintain quality of the fresh root for future use. High quality cassava flour is a potential product for composite flour in baking of high grade food such as bread. Studies have proven cassava flour to be a good partial substitute for wheat flour in food processing (Nwabueze & Anoruoh, 2011; Sanful & Darko, 2010). This is attributed to its high energy value, gluten-free nature, low cost of production and its availability. Consequently, cassava flour promotes the utilisation of cassava root for human, animals feed as well as for industrial purposes (Falade & Akingbala, 2010). Furthermore, commercialisation of cassava flour will promote the growing economy of most developing countries and reduce dependency on wheat flour. This will offer more job opportunities for farmers and food processors (Gyedu-Akoto & Laryea, 2013).

One major postharvest challenge limiting the shelf-life and quality of flour is the use of improper packaging materials. In particular, flour is a hygroscopic product, which could be exposed to high humidity during shipping or storage. Some packaging materials are not permeable to gases and moisture, hence, moisture as well as environmental gases such as oxygen is trapped inside with the flour. This consequently may result to rancidity, mouldy or microbial proliferation in the flour thereby reducing shelf-life, commercialisation, exporting of cassava flour and most importantly reduce its utilisation (Ogiehor & Ikenebomeh, 2006). Food packaging offers the benefit of containment, easy communication, convenience and the protective measures from contamination associated with postharvest handling (Opara & Mditshwa, 2013). Optimum storage condition and packaging help to reduce incidence of postharvest losses, add value, extend shelf-life, maintain quality and wholesomeness of products, improve market standard and food safety (Inyang *et al.*, 2006; Opara & Al-Ani, 2010). However, the product quality and shelf-life can be influenced by the type of package used and the characteristic features of the packaging material. This means that a particular product has a specific packaging material suitable for it.

Various packaging materials have been reported in literature for flour storage and these include: paper bags, plastic buckets, polyethylene bags, sack bags or a combination of one or two of the materials such as laminated paper bags and jute bags with polyethylene lamination (Ogiehor & Ikenebomeh, 2006; Ogugbue & Gloria, 2011). Ogiehor and Ikenobomeh (2006) evaluated the effect of different packaging materials (LDPE bags, Hessian bag, HDPE bags, and plastic buckets) on the stability of garri stored at 30 °C for 24 weeks and found that bacteria count increased gradually from 3.0 x 10^1 to 9.9 x 10^3 cfu/ g in LPDE, and to 1.10 x 10^3 cfu/ g in plastic buckets. One major observation the authors noticed was increase in moisture content (12.15 ± 0.01 to 19.18 ± 0.25%), which gave rise to higher microbial load in the stored garri for all the packages. This observation was attributed to the inability of the LDPE and plastic bucket to permeate water and gases out from the package; instead the gases were absorbed by the garri samples. The authors also found that hessian bag gave the least microbial count (1.01 x 10^3 cfu/ g) and was recommended for best keeping quality of garri. Similarly, Ogugbue and Gloria (2011) stored garri under tropical market conditions for 9 weeks in gunny sack, HDPE bag, LDPE bag and plastic container and observed progressive increase in moisture in all the packages. The authors reported that the openly displayed garri had the highest moisture content (18.0%) and fungi count (5.5 ± 0.3 x 10^5 cfu/ g) while HDPE bags with lowest moisture (10.52%) best maintained microbial stability at the end of the 9 weeks storage. The study revealed that packaging type and storage duration had significant impact on the shelf-life stability of garri. Therefore, critical evaluation of effect of package type on product quality is essential. However, to date there is limited information on shelf-life stability of cassava flour stored in different packaging materials over long term period. Hence, this work was designed to evaluate the impact of three selected packaging materials (plastic bucket, low density polyethylene bag (LPDE) and brown paper bag) on the proximate, physicochemical and microbial stability of cassava flour stored at ambient temperature (23 ± 2 °C, 60% RH) for 12 weeks.

Materials and methods

Plant materials and processing

One of the newly released yellow cassava cultivars ‘UMUCASS 36’ and ‘TME 419’ (white root) were used for this research. Fresh roots of about 30 kg from each cassava cultivar were harvested after 12 months of planting from a randomised complete block design field plot at National Root Crops Research Institute (NRCRI) Umudike (5°28′33″N 7°32′56″E), Abia state, Nigeria.

The freshly harvested cassava roots were sorted, washed, peeled and re-washed with clean running tap water. Processing into chips was done immediately after washing to avoid enzymatic browning, fermentation or on set of deterioration. A chipping machine (YS QS400, Shandong, China) was used to cut the roots into chips of about 2 - 3 cm smaller surface area to enhance drying. The chips were thinly spread on clean mat (Fig. 1) and sun-dried for three days to constant moisture level 13 %. Chips were shuffled at interval to avoid browning during the drying process and for uniformity in drying (Falade & Akingbala, 2010). The dried chips were packaged in air-tight polyethylene bags and transported to Stellenbosch University where they were milled into flour with a Cyclone Laboratory mill (Model 3100, Perten Instruments, Hagersten, Sweden) with 0.5 mm sieve size.

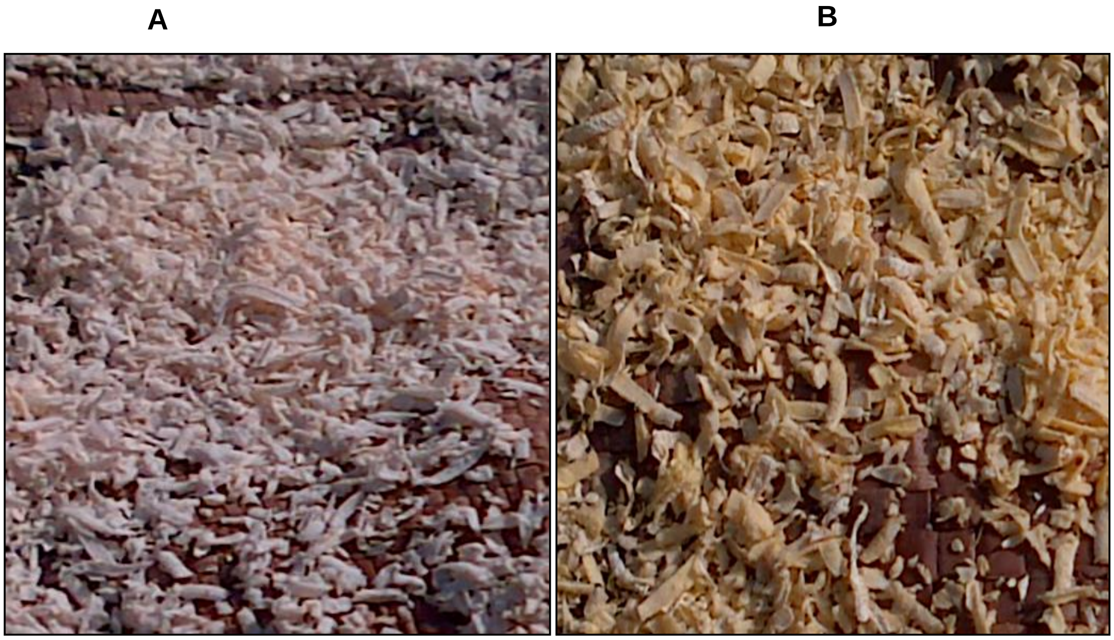

Figure 1 Cassava chips obtained from processing the roots of both cultivars (A) TME 419 and (B) UMUCASS 36.

Flour packaging and storage

Fresh processed cassava flour from both cultivars was weighed (200 g) and packaged in three selected packaging materials (plastic bucket, LDPE bag (50 micron) and brown paper bag (150× 250 mm)) (Fig. 2). The packaged cassava flour was stored at ambient temperature 23 ± 2 °C, 60% RH for 12 weeks. Samples were taken for analysis every 4 weeks and proximate, physicochemical and microbial quality was evaluated in triplicates on each sampling day starting from the week zero (fresh, unpackaged flour).

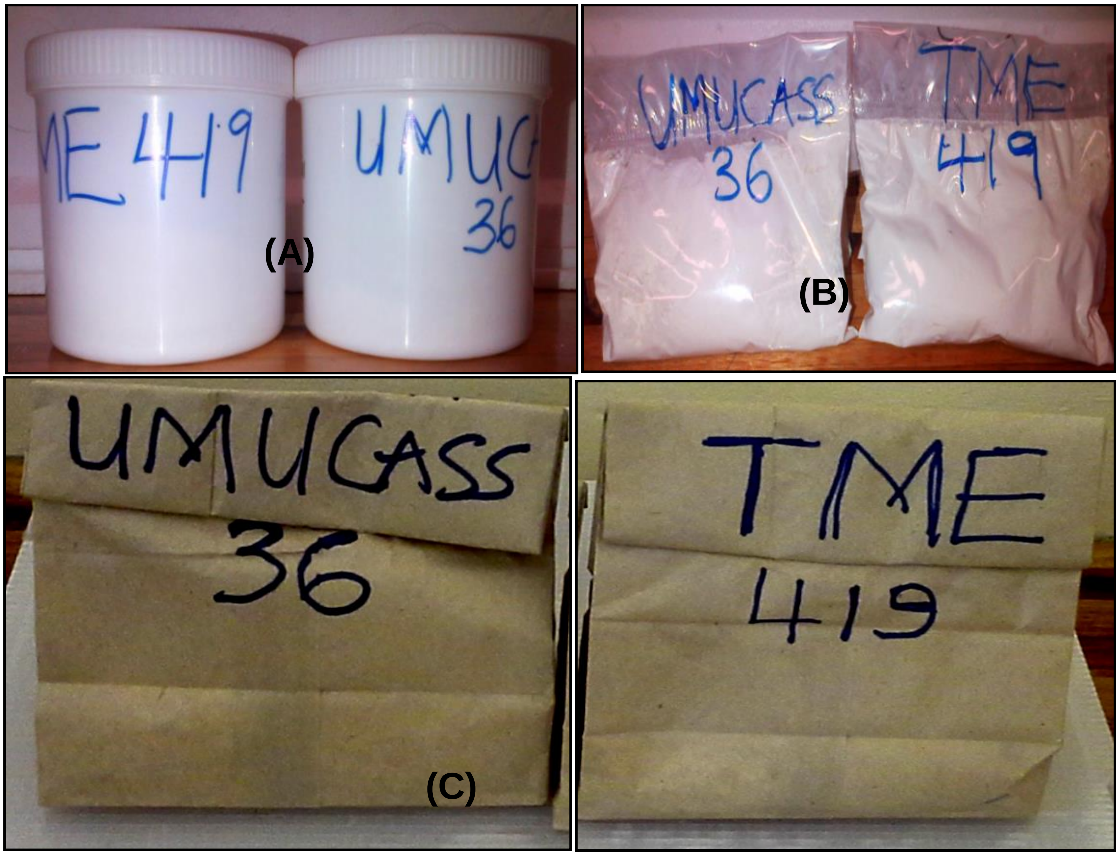

Figure 2 Package types used for cassava flour storage. (A) Plastic buckets, (B) LDPE bags, (C) Paper bags

Proximate composition of flour

Moisture content was determined by loss on drying method using force draft Pro-Lab oven and incubator (OTE 160L Lab tech. separation scientific, South African). Ash content was done following the direct method (923.03) (AOAC, 2012), with minor modifications. Cassava flour (2 g) was weighed into a pre-weighed crucible and put in an electric muffle furnace (LEF 230 PE (1), Daihan Lab Tech, India) set at 100 ºC for 2 h after which the furnace was ignited at 550 ºC for 12 h. The ash was cooled in desiccators and weighed. The percentage ash was calculated for all cassava flour treatments. Protein analysis was carried out following the Kjeldahl method 920.152 (AOAC, 2006). Cassava flour samples (2 g) were digested at 400 °C in sulphuric acid using Kjeldahl tablet (Merck, South Africa) as catalyst. The digests were distilled through a distillation unit (UDK129, Italy) into a boric acid solution containing methyl red and bromocrescyl green indicator (Merck, South Africa). Sodium hydroxide about 35% was used to neutralize the reaction process. Afterwards greenish boric acid solution was titrated against 0.1 M HCl until a permanent light pink colour appeared. The titre values for all the treatments were used to calculate the % nitrogen (N) and protein content was calculated from % N using a conversion factor 6.25 (N × 6.25). Soxhlet extraction method was used to determine the percentage fat content, using petroleum ether as the extraction solvent (AOAC, 2006). Percentage carbohydrate content was calculated by difference with equation 1 (Nwabueze & Anoruoh, 2009). Energy value was calculated using Atwater factor equation 2 (Akubor *et al*., 2003). The dry matter was determined using equation 3 (100-MC) (Etudaiye *et al.*, 2009)

$$\mathrm{CHO}(\%) = 100 - \%\ (\mathrm{protein} + \mathrm{fat} + \mathrm{Moisture} + \mathrm{Ash}) \quad (1)$$

$$\mathrm{Energy\ value\ (KJ/g)} = (\mathrm{protein} \times 4 + \mathrm{Fat} \times 9 + \mathrm{CHO} \times 4) \quad (2)$$

$$\mathrm{Dry\ matter} = (100 - \mathrm{moisture\ content}) \quad (3)$$

Physicochemical analysis

Colour difference

Colour was measured using CIE*LAB* colour coordinates. The colour values expressed as *L** (lightness), *a** (green) and *b** (yellowness/blueness) were measured using a Minolta chroma meter (Model CR-400/410, Konica Minolta sensing Inc., Japan). The instrument was calibrated against a white plate before snapshots were taken on the sample. Flour was poured into a dry dish and colour snapshots were taken at 3 different points for each treatment. The mean of the nine measurements were calculated accordingly with equation 4 (Rhim & Hong, 2011; Pathare *et al*., 2013).

$$\Delta\mathrm{E} = \sqrt{\Delta\mathrm{a}*^2 + \Delta\mathrm{b}*^2 + \Delta\mathrm{L}*^2} \quad (4)$$

Whiteness and yellowness index

The indices are usually used to estimate the level of colour variations in products. Whiteness indices (WI) signify the total whiteness of flour or other food products and can also denote the degree of discolouration or degradation of whiteness in products during storage (Hsu *et al.*, 2003; Lin *et al.*, 2009). Whiteness has an impact on flour appearance, quality of flour during storage and flour quality influences the acceptance of the end products (Rodriguez-Aguilera *et al.*, 2011). Whiteness is based on the scale of 0-100. Equation 5 was used to calculate WI of cassava flour (Pathare *et al*., 2013).

$$\mathrm{WI} = 100 - \sqrt{(100 - \mathrm{L}*^{2}) + \mathrm{a}*^{2} + \mathrm{b}*^{2}} \tag{5}$$

The yellowness index (YI) was measured to quantify the level of yellow colouration especially in the carotenoid cultivar during storage. Studies have shown that packaging and storage for longer period can degrade the yellowness in flour and/or processed products (Rodriguez-Amaya *et al.*, 2011). Flour YI was calculated using equation 6 (Pathare *et al*., 2013).

$$\mathrm{YI} = \frac{142.86 \times \mathrm{b}*}{\mathrm{L}*} \tag{6}$$

Total carotenoids

The total carotenoid content of cassava flour was determined using UV-Vis spectrophotometer as described by Opiyo and Ying (2005) with minor modifications. The n-hexane: acetone 14 mL solution prepared in the ratio of (3:2 v/v) was used to homogenise 1 g of cassava flour. The whole mixture was agitated for 1 min before centrifuging for 10 min at 6000 rpm and 4 ºC in eppendorf centrifuge (Mark Chemicals, (Pty) Ltd, South Africa). Absorbance was read immediately at 450 nm using a spectrophotometer (Helios Omega UV-Vis Thermo Scientific, USA). Carotenoid products are sensitive to light and as a result of that, this experiment (starting from sample preparation to absorbance reading) was done under less intense (not bright) light in the laboratory. Total carotenoids content were calculated for all packaging materials and expressed in (mg/ 100g) dry weight basis using the equation 7 (Opiyo & Ying, 2005; Opara & Al-Ani, 2010).

$$\text{Total carotenoid (mg/100g)} = \frac{\mathrm{Ab}^{450} \times 4}{\text{Sample weight}} \times 1000 \tag{7}$$

***Hydrogen Cyanide Determination* (HCN)**

Hydrogen cyanide was determined with minor modification using the alkaline picrate method (Onwuka, 2005). Cassava flour (5 g ± 0.02) was taken from each package into a conical flask. The flour was mixed with 50 mL distilled water and kept at room temperature overnight. Later the solution was filtered using Whatman filter paper No 1. Using a pipette, 1 mL was taken from the filtrate and added to 4 mL of alkaline picrate prepared by dissolving 5 g Na_2CO_3 and 1 mL of picric acid with warm water in 200 mL volumetric flask. The whole set up was incubated in a water bath at 50 °C for 5 min, a light reddish brown colour was observed. The absorbance was read using a UV-Vis spectrophotometer at 490 nm (Helios Omega UV-Vis Thermo Scientific, USA) against a reagent blank containing 1 mL water and 4 mL alkaline picrate. Standard curve (Fig. 3) was used to extrapolate the concentration of HCN with the standard curve equation $y = (2.4x + 0.216)*10$. Where y= concentration, 2.4= slope of curve, X= absorbance, 0.216 = intercept and 10 is the dilution factor (R^2= 0.917). The mean values of the concentration were expressed in µg/ mL.

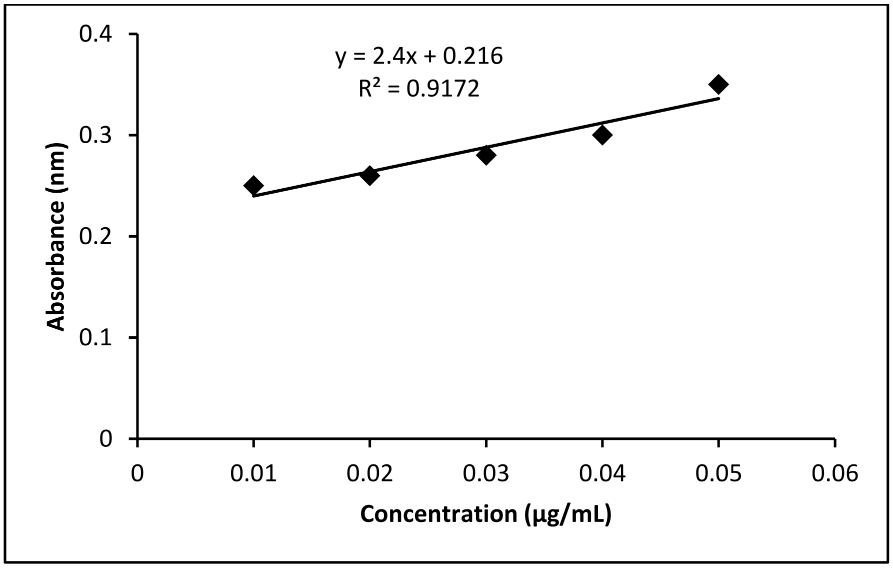

Figure 3 Standard curves for hydrogen cyanide prepared by diluting potassium cyanide (KCN) in water acidified with HCL

Determination of pH

Cassava flour from each treatment was dissolved with distilled water in a ratio of 10 g to 10 mL. The dissolved flour samples were allowed to stand for 1 h and the residue was filtered (Ogiehor & Ikenebomeh, 2006). The pH levels were then determined with a reference glass electrode pH meter basic 20^+ (Crison, 52-01 PI1000, Spain) by placing the electrode in the filtrates.

Water activity

Water activity (a_w) of cassava flour was measured throughout the storage period in all treatments using Pawkit water activity meter (Decagon Devices, Inc., Pullman, Washington, USA). Water activity for each treatment was taken in approximately 5 min following the pawkit standard procedure as described. The sample cups were half-filled (to avoid sample covering the water meter sensor) with cassava flour samples from the different packaging materials. The instrument was placed on the cup containing the cassava flour samples and measurement taken at a constant temperature of 21 °C (Rhim & Hong, 2011). Measurement was done at 4 weeks interval for 12 weeks starting from the week zero

Microbial analysis

Microbial analysis was done to determine the influence of different packaging materials on the microbial load of cassava flour over time starting from the baseline (week zero). Potato dextrose agar (PDA) was used for yeast and mould count, while plate count agar (PCA) was used for total aerobic mesophilic plate count. Physiological solution (PS) containing 9 g of NaCl in 1000 mL distilled water was used for the serial dilutions. From each package, 10 g of cassava flour was aseptically taken and homogenised in 90 mL of previously sterilised physiological solution. From homogenised flour solution, 1 mL was taken and transferred into 9 mL PS and fivefold dilution was done. The solution was then plated out in triplicates on PCA and PDA plates, for bacteria and fungi, respectively using the pour plate technique. Plates were allowed to solidify, and placed at inverted position to avoid water condensing into the medium. The PCA plates were incubated at 37 °C for 48 h and the PDA plates were incubated at 26 °C for 72 h (Akhtar *et al.*, 2008). The observed colonies were counted after incubation and expressed as log cfu/ g. To ascertain the microbial state of the environment, sterile plates without samples were left open for 10 min in the laboratory and on the work area

Statistical analysis

Statistical analyses were carried out according to the factorial design. Three replicates of cassava flour were done per treatment and experimental data were analysed using Statistica software, Version 12.0 (Statistica, Statsoft, USA). The effects of packaging material, storage duration, and cultivar as well as their interaction on the quality attributes of cassava flour were analysed using a three-way factorial Analysis of Variance (ANOVA). Significant differences were established at $p \leq 0.05$ according to Duncan's multiple range tests. Data obtained were reported as mean and ± standard deviations for each treatment.

Results and discussion

Proximate composition of cassava flour

The influence of the packaging materials on the proximate composition of cassava flour (cvs. 'TME 419' and 'UMUCASS 36') are presented in Table 1. Package and storage duration had significant impact on the proximate composition of both cassava flour cultivars ($p < 0.05$). Progressive decrease in MC during the storage period was observed in both cultivars across all the package types. Moisture content of cassava flour ranged from 9.1 ± 0.01 to 10.9 ± 0.02% in paper bags, from 9.1 ± 0.12 to 11.3 ± 0.03% in LDPE bags, and from 9.2 ± 0.13 to 11.4 ± 0.02% in plastic buckets. Flour packed in paper bag had the lowest MC (10.9 ± 0.02%) while flour packed in plastic bucket had the highest MC at the end of the 12 weeks storage (11.4 ± 0.02%). The MC range corresponds with the recommended moisture content (12-13%) for flour stability and in line with the reported range (9.2 to12.3%) for MC of different cultivars of cassava flour (Charles *et al*., 2005).

Lower moisture content in flour is a good indication of microbial stability and may also contribute to reducing the tendency of staling in baked food products (Ogiehor & Ikenebomeh, 2006). Furthermore, the high moisture noticed in flour packed in plastic bucket after 12 weeks storage period would be related to its low water vapour transmission rate as well as the higher microbial activities (Agrahar-Murugkar & Jha, 2011). Results from this research are in the range of the values reported for 31 different cassava flour cultivars (3.21 to 11.75%) by Aryee *et al*. (2006). It also corresponds with the values (9.8-11.8%) reported in the study on the effect of moisture and packaging materials on the shelf-life of wheat flour (Butt *et al.*, 2004). Butt *et al.* (2004) observed low moisture level (10.9%) after the 60 d storage period in paper bags and recommended that paper bags would be more appropriate for stabilising shelf-life, controlling microbial infestations and maintaining quality of wheat flour. Based on the result from the present study, paper bag would be most effective for shelf-life stability of cassava flour stored at ambient conditions (23 °C, 60% RH). Statistical results showed that the interaction of cultivar, storage duration and package types had a significant effect on MC ($p < 0.05$) (Fig. 4).

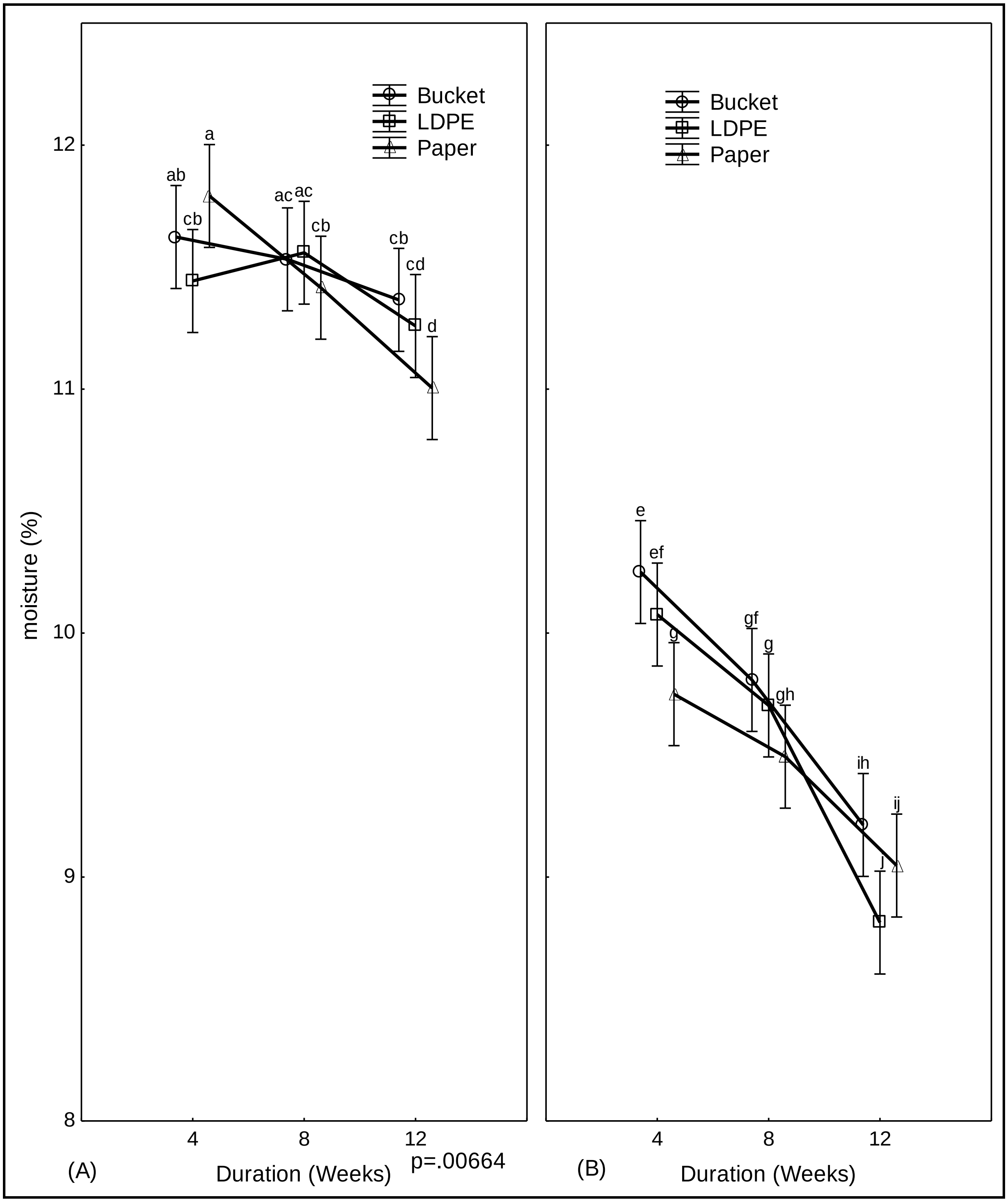

Figure 4 Effects of the interaction of the package types, storage duration and cultivar on the moisture content of cassava flour. (A) 'TME 419', (B) 'UMUCASS 36'. Different superscript letters show significant differences ($p < 0.05$) between data points

Table 1 Effects of packaging and duration on the proximate composition of cassava flour (cvs. 'TME 419' and 'UMUCASS 36') (dry weight)

Proximate composition	Cassava flour	Package type	Week 0	Week 4	Weeks 8	Week 12
***Moisture* (%)**	TME 419	Bucket	12.0 ± 0.11^{a}	11.6 ± 0.1 3^{ab}	11.5 ± 0.23^{ac}	11.4 ±0.02^{cd}
		LDPE	12.0 ± 0.11^{a}	11.4 ±0.45^{bc}	11.6 ± 0.11^{ac}	11.3 ± 0.03^{cd}
		Paper bag	12.0 ± 0.11^{a}	11.8 ± 0.08^{a}	11.4 ± 0.35^{bc}	10.9 ± 0.02^{d}
	UMUCASS 36	Bucket	9.8 ± 0.15^{g}	10.3 ± 0.20^{e}	9.8 ± 0.21^{gf}	9.2± 0.13^{ih}
		LDPE	9.8 ± 0.15^{g}	10.1 ± 0.02^{ef}	9.7 ± 0.07^{g}	9.1 ± 0.12^{ih}
		Paper bag	9.8 ± 0.15^{g}	9.8± 0.2^{g}	9.5 ± 0.05^{gh}	9.1 ± 0.01^{i}
***Ash* (%)**	TME 419	Bucket	1.8 ± 0.03^{a}	1.7± 0.06^{a}	1.6 ± 0.04^{abc}	1.8 ±0.09^{ab}
		LDPE	1.8 ± 0.03^{a}	1.8 ± 0.16^{a}	1.8 ±0.20^{a}	1.7±0.10^{ab}
		Paper bag	1.8 ± 0.03^{a}	1.8 ± 0.21^{a}	1.6 ± 0.06^{abc}	1.6 ±0.07^{abc}
	UMUCASS 36	Bucket	1.6 ± 0.03^{b}	1.5 ± 0.14^{ab}	1.4 ± 0.12^{cde}	1.3 ±0.07^{de}
		LDPE	1.6 ± 0.03^{b}	1.5 ±0.10^{bd}	1.5 ± 0.05^{bd}	1.2 ±0.00^{ef}
		Paper bag	1.6 ± 0.03^{b}	1.5 ± 0.10^{db}	1.4 ± 0.14^{cde}	1.1 ±0.13^{f}
***Protein* (%)**	TME 419	Bucket	2.0 ± 0.07^{c}	1.7 ± 0.07^{c}	1.7 ± 0.07^{c}	1.4 ±0.00^{d}
		LDPE	2.0 ± 0.07^{c}	1.7 ± 0.04^{c}	1.7 ± 0.07^{c}	1.4 ±0.00^{d}
		Paper bag	2.0 ± 0.07^{c}	1.8 ± 0.02^{c}	1.7 ± 0.08^{c}	1.3 ±0.00^{d}
	UMUCASS 36	Bucket	3.0 ± 0.05^{a}	2.7 ± 0.03^{b}	2.8 ± 0.05^{a}	2.7 ±0.00^{b}
		LDPE	3.0 ± 0.05^{a}	2.7 ±0.03^{b}	2.9 ±0.00^{a}	2.7 ±0.00^{b}
		Paper bag	3.0 ± 0.05^{a}	2.8 ±0.05^{a}	2.9 ±0.00^{a}	2.7 ±0.00^{b}
***Fat* (%)**	TME 419	Bucket	1.0 ± 0.07^{g}	1.0 ± 0.01^{h}	0.9 ± 0.02^{j}	1.0± 0.00^{h}
		LDPE	1.0 ± 0.07^{g}	0.9± 0.0^{k}	0.8 ± 0.01^{i}	0.8± 0.00^{m}
		Paper bag	1.0 ±0.07^{g}	0.9 ± 0.03^{h}	0.8± 0.00^{m}	0.7± 0.00^{n}

Table 1 continues

Proximate composition	Cassava flour	Package type	Week 0	Week 4	Weeks 8	Week 12
	UMUCASS 36	Bucket	1.2 ± 0.05^{d}	1.1 ± 0.00^{g}	1.1 ± 0.00^{f}	1.2 ±0.00^{d}
		LDPE	1.2 ± 0.05	1.4 ± 0.00^{b}	1.5 ± 0.00^{a}	0.8 ±0.00^{j}
		Paper bag	1.2 ± 0.05	1.2 ± 0.00^{d}	1.2 ± 0.00^{c}	0.8 ± 0.00^{i}
***CHO* (%)**	TME 419	Bucket	83.6 ± 0.09^{fg}	82.9 ± 0.2^{jk}	83.2 ± 0.23^{hi}	83.2 ± 0.10^{gh}
		LDPE	83.6 ± 0.09^{fg}	83.2 ± 0.34	83.1^{ji} ± 0.26^{hi}	83.6 ± 0.07^{fg}
		Paper bag	83.6 ± 0.09^{fg}	82.7 ± 0.19^{k}	83.5 ±0.24^{hg}	83.9 ± 0.14^{f}
	UMUCASS 36	Bucket	85.5 ± 0.17^{b}	84.5 ± 0.22^{cd}	84.9 ± 0.13^{c}	85.7 ± 0.18^{b}
		LDPE	85.5 ± 0.17^{b}	84.3 ± 0.11^{e}	84.4 ± 0.04^{e}	86.2 ± 0.12^{a}
		Paper bag	85.5 ± 0.17^{b}	84.8 ± 0.33^{cd}	85.0± 0.11^{c}	86.4 ± 0.14^{a}
***Dry matter* (%)**	TME 419	Bucket	88.8 ± 0.11^{gh}	88.4 ± 0.13^{ih}	88.5 ±0.23^{ih}	88.6 ± 0.02^{gh}
		LDPE	88.8 ± 0.11^{gh}	88.6 ± 0.45^{ih}	88.4 ± 0.11^{ih}	88.7 ± 0.02^{gh}
		Paper bag	88.8 ± 0.11^{gh}	88.2 ± 0.08^{i}	88.6 ± 0.35^{ih}	89.0 ± 0.02^{g}
	UMUCASS 36	Bucket	90.2 ± 0.15^{d}	88.7 ±0.23^{gh}	90.2 ± 0.38^{c}	90.8 ± 0.18^{a}
		LDPE	90.2 ± 0.15^{d}	89.7 ± 0.30^{f}	90.3 ± 0.03^{c}	90.9 ±0.22^{ab}
		Paper bag	90.2 ± 0.15^{d}	90.2 ± 0.30^{d}	90.5 ± 0.03^{e}	90.9 ± 0.14^{a}
***Energy value* (KJ/kg)**	TME 419	Bucket	1487.8 ± 0.05^{d}	1451.8 ± 1.16^{ef}	1451.3 ± 0.90^{e}	1450.6 ±0.38^{ef}
		LDPE	1487.8 ± 0.05^{d}	1453.9 ± 1.38^{ef}	1451.0 ± 1.13^{ef}	1449.3 ± 0.28^{f}
		Paper bag	1487.8 ± 0.05^{d}	1448.9 ± 0.42^{f}	1454.8 ± 1.12^{ef}	1452.7 ± 0.57^{ef}
	UMUCASS 36	Bucket	1508 3 ± 0.34^{c}	1499.5 ± 0.79^{d}	1510.0 ± 0.37^{c}	1522.5 ± 0.73^{a}
		LDPE	1508 3 ± 0.34^{c}	1509.1 ± 0.34^{c}	1517.9 ± 0.15^{ab}	1521.7 ± 0.47^{c}
		Paper bag	1508 3 ± 0.34^{c}	1509.6 ± 1.20^{b}	1516.6 ± 0.44^{ab}	1521.7 ± 0.56^{c}

Values are given as means of triplicate determinations ± standard deviation.

Different letters in columns are significantly different ($p < 0.05$).

A significant decrease in ash content of the flour was observed in all the packaging materials across the storage durations. The decrease was in the order, paper bag (1.1 ± 0.13%) > LDPE (1.2 ± 0.001%) > plastic bucket (1.3 ± 0.07%) at the end of 12 week storage. However, the effect of packaging and duration was not significant in ash content for both flour cultivars (p = 0.84). Similar result was reported in the study on the effects of different packaging materials on the ash content of garri (Ogiehor & Ikenebomeh, 2006). Ogiehor and Ikenebomeh (2006) observed the ash content of garri to range from 0.2 to 1.8% after 24 weeks of storage in different package types. The observed results are in the range 1.45 ± 0.31 to 2.35 ± 0.35% reported for other cassava flour cultivars (Eleazu & Eleazu, 2012).

Protein and fat contents slightly decreased during the storage period in both cultivars and were consistent in all the packaging materials. Flour packaged in plastic bucket had the highest percentage protein content at the end of 12 weeks storage, which was as a result of effect of moisture as well as the activities of the microbes in the package. The percentage decrease in protein (dry basis) was between 3.0 ± 0.05 - 2.7 ± 0.001% in cv. 'TME 419' and between 2.0 ± 0.07 - 1.3 ± 0.001% in cv. 'UMUCASS 36'. Significant difference was observed between the packaging materials used and in both cassava cultivars. Similarly, fat content of cassava flour decreased across the storage duration and in all package type in the trend LDPE (0.8 ± 0.001%) > paper bag (0.08 ± 0.001%) > bucket (1.2 ± 0.001%), which was consistent in both cultivars. This decrease could be attributed to the possible high proteolytic and lipolytic activities of the corresponding enzymes which in turn led to the loss in the nutrients (Agrahar-Murugkar & Jha, 2011). Previous research on the storage of other flour products also reported decrease in protein and fat contents (Ogiehor & Ikenebomeh, 2006; Agrahar-Murugkar & Jha, 2011). In addition, the decrease could also be due to the microbial activities inside the different packaging types during storage which catalysed the release of organic acids. Similar findings have been reported on garri (Ogiehor & Ikenebomeh, 2006). However, cultivar differences had significant impact in both fat and protein contents of flour, but the interaction between treatments showed no significance in the protein content ($p > 0.05$).

Cassava flour of the two cultivars showed high carbohydrate and dry matter contents of flour in all the packaging materials during the 12 weeks storage period. The highest CHO was observed in flour packed in paper bag for both cultivars (83.9 ± 0.14) for 'TME 419' and 86.4 ± 0.14%) for 'UMUCASS 36' at 23 °C, 60% RH storage condition. Flour in plastic bucket had the lowest CHO value (83.2 ± 0.10 and 85.7 ± 0.18%) for 'TME 419' and 'UMUCASS 36', respectively. This was in the range reported for percentage CHO of cassava flour cultivars (92b/00061, 95/0289, 92/0057, 96/1632, 98/0505, 97/2205, TME 419 and 92/0326) in Nigeria (84.1 ± 0.06-85.4 ± 0.07%) (Nwabueze & Anoruoh, 2009).The dry matter content starting from the zero week ranged from 88.8 ± 0.11 to 89.0 ± 0.02% in cv. 'TME 419' and from 90.2 ± 0.15 to 90.9 ± 0.14% in cv. 'UMUCASS 36'. A slight increase was noticed in the dry matter in both flour cultivars at the end of the 12 weeks storage period in all the packaging material. This could be attributed to the loss of moisture content in the flour. However, paper bag maintained the highest dry matter content of flour (89.0 ± 0.06 and 90.9 ± 0.14%) throughout the storage period. Change in CHO was observed to have a strong negative correlation with percentage MC ($r = -0.9631$). This finding suggests that at lower moisture content, percentage CHO could increase depending on the percentage of all other components (protein, fat, and ash). This corresponds with the literature on the effect of different packaging materials on garri quality (Ogiehor & Ikenebomeh, 2006). The authors observed that package with the highest moisture content gave a corresponding decrease in percentage CHO and vice-versa. Therefore, cassava flour packaged in paper bags best maintained dry matter content, CHO, as well as the moisture content stability, which is a clear indication of longer shelf stability with paper bag storage.

Effects of packaging on physicochemical properties of cassava flour

Colour quality

The physicochemical properties of cassava flour after 12 weeks storage in 3 different packaging materials is presented in (Table 2). Packaging materials had significant impact on the colour attributes of both cassava flour cultivars ($p < 0.05$). Lightness *L** value usually presented on the scale of 0 - 100 (black to white) shows an approximate measurement of luminosity, upon which a distinction between black and white coloration in food products is made (Granato & Masson, 2010). The lightness values (90.2 ± 0.73 and 87.7 ± 1.68) for both cultivars were significantly high irrespective of the length of storage. Although 'TME 419' cassava flour had higher values (90.2 ± 0.73) before storage, there was a slight drop in lightness after 12 weeks storage in all the packages. In contrast, 'UMUCASS 36' flour which is the yellow cultivars showed an increase in lightness values across the packaging materials, this observation could be due to the loss in the yellow pigment in the flour. However, the higher influence in lightness was observed in flour packed in paper bag for both cultivars after 12 weeks of storage.

The total colour difference increased with the increase in storage period and was consistent in both cultivars and packaging materials. Colour difference (ΔE^*) at the end of 12 weeks storage was highest in flour packed in paper bag, with 3.6 ± 0.35 for 'TME 419' and 4.9 ± 0.22 for 'UMUCASS 36'. Cassava flour packed in plastic bucket had the lowest ΔE^* value of 3.2 ± 0.42 for cultivar 'TME 419' and 4.1 ± 0.87 for 'UMUCASS 36' (Table 2). This shows that the rate of change in colour of flour is a function of the packaging materials used for storage. Thus package and cultivar effects were significantly different ($p < 0.05$) but the effects of package and storage duration were not significant ($p > 0.05$). Consequently, the increase in ΔE^* resulted to a decrease in whiteness (WI) of flour for 'TME 419' and a decrease in yellowness (YI) for 'UMUCASS 36'. This observation correlates with the literature on the effect of drying on the whiteness of yam flour (Hsu *et al.*, 2003). The authors reported decrease in WI of white yam flour, which they attributed to the drying method (sun-drying) employed during processing. Therefore, the colour variations observed in the cassava flour during this study could be due to the length of storage, cultivar differences and the drying method since similar methods of drying were used for both cultivars.

In addition, package types had an impact on changes in the whiteness indices of flour (Table 2). Cassava flour stored in paper bag had 1.6% loss in whiteness for 'TME 419' while the loss in yellowness in 'UMUCASS 36' flour was up to 32% in flour packed in paper bags giving about 8 times loss compared to 'TME 419' flour. This clearly showed that the rate of degradation in colour was higher in the yellow cultivar flour than in the white flour. This could be due to the oxidation of carotenoid pigment in the yellow cultivar. However, the interaction of package types and storage duration had no significant impact ($p > 0.05$) on the observed changes in colour difference of both cassava flour cultivars.

Table 2 Changes in the physicochemical properties of cassava flour in different packaging materials at the end of 12 weeks of storage under ambient condition

		Colour parameter					
Cultivar	**Packaging Materials**	***L****	**ΔE***	**WI**	**% Δ in WI**	**YI**	**% Δ in YI**
TME 419	**Initial**	90.2 ± 0.73^{a}	-	85.1 ± 0.02^{a}	-	17.8 ± 0.86^{c}	-
	Plastic bucket	88.8 ± 0.94^{b}	3.2 ± 0.42^{dc}	84.5 ± 0.26^{b}	0.7	15.8 ± 0.14^{d}	11
	LDPE	88.5 ± 0.98^{b}	3.2 ± 0.36^{cd}	84.5 ± 0.65^{b}	0.7	15.6 ± 0.16^{d}	12.4
	brown paper bag	86.1 ± 0.94^{d}	3.6 ± 0.35^{c}	83.7 ± 0.91^{bcd}	1.6	15.4 ± 0.05^{d}	13.5
UMUCASS 36	**Initial**	87.7 ± 1.68^{cb}	-	78.8 ± 1.86^{f}		28.2 ± 2.31^{a}	
	Plastic bucket	87.9 ± 0.80^{cb}	4.1 ± 0.87^{ab}	82.6 ± 0.95^{cde}	-4.8	20.3 ± 1.09^{b}	28
	LDPE	88.4 ± 0.71^{b}	4.3 ± 0.40^{b}	83.4 ± 1.84^{bcd}	-5.8	20.1 ± 1.29^{b}	28.7
	brown paper bag	89.6 ± 0.13^{b}	4.9 ± 0.22^{a}	83.9 ± 1.41^{bcd}	-6.5	18.9 ± 1.69^{bc}	32

Effects	**p (ΔE)**	**p (WI)**	**p (YI)**
Cultivars	0.000000	0.000402	0.000000
Packaging	0.000172	0.000763	0.000107
Duration	0.000000	0.000015	0.000000
Cultivars*Packaging	0.823257	0.000012	0.003604
Cultivars*duration	0.026321	0.001845	0.000000
Packaging*duration	0.464205	0.220810	0.808941
Cultivars*Packaging*duration	0.767676	0.085033	0.696285

The values are given as means of triplicate values ± standard deviation.

Different letters in a column are significantly different ($p < 0.05$).

Change in pH

The pH of the flour of both cassava cultivars before packaging and storage ranged from 5.9 to 5.7. The 'TME 419' was higher (5.76 ± 0.02) compared to 'UMUCASS 36' flour (5.54 ± 0.05). During the storage period in the different package types, a slight increase was observed. The highest increase was noted in flour packed in paper bag from 5.76 ± 0.02- 6.21 ± 0.14 in 'TME 419'. The trend of increase was similar to both cultivars. This could be attributed to the loss of moisture content during storage which consequently gave rise to the increase in pH concentrations. However, value before and after storage was within the range (4.99 - 8.15) reported for different cassava cultivars (Aryee *et al*., 2006). The values were ideal for composite flour used in baking as research had proven that acidity in flour limits the substitution level of flour while near neutral pH (7) contributes in improving the quality of flour (Aryee *et al.*, 2006). The interaction between cultivars and duration had significant effects on the pH value of the cassava flour (Table 3). However, package types and duration had no significant effect but package alone was significantly different ($p < 0.05$). From this result it clearly shows that duration is a major factor influencing pH concentration during flour storage.

Table 3 Effects of package type and storage duration on pH of cassava flour cultivars during storage.

		Storage duration (weeks)			
Cultivars	**Package**	**0**	**4**	**8**	**12**
TME 419	bucket	$5.76 \pm 0.02^{c}_{A}$	$5.57 \pm 0.19^{d}_{B}$	$5.96 \pm 0.14^{ab}_{AB}$	$6.04 \pm 0.02^{a}_{B}$
	LDPE	$5.76 \pm 0.02^{b}_{A}$	$5.65 \pm 0.18^{c}_{A}$	$5.99 \pm 0.03^{a}_{B}$	$5.99 \pm 0.06^{a}_{B}$
	Paper	$5.76 \pm 0.02^{c}_{A}$	$5.75 \pm 0.27^{c}_{A}$	$6.12 \pm 0.10^{ab}_{A}$	$6.21 \pm 0.14^{a}_{A}$
UMUCASS 36	bucket	$5.54 \pm 0.05^{c}_{B}$	$5.62 \pm 0.10^{bc}_{A}$	$5.65 \pm 0.07^{bc}_{D}$	$5.82 \pm 0.04^{a}_{C}$
	LDPE	$5.54 \pm 0.05^{c}_{B}$	$5.58 \pm 0.18^{c}_{B}$	$5.80 \pm 0.06^{a}_{C}$	$5.72 \pm 0.01^{b}_{D}$
	Paper	$5.54 \pm 0.05^{d}_{B}$	$5.68 \pm 0.03^{c}_{B}$	$5.71 \pm 0.14^{b}_{DC}$	$6.03 \pm 0.01^{a}_{B}$

The values are given as means of triplicate determinations ± standard deviation.
Similar superscript letters in rows are not significant across duration, and similar subscripts letters in a column are not significant across package types and cultivars ($p > 0.05$).

Hydrogen cyanide (HCN) *concentration*

The concentration of cyanide in cassava root and products is a major concern limiting the utilisation of minimally processed cassava root in the food industries. Although, processing helps to detoxify the root, some consumers are still in doubt of the use of cassava produce

(Orjiekwe *et al*., 2013). From this study the concentration of cyanide retained in the flour after processing was between 3.9 ± 0.06 and 4.9 ± 0.21 µg/ mL for 'TME 419' and 'UMUCASS 36' cultivars respectively. The yellow root (UMUCASS 36) had higher HCN than 'TME 419'. This result supports previous report of higher cyanide content in yellow cultivars as they are characterised with sour or bitter taste (Eleazu & Eleazu, 2012). Cassava roots are classified based on bitter and sweet taste and those with bitter taste have been reported to have higher cyanide level (Chiwona-Karltun *et al.*, 2004; Charles *et al.*, 2005). The higher content of HCN in the flour of 'UMUCASS 36' cultivar could be attributed to the sour taste attribute of the root, although there is no scientific justification to this yet but since acids are characterised with sour taste, it is speculated that the HCN in 'UMUCASS 36' was higher since the bitter type of cassava is characterised with high HCN. However values for both cultivars were below the safe level (50 µg/ mL) for HCN in food (Akiyama *et al.*, 2006). Furthermore, pH values of both flour cultivars could have contributed to the variation in the HCN content. This was established in the study on the effect of pH on HCN retention (Cumbana *et al.*, 2007). The authors in their research observed a minimal retention of cyanide at pH range 6.3 to 6.7 due to the increased rate of hydrolysis of linamarin to acetone cyanohydrins. The pH less than 6 led to more retention of cyanide in the flour due to decreased rate of linamarin breakdown.

At the end of 12 weeks storage, a slight decrease in HCN was noticed in all the package types and was consistent in both cultivars. The 'TME 419' cultivar decreased from 3.9 µg/ mL to 3.3 µg/ mL after storage; this implies that storage duration could also facilitate in the breakdown of the cyanogenic compounds in the flour. The observation from this study thus creates an assurance of the safety of cassava flour and for its use in the food industries. No significant effect was observed in the different packages across storage duration (Fig. 5), but the effects of cultivar differences and storage duration was significantly different ($p < 0.05$). The interaction between all the treatments however was not significant.

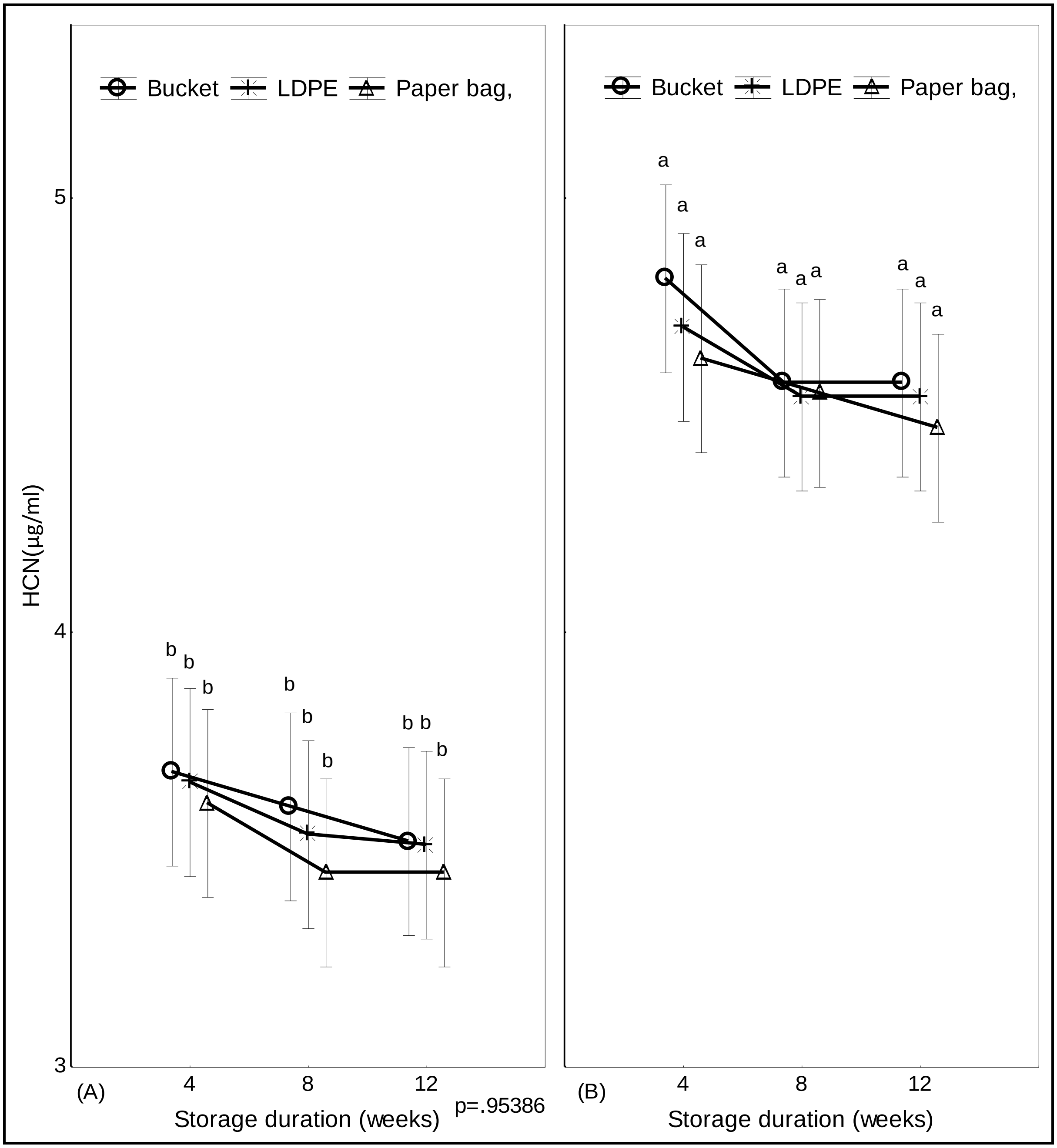

Figure 5 Effects of package and duration on the HCN concentration. (A) 'TME 419', (B) 'UMUCASS 36' Different superscript letters indicate significant differences ($p < 0.05$) between data points

Total carotenoids content

Carotenoid concentration decreased with the storage duration in all the packaging materials used. Both cultivars followed similar trends of decrease in the order of brown paper < LDPE <

plastic bucket. Flour packed in plastic bucket had the least percentage loss in total carotenoid from 1.84 ± 0.10 to 1.01 ± 0.03 mg/ 100g giving about 8% loss for cultivars 'TME 419' and from 2.5 ± 0.09 to 1.99 ± 0.01mg/ 100g giving 24% loss in carotenoid for 'UMUCASS 36'. Cultivar differences and storage duration had a significant effect on total carotenoid level in cassava flour ($p < 0.05$), while the packaging type had no significant impact on the observed change in carotenoid content ($p > 0.05$). Degradation of carotenoid content during processing and storage is one major challenge associated with maintaining the health benefits of high carotenoid products (Krinsky & Johnson, 2005). Reduction in total carotenoid content observed from this study was experienced in two ways. First was during processing of flour which includes grating/chipping, sun-drying, and milling. These processing methods have been reported to be major constituting factors exacerbating carotenoid degradation in cassava flour (Chávez *et al.*, 2007). Oliveria *et al*. (2010), describe the effect of heat exposure during flour processing as a catalyst to oxidation thus leading to carotenoid degradation. The authors also observed a continuous decrease (6.07 ± 0.01 - 0.16 ± 0.09) in carotenoid concentration during storage of cassava flour and recording a percentage loss of about 86% on the 19th day of storage. These investigations agree with the second means of carotenoid reduction trend observed from this study and therefore justify that carotenoid retention is a function of the length of storage such that the longer the storage period the higher the percentage loss.

Furthermore, from this study, it was observed that the increase in total colour difference (ΔE^*) had significant impact in carotenoid retention of flour from both cassava cultivars (Fig. 6a and 6b). A strong negative correlation ($r = -0.8280$) was observed between them, thus increase in total colour difference resulted to a decrease in carotenoid content. This is because carotenoids are denoted with a yellow or orange colour pigment and contributes to the flour colour. Hence, the degradation of carotenoid content would result in colour change (Oliveira *et al.*, 2010). Carotenoid pigments as provitamin A is associated with good human vision. The pigment accumulates in the retina and helps to prevent retinal degeneration and ageing (Krinsky & Johnson, 2005). Therefore, it is important that the level or rate of carotenoid degradation in cassava flour be minimized with an effective packaging system and optimum storage condition.

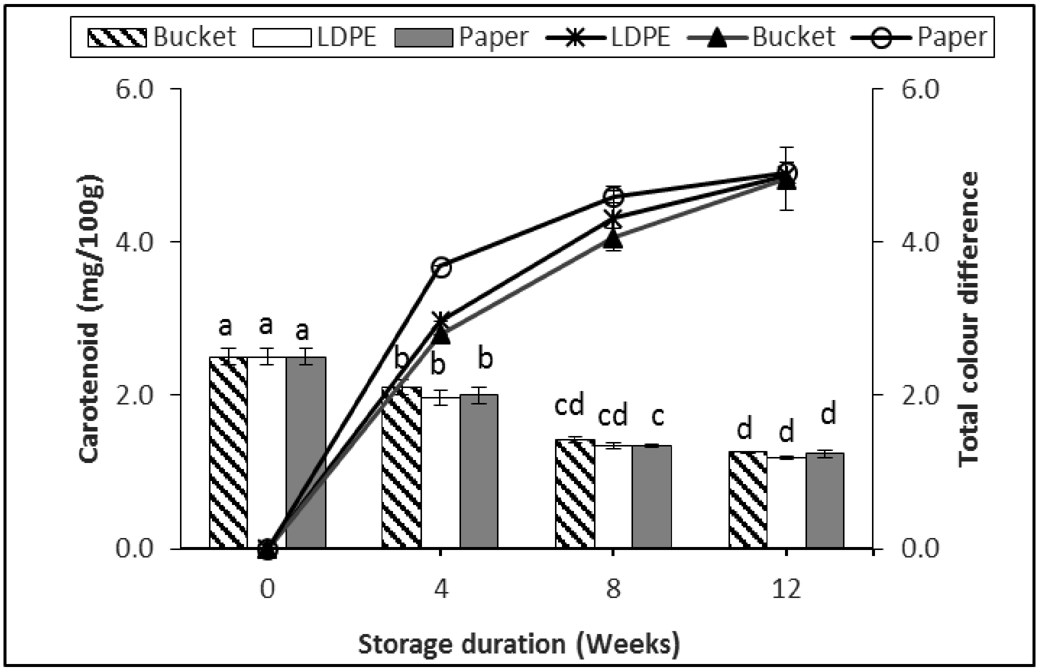

Figure 6a Changes in total colour difference and carotenoid concentration of cassava flour cultivar 'UMUCASS 36' during storage. Similar superscript letters are not significantly different between data points ($p > 0.05$). Line graphs show the total clour different, bars show the carotenoid concentration

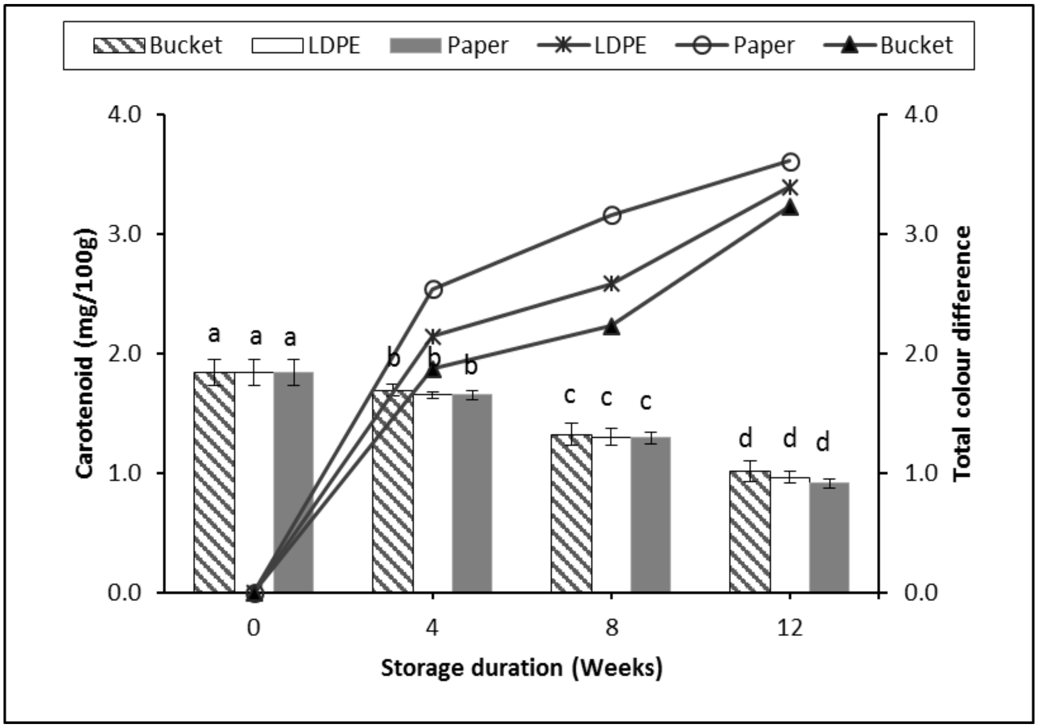

Figure 6b Changes in total colour difference and carotenoid concentration of cassava flour cultivar 'TME 419' during storage. Data point letters that are similar are not significantly different ($p > 0.05$). Line graphs show the total clour different, bars show the carotenoid concentration.

Microbial quality and water activity of cassava flour

Microbial load of cassava flour stored for 12 weeks in different packaging materials under ambient condition (23 °C, 60% RH) was influenced by moisture content and water activity level of the flour in this study. Results from this experiment showed a slight and progressive decrease in total plates count for aerobic mesophilic bacteria and fungi in all the packaging materials. This could be due to the decrease in moisture content during the storage period. Similar observation was reported in previous study on garri stored in different packaging materials under tropical market conditions (Ogugbue & Gloria, 2011). The result of Ogugbue and Gloria (2011) corroborate with the result in this present research. The authors observed increase in moisture content (18.7%) in exposed garri samples resulting to a corresponding increase in microbial count (5.7 ± 0.3 log cfu/ g) in their garri samples. Therefore, decrease in microbial count was consistent as moisture content decreased from the baseline (12.0 ± 0.11%). Similarly, the variation in water activity had significant effect on the microbial load of the stored cassava flour (Fig. 7a and 7b). The decline in a_w level of both flour cultivars (0.58 to 0.54) for 'TME 419' and (0.56 to 0.54) in 'UMUCASS 36' led to a slight reduction in the microbial count and was significant across the storage duration.

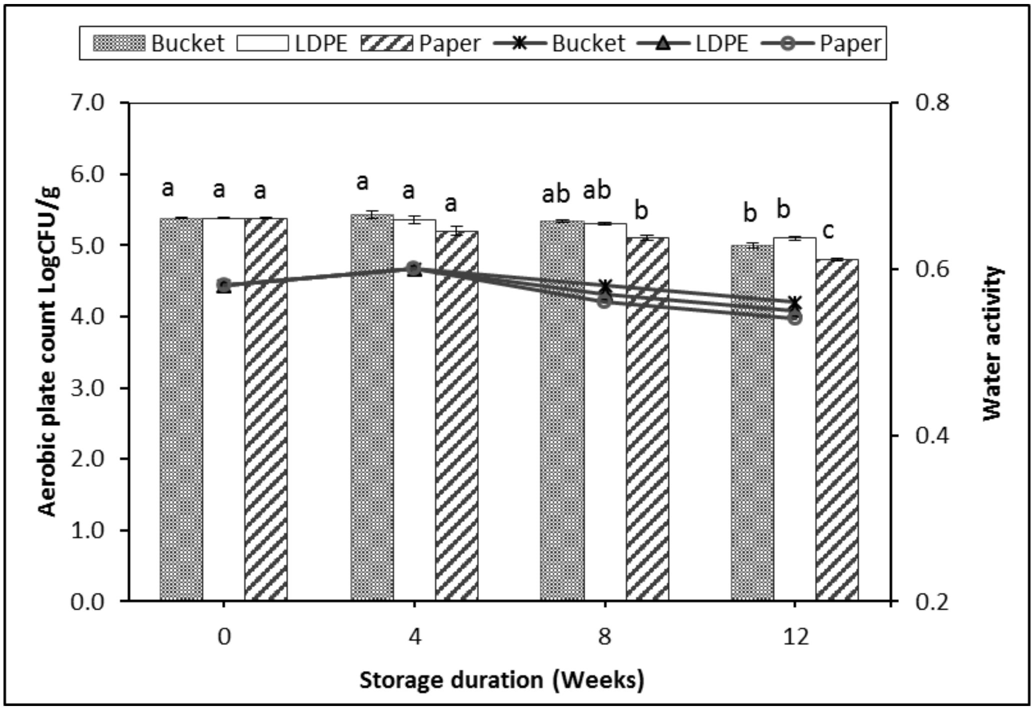

Figure 7a Changes in water activity on the aerobic plate count of 'TME 419' cassava flour stored under ambient conditions. Different superscript letters show significant differences ($p < 0.05$) between data points. Line graphs show the water activity and the bars show aerobic plate count.

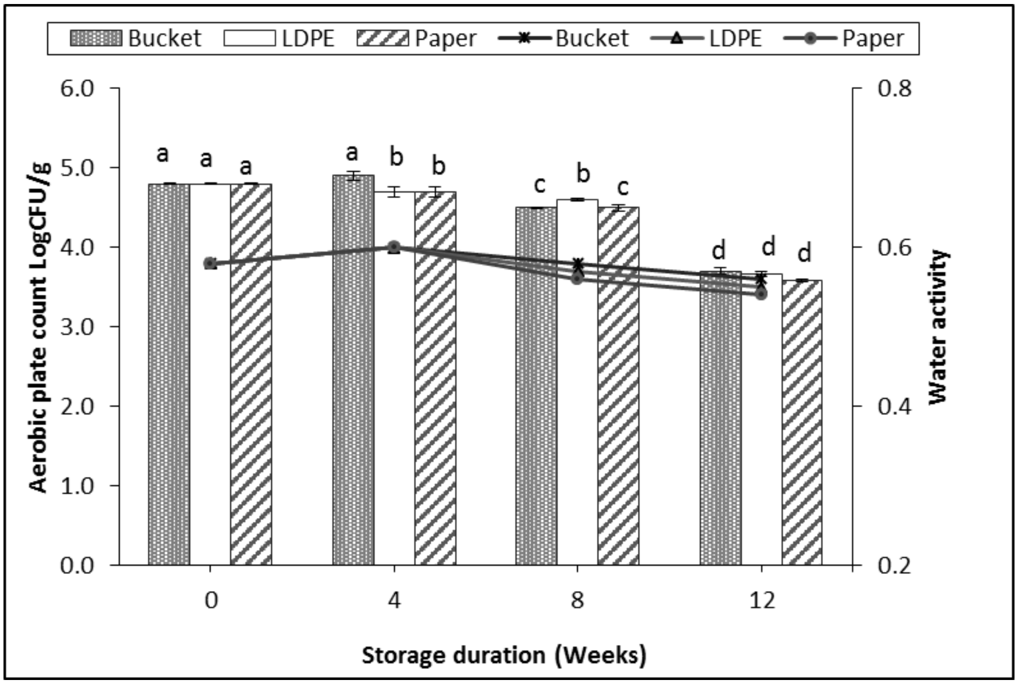

Figure 7b. Changes in water activity on the aerobic plate count of 'UMUCASS' 36 cassava flour stored at 23 °C. Different superscript letters show significant differences (*p* < 0.05) between data points. Line graphs show the water activity and the bars show aerobic plate count.

In addition, Butt *et al*. (2004) evaluated the effect of moisture and packaging on the shelf-life of wheat flour and noted that flour packed in paper bag which had the lowest moisture content showed higher shelf-life stability of wheat flour. This was consistent with the report in this study. The lowest counts (3.39 ± 0.02 log cfu/ g) was observed in cassava flour stored in paper bag which also had the lowest moisture content and water activity level after the 12 weeks storage period. However, the influence of packaging material and duration gave no significant effect at p > 0.05 but the paper bag could be assumed to have better shelf-life stability than the other materials. Bacterial counts decreased from 5.4 ± 0.01 log cfu/ g on week zero to 5.0 ± 0.03 log cfu/ g at the end of 12 weeks storage in 'TME 419', while 'UMUCASS 36' decreased from 4.7 ± 0.06 to 3.4 ± 0.07 log cfu/ g. Similar trends of decrease was observed for fungi growth (Fig. 8a and 8b) and were consistent in both cassava flour cultivars. The TME 419 cv. decreased from 5.3 ± 0.03 - 4.3 ± 0.05 log cfu/ g and 'UMUCASS 36' decreased from 4.8 ± 0.03 - 3.4 ± 0.05 log cfu/ g. However, aerobic bacteria counts were higher during the storage period than the fungi. Being that fungi thrive more in lower water activity level, the count for yeast and mould should have been higher than the bacterial count.

This observation could be as a result of different factors such as oxygen and pH level of the flour other than water activity. An update on food mycotoxins reported that yeasts and moulds can thrive better in low pH (less than 4) of food products (Murphy *et al.*, 2006). A study on maize and wheat flour confirms the low fungi count with pH ranged from 5.0 to 6.5 (close to neutral) (Victor *et al.*, 2013). The report of the authors agrees with the findings in this research and since the pH of the flour ranges from 5.54 ± 0.05 – 6.21 ± 0.14, same reason is adopted for why the lower yeast and mould than bacterial counts recorded during the 12 weeks storage. In addition, the variation in the microbial count could also be attributed to the permeability attribute of the package types to gases and moisture. Paper bag is more permeable to moisture compared to the plastic bucket and LDPE. Studies have also shown that some packaging films are more permeable than others and can allow the free passage of the atmospheric gases, water vapour, oxygen and other gases (Butt *et al.*, 2004; Ogiehor & Ikenebomeh, 2006). While these gases are trapped in some materials and they contribute to increasing the moisture content hence increasing the microbial load in such packaging materials. Therefore, the higher counts observed in flour packed in plastic bucket (5.17 ± 0.01 log cfu/ g in cv. 'TME 419' and 3.42 ± 0.02 log cfu/ g in 'UMUCASS 46') could be the inability of the plastic bucket to readily permeate water vapour and other surrounding gases.

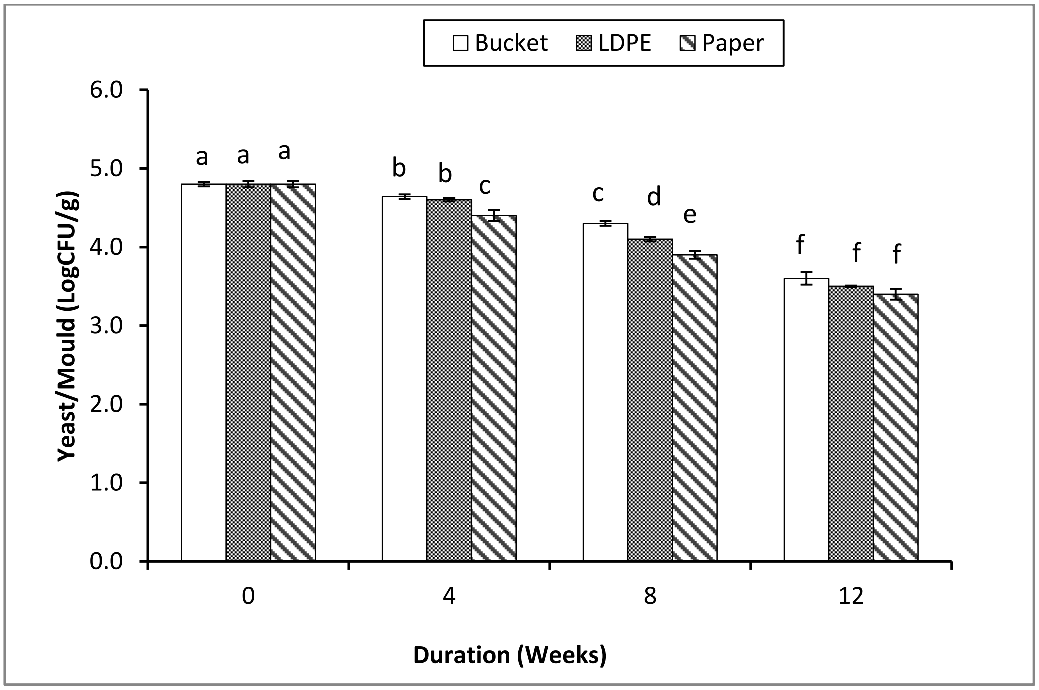

Figure 8a Effects of packaging materials and duration on yeast and mould count of 'UMUCASS 36' cassava flour stored under storage condition. Similar letters indicate no significantly different (*p* > 0.05) between data points.

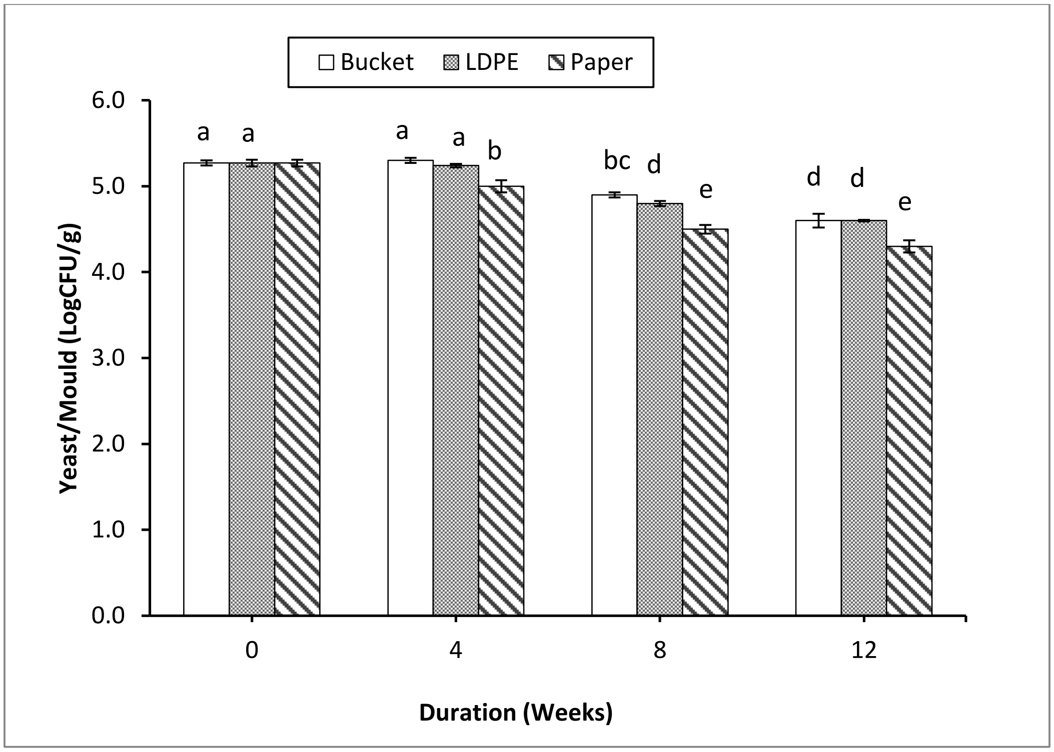

Figure 8b Effects of packaging materials and duration on yeast and mould count of 'TME 419' cassava flour stored under ambient condition. Different superscript letters indicate significant differences (*p* < 0.05) between data points.

The results from this study closely corroborate the assessment on the effects of different packaging materials on the shelf stability of garri kept for 24 weeks (Ogiehor & Ikenebomeh, 2006). The authors also recorded high microbial count in the product stored in plastic bucket during their storage period. These reports therefore give an indication that paper bags having the lowest microbial count after 12 weeks storage could best maintain microbial stability in flour during longer storage at 23 °C while maintaining other quality attributes of flour such as carbohydrate and dry matter.

Conclusion

The present study showed that packaging is an important factor to be considered in establishing the shelf-life stability of cassava flour. Packaging materials had significant impact on the moisture content of the two cassava flour cultivars studied, and this significantly influenced the microbial stability as well as the physicochemical properties during storage. Lower moisture content in flour corresponds with lower microbial growth and hence longer shelf-life stability and better quality attributes. Low moisture content also resulted in higher dry matter content, which is a good attribute for the food industries and cassava flour processing.

The influence of the different packaging materials on the physicochemical properties during the storage period was significant. Paper bags provided the lowest moisture content and the maximum stability of microbial quality of flour during the 12 weeks storage period. This could be attributed to the reported high permeability of paper material to moisture and atmospheric gases. Flour stored in paper bag showed the lowest HCN retention (3.4 ± 0.23 and 4.5 ± 0.13 µg/ mL) and had the highest pH level (6.2 ± 0.14 and 6.0 ± 0.01) for cvs 'TME 419' and 'UMUCASS 36' respectively. Cassava flour in plastic bucket had the lowest percentage loss of carotenoid with about 8% in 'TME 419' and 23% in 'UMUCASS 36' followed by flour in LDPE bags, and flour in paper bags had the highest loss. However, based on the overall quality and microbial safety, paper bag would be more effective in long term storage of cassava flour to maintain microbial stability and the quality attributes essential for the food industries. Therefore this information could be useful in packaging, storage, exporting and commercialisation of cassava flour and cassava-based products.

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CHAPTER 5

CHANGES IN FUNCTIONAL AND PASTING PROPERTIES AND MINERAL CONTENTS OF CASSAVA FLOUR (CVS. 'TME 419' AND 'UMUCASS 36') UNDER DIFFERENT PACKAGING AND STORAGE CONDITIONS

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Summary

The functional and pasting properties and mineral contents of the flour of two cassava cultivars (‘TME 419’ and ‘UMUCASS 36’) packaged in plastic buckets, LDPE bags and paper bags and stored under ambient (23 °C, 60% RH) and hot (38 °C, 60% RH) conditions were investigated for 12 weeks. Swelling power and solubility of ‘TME 419’ flour (12.1 ± 0.13% and 9.51 ± 1.59%) was significantly higher than that of ‘UMUCASS 36’ flour (10.1 ± 0.22%). The highest values in swelling power at the end of 12 weeks of storage were observed in cassava flour packed in paper bag and stored under higher condition. The peak, trough, final, breakdown and setback viscosities were higher in cv. ‘TME 419’ than cv. ‘UMUCASS 36’. Viscosities were generally influenced by the storage condition, packaging materials and varietal differences. Correlation analysis revealed a strong positive and significant correlation (r = 0.94, $p \leq 0.05$) between peak viscosity and breakdown viscosity. The shortest pasting time (3.9 ± 0.06 min) and lowest pasting temperature (84.9 ±0.36 °C) after 12 weeks was observed in the flour packed in paper bag under higher condition for both cultivars indicating lesser processing energy, time and therefore reduced cost of production.

Introduction

The use of high quality (unfermented) cassava flour as composite and non-gluten flour for bread making has recently been practised across West African countries. This innovation is due to some economic, social and health reasons noted in cassava flour such as diet for the celiac patients (Biagi *et al*., 2009). The prevalence of celiac disease (gluten intolerance) and the high cost of wheat flour, gave rise to the need of partial substitution of wheat flour with cassava flour and/or other gluten-free flour (Shittu *et al.*, 2009). Apart from using cassava flour as composite flour in bread making, it can be utilised for other purposes in the food industry such as the production of baby foods, noodles (Nwabueze & Anoruoh, 2011), other confectionery like doughnuts, biscuits, muffins, cakes and cookies (Eddy *et al.*, 2007; Gyedu-Akoto & Laryea, 2013), and in the textile industries (Moorthy, 2002). These uses of cassava flour are attributed to its distinctive functional properties which are desirable in industrial applications. Some of the functional properties are common with certain cassava cultivars such as low amylose or amylose-free (waxy cassava) (Sánchez *et al.*, 2010).

High amylose starch is associated with paste retrogradation, which is undesirable for many applications of starch paste as well as composite flour for baking purposes (Koehorst-van Putten *et al.*, 2012). On the other hand, low amylose content of some cassava flour cultivars offer the benefit of its uses in the formulation of frozen foods since the gels show little or no syneresis during storage even at low temperatures (20 °C) (Sánchez *et al.*, 2010). Similarly, these cultivars need no modification with expensive chemicals like hexamethylene diisocyanate derivatives and phenyl isocyanate to enhance stability because they form stable gels (Wilpiszewska & Spychaj, 2007). Based on this, there is on-going research focused on breeding of more cassava flour cultivars with waxy starch for industrial purposes (Zhao *et al.*, 2011).

Functional properties of cassava flour can be assessed based on their particle size distribution which is a reflection of the breaking phase of the intermolecular hydrogen bond holding the flour/starch granules (Shimelis *et al.*, 2006). For instance, the low swelling power in cassava flour is a clear indication of a strong binding force, high resistance to breaking during cooking and a stable gel at cooling phase (Shimelis *et al.*, 2006). On the other hand high swelling capacity can lead to shear when the flour is cooked in water. Solubility, which is the ability of solid particles to dissolve in aqueous solution usually water is also influenced by the granular size thus that smaller granular size results to lower solubility pattern (Singh *et al.*, 2003). Swelling capacity and solubility of flour and starch are directly correlated, in that increase in swelling power results to increased solubility pattern. The ability of food products to take up and retain water either by adsorption or absorption contributes enormously to the easy handling of dough during food preparation and baking (Doporto *et al.*, 2012). This hydration phase of the flour is expressed as the water binding capacity (WBC) and it is influenced by the extent of starch disintegration (Falade & Okafor, 2013). Pasting profile of cassava flour is essential for characterisation and its use in the food industries. It enables the food processors to identify how gelatinisation processes during cooking affect the texture and stability of the food products (Iwe & Agiriga, 2014). High viscous flour is useful in the production of jelly, food thickener and binder, while flours with low viscosity could be suitable for weaning food production (Tsakama *et al.*, 2010).

Despite the detailed reports on the functionality and pasting properties of cassava flour and other cassava products, there is little information on the changes in functionality of cassava flour during storage under different packing and storage conditions. Hence, there is need to examine the functional properties during storage as this could influenced cassava flour utilisation, large scale production, market value, and consumers' acceptability. The objective of this study was to investigate the changes in functional, pasting properties and mineral contents of the flour of two cassava cultivars packed in plastic buckets, LDPE bags and paper bags and stored at 23 and 38 ± 2°C, 60% relative humidity (RH) for 12 weeks duration. The outcome from this study would be valuable for commercialisation of cassava flour and informative for food processors in making choices on the cassava cultivars for a definite purpose. In addition, results from this study will serve as a guide in estimating the stability and cooking quality of cassava flour.

Materials and methods

Plant materials

Two cassava cultivars 'TME 419' a white cultivar and 'UMUCASS 36' newly released yellow root cultivar (Fig. 1), were used for this study. Both cultivars were harvested at 12 months after planting (commercial maturity) from National Root Crops Research Institute (NRCRI) Umudike (5º28′33"N 7º32′56"E), Abia state, Nigeria. The cultivars used in this study were selected based on their flour yield and desirable functional properties for food industries (Nwabueze & Anoruoh, 2009; Eleazu & Eleazu, 2012).

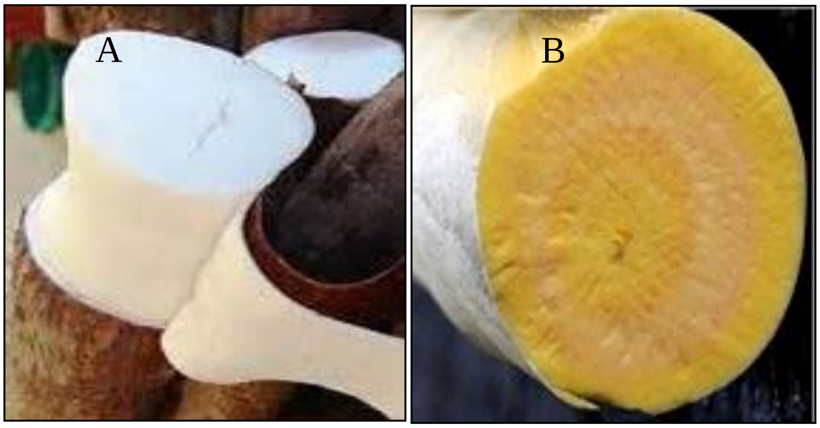

Figure 1 Cross sections of cassava root obtained from cultivars (A) 'TME 419' and (B) 'UMUCASS 36'

Cassava processing, packaging and storage

The freshly harvested cassava root were sorted, washed, peeled and re-washed with clean, running tap water. The cassava roots were then sliced into chips with an electric stainless steel chips making machines (YS QS400, Shandong, China). Chips were sun-dried for three days to approximately 13% final moisture content (MC). The dried cassava chips obtained from the two cultivars were then packaged separately in sterile polyethylene bags and transported to Department of Food Science, Stellenbosch University, South Africa. The chips were stored at room temperature prior to milling and further analysis. The chips were milled into flour using Cyclone Laboratory milling machine (Model 3100. Perten Instruments, Hagersten, Sweden) fitted with a sieve of 0.5 mm size. The milled cassava flour from both cultivars was analysed for functional, pasting properties and mineral contents before and during 12 weeks storage period at 4 weeks interval.

Cassava flour (200 g) freshly processed from both cultivars was packaged in the following packaging materials: Plastics bucket; low density polyethylene bag (LDPE) 50 micron; and brown paper bag and stored under ambient condition (23 ± 2 °C; 60% RH), and higher condition (38 ± 2 °C; 60% RH) for 12 weeks using environmental test chamber (MLR-351H, Sanyo, Japan). Samples were taken for analysis at 4 weeks interval. Functional, pasting properties and mineral contents were performed in triplicate samples on every sampling days.

Functional properties

Swelling power and solubility pattern

The swelling power and solubility of all the treatments of cassava flour were determined according to the method described by Crosbie (1991) with modifications. Swelling power is used to determine the hydration capacity of flour samples under specific temperature (Shimelis *et al.*, 2006). This was measured by calculating the weight of centrifuge swollen granules and dividing by the initial weight of flour used to make the paste. Cassava flour (1 g) was measured into a 50 mL graduated centrifuge tube and 40 mL distilled water was added to form a paste. The whole set up was stirred and incubated in a water bath (TMK 14R20, FMH instruments, India) at 95 °C. The suspension was stirred at interval of 30 min to keep the flour granules suspended. The tubes were then rapidly cooled to room temperature and centrifuges for 15 min at 2200 rpm to separate the gel and supernatant.

The aqueous supernatant was poured into a tarred dish and kept in force draft oven at 100 °C for 4 h to evaporate. Water solubility index was determined from the amount of dried solids recovered after evaporating the supernatant, and was expressed as grammes dried-solids per gramme of sample. The solubility and swelling power (SP) on dry basis were calculated with the following equations:

$$\text{Solubility } (\%) = \frac{\text{weight of soluble matter in supernatant}}{\text{weight of sample (g.dry basis)}} \times 100 \qquad (1)$$

$$\text{Swelling power } (\%) = \frac{\text{weight of swollen matter}}{(\text{weight of sample}) \times (100 - \text{solubility})} \times 100 \qquad (2)$$

Water binding capacity (WBC)

The water binding capacity of the different flour treatments were determined with little modifications following the method by Aryee *et al*. (2006). Approximately, 2 g of cassava cultivar flour from each treatment was suspended in 40 mL of distilled water in a 50 mL graduated centrifuge tube. The suspension was agitated for 1 h, centrifuged at 2200 rpm for 10 min and the free water was drained from the paste while the wet flour in the tube was weighed. The % WBC was calculated using the equation 3:

$$\text{WBC (\%)} = \frac{\text{Weight of paste after drain}}{\text{weight of sample}} \times 100 \qquad (3)$$

Starch determination

Total starch was determined enzymatically using the total starch assay kit (Megazyme International, Ireland) following the manufacturers manual. About 100 mg of cassava flour samples, wheat and corn (control starches) were weighed and washed with 80% ethanol. The samples were digested with thermo-stable α-amylase and amyloglucosidase and incubated at 50 °C for 30 min. The glucose released was determined using an enzymatic glucose assay reagent (GOPOD method), and the absorbance was measured with a UV-Vis spectrophotometer at 540 nm (Helios Omega UV-Vis Thermo Scientific, USA). The total starch was calculated using equation 5:

$$\text{Total starch \%} = \mathrm{A} \times \mathrm{F} \times 1000 \times \frac{1}{1000} \times \frac{100}{\mathrm{W}} \times \frac{162}{180} \qquad (4)$$

$$= \mathrm{A} \times \frac{\mathrm{F}}{\mathrm{W}} \times 90 \qquad (5)$$

where: A = absorbance against reagent blank; F = factors to convert absorbance values to µg glucose; W= weight of sample; 162/180 = factor to convert from free glucose to starch; and 100/W = conversion to 100mg test portion.

Determination of pasting properties

The pasting properties of cassava flour subjected to different storage conditions were determined using a Rapid Visco Analyzer (RVA) (Newport Scientific Pty. Ltd., Australia) according to Noda *et al.* (2004) method with minor modification. Cassava flour (2.8 g) from each package was weighed into a dry empty canister and 25 mL distilled water was added to make an 8% suspension. The solutions were mixed and the canister carefully fixed on the RVA instrument. The whole profile took 13.13 min, each flour was kept at 50 °C for 1 min before heating up to 95 °C at the rate of 12.2 °C per min and held for 2.5 min at 95 °C. The paste formed was then cooled to 50 °C at 11.8 °C per min and kept for 2 min at 50 °C. Peak viscosity, trough, breakdown, final viscosity, setback, peak time and pasting temperature were obtained from the pasting profile using thermocline software (Newport Scientific 1998), and the results were expressed in rapid visco unit (RVU).

Minerals determination

The minerals compositions of the cassava flour across the different treatments were analysed by Bemlab, Somerset West, South Africa. The following mineral contents were investigated: Nitrogen (N); phosphorus; (P); potassium (K); calcium (Ca); magnesium (Mg); sodium (Na); manganese (Mn); iron (Fe); copper (Cu); zinc (Zn); boron (B); and Nickel (Ni).

Statistical analysis

All experiments for cassava flour treatments were done in triplicates. Analysis of variance (ANOVA) was conducted on all data using Statistica software, (Version 12.0, Statistica, Statsoft, USA) to determine the effects of package types, storage conditions and duration. Significant differences were established at $p \leq 0.05$ according to Fisher LSD test and data obtained were reported as mean and ± standard deviations for each treatment.

Results and discussion

Functional properties

Swelling power and solubility pattern

Swelling power of flour is described as the ability of flour to absorb water and enlarge/expand under a particular temperature at a given time (Falade *et al.*, 2014). The hydration capacity of the flour enables measurement of the intermolecular force of starch granules. For instance, high swelling power implies that the starch granules have weak binding forces and high amylose content, while lower swelling power indicates a strong binding force (Shimelis *et al.*, 2006). In this study, the swelling power of both cassava flour cultivars ranged from 8.48 ± 0.55 to 12.11 ± 0.13% (Table 1). The 'TME 419' flour cultivar had significantly higher (12.11 ± 0.13%) swelling power (SP) than the 'UMUCASS 36' (10.07 ± 0.22%) after processing. The range of SP from this study agrees with the data obtained from other cassava cultivars (Aryee *et al.*, 2006). The authors also observed the influence of cultivar difference amongst 31 different cassava flour cultivars including TME 1, 91/0055, Tek Bankye, 92/0035, 94/0050 on the cassava flour SP. The higher SP in the 'TME 419' suggests a weaker associative binding force, while starch granules in the 'UMUCASS 36' flour had a more orderly and compact arrangement hence stronger force. Sanni *et al.* (2006) reported a higher swelling power for dried fufu flour resulting to a corresponding lesser binding force.

During the 12 weeks storage period a decrease in swelling power was observed and this was consistent across all the treatments. The lowest swelling power in both cultivars was observed in cassava flour packed in plastic bucket (8.48 ± 0.55%) for 'TME 419' and (8.2 ± 0.46%) for 'UMUCASS 36', followed by LDPE (9.10 ± 0.13% and 8.64 ± 0.10%) and paper bag had the highest SP (9.10 ± 0.13% and 8.64 ± 0.10%) for 'TME 419' and 'UMUCASS 36', respectively. The higher SP observed in flour packed in paper bag could be attributed to the increase in the dehydration capability of the flour as a result of the effect of storage duration. Sánchez *et al*. (2013) also reported a decline in swelling power in their study on the changes in extended shelf-life of cassava roots during storage in ambient temperature. The authors concluded that a decline in swelling power is consistent with low amylose or amylose-free cassava cultivars. In addition the reduced swelling power at the end of storage could be as a result of the formation of protein amylose complexes in the flour (Shimelis *et al.*, 2006; Sánchez *et al.*, 2013). The authors reported a lower swelling as well as solubility due to the formation of these complexes in beans flour. These protein complexes are stimulated during gelatinisation and they have been reported to cause a decline or restriction in swelling capacity (Shimelis *et al.*, 2006).

Storage temperature also had significant influence on the swelling power of the cassava flour cultivars. The highest values SP (8.72 ± 0.20 and 9.57 ± 0.20%) were recorded in 'UMUCASS 36' and 'TME 419' flour stored at 38 °C, respectively. However, the interaction of temperature and packaging had no significant effect ($p > 0.05$) on the swelling power. The low values of swelling power recorded for both cassava flour cultivars are attributes for highly restricted swelling which is commended for use in the food industries where stable gels are essential. This is because the paste shows shear resistance during cooking while high swelling power (≥ 30%) are indications of fragile and less stable starch which tend to breakdown upon cooking in water (Shimelis *et al.*, 2006).

Similarly, the solubility of cassava flour declined over time (Table 1). Solubility pattern is the ability of solid substances to dissolve in aqueous solution usually water (Falade & Okafor, 2013). Solubility could be influenced by the granular size. Singh *et al*. (2003) reported lower solubility in smaller granules of starch from botanical sources. The solubility ranged from 6.7 ± 0.27 to 9.5 ± 1.59% in 'TME 419' flour and from 6.1 ± 0.11 to 9.2 ± 0.24% in 'UMUCASS 36'.

The lowest value of solubility 6.14 ± 0.11% was noticed in ‘UMUCASS 36’ flour packed in plastic bucket and stored under ambient condition, while the highest 7.0 ± 0.11% was recorded in ‘TME 419’ flour cultivar stored under higher condition in paper bag. The obtained values were within the range (6.2 - 13.6%) reported for cassava, sweet potato and cocoyam starches (Ikegwu *et al.*, 2009; Abegunde *et al.*, 2013; Falade & Okafor, 2013). The low solubility observed could indicate smaller starch granules. A study on potato starch affirmed that low solubility is a function of granular size (Kaur *et al.*, 2002). Swelling power and solubility showed a positive correlation (r = 0.64) in this study, such that decrease in swelling power led to a decrease in solubility pattern. This corroborates the observation in the study of the physicochemical properties of cocoyam starch (Falade & Okafor, 2013).

***Water binding capacity* (WBC)**

The water binding (WBC) of the two cassava flour cultivars after processing ranged from 128 ± 0.14 in ‘TME 419’ to 132.82 ± 1.42 in ‘UMUCASS 36’(Table 1). The higher water binding value in ‘UMUCASS 36’ could be ascribed to the nature of the starch. Higher water binding is an essential quality in product development as it allows easy handling of dough during baking processes. During the 12 weeks storage, the WBC of the flour from both cassava cultivars was observed to increase with about 4.2% in ‘TME 419’ and 4.1% in ‘UMUCASS 36’. The highest values at the end of 12 weeks storage were noted in the flour packed in LDPE bags for ‘TME 419’ and in flour packed in paper bag for ‘UMUCASS 36’. However, package types, cultivars and temperature effects did not show significant difference but cultivars and storage duration were significant. This observation therefore explains that cultivar differences are major factors influencing the water binding capacity of cassava flour. The values obtained fall within the range (128.7 ± 0.40 - 201.9 ± 0.20) reported for other cassava flour cultivars (Aryee *et al.*, 2006).

The WBC values obtained from this study across all treatments were higher than the value (110 ± 2.1) reported by Doporto *et al*. (2010), but relatively lower than that of water yam-cassava flour (Babajide & Olowe, 2012). Babajide and Olowe (2012), evaluated the water binding capacity of yam-cassava flour at different ratios, the range (278.51 ± 12.85-421 ± 28.63) was higher than the observation from this study. The reason for the variations in WBC could be attributed to some factors such as cultivars, starch granular size, shape and binding force of granules (Aryee *et al.*, 2006; Babajide & Olowe, 2012).

Table 1 Effects of storage conditions and packaging types on the functional properties of two cassava flour cultivars

			Package type					
Composition (%)	**Cultivar**	**Duration (Weeks)**	**Bucket**		**LDPE**		**Paper bag**	
			23 °C	**38 °C**	**23 °C**	**38 °C**	**23 °C**	**38 °C**
Swelling power	**TME 419**	0	12.11 ± 0.13^{a}	12.11 ± 0.13^{a}	12.11 ± 0.13^{a}	12.11 ± 0.13^{a}	12.11 ± 0.13^{a}	12.11 ± 0.13^{a}
		4	11.07 ± 0.29^{b}	11.49 ± 0.25^{b}	11.17 ± 0.61^{b}	11.59 ± 1.07^{b}	11.46 ± 0.03^{b}	11.80 ± 0.79^{b}
		8	10.05 ± 0.61^{c}	10.62 ± 0.47^{c}	10.00 ± 0.11^{c}	10.68 ± 0.35^{c}	10.31± 0.41^{d}	10.81 ± 0.23^{c}
		12	8.48 ± 0.55^{f}	9.36 ± 0.40^{e}	9.10 ± 0.13^{e}	9.47 ± 0.64^{e}	9.32 ± 0.41^{e}	9.57 ± 0.20^{f}
	UMUCASS 36	0	10.07 ± 0.22^{c}	10.07 ± 0.22^{d}	10.07 ± 0.22^{c}	10.07 ± 0.22^{d}	10.07 ± 0.28^{d}	10.07 ± 0.22^{d}
		4	9.64 ± 0.08^{d}	9.73 ± 0.47^{e}	9.52 ± 0.25^{d}	9.71 ± 0.39^{e}	9.64± 0.43^{e}	9.81 ± 0.28^{e}
		8	9.49 ± 0.14^{e}	9.61 ± 0.25^{e}	9.15 ± 0.22^{e}	9.50 ± 0.15^{e}	9.53± 0.08 ^{e}	9.87 ± 0.13^{e}
		12	8.21 ± 0.46^{f}	8.32 ± 0.11^{f}	8.64 ± 0.10^{f}	8.66 ± 0.30^{f}	8.68 ± 0.26^{f}	8.72 ± 0.20^{g}
Solubility	**TME 419**	0	9.51 ± 1.59^{a}	9.51 ± 1.59^{a}	9.51 ± 1.59^{a}	9.51 ± 1.59^{a}	9.51 ± 1.59^{a}	9.51 ± 1.59^{a}
		4	8.57 ± 0.18^{dc}	8.65 ± 0.10^{d}	8.65 ± 0.40^{c}	8.77 ± 0.13^{c}	8.64 ± 0.18^{c}	8.83 ± 0.37^{c}
		8	8.05 ± 0.57^{d}	8.15 ± 0.24^{d}	7.93 ± 0.26^{d}	8.11 ± 0.10^{d}	8.10 ± 0.19^{d}	8.17 ± 0.14^{d}
		12	6.77 ± 0.40^{e}	6.85±0.10^{f}	6.66 ± 0.27^{e}	6.80 ± 0.13^{e}	7.04 ± 0.11^{e}	7.09 ± 0.12^{fe}
	UMUCASS 36	0	9.15 ± 0.24^{b}	9.15 ± 0.24^{b}	9.15 ± 0.24^{b}	9.15 ± 0.24^{b}	9.15 ± 0.24^{b}	9.15 ± 0.24^{b}
		4	8.75 ± 0.09^{c}	8.93 ± 0.33^{c}	8.43 ± 1.20^{c}	8.82 ± 0.70^{c}	8.77 ± 0.15^{c}	8.84 ± 0.50^{c}
		8	8.71 ± 0.40^{c}	8.00 ± 0.02^{e}	8.07 ± 1.28^{c}	8.13 ± 0.33^{d}	7.95 ± 0.31^{d}	8.20 ± 0.26^{d}
		12	6.14 ± 0.11^{f}	6.25 ± 0.19^{gf}	6.37 ± 0.24^{e}	6.95 ± 0.10^{e}	6.83 ± 0.10^{f}	7.27 ± 0.07^{e}
WBC	**TME 419**	0	128.81 ± 0.14^{f}	128.81 ± 0.14^{g}	128.81 ± 0.14^{g}	128.81 ± 0.14^{f}	128.81 ± 0.14^{f}	128.81± 0.14^{f}
		4	128.86 ± 1.97^{f}	130.13 ± 2.01^{ef}	129.46 ± 0.94^{f}	130.37 ± 1.66^{ef}	129.25 ± 0.83^{e}	130.84 ± 0.76^{e}
		8	130.88 ± 0.87^{e}	131.30 ± 0.29^{e}	131.18 ± 0.47^{e}	131.82 ± 0.24^{e}	132.01 ±0.71^{d}	132.87 ± 0.29^{d}
		12	133.81 ± 0.50^{c}	134.32 ± 0.51^{c}	133.90 ± 0.44^{c}	134.34 ± 0.56^{cd}	134.01± 0.16^{c}	134.12 ± 0.35^{c}
	UMUCASS 36	0	132.82± 1.42^{d}	132.82 ± 1.42^{d}	132.82 ± 1.42^{d}	132.82 ± 1.42^{d}	132.82 ± 1.42^{d}	132.82 ± 1.42
		4	133.03 ± 2.43^{cb}	134.02 ± 0.28^{c}	133.51 ± 0.55^{c}	134.00± 0.21^{d}	134.13± 0.28^{c}	134.15 ± 0.12^{c}
		8	135.00 ± 0.32^{b}	135.18 ± 0.37^{b}	135.13 ± 0.22^{b}	135.17^{b} ± 0.17	135.04 ± 0.25^{b}	135.29 ± 0.59^{b}
		12	136.08 ± 0.34^{a}	136.28 ± 0.28^{a}	136.58 ± 0.32^{a}	136.65 ± 0.32^{a}	136.95 ± 0.63^{a}	137.46 ± 0.51^{a}
Starch	**TME 419**	0	89.82± 0.53^{a}	89.82± 0.53^{a}	89.82± 0.53^{a}	89.82± 0.53^{a}	89.82± 0.53^{a}	89.82± 0.53^{a}
		4	88.38 ± 0.28^{b}	87.66 ± 1.08^{b}	88.38 ± 0.34^{b}	88.27 ± 0.38^{b}	87.58 ± 1.02^{b}	88.30 ± 0.23^{b}
		8	87.58 ± 0.25^{c}	87.89 ± 0.63^{b}	88.22 ± 0.20^{b}	87.99 ± 0.39^{b}	87.78 ± 0.12^{b}	88.14 ± 0.60^{b}
		12	85.98 ± 0.81^{de}	86.29 ± 0.81^{c}	86.03 ± 0.72^{d}	85.93 ± 0.42^{c}	86.47 ± 0.45^{c}	86.96 ± 0.73^{c}
	UMUCASS 36	0	87.36 ± 0.92^{c}	87.36 ± 0.92^{b}	87.36 ± 0.92^{c}	87.36 ± 0.92^{b}	87.36 ± 0.92^{b}	87.36 ± 0.92^{bc}
		4	85.37 ± 0.29^{e}	84.59 ± 0.97^{d}	85.18 ± 0.32^{e}	85.41 ± 0.20^{c}	85.60 ± 0.35^{d}	86.06 ± 0.73^{c}
		8	83.80 ± 0.30^{f}	84.10 ± 0.37^{e}	83.84 ± 0.17^{f}	84.30 ± 0.56^{d}	84.76 ± 0.22^{e}	85.00 ± 0.27^{d}
		12	82.86 ± 0.83^{g}	83.36 ± 0.31^{f}	82.35± 0.20^{g}	83.30 ± 0.47^{e}	83.76 ± 0.23^{f}	84.47 ± 0.51^{e}

Values are given as means of triplicate determinations ± standard deviation. Different letters in a column are significantly different ($p \leq 0.05$).

Starch content

Starch as a major component in cassava flour is useful for two vital purposes: as raw material for food processing and as a food additive in food (Aina *et al.*, 2012). During the storage of food, starch could be functional in maintaining food quality, stabilising moisture content and regulating water mobility (Abegunde *et al.*, 2013). The starch content of the flour of the cassava cultivars ranged from 87.36 ± 0.92% in 'UMUCASS 36' to 89.82 ± 0.53% in 'TME 419' (Table 1). Both cultivars gave reasonably high starch content, although 'TME 419' was higher than the 'UMUCASS 36'. This is probably because of the inherent genetic differences such as granules size and the molecular structure (Tsakama *et al.*, 2010). Therefore, the differences in starch content due to variation in granular sizes which consequently had influence on the swelling capacity and paste viscosity were reported by Tsakama *et al.* (2010).

The high starch values obtained in both cultivars suggest that starches from these cultivars will be useful in the food industries as a thickener, stabiliser and emulsifier or in the production of pasta and confectionery (Aina *et al.*, 2012). This observation corroborates with the range (83.6 ± 0.01 - 86.0 ± 0.33%) reported for twelve cultivars of cassava flour with low cyanide content (Charoenkul *et al.*, 2011). During the 12 weeks storage period, the starch contents decreased slightly over time with the lowest values (82.35 ± 0.20% and 85.93 ± 0.42%) observed in flour packed in LDPE bags for 'UMUCASS 36' and 'TME 419', respectively. The variations observed in the starch content during storage could be attributed to the differences in the flour inherent properties such as carbohydrate. The interaction effect of storage temperature, duration and package had no significant effect ($p > 0.05$) on starch content in both cassava flour cultivars during storage (Fig. 2), whereas, cultivar and storage duration effects with package and duration effects had a significant impact on the starch content ($p < 0.05$).

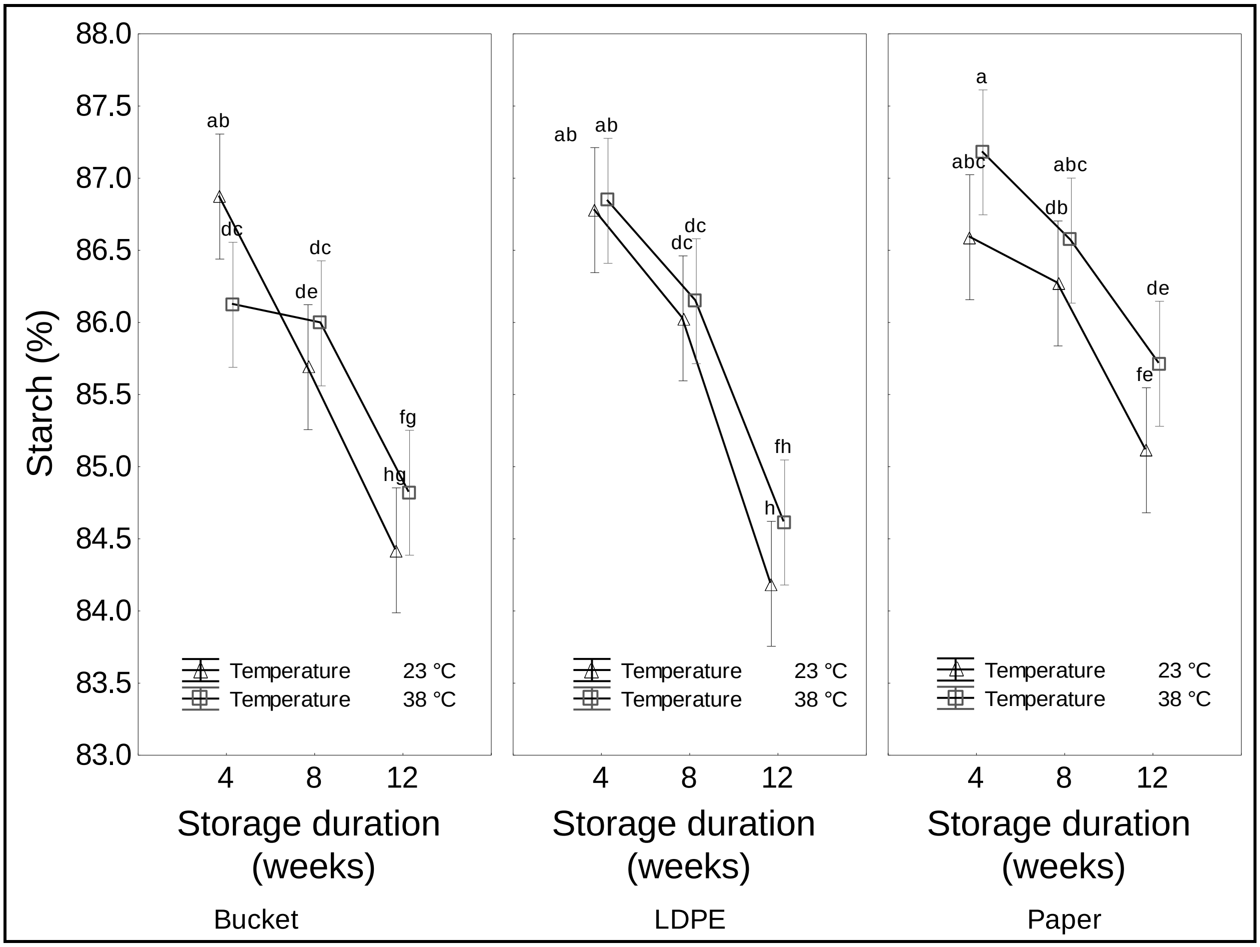

Figure 2 Effects of temperature, package types and duration on starch content of the flour of both cassava cultivars. Similar letters are not significantly different according to Fisher LSD test (*p* > 0.05).

Mineral compositions of cassava flour cultivar under storage at different conditions

The mineral contents of both cultivars were generally low (Table 2) compared to flour from other root crops such as sweet potato, and this confirms the deficiency of mineral in cassava (Charles *et al.*, 2005). There was significant difference in the mineral contents between both cultivars 'TME 419' and 'UMUCASS 36'. The cv. 'TME 419' had higher content in most of the measured minerals such as K, Na, Cu, Fe, and Zn. This implies that some cultivars have more mineral content than others. The summary of the mineral compositions shows that none of the treatments in this study had significant influence on the mineral content of the flour of both cassava cultivars. However, increase in storage duration observed resulted in significant loss of the essential minerals (Na, Mn, Fe, Zn, and B) (p < 0.05). Similarly, Charles *et al*. (2005) observed significant differences in cultivars in their study on the mineral compositions of five different cassava cultivars (Rayong 5, Kaesetsart 50 (KU50), Rayong 2, Hanatee, and KMUL 36-YOO2 (YOO2).

Table 2 Statistical summary of treatment effects on mineral contents of cassava flour.

Effect	**p values**
Cultivars	0.000000
Package	0.009506
Temperature	0.001536
Duration	0.000000
Cultivars*Package	0.985040
Cultivars*Temperature	0.183371
Package*Temperature	0.862583
Cultivars*Duration	0.002567
Package*Duration	0.986491
Temperature*Duration	0.259098
Cultivars*Package*Temperature	0.914218
Cultivars*Package*Duration	0.180865
Cultivars*Temperature*Duration	0.629357
Package*Temperature*Duration	0.980652
Cultivars*Package*Temperature*Duration	0.844847

Table 3 presents the summary of the influence of storage conditions and package on the mineral compositions of cassava flour during storage. A slight increase was observed at the end of storage in (N, P, Mg and Ca) while the other measured minerals showed a slight decrease after storage. Sodium declined from 680 - 525 mg/ kg in ‘TME 419’ and from 556 - 523 mg/ kg in ‘UMUCASS 36’. This observation was noted in flour packed in paper bag under higher condition (38 °C, 60% RH). Similarly the lowest values in iron (19 mg/ kg and 14mg/ kg), manganese (6 mg/kg and 5 mg/ kg) and calcium (0.03% and 0.03%) were also noticed in flour packed in paper bag under higher condition for both cultivars. Storage conditions and package effects as well as temperature and storage duration were insignificant on the measured minerals (Table 2). Nitrogen content ranged from 0.22% in ‘TME 419’ to 0.45% in ‘UMUCASS 36’. All the treatments showed little variations in nitrogen content of the flour during storage but were not significantly different ($p > 0.05$). Potassium content of ‘TME 419’ flour cultivar was significantly higher (0.30%) than the ‘UMUCASS 36’ flour cultivars (0.17%).

A slight decline in percentage potassium (0.26%) in ‘TME 419’ flour and (0.12%) in ‘UMUCASS 36’ was observed at the end of 12 weeks storage period. This drop was consistent in all the packages as well as storage temperature but the lowest was noted in flour packed in paper bag (0.03%). However, no significant effect was observed on the interactions between all treatments (package, duration, temperature and cultivars) after the 12 weeks storage period in the present study. Most of the reported minerals were lower than the observation by Charles *et al*. (2005) who evaluated the proximate and mineral compositions of five different cassava flour cultivars. This could possibly be because of the low ash content (1.2 ± 0.02-1.6 ± 0.03%) found in the cassava flour cultivars used for this study compared with the range (1.4 ± 0.52-2.4 ± 0.35%) reported by Eleazu and Eleazu (2012) since ash content gives an indication of the amount of mineral contents in the flour.

Table 3 Effects of storage conditions (ambient and higher conditions) and package on the minerals compositions of cassava flour after storage (dry weight base)

Cultivars	Duration (Weeks)	Package type	Temp (°C)	N (%)	P (%)	K (%)	Ca (%)	Mg (%)	Na (mg/kg)	Mn (mg/kg)	Fe (mg/kg)	Cu (mg/kg)	Zn (mg/kg)	B (mg/kg)	Ni (mg/kg)
TME 419	0			0.22e	0.05b	0.30b	0.03a	0.05a	680a	40a	93.5a	15a	23a	62a	2.02a
	12	Bucket	23 °C	0.23e	0.05b	0.31ab	0.04a	0.06a	583ab	19b	83b	10c	11b	40b	2.25a
		Bucket	38 °C	0.26d	0.05b	0.29b	0.04a	0.06a	554c	12c	66c	14ab	7c	24c	0.71d
		LDPE	23 °C	0.25de	0.04b	0.28b	0.03a	0.04b	533d	8d	43e	13bc	2d	14d	0.58e
		LDPE	38 °C	0.28d	0.06ab	0.34a	0.05a	0.07a	534d	8d	51d	16a	3d	13de	0.72d
		Paper	23 °C	0.27d	0.04b	0.26bc	0.04a	0.05ab	526e	6e	29gh	14a	2d	13de	0.81b
		Paper	38 °C	0.25e	0.04b	0.28b	0.03a	0.05ab	525e	6e	19i	14a	6c	6f	0.62de
UMUCASS 36	0			0.45ab	0.04b	0.17d	0.04a	0.04b	556c	7de	34f	6e	3d	7f	0.53e
	12	Bucket	23 °C	0.40c	0.04bc	0.17d	0.04a	0.04bc	536d	7de	26h	11c	0	5f	0.55e
		Bucket	38 °C	0.43b	0.03c	0.14ef	0.04a	0.04bc	532d	7de	26h	13b	1e	4f	0.75d
		LDPE	23 °C	0.48a	0.06a	0.17d	0.05a	0.04bc	545cd	8d	22h	17a	5c	6f	0.78d
		LDPE	38 °C	0.45b	0.04bc	0.16de	0.04a	0.04bc	531d	8d	25h	16a	4d	11e	0.83b
		Paper	23 °C	0.41bc	0.04b	0.15e	0.03a	0.04bc	534d	6e	30g	8d	2d	5f	0.64d
		Paper	38 °C	0.44b	0.03c	0.12f	0.03a	0.03c	523e	5f	14j	7de	0	10e	0.55e

Values are given as means of triplicate determinations. Similar letters in columns are not significantly different ($p > 0.05$)

Changes in pasting properties of cassava flour during storage

The determination of pasting behaviour of flour or starch extract is important because it helps in the characterisation of flour and estimating its suitable applications in the food industries (Shimelis *et al.*, 2006; Aviara *et al.*, 2010). In product development, gelatinisation and pasting processes contribute to influence the stability as well as the texture of the products (Iwe & Agiriga, 2014).

Peak viscosity of both cassava flour cultivars at the initial week before storage was 238.57 ± 4.35 RVU and 266.20 ± 4.35 RVU for 'UMUCASS 36' and 'TME 419', respectively. Flour from 'TME 419' had the higher peak viscosity compared to 'UMUCASS 36' (Fig. 3). Variations in viscosity have been reported to be influenced by variety and time of harvest (Iwe & Agiriga, 2014). In the present study both cultivars were harvested 12 months after planting, hence, the observed variation could only be associated with varietal differences and/or the inherent properties of the flour such as amylose contents (22.9 - 24.3%) (Adebowale *et al*., 2008). For instance, Ikegwu *et al*. (2009) made similar observation of cultivar differences with 13 different cultivars of cassava flour. Also the degree of starch damage and the binding force between the flour particles can contribute to influence the peak viscosity (Zaidul *et al.*, 2007; Kaur *et al.*, 2013). The high viscosity in flour are associated with weak starch molecules, this is because starch molecules penetrate easily into their granules leading to high viscosities and reduced resistant to shear. The weak force makes the paste from such flour more susceptible to breakdown during cooking (Etudaiye *et al.*, 2009). Similarly, Liu *et al.* (2006) in the study of pasting properties of cocoyam affirmed that high peak viscosity is an evidence of the maximum swelling power of the starch granules before disintegrating Therefore, this suggests that flour from 'TME 419' with higher peak viscosity (Fig. 3) will be best for making jelly, thickener or binders and products that cannot withstand higher temperature during processing, while 'UMUCASS 36' flour with lower peak viscosity would be recommended in making weaning food (Tsakama *et al.*, 2010).

Trough or hot paste viscosity is referred to as the viscosity at the end of the holding period at 95 °C and this gives an indication of the ability of the paste to withstand shear during gelatinisation processing at high temperature (Jimoh *et al.*, 2010). Trough of flour can be influenced by the nature of starch leaching, and the rapid leaching of flour amylose component to the aqueous phase during pasting causes a quick granular realignment resulting to higher trough (Singh *et al*., 2006; Iwe & Agiriga, 2014).

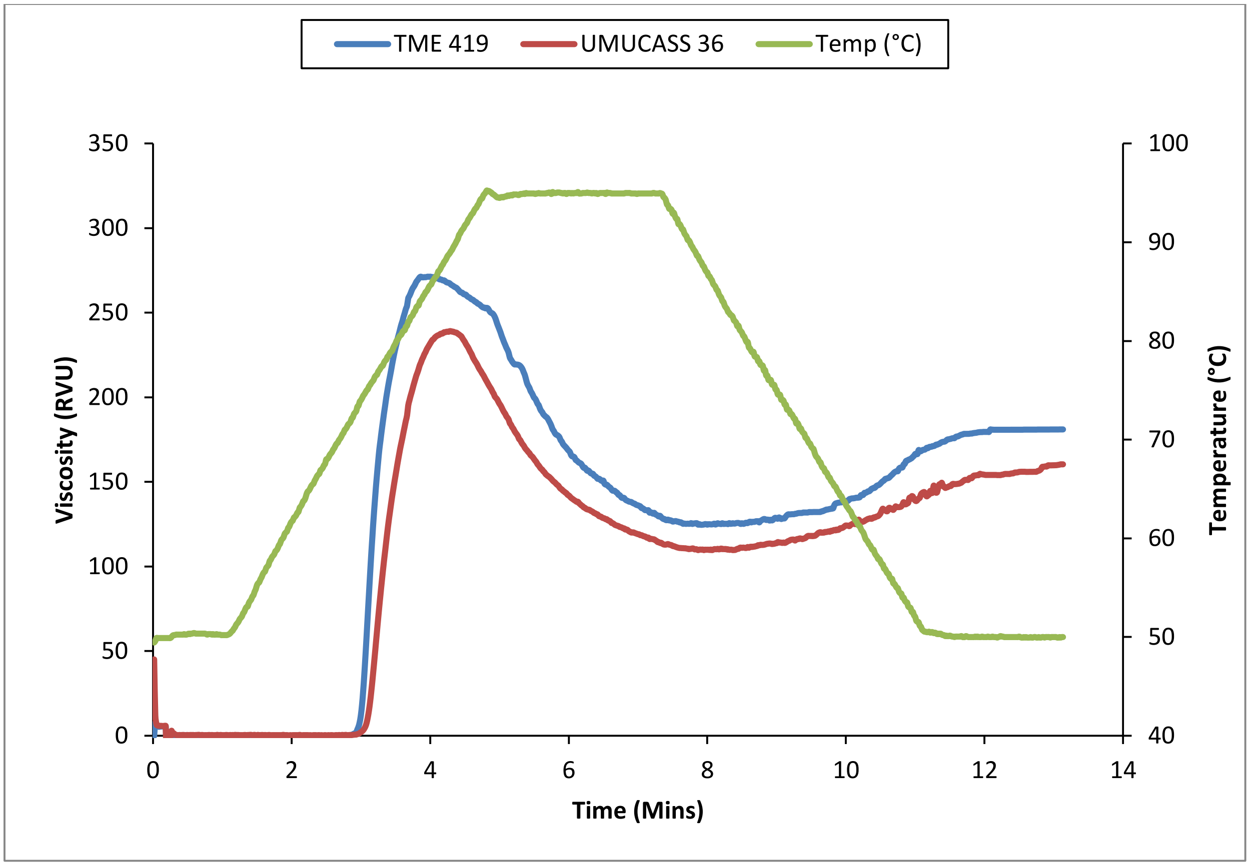

Figure 3 Typical pasting curves for two cassava flour cultivars (cvs. 'TME 419' and 'UMUCASS 36') after processing

The viscosity of hot paste of 'TME 419' flour (123.67 ± 1.06 RVU) was significantly higher ($p < 0.05$) for 'UMUCASS 36' (107.47 ± 2.01 RVU) as shown in Fig. 3. Similar significant varietal differences in hot paste were observed in five different varieties of cocoyam flour (Falade & Okafor, 2013). The values obtained for both cassava cultivars in this present study were within the range (108 - 157 RVU) reported for blend of water yam and cassava (Babajide & Olowe, 2012). Higher trough observed in 'TME 419' flour could also be attributed to the higher swelling power which allowed the granules to reach its highest peak faster. A study on the pasting behaviour of blends of resistant starch and wheat flour supports this observation of high trough viscosity being a function of the swelling power (Fu *et al.*, 2008); hence, could lead to decrease in shear resistance force at high temperature.

After the 12 weeks storage period, the peak viscosity and the trough increased significantly in all the packaging materials and storage conditions. The values ranged from (266.2 ± 0.51 to 302 ± 9.5 RVU) in 'TME 419' flour and from (228.5 ± 1.36 to 246.3 ± 2.91 RVU) in ‘UMUCASS 36’ for peak viscosity. Trough ranged from (129.8 ± 1.19 to 145.3 ± 2.23 RVU) in ‘TME 419’ while ‘UMUCASS 36’ ranged from (115.1 ± 1.52 to 127.3 ± 1.78 RVU) (Table 4). The lowest viscosities were observed in the flour stored in plastic bucket under ambient condition for both cultivars. This could be as a result of the fat content reported in this package type. Fat content in flour as well as protein have been reported to form complexes with amylose during heating and prevent the swelling of the starch thereby reducing the viscosity of the flour (Shimelis *et al.*, 2006). Similar observation was reported for wheat flour as flour with the highest fat gave the lowest peak viscosity, while the reverse was the case of flour samples with reduced fat content (Salman & Copeland, 2007). In addition, viscosity can be influenced by other factors such as phosphorus content of flour, and some added ingredients like salt, pH modifier and sugar. This was proven in the study of pasting profile of wheat flour in combination with root and tuber flour blends (Zaidul *et al.*, 2007). The authors observed that potato flour with the highest phosphorus content showed the highest viscosity compared to the other root and tuber crops as well as those mixed with wheat flour. Furthermore, the authors reported that potato flour with the highest peak viscosity showed higher resistance to retrogradation and could permit substitute with wheat flour up to 50%. This information therefore implies that flour kept in paper bag which showed the highest viscosity will best substitute with wheat flour as composite flour in the food industries. The effect of the interaction of package and storage condition had significant impact on the viscosity of cassava flour ($p < 0.05$).

After gelatinisation when the highest peak is reached, the swollen starch granules begin to disintegrate and this causes the amylose molecules to leach into the aqueous solution (water) and viscosity begins to decrease. This phenomenon is known as breakdown viscosity (Tsakama *et al.*, 2010). This can be determined from the difference between the peak viscosity and the trough. The mean breakdown viscosity of flour was significantly higher for ‘TME 419’ (142.53 ± 3.35 RVU) than ‘UMUCASS 36’ (121.10 ± 1.57 RVU). Low breakdown viscosity has been reported to show better resistance to shear during heating and the formed paste will be stable under hot or heating condition (Adebowale *et al*., 2005; Abiodun *et al.*, 2009). Falade and Okoafor (2013) added that low breakdown indicates a low peak viscosity. Therefore, it could be expected that flour obtained from ‘UMUCASS 36’ cassava cultivar will show greater resistance to shear during heating compared to flour from ‘TME 419’ cultivar because of the lower viscosity.

Table 4 Pasting profile of two cultivars of cassava flour after storage

Cultivar	Package type	Temp (°C)	Duration (weeks)	Peak viscosity (RVU)	Trough (RVU)	Breakdown (RVU)	Final viscosity (RVU)	Setback viscosity (RVU)	Peak time (min)	Peak temp (°C)
TME 419			0	266.20 ± 4.35^{b}	123.67 ± 1.06^{h}	142.53 ± 3.35^{b}	181.00 ± 1.92^{de}	51.00 ± 0.95^{a}	4.00 ± 0.00^{c}	85.43 ± 0.15^{f}
	Bucket	23	12	260.23± 0.51^{d}	130.80 ± 2.46^{e}	129.43 ± 1.96^{e}	179.20 ± 1.71^{f}	48.40 ± 0.75^{b}	4.07 ± 0.06^{b}	86.10 ± 0.00^{e}
		38		264.43 ± 0.50^{c}	138.50 ± 0.75^{c}	125.93 ± 0.29^{f}	184.60 ± 0.75^{c}	46.10 ± 0.01^{c}	3.97 ± 0.12^{c}	86.30 ± 0.10^{e}
	LDPE	23		263.67 ± 4.04^{cd}	129.77 ± 1.19^{ef}	133.90 ± 2.86^{c}	180.23 ± 2.61^{de}	50.47 ± 3.30^{ab}	4.03± 0.06^{b}	85.77 ± 0.31^{f}
		38		292.30 ± 2.82^{a}	148.80 ± 2.69^{a}	143.50 ± 5.28^{b}	190.30 ± 0.82^{b}	41.50 ± 3.15^{f}	3.97 ± 0.08^{c}	86.25 ± 0.57^{e}
	Paper	23		265.93 ± 0.50^{b}	133.33 ± 0.40^{d}	132.60 ± 0.56^{d}	182.00 ± 1.87^{d}	48.67 ± 2.10^{b}	4.03 ± 0.06^{b}	85.07 ± 0.12^{g}
		38		302.13 ± 9.52^{a}	145.30 ± 2.23^{b}	156.83 ± 7.45^{a}	193.07 ± 5.46^{a}	47.77 ± 3.23^{bce}	3.87 ± 0.08^{d}	84.90 ±0.36^{g}
UMUCASS 36			0	228.57± 0.46^{f}	107.47 ± 2.01^{k}	121.10 ± 1.57^{g}	160.35 ± 4.10^{k}	47.80 ± 2.19^{bc}	4.20 ± 0.00^{a}	88.60 ± 0.35^{a}
	Bucket	23	12	228.47 ± 1.36^{h}	115.03± 1.52^{j}	117.17 ± 3.09^{h}	162.30 ± 3.05^{ij}	46.27 ± 1.58^{c}	4.00 ± 0.06^{bc}	88.80 ± 0.26^{a}
		38		229.30 ± 1.13^{h}	120.83 ± 1.63^{i}	108.4 ± 2.50^{k}	170.20 ± 0.85^{g}	49.37 ± 2.29^{ab}	4.03± 0.06^{b}	88.37 ± 0.15^{b}
	LDPE	23		230.57 ± 2.40^{g}	115.27 ± 0.85^{j}	113.20 ± 0.53^{j}	160.90 ± 2.59^{jk}	45.63 ± 2.95^{e}	4.00 ± 0.00^{c}	88.57± 0.15^{a}
		38		232.20± 4.53^{g}	120.70 ± 2.04^{i}	109.87 ± 0.50^{k}	161.93± 2.70^{j}	41.23 ± 0.71^{f}	4.00 ± 0.10^{c}	88.10 ± 0.10^{bc}
	Paper	23		239.10 ± 1.47^{f}	124.07 ± 1.77^{g}	115.03 ± 1.4^{i}	164.90 ± 2.95^{i}	40.83 ± 1.59^{fg}	3.93 ± 0.06^{c}	87.93 ± 0.15^{c}
		38		246.30 ± 2.91^{e}	127.30 ± 1.78^{f}	119.00 ± 3.44^{hg}	167.57 ± 0.58^{h}	40.27 ± 1.16^{fg}	3.87 ± 0.06^{d}	87.50 ± 0.16^{d}

Values are given as means of triplicate determinations ± standard deviation. Similar letters in a column are not significantly different ($p > 0.05$)

This observation closely agrees with the study on 13 cultivars of cassava starches, where the authors also reported lower breakdown viscosities in cultivars with lower peak viscosities (Ikegwu *et al.*, 2009). Correlation analysis revealed a strong positive and significant correlation (r = 0.94, p ≤ 0.05) between peak viscosity and breakdown (Fig. 4). The shear resistance of the flour under heat could be predicted using this relationship, as flour with high peak will exhibit high breakdown and low resistance (Tsakama *et al.*, 2010). This implies that weaker gel will be formed from such flour samples with higher viscosities.

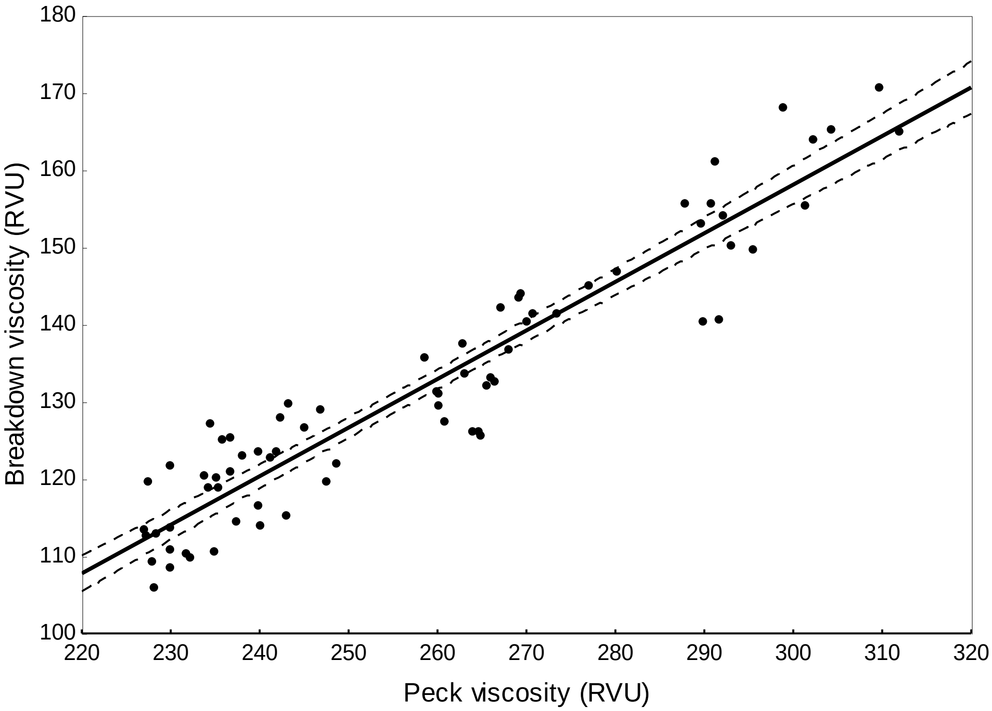

Figure 4 Relationship between peak viscosity and breakdown viscosity of cassava flour during storage ($p \leq 0.05$).

Another viscosity parameter measured was the setback viscosity. This parameter describes the stability of flour paste after cooking, which is the cooling phase in the pasting profile (Sanni *et al.*, 2006). The initial values before storage of the cassava flour for the setback viscosity were 51.00 ± 0.95 RVU and 47.80 ± 2.19 RVU for 'TME 419' and 'UMUCASS 36', respectively. These values closely correspond to the values reported by Etudaiye *et al*. (2009) who evaluated the pasting properties of 43 cultivars of cassava fufu (fermented) flour and reported a range of setback viscosity from 70.42-28.17 RVU. Low setback values imply the flour would have high paste stability and resistance to retrogradation on cooling (Sanni *et al.*, 2006; Etudaiye *et al.*, 2009). A decrease was observed in both flour cultivars after storage across all the packaging materials from 40.27 ± 1.16 RVU in 'UMUCASS 36' to 51.00 ± 0.95 RVU in 'TME 419' flour. However, LSD results explained that the interaction of package, temperature and duration had no significant impact on setback viscosity of the flour cultivars, whereas, cultivar and package effects had significant effect on setback $p \leq 0.05$.

Final viscosities of both cultivars were between 181.00 ± 1.92 RVU for 'TME 419' and 160.35 ± 4.10 RVU for 'UMUCASS 36'. Final viscosity has been reported as the ability of flour to form gel after cooking and cooling, giving an indication of flour paste stability (Shimelis *et al.*, 2006). Both cultivars used for this study differed significantly in their final viscosities ($p < 0.05$). Iwe and Agiriga (2014) also reported significant difference in the final viscosity of cassava flour cultivars. These variations amongst cultivars were supported by Ikegwu *et al*. (2009) and the authors attributed the differences to be the kinetic effect of cooling and the reorganisation of the starch molecules. Furthermore, the lower final viscosity in the 'UMUCASS 36' could be attributed to the amylose content in the flour. Higher amylose starches are known to reorganise more rapidly because the linear chain allows the molecules to tightly arrange and held together by hydrogen bond (Shimelis *et al.*, 2006; Fu *et al.*, 2008). Flour samples with higher final viscosities are known for better paste or gel stabilities. The notion was affirmed by Iwe and Agiriga (2014) who reported a more stable gel in cassava flour cultivar with high final viscosity. Hence, the 'TME 419' flour with higher final viscosity could be more preferred in the food industries for the production of salad dressings, confectionery and in other products where high viscosity is required (Shimelis *et al.*, 2006).

The final viscosity increased significantly after 12 weeks storage with values ranging from 160.90 ± 2.59.to 193.07 ± 5.46 RVU. Temperature and package effects were significant in both cultivars at the end of 12 weeks of storage. Cassava flour cultivars stored under higher condition had the highest final viscosities at the end of the storage period and was consistent in all the package type. The observed values ranged from 161.93 ± 2.70 RVU in LDPE to 193.07 ± 5.46 RVU in paper bag. This effect of increase in temperature was also reported by Salman and Copeland (2007) who evaluated wheat flour samples at 30 °C and 4 °C and the highest final viscosity was seen in flour stored at 30 °C compared to the samples at the lower temperature However, the results (Table 5) did not report significant difference on the effects of cultivar, package and duration ($p > 0.05$).

Table 5 Statistical summary of the treatment effects on final viscosities of cassava flour

Effects	SS	MS	F	p
Duration	26	26	4.0	0.052359
Cultivar*package	38	19	2.9	0.064730
Cultivar*temperature	5	5	0.7	0.403402
Package*temperature	64	32	4.8	0.012147
Cultivar*duration	20	20	3.0	0.088054
Package*duration	4	2	0.3	0.749691
Temperature*duration	35	35	5.3	0.026271
Cultivar*package*temperature	92	46	7.0	0.002113
Cultivar*package*duration	21	10	1.6	0.217120
Cultivar*temperature*duration	39	39	6.0	0.018013
Package*temperature*duration	41	20	3.1	0.054223
Cultivar*package*temperature*duration	14	7	1.1	0.354408
Error	315	7		

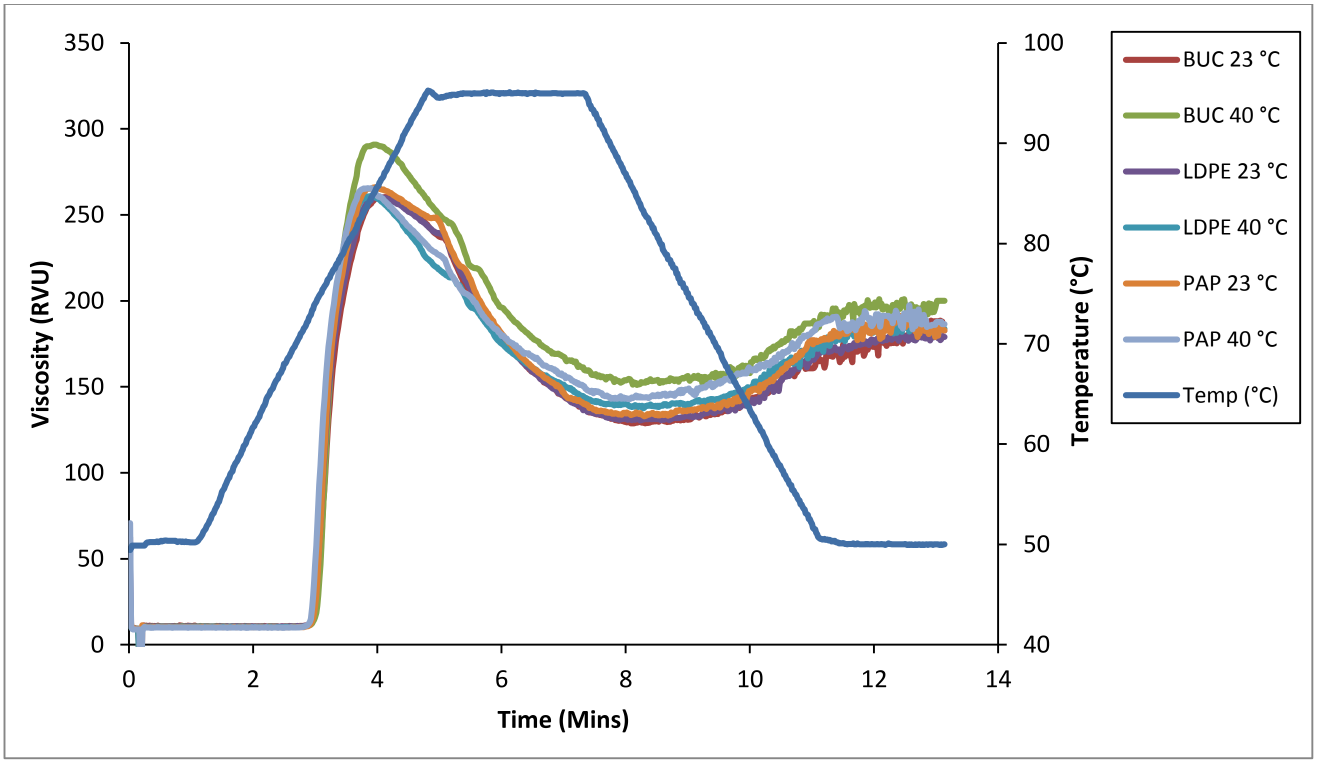

Figure 5 Typical pasting curves for ‘TME 419’ cassava flour at the end of 12 weeks storage.

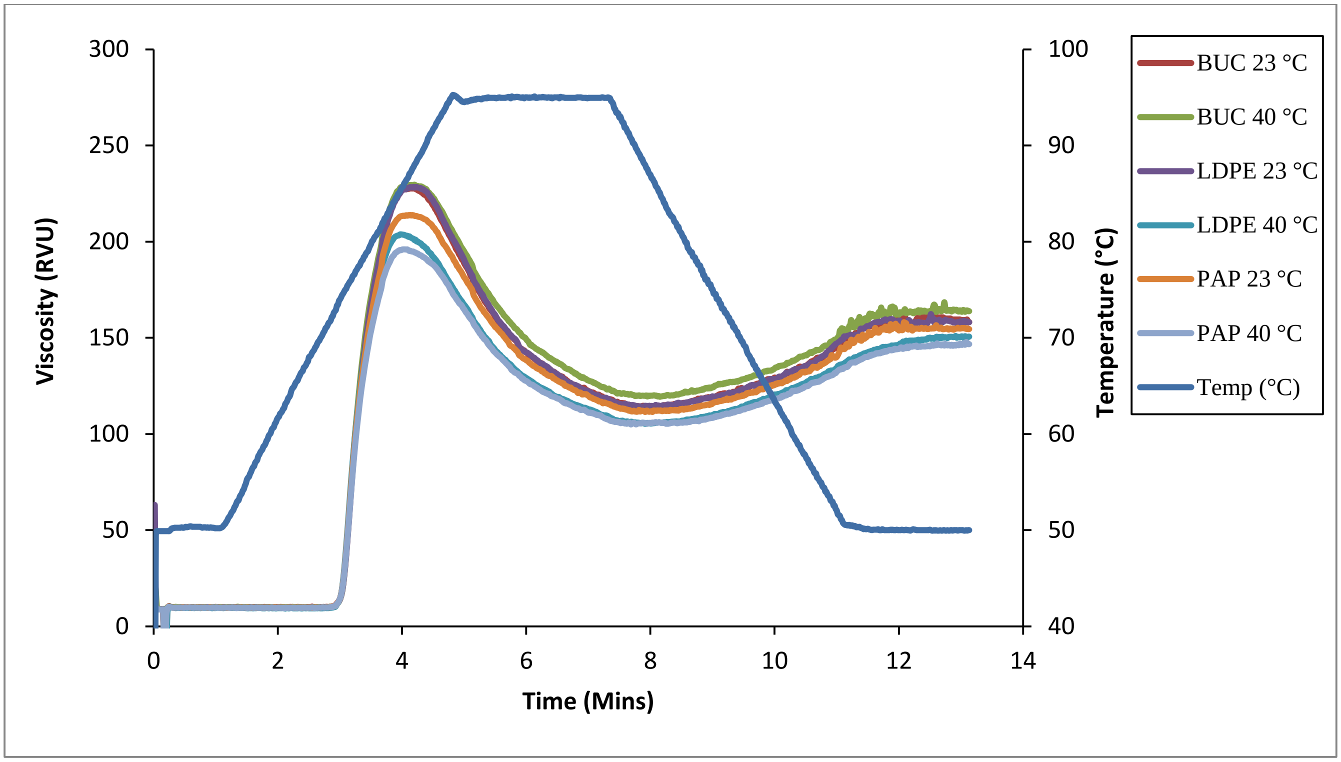

Figure 6 Typical pasting curves for 'UMUCASS 36' cassava flour at the end of 12 weeks storage.

The pasting time is the set time when the paste reaches its peak viscosity and forms gel at a particular temperature (Etudaiye *et al.*, 2009). It can be used to estimate the actual cooking time for flour based food product (Adebowale *et al.*, 2005). The pasting time for the cassava flour in this study ranged from 3.87 ± 0.06 min - 4.20 ± 0.001 min. The lowest pasting time (3.87 ± 0.06 min) for the cassava flour after storage was recorded in the flour packed in paper bag under higher condition for both cultivars. No significant difference was noticed in the pasting time for all treatments (cultivars, temperature, package and duration). However, cassava flour with shorter pasting time could be expected to use lesser energy during processing thus reducing the production time and cost. The 'TME 419' flour had shorter pasting time (4.0 ± 0.001) than 'UMUCASS 36' flour (4.02 ± 0.001), however, after storage the pasting time decreased. Although cultivars difference was significant at the initial week, no significant effect was noticed at the end of 12 weeks storage. Similar observation was reported by Iwe and Agiriga (2014), who concluded that sample with short peak time, would show low resistance to shear. In addition, Tsakama *et al*. (2010) reported that samples with short peak time would swell quickly which will lead to fast disassociation of the granules. Therefore, this agrees with the result from this present study as flour treatments with the shortest peak time gave the highest viscosities and swelling power.

The peak temperature ranged from 85.07 ± 0.12 °C to 88.80 ± 0.26 °C after the storage. Peak temperature indicates the minimum temperature required to cook a product as well as the energy cost involved (Ikegwu *et al.*, 2009) The 'UMUCASS 36' was higher (88.60 ± 0.35 °C) than the TME (85.43 ± 0.15). Therefore, it clearly shows that flour from 'TME 419' would require less energy during cooking. Etudaiye *et al*. (2009) reported pasting temperature from 77.55-81.60 °C in fufu flour while Babajide and Olowe (2013) reported a range from 68.40-82.35 °C for water yam-cassava composite flour. The lowest temperature (84.90 ± 0.36 °C) after storage was observed in the cassava flour sample packed in paper bag at 38 °C for both cultivars. Also, the effects of all the interactions (cultivar, temperature, package and duration) gave no significant difference ($p > 0.05$) in the pasting temperature. However, flour under such treatments of low temperature could be expected to use lesser energy during processing thus reducing cost of production.

Conclusion

Results from this study showed the influence of selected package types and storage conditions on the mineral contents, functional and pasting properties of cassava flour. The variations noticed in swelling power and solubility was influenced by the amylose content in both cultivars. The swelling power of both cultivars was within the highly restricted starch level (≤ 16) and would exhibit shear resistance during cooking. The highly restricted flour is desirable for substitution with cereals and in the production of noodle, bread and other confectionery, thus highlighting the potential of both cassava cultivars as sources of flour in the food processing industry because of the strong force of attraction between the molecules. However, 'UMUCASS 36' flour samples packed in plastic bucket and stored under ambient condition with lower swelling power would form more stable paste which is a desirable attribute in the industries for the production of weaning foods and noodles. Furthermore, higher water binding capacity indicates easy handling of dough during product development.

Higher viscosities observed in 'TME 419' flour packed in paper bag and stored under higher conditions (38 °C, 60% RH) show a weaker binding force, low resistance to shear during heating and less paste stabilities, making potential useful raw materials in the production of thickeners, jelly or in other products where high viscosities are recommended. However, for the lower viscous cassava flour samples, use in the production of weaning food and in the paper and textile industries will be ideal. Also lower breakdown in 'UMUCASS 36' shows better paste stability against paper package with higher breakdown after storage. Shorter peak time indicates reduction in energy and invariably low production cost for the produce. The strong positive and significant correlation ($r = 0.94$, $p \leq 0.05$) observed between peak viscosity and breakdown could be useful in assessing paste stability. The differences in the functional and pasting properties allows cassava flour to be used for various industrial purposes such as baking, production of noodles, weaning food, thickeners, sugar syrup and many others.

Therefore, based on the information from this study, cassava flour packed in paper bag and stored in typical higher condition found in many tropical and sub-tropical climates (38 °C, 60% RH) would be more suitable for cassava flour storage. This finding is particularly useful for storage of cassava flour as composite flour for baking given the relevant functional properties and other quality attributes.

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CHAPTER 6

GENERAL DISCUSSION AND CONCLUSIONS

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Cassava (*Manihot esculenta* Crantz*)* is a tuberous root crop, commonly grown in the tropics. The tuberous root, rich in carbohydrate and a good calorie source is the major edible part of the plant (Lebot *et al.*, 2009). Cassava root serves as a major staple food in some continents such as African, Asia and in South America. It is an energy giving food and supplies calorie to population of more than 500 million people daily (Opara, 1999; Morante *et al.*, 2010). Cassava root is widely used for industrial and domestics purposes. However, the application is limited by the rapid postharvest physiological deterioration, which starts within 2 - 3 days after harvest of the root (Sánchez *et al.*, 2006). As a result of this challenge, processing cassava root into other food forms is essential as it also helps to extend the postharvest life, improve palatability and add value to the root (Taiwo, 2006; Fadeyibi, 2012). The primary products obtained from the root are classified into fermented (fufu, garri, starch, lafun) and unfermented (flour, tapioca, chips and pellets). These products could be further processed into other products like cassava bread, weaning foods, beer, and paper.

Most of the advanced technologies to prevent the onset of postharvest physiological deterioration such as freeze-drying, coating with paraffin wax and storage in box with moist sawdust are not being fully harnessed because they are expensive to acquire and maintain (Reilly *et al.*, 2004). These processing and preservation techniques are generally inefficient in controlling postharvest deterioration because they could only extend the shelf-life and maintain quality of the root for few weeks (Zidenga *et al.*, 2012). Furthermore, in-ground storage of the root which is a preferred method to extend shelf-life could result to loss of essential nutrients, root lignifications and even rotting of the root if left longer in the ground (Westby, 2002). However, minimally processed products like flour, garri, and chips offer the benefit of longer shelf-life, flexibility in transportation, and variety in utilisations (Falade & Akingbala, 2010; Opara & Mditshwa, 2013)

High quality cassava flour, an unfermented, minimally processed product from the root is recommended as a potential product for composite flour in baking of high grade food such as bread (Sanful & Darko, 2010; Nwabueze & Anoruoh, 2011). This is attributed to its health benefit as gluten-free flour, economic benefits and its availability (Biagi *et al.*, 2009; Kulchan *et al.*, 2010; Gyedu-Akoto & Laryea, 2013). Cassava flour, usually from the West African countries, is readily available and cost of production is low. As a result, it reduces the dependency on the imported wheat flour which is expensive and not readily available. However, the use of cassava flour is still strange in some developed countries across the world, thus there is need for advocating its use. This could be possible by estimating an optimum packaging and storage conditions for cassava flour in order to harness exportation to countries where cassava is not grown and commercialisation to meet consumers' demand of the produce. The knowledge of this will also help to stabilise shelf-life and maintain the quality attributes of the root.

Flour products are globally handled in various packaging materials such as paper bags, plastic buckets, polyethylene, sack bags or a combination of one or two of the materials like the laminated paper bags and stored at ambient storage conditions 20 - 28 °C, 40 - 60% RH (Butt *et al.*, 2004; Daramola *et al.*, 2010; Agrahar-Murugkar & Jha, 2011; Ogugbue & Gloria, 2011). However, studies on the effect of packaging and storage on the quality of cassava flour as well as the changes in functional properties over storage period are limited. Therefore a comprehensive study was done to investigate the effects of storage conditions (15 °C, 90% RH) cool storage, (23 ± 2 °C, 60% RH) ambient condition, and (38 ± 2 °C, 60% RH) higher condition on the quality of two cassava flour cultivars 'TME 419' and 'UMUCASS 36'. Also the influence of selected packaging materials (brown paper bag, low density polyethylene bag and plastic bucket) was evaluated. Furthermore, the study investigated the changes in functional and pasting properties under different storage conditions over time. Information generated from this research would be useful for maintaining quality, extending shelf-life and improving the commercialisation and utilisation of cassava flour.

The influence of storage conditions, cultivar and their interactions showed significant difference on the proximate compositions, physicochemical and microbial quality of cassava flour during the storage period. The temperature effect clearly showed that storage at cool conditions (15 °C, 90% RH) is not appropriate for flour packed inside paper bag because of the moisture increase and visual observation of decay noticed on the package before the fourth week of storage. In the study of whole grain wheat flour storage, Kolmanič *et al*. (2010) also observed high moisture content in paper bag kept at 10 °C which led to microbial instability. The studies are in agreement with the observation in this study as higher microbial counts was recorded in flour kept in paper bag under cool conditions after the first two weeks of storage. This could be attributed to the higher permeability of paper to moisture compared with the other packaging materials studied, especially in humid environment. Similarly, a study on the effect of storage conditions on the wall friction of three food powders concluded that increase in moisture content of the powders stored at 5 and 15 °C caused the particles to become sticky (Iqbal & Fitzpatrick, 2006). This report supports the sticky nature observed with paper package under cool condition as a result of increased moisture content in the flour. On the other hand, flour stored under ambient and higher condition had lower moisture content; therefore shelf-life of cassava flour would be expected to be more stable at higher storage temperature and lower RH conditions. In addition, the ambient condition (23 °C, 60% RH) was observed to best maintain nutritional and physicochemical quality of cassava flour during the 12 weeks storage duration. This could be as a result of higher proteolytic and lypolytic activities of their corresponding enzymes observed in the flour stored under higher condition thus leading to higher reduction in percentage protein and fat. A study on soybean flour by Agrahar-Aurugkar and Jha (2011) agrees with this observation as the authors also reported higher decrease (18.7% to 17.2%) in percentage fat content of flour kept at higher temperature after storage.

Previous research on the storage roots of cassava clones reported a range from 120 mg/ kg to 1941 mg/kg of hydrogen cyanide (Iglesias *et al.*, 2002). About 43% of the clones evaluated by the authors recorded more than 800 mg/ kg of cyanide content. This observation implies that consumption of root without processing especially for the high cyanide cultivars would be toxic to human body. Other studies also confirm that some processing method employed in this study such as chipping and drying facilitated the breakdown of the linamarase and releasing the cyanogenic compound from the root (Cumbana *et al.*, 2007; Nwabueze & Anoruoh, 2011). The 'UMUCASS 36' flour had higher retention of cyanide (4.9 ± 0.21µg/ mL) after processing because of the inherent genes in the yellow root. Yellow roots are known with bitter taste and this is synonymous with higher cyanide level (Eleazu & Eleazu, 2012). Eleazu and Eleazu (2012) also noted higher cyanide level in yellow cassava flour cultivars. In addition, the milling process of flour from cassava chips after drying also contributed in reducing the cyanide concentration. This is because more surface area was generated during milling which facilitated the breaking of the cyanogenic compound in the flour. Other reported methods to reduce cyanide include the wetting method. Cassava flour mixed with water and allowed to stand for 5 h gave up to 50% decline in cyanide concentration (Burns *et al.*, 2012). The observation from this study corresponds with the previous study as the concentration of the flour reduced after mixing with water and allowing mixture to stand for 1 h. The total cyanide retained after storage in all the conditions falls within the safe limit (10 ppm) of cyanide in food FAO/WHO (1995) and (0 - 50 mg/ kg) for human consumption (Akiyama *et al.*, 2006). Hence, cassava root processing should be properly handled as substantial amount of the cyanide is being eliminated during the process.

The impact of the different packaging materials used in the study was significantly different on the moisture content of the cassava flour samples. This consequently influenced the microbial stability as well as the physicochemical properties during storage. The package with the highest moisture (plastic bucket) showed the highest total mesophilic and fungi counts, which imply less stability in such package type. This observation was possible because of the permeable nature and water vapour transmission rate of the different package types. Paper bag was more permeable and easily allowed the passage of water vapour, oxygen and other gases out of the package, thus showing better microbial stability. Microbes were less active at optimum storage condition (38 °C, 60% RH) and package material (paper bags), because moisture content which could exacerbate the growth of microbes was stabilised (Kolmanič *et al.*, 2010). The other packages trapped the gases and caused the higher microbial counts in cassava flour packed in them. This observation was confirmed in earlier studies where the package with the highest moisture gave the highest microbial infestation (Butt *et al.*, 2004; Ogiehor & Ikenebomeh, 2006).

Furthermore, the decrease in fat and protein content could be because of the loss of moisture content and the enzymatic activities of lipase and protease enzymes. However, plastic bucket best retained the percentage fat and protein concentration (dry weight basis) in the flour, reason being because of the higher moisture content in the container. Another reason could be due to the microbial activities which utilize the carbohydrate and protein thus increasing the moisture content and microorganism infestations in the package. Ogiehor and Ikenebomeh (2006) noticed an increase in moisture due to the microbial infestation in the packages which were suspected to have low water vapour transmission rate, thus leading to higher lipolytic and proteolytic activities and losses in protein and fat content. This is because the microorganisms utilised the carbohydrates as the energy source, protein as nitrogen and hydrolysing the lipids thus releasing water which result to increase in moisture content.

Retention of total carotenoid after processing and during storage of flour is a major challenge in the food industries. This present study justified the notion of carotenoid degradation during long term storage, due to progressive decline in total carotenoid observed over time. Oliveria *et al*. (2010) described the effect of heat exposure during flour processing as a catalyst to oxidation thus leading to carotenoid degradation. The authors of the assessment and degradation of carotenoid also observed a continuous decrease (6.1 ± 0.01 to 0.2 ± 0.09 µg/ g) in carotenoid concentration during the storage of cassava flour. Degradation during processing was basically due to the sensitivity of carotenoid to light, since the chips were exposed to light during the drying process. After the 12 weeks storage period, flour packed in plastic bucket had the least percentage loss with about 8% loss in ‘TME 419’ and 23% in ‘UMUCASS 36’ compared to flour packed in paper bag which had more percentage losses. This was because the container (plastic bucket) created a barrier to light penetration. Similarly, the colour change was lower in flour packed in plastic bucket and stored under ambient condition, because the carotenoid pigment denoted with yellow colour which contributes to the brightness of products was retained more in flour packed in plastic buckets. Therefore, caution should be taken during the postharvest handling of flour in order to reduce degradation of carotenoid pigments and maintain the quality attributes. The functional and pasting profile of the different cassava flour cultivars were significantly influenced by cultivar differences while temperature and package effects had no significant effect on most of the measured variables. This could be attributed to the different inherent genes, amylose content as well as the size of the starch granules (Shimelis *et al.*, 2006; Sánchez *et al.*, 2013). These investigations revealed that the application of cassava flour in the industries is basically based on their functional attributes. For instance, the low values < 16% of swelling power recorded in Chapter 5 falls within the highly restricted swelling power which showed strong binding force and shear resistance during cooking. Highly restricted flour is desirable for substitution with cereals and in food industries for baking and production of noodles, thus highlighting the potential of both cassava cultivars as sources of flour in the food processing industry According to Shimelis *et al*. (2006), this type of swelling is associated with flour with low amylose and low protein contents. The protein contents are responsible for forming complexes during starch pasting and gelatinisation process thereby resisting volume expansion in the flour. Furthermore, low swelling power results to low viscosities as was observed from this study.

Lower functional attributes could favour the food industries in the production of baby food and noodles because they exhibit strong binding force and easy digestibility than flour with higher viscosity and swelling capacity. Conversely, flour with higher viscosity and swelling capacity are known with weaker binding force, low resistance to shear during heating and less paste stabilities. Previous studies supported this and recommended that high viscous flours would be appropriate for the production of thickeners, binding agent and jelly since the paste is less stable to heat (Liu *et al.*, 2006; Etudaiye *et al.*, 2009; Tsakama *et al.*, 2010). In addition, flour with low peak viscosities showed lower breakdown. This correlates with the literature observation for 13 cultivars of cassava starches, where the authors also reported lower breakdown viscosities in cultivars with lower peak viscosities (Ikegwu *et al.*, 2009) Therefore, this implies a positive correlation between peak viscosities and breakdown. With this observation it would be expected that such flour would show greater resistance to shear during heating and form stable paste. Tsakama *et al*. (2010) also observed similar trend in sweet potato and supported better shear resistance of the flour under heat.

This study showed the importance of packaging, storage conditions (temperature and relative humidity) on the proximate, physicochemical, functional and microbial stability of cassava flour over time. Results indicated that longer shelf-life of cassava flour while maintaining good quality attribute for industrial use could be achieved through optimum packaging and storage conditions. Storage of cassava flour with plastic bucket at ambient conditions (23 - 25 °C, 60% RH) is suitable for maintaining nutritional quality, while hot storage condition (38 °C, 60% RH) with paper bag would be appropriate for better shelf-life stability.

Furthermore, the differences observed in the functional properties clearly indicate that cassava flour could be utilised for different purposes in the food and pharmaceutical industries other than for baking purposes. However, additional research should be done to evaluate the interaction of cassava flour with wheat flour at different proportions and assessing the texture and stability of paste from stored flour. Thus sensory studies would be required to evaluate the general acceptability of the product formed by baking with the cassava flour stored for a period of time.

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