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Developing and Persuading Diabetic Mellitus Patients to Adhere to an Improved Dietary Regime

Tochukwu Arinze Ikwunne[1]*, Chioma Lilian Ugwu[2], Rita Orji[3] and Samuel Okide[4]

[1]*Department of Mathematics/Computer Science/Statistics/Informatics, Federal University Ndufu-Alike, Nigeria*

[2]*Department of Food Science and Technology, University of Nigeria, Nigeria*

[3]*McGill University, Canada*

[4]*Department of Computer Science, NnamdiAzikiwe University, Nigeria*

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***Corresponding author:** Tochukwu Arinze Ikwunne, Department of Mathematics/ComputerScience/Statistics/Informatics, Federal University Ndufu-Alike, Ikwo, Nigeria, Tel: +2348038653851; Email: tochukwuikwunne@yahoo.com

Abstract

This study aims to investigate how to process and persuade the intake of a new dietary. Persuasive messages were sent to 30 diabetic patients to remind them of the need of taking the new dietary regime regularly. We produced flour from tiger nuts and blended it with yellow cassava flour at different levels of substitution. The flour blends were analyzed for their functional, proximate, anti-nutritional and microbial properties using standard analytical methods for ten weeks. For patients to maintain regular dietary intake, sensory properties of the new diet produced from the different flour blends were also determined by the patients.

Keywords: Dietary fibre; Yellow cassava flour; Computer-tailored message; Diabetic patients

Abbereviation : DM: Diabetes Mellitus; CDC: Centers for Disease Control; NCDs: Non-Communicable Diseases; YCF: Yellow Cassava Flour; GI: Glycaemic Index; UNTH: University of Nigeria Teaching Hospital Ituku-Ozalla; FETHA: Federal Teaching Hospital Abakaliki; FMC: Federal Medical Center, FMCABA: Federal Medical Centre, Abakaliki; NAUTH: NnamdiAzikiwe University Teaching Hospital; GMPs: Good Manufacturing Practices; CRD: Completely Randomized Design; LSD: Least Significant Difference; NNRCRC: National Root Crop Research Centre, Umudike; BD: Bulk Density (BD); WAC: Water Absorption Capacity; OAC: Oil Absorption Capacity; LGC: Least gelation concentration; SDA: Sabourand Dextrose Agar; GAE: Gallic Acid Equivalents

Introduction

The word "Diabetes Mellitus (DM)" has been defined by many authors from different viewpoints. However, the similarity in these definitions is that diabetes is a chronic metabolic disorder, characterized by high blood glucose (hyperglycaemia), associated with unpaired carbohydrate, fat, and protein metabolism, resulting from either insufficient or no release of insulin in the body by the pancreas [1-3]. DM is a life-long disease that has an assuming pandemic proportion worldwide. As at 2010, the Centers for Disease Control (CDC) and Prevention estimated that there are 18.8 million people affected with diabetes in the United States with another 7 million people with undiagnosed diabetes [4]. In addition, almost 2 million people older than the age of 20 were diagnosed with diabetes in 2010 [4]. Diabetes was estimated to cost more than $174 billion in 2007 in the United States when taking into account medical costs and loss of productivity [5]. Similarly, diabetes is increasingly being recognized as a major health problem in developing countries. For instance, in Africa, the estimated prevalence of diabetes is 1% in rural areas, and ranges from 5% to 7% in urban Sub-Saharan Africa [6].

DM is the predominant form of diabetes in Sub-Saharan Africa, accounting for over 90% of cases. The International Diabetes Federation [7] estimated that 10. 8million people have DM in sub Saharan Africa in 2006 and this would rise to 18.7 million by 2025, an increase of 80%, as such exceeding the predicted worldwide increase of 55% [8]. This rise in diabetes cases are due to rapid urbanization as well as fast changing diets [9]. Nigeria is among the top 5 countries that have the highest number of people affected by DM in sub-Saharan Africa. Some sporadic figures on prevalence rates of diabetes in Nigeria were published [10-12]. Notwithstanding that, a good number of people still live with it undiagnosed. In general, it was estimated in 2013 that over 382 million people throughout the world had diabetes [13]. Currently, diabetes ranks fourth worldwide among non-communicable diseases

(NCDs); cardiovascular diseases, cancer, chronic respiratory diseases and diabetes with prevalence rates of 30%, 13%, 7 % and 2%, respectively [14].

Conventionally, DM is controlled with diet alone or with a combination of diet, hypoglycaemic drugs, and exercise. A diet is a selection of foods people eat to exist or live, or use for therapeutic purposes. Diet is known for many years to play a key role as risk factor for chronic disease [15]. Staple diets in Nigeria especially in the Eastern and Southern part consist of a wide variety of foods such as cassava (made into fufu or gari which is the most common), yam, sweet potatoes, corn starch, plantain, water yam, cocoyam, grains and vegetables, fruits, meat, and fish. Most DM patients in this area are constrained to take mostly plantain as the major and most often the only source of carbohydrate among other carbohydrate sources to help them control their blood glucose levels [16].

The problem with the prescription of plantain as the only carbohydrate source is that it is restrictive, monotonous, and most patients react negatively, therefore, resulting in low compliance and poor glycaemic control [16]. Although many studies have focused on the role of single nutrient diet or diet groups in disease prevention and management, emerging research suggests that there are health benefits from food patterns that include mixtures of diets containing multiple nutrients [17-21]. Specifically, the objectives of dietary management of diabetes are to: achieve optimal blood glucose and blood lipid concentrations; provide appropriate energy for reasonable weight; prevent, delay, and treat diabetes-related complications; and improve health through optimal nutrition [22].

Fortunately, there are three yellow root cassava varieties, UMUCASS 36, UMUCASS 37, and UMUCASS 38 that are being grown (under the Harvest Plus Project) [23,24] in Nigeria for their high concentrations of β-carotene. β-carotene is a precursor to Vitamin A. Fufu which is often made with cassava flour and served alongside soup, is a national dish of Nigeria [25]. On the other hand, tiger nuts (Cyperusesculentus) which is also produced in Nigeria has been reported to be high in dietary fibre content, which implies that it could be effective in the treatment and prevention of many diseases including colon cancer, coronary heart disease, obesity, diabetes, and gastro intestinal disorders [1]. Tiger nut flour has been demonstrated to be a rich source of quality oil and contains moderate amount of protein. Tiger nut reduces the levels of triglycerides in blood and the risk of forming bloody clots, thereby preventing arteriosclerosis [26].

It is also an excellent source of some useful minerals such as iron, and calcium which are essential for body growth and development [27]. Tiger nut has been used extensively in Spain, mainly for human consumption [28]. The flour is used to make cakes, breads and biscuits, and the oil is used for cooking and is a superb bait for carp fishing in the United Kingdom [29]. A diabetes diet is simply a healthy eating plan that is high in nutrients, low in bad fat and moderate in calories. It is a healthy diet for anyone.

The only difference is the need to pay more attention to some of the food choices most notably the type and quantity of carbohydrates consumed. It is best to limit highly refined carbohydrates like white bread, rice, snack foods, carbonated drinks, candy etc; focusing on high-fibre complex carbohydrates (also known as slow-release carbohydrates) instead. Slow-release carbohydrates help keep blood sugar levels even because they are digested more slowly, thus preventing the body from producing too much insulin. Increasing fibre content decreases the glycaemic index (GI) of foods [30].

Decreased GI leads to smaller increases in blood glucose and thus, reduces blood glucose glycosylated hemoglobin (HbA1c) levels. Fibre-rich foods are one of the specific diets highlighted by the American Diabetes Association as part of medical nutrition therapy for secondary and tertiary prevention of diabetes mellitus [31]. There are two main objectives of this study: One, to produce a new dietary regime (consisting of cassava fufu fortified with tiger nuts) for Type 2 diabetic patients and test its suitability and acceptability by diabetic patients. Two, to investigate how persuasive technology can be used the improve the intake of the new dietary regime in a sample of 30-diabetes mellitus patients in Nigeria by sending them a computer-tailored persuasive message that reminds them the need to take the new dietary regime regularly and highlights the benefits of the new dietary regime. To achieve this, we produced flour from tiger nuts(TNF) and blended it with yellow cassava flour(YCF) at different levels of substitution (YCF:TNF - 100:0, 90:10, 80:20, 70:30, 60:40, 50:50).

The flour blends were analyzed for their functional, proximate, anti-nutritional and microbial properties using standard analytical methods for ten weeks. The sensory properties (color, flavour, taste, aftertaste, mouthfeel and overall acceptability) of the improved fufu produced from the different flour blends were also determined. To ensure that patients maintain regular dietary intake for 4 weeks, a tailored persuasive messages was sent to the patients as a follow up strategy.

There were significant differences ($p<0.05$) in their proximate composition (moisture; 5.38±0.06 to 7.56±0.08, protein; 4.50±0.16 to 10.10±0.09, fat; 1.51±0.01 to 6.04±0.03, ash; 1.00±0.03 to 2.92±0.01, fibre; 3.95±0.05 to 6.00±0.01, carbohydrate; 82.51±0.21 to 70.38±0.02). From the sensory analysis carried out by the patients, sample F (YCF: TNF - 50:50), had the highest rating in all the parameters tested including their overall acceptability. We conclude that yellow cassava could be improved with tiger nuts flour and made

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suitable and acceptable by the diabetics' patients and that computer-tailored persuasive messages can be used to improve patients' acceptance and ensure patients adherence to their dietary regime.

This paper made the following contributions: First, we produce flour from tiger nuts and obtained flour blends at various levels of substitution of yellow cassava flour with tiger nuts flour (YCF:TNF - 100:0, 90:10, 80:20, 70:30, 60:40, 50:50). Second, we determine physicochemical, proximate, toxicological and microbial properties of the flour blends, and produced improve and 'instant' yellow cassava fufu fortified with tiger nuts from the different flour blends. To determine the acceptability of the various flour blends, we administer them to a sample of 30 diabetic patients that were randomly selected from five hospitals in Nigeria; University of Nigeria Teaching Hospital Ituku-Ozalla (UNTH) Enugu, Federal Teaching Hospital Abakaliki (FETHA), Federal Medical Center (FMC) Owerri, Federal Medical Centre, Abakaliki, (FMCABA) and NnamdiAzikiwe University Teaching Hospital(NAUTH) Nnewi. Fourth, to encourage the patients to maintain a regular intake of the new dietary regime, we delivered a computer-tailored persuasive message that reminds them the need to take the new dietary regime and highlights the benefits after the patients have determined the sensory properties of the improved products.

The use of persuasive technology to tailor health messages to characteristics of individuals has been applied to various health related behaviors, most often in smoking cessation interventions [32-34], but also to increase participation in breast cancer screening [35], and to help people to adopt healthier diets [36]. Researchers who invited people with diabetes to describe their experiences have concluded that self-management is culturally embedded, dependent on core knowledge and understanding, and improves with family and social support [37-45]. In this research, the process of providing the 30-sampled diabetic patients with a computer-tailored persuasive message follows similar approach that was described by Brug et al. [46].

The researchers believe that this research will serve as an eye opener to the general public on the nutritional and therapeutic value of both yellow cassava and tiger nuts and how dietary fibre from tiger nuts can be used as an alternative to other food sources of dietary fibre. Also, the Good Manufacturing Practices (GMPs), required for the production and detoxification of cassava and tiger nuts is made known. We also determine the proximate, physicochemical, microbial, and toxicological assessment of the composite flour and the sensory evaluation of the finished product. The study also demonstrates the capability of computer-tailored persuasive messages for improving adherence and acceptability of dietary regime among diabetic patients

Materials and Methods

A Completely Randomized Design (CRD) was used to study the chemical composition of the blended flours and the sensory properties of the Cupcakes produced from the flour blends. The researchers used One-way Analysis of Variance (ANOVA) to measure the physicochemical, proximate, toxicological and microbial properties across the flour blends of yellow cassava with tiger nuts, and the sensory survey scores from the patients about the improved cassava fufu fortified with tiger nuts from the different flour blends. Means that where significantly different were separated by the Least Significant Difference (LSD) test and the significance level of $p<0.05$ was used. Prior to conducting the analysis, the researchers ensured that there were no missing values and screened for ANOVA assumption violations using the SPSS 16 software (SPSS Inc., Chicago, IL, USA).

We used approximately 10 weeks to test the physicochemical, proximate, toxicological and microbial properties across the flour blends of yellow cassava with tiger nuts. Following from that, we conducted follow-up evaluations of all the diabetic patients at their different hospitals over a period of 4 weeks. The evaluation involved sensory evaluation of the improved cassava fufu by the patients and completing the self-report questionnaires to measure the suitability and acceptability of a new dietary. The survey results were entered into a data file and computer software was written that linked the data with a feedback source in form of a tailored persuasive message for each patient.

In addition to the usual laboratory equipment, the following apparatus were used: Centrifuge was used for determining water absorption capacity and oil absorption capacity; Hammer mill was used for milling or grinding of tiger nuts; Water bath was used for boiling water; Muffle furnace was used for determining ash content of the flour samples; Spectrophotometer was used for determining the toxicological content of the flour samples and Reagents was used for determining protein content of the flour samples.

Production of improved cassava fufu

Cassava fufu is known to be low in nutrients, except for its carbohydrate content. Therefore, improving cassava fufu, made from yellow cassava (Pro-vitamin A cassava) by fortifying with whole tiger nuts flour will be of great benefit to Nigerians, especially the diabetic patients. According to Proft [47], a project called "BioCassava Plus" which involves developing a cassava with lower cyanogen glucosides and fortified with vitamin A, iron, and protein could help the nutrition needs of people in sub-Saharan Africa. The diagram of conventional cassava and vitamin A cassava is shown in Figure 1 below.

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Figure 1: White and Yellow Cassava, Egesi [24].

Dried tiger nuts were purchased from the local/street vendors. Yellow cassava was gotten from Nigeria's National Root Crop Research Centre, Umudike (NNRCRC). The dried tiger nuts were sorted to remove bad and damaged tubers and extraneous materials - stones, metal scraps etc. The clean tubers were thoroughly washed with clean water and sundried for one week. After sun drying, the clean and dried tubers were broken using a hammer mill. The broken tiger nuts were reconstituted with water and dried in an oven for 2 hours at 60 °C. The pre-gelled, broken and dried tubers were cooled and milled into flour.

Yellow cassava flour on the other hand was reconstituted on the basis (200g of flour with about 300mls of water), to form a paste. The yellow cassava paste was then oven-dried at 60 °C for 2 hours. The pre-gelled and dried yellow cassava was then pulverized/milled into flour. The flow diagram below in Figure 2 represents the formulation of improved cassava fufu. The two flours (i.e. Tiger nuts and yellow cassava flours) were blended as shown in Figure 2 and Figure 3 below, to get improved cassava flour for fufu at various levels of substitution.

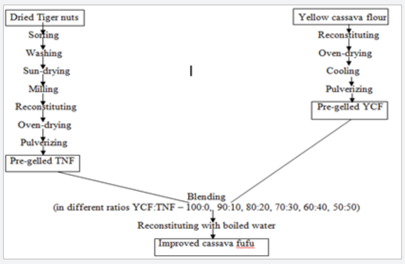

Figure 2: shows the formulation of improved cassava fufu

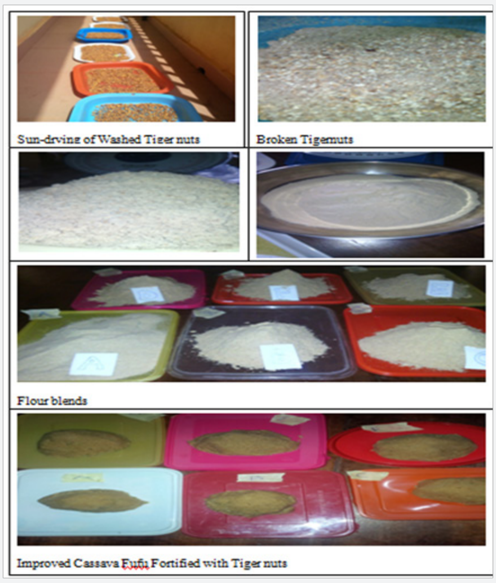

Figure 3: Pictures of the stages of formulation of improved yellow cassava fufu fortified with tiger nuts.

Evaluation of the flour samples

We evaluated the flour blends to determine the physicochemical properties, proximate composition, microbial analysis, toxicological/anti-nutrient properties, and sensory evaluation and the effect of the persuasive technology (Computer-tailored Messages) on the acceptance and regular intake of the new flour blends. The physicochemical properties of the flour blends were determined by calculating their bulk density (BD), water absorption capacity (WAC), oil absorption capacity (OAC) and least gelation concentration (LGC) respectively as shown in Table 1 below. First, bulk density was determined using the method described in Onwuka [48]. Flour sample (50g) was put into a 100ml measuring cylinder. The cylinder was tapped several times on a laboratory bench to a constant volume. The volume of sample recorded.

$$\text{Bulk density } (g/cm^3) = \frac{\text{Weight of sample}}{\text{Volume of sample after tapping}} \quad (1)$$

Table 1: Physicochemical Properties of the Flour Samples.

Sample(YCF:TNF)	Bulk Density (g/cm3)	WAC (ml)	OAC (ml)	LGC (%)
A(100:0)	0.94^{d} ± 0.01	3.50^{a} ± 0.49	1.15^{a} ± 0.70	18.00^{b} ± 0.00
B (90:10)	0.81^{c} ± 0.03	3.35^{a} ± 0.21	1.05^{a} ± 0.21	18.00^{b} ± 0.00
C (80:20)	0.81^{c} ± 0.00	3.25^{a} ± 0.21	1.00^{a} ± 0.14	18.00^{b} ± 0.00
D (70:30)	0.79^{bc} ± 0.00	3.20^{a} ± 0.28	0.90^{a} ± 0.14	16.00^{a} ± 0.00
E (60:40)	0.78^{b} ± 0.01	2.80^{a} ± 0.57	0.90^{a} ± 0.14	16.00^{a} ± 0.00
F (50:50)	0.71^{a} ± 0.01	2.65^{a} ± 0.49	0.85^{a} ± 0.21	16.00^{a} ± 0.00

* Values are means of duplicate samples ± standard deviation. Means with the same superscript in the same column are not significantly (p>0.05) different. The mean different is significant at the 0.05 level. WAC= water absorption capacity, OAC= oil absorption capacity, LGC= least gelation concentration.

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Second, water absorption capacity (WAC) was determined using the procedure described in Sathe and Salunkhe [49]. Distilled water (15ml) was added to 1g of the flour in a weighed 25ml centrifuge tube. The tube was agitated on a vortex mixer for 2min. It was centrifuged at 4000rpm for 20min. The clear supernatant was decanted and discarded. The adhering drops of water were removed and then reweighed. Water absorption capacity is expressed as the weight of water bound by 100g dried flour. (This is to determine the ability of the flour to entrap or absorb water, it shows if the flour has good binding water capacity). Third, oil absorption capacity was determined using the procedure described in Sathe and Salunkhe [49]. One-gram flour was mixed with 10ml refined vegetable oil and allowed to stand at ambient temperature for 30min. The centrifuged for 30min at 2000rpm. Oil absorption capacity is expressed as % water or oil bound per gram flour. (It determines the ability of the flour to absorb oil). Fourth, least gelation concentration was determined by a modified method of Coffman and Garcia [50]. Appropriate sample suspensions 2, 4, 6, 8, 10, 12, 14, 16, 18 and 20% (W/V) was prepared in distilled water. The test tubes containing these suspensions were heated to 90 °C for one hour in a water bath, followed by rapid cooling under running tap water. The test tubes were is cooled at 10±2 °C for 2 hours. The least gelation concentration was taken as the concentration when the sample from inverted test tube did not fall down or slip. (Significance: it determines the index of gelation capacity of the flour).

Proximate composition of the flour blends were determined by calculating the moisture, protein, fat, protein, fiber, carbohydrate, and ash contents of the flour blends respectively as shown in Table 2. First, moisture content was determined by the oven method as described in AOAC [51]. Five grams of the prepared samples were weighed in triplicate into Petri-dishes of known weigh and covered immediately. These were transferred into the oven, uncovered and heated at 105±2 °C for 2 hours. The samples were then removed and placed in desiccators and allowed to cool for 15 minutes before weighing. This was repeated until constant weights are recorded. The loss in weight from the original weight (before drying) was reported as the moisture content.

$$\% \text{ Moisture Content} = \frac{w_1\ w_2}{w_1} x\,100 \quad \ldots\ldots\ldots\ldots\ldots\ldots (2)$$

Where, W1 = Weight of wet sample, W2 = Weight of dried sample.

Table 2: Proximate Composition of the Composite Flour Samples.

	Moisture(%)	Protein(%)	Fat(%)	Ash(%)	Fibre(%)	CHO(%)
A(100:0)	5.84^{B} ± 0.05	4.50^{A} ± 0.16	1.51^{A} ± 0.01	1.00^{A} ± 0.03	3.95^{A} ± 0.05	82.51^{F} ± 0.21
B (90:10)	7.56^{E} ± 0.08	4.94^{B} ± 0.04	3.81^{B} ± 0.02	1.01^{A} ± 0.03	4.65^{B} ± 0.06	75.13^{E} ± 0.06
C (80:20)	6.30^{C} ± 0.03	6.91^{C} ± 0.01	4.48^{C} ± 0.04	1.07^{A} ± 0.10	4.73^{B} ± 0.06	77.89^{E} ± 0.15
D (70:30)	5.38^{A} ± 0.06	8.49^{D} ± 0.08	5.84^{D} ± 0.06	1.95^{B} ± 0.01	5.74^{C} ± 0.04	71.44^{C} ± 0.11
E (60:40)	6.60^{D} ± 1.56	9.89^{E} ± 0.08	5.96^{E} ± 0.04	1.99^{B} ± 0.02	5.95^{D} ± 0.01	70.38^{A} ± 0.02
F (50:50)	6.53^{D} ± 0.06	10.10^{E} ± 0.09	6.04^{E} ± 0.03	2.92^{C} ± 0.01	6.00^{D} ± 0.01	71.06^{B} ± 0.16

*Values are means of duplicate samples ± standard deviation. Means with the same superscript in the same column are not significantly (p>0.05) different. The mean different is significant at the 0.05 level. (YCF: TNF) - Yellow cassava flour - Tigernut flour, CHO - Carbohydrates.

Second, the Kjeldahl procedure outlined by the AOAC [51] was used to digest the prepared samples system under a fume chamber. The digested samples were then allowed to cool and then distilled in boric acid containing Bromocresol indicator, after being appropriately diluted first with water and later with Sodium Thiosulphate. The samples were then titrated against 0.1M Hcl solution. Crude protein content was estimated by titration and its percentage was calculated as following:

$$\% \text{ Crude Protein} = \frac{(Title\ value\ x\,14.01\ x\ mol\ of\ acid\ (0.1N)\ x\ 6.25\ x5}{Weight\ of\ sample} \quad \ldots\ldots\ldots(3)$$

Third, the Fat content was determined using the method described in [51]. Five grams of the prepared samples were weighed into extraction thimbles and fixed into extraction flask of known weights. Extraction was carried out using petroleum ether on electro-thermal model equipment for eight hours. At the completion of the extraction, the petroleum ether was removed by evaporation on an electrical bath and the remaining fat in the flask dried at 60 °C for 30 minutes in the oven, cooled for 15 minutes and weighed. The fat content (%) was calculated as follows:

$$\% \text{ Fat Content} = \frac{w_3 w_1}{w_2} x\,100 \ldots\ldots\ldots\ldots\ldots\ldots(4)$$

Where: W1 = initial weight of dry soxhlet flax, W2 = weight of sample taken, W3 = final weight of dry flask + fat.

Fourth, the Crude fibre content was determined following the procedure outlined in [51]. Five grammes of the samples were extracted using petroleum ether. This was followed by sequential digestion of sample with 1.25% H2SO4 and 1.25% NaOH; after which the samples were filtered through the California Buchner System. The resulting residue was dried at 105±2 °C for 1 hour, cooled in a dessicator and weighed. The dried, cooled and weighed residue were then transferred into a muffle furnace and ignited at 600±2oC for 30 mins, cooled and re-weighed. % Crude fiber = $\frac{w_3 w_1}{w_2} x\,100$ .... (5)

W1 = Weight of the crucible, W2 = Weight of sample use, W3 = Weight of crucible and fiber.

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Fifth, the Ash content was determined using the method described in [51]. Five grams of sample were weighed in triplicates into ash dishes that had been previously weighed. The dishes were placed in the furnace and ignited at 550±2oC for 5 hours, cooled and weighed to constant weight.

The resulting ash was calculated as follows;

$$\% \; Ash = \frac{w_1 w_2}{w_3} x\, 100 \text{ ....(6)}$$

Where, W1=Weight after ash sample, W2 = Weight of crucible, W3 = Weight of sample used.

Sixth, the Carbohydrate was calculated by difference as described by Rampersad et al. [52]. 100 - (% Moisture+%Crude protein+%Crude fiber+% Ash+%Crude fat). They were separated by the Least Significant Difference (LSD) test and the significance will be accepted at p<0.05, [53]. Microbiological analysis were carried out on the sample flour blends by determining the total viable (TVC), coliform, and mould count respectively as shown in Table 3 below. For determining, total viable count, the pour plate method as described in Harrigan and McCance [54] was used.

Table 3: TVC, Coliform count and Mould count (Microbial Analysis).

Samples (YCF:TNF)	*TVC (cfu/ml)	Mould Count (cfu/ml)	Coliform Count(cfu/ ml)
A(100:0)	1.4 X 106	-	-
B(90:10)	1.9 X 106	-	-
C(80:20)	1.2 X 106	3.5 X 105	-
D(70:30)	7.5 X 105	-	-
E(60:40)	5.5 X 105	4.5 X 105	-
F(50:50)	2.1 X 105	-	-

Key=> (YCF: TNF) - Yellow cassava flour: Tigernut flour. *TVC - total viable count. The mean different is significant at the 0.05 level.

One gram of the samples was macerated into 9ml of Ringers solution and mixed thoroughly by shaking. This was further diluted to obtain 10-2 and 10-3 concentration. Then 0.1ml dilution was transferred from each dilution bottle into the corresponding plate and 15ml of sterile nutrient agar medium was poured and mixed thoroughly with the inoculums by rocking the plates. The plates were incubated at 38 °C for 24hours after which the colonies formed were counted and expressed as colony forming units per gram (cfu/g). Similarly, the same method was used to determine coliform count by putting Nine milliliters of sterilized violet red bile agar into each plate containing 1ml of inoculums from 10-3 dilution.

The plate was shaken gently to mix with the content properly and then it was allowed to set and subsequently incubated at 37 °C for 72 hours. After the incubation, the number of colonies which appeared in dark red or pink centres was counted. This was expressed as colony forming units per gram (cfu/g). In the same manner, the sample dilution weighing 0.1ml was transferred from each dilution into corresponding plates and 15ml of sterile Sabourand Dextrose Agar (SDA) medium was poured and mixed thoroughly with the inoculums by rocking the plates. The plates were incubated at ambient temperature for three days after each colony formed were counted and expressed as colony forming units per gram (cfu/g).

The Tannin, Phenols and Hydrogen Cyanide contents respectively were determined from the flour sample as shown in Table 4 below. Tannin content was determined by the Folin - Denis colorimetric method described in Kirk and Sawyer [55]. Five-gram sample was dispersed in 50ml of distilled water and shaken. The mixture was allowed to stand for 30mins at 28 °C before it was filtered through Whatman No. 42 grade of filter paper. Two millilitres of the extract were dispersed into a 50ml volumetric flask. Similarly, 2ml standard tannin solution (tannic acid) and 2ml of distilled water were put in a separate volumetric flask to serve as standard and reagent was added to each of the flask and then 2.5ml of saturated Na2CO3 solution was added. The content of each flask was made up to 50ml with distilled water and allowed to incubate at 28 °C for 90mins.

Table 4: Toxicological/ Anti-nutrient properties of the flour samples.

Samples (YCF:TNF)	Hydrogen Cyanide (HCN) (ppm)	Tannin (%)	Phenol (%)
A(100:0)	25.14a ± 0.31	0.07a ± 0.00	0.027b ± 0.00
B(90:10)	447.59e ± 3.93	0.06a ± 0.00	0.046c ± 0.00
C(80:20)	178.02d ± 0.57	0.07a ± 0.00	0.04bc ± 0.00
D(70:30)	141.83b ± 1.75	0.09b ± 0.00	0.01a ± 0.00
E(60:40)	16 1.58c ± 1.79	0.11c ± 0.00	0.05c ± 0.00
F(50:50)	182.52d ± 0.12	0.11c ± 0.01	0.04bc ± 0.01

*Values are means of duplicate samples ± standard deviation. Means with the same superscript in the same column are not significantly (p>0.05) different. (YCF: TNF) - Yellow cassava flour: Tiger-nut flour; ppm - parts per million.

Their respective absorbance was measured in the spectrophotometer at 260nm using the reagent blank to calibrate the instrument at zero. Similarly, the total phenolic content of the flour samples was determined with Folin-Ciocalteu reagent according to the method described in Singleton & Rossi [56], with slight modification. A 25-μL aliquot of each beverage was mixed with 0.25mL of Folin-Ciocalteu reagent and incubated at room temperature. After 5min, 0.25mL of 10% sodium bicarbonate (Na CO) solution was added, and the mixture was allowed to stand in the dark for 30 min at 37 °C. The absorbance of the incubated mixture was measured at 750nm using a JENWAY UV-visible spectrophotometer (JENWAY Inc., Staffordshire, UK).

A standard curve was prepared and used to translate the measured absorbance values to gallic acid equivalents (GAE). In addition, the total cyanide content of the flour samples was also determined in triplicates by the colormetric Alkaline Picrate method of Ikediobiet et al. [57]. The yellowish

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alkaline picrate was prepared by dissolving 1g picric acid and 5g sodium carbonate in distilled water. The liquid filtrate (1.0ml) from the cyanide extraction process was added to 4.0ml alkaline picrate solution in a test tube and corked. The mixture was incubated at 50 ºC for 5minutes to allow for color development. After color development (from yellowish color to reddish color) and cooling, the absorbance was read at 490nm wavelength with UV/Visible spectrophotometer (Jenway 6405, England). Diluted potassium cyanide (KCN) was used to prepare the standard curve that was employed in calculating the cyanide content of the experimental samples.

Sensory analysis was carried out on the improved cassava fufu produced from the flour blends using 9-point hedonic scale rating (where '9' was regarded as extremely like and '1' as extremely dislikes). 30 diabetic patients were sampled from the four hospitals in Nigeria (University of Nigeria Teaching Hospital Ituku-Ozalla (UNTH) Enugu, Federal Teaching Hospital Abakaliki (FETHA), Federal Medical Center (FMC) Owerri, Federal Medical Centre, Abakaliki, (FMCABA) and NnamdiAzikiwe University Teaching Hospital(NAUTH) Nnewi) and they evaluated and scored the product based on color, flavour, taste, aftertaste, mouthfeel/texture and overall acceptability. The patients were surveyed with a baseline questionnaire, and the survey results were entered into a data file. The survey results are shown in Table 5 below.

Table 5: The result of the sensory attributes of the improved fufu samples.

Samples (YCF:TNF)	Colour	Flavour	Taste	Aftertaste Texture	Mouthfeel/ Acceptability	Overall
A(100:0)	6.57a ± 2.11	7.00a ± 1.11	5.77a ± 1.63	5.83a ± 1.56	4.97a ± 1.69	6.27a ± 1.55
B(90:10)	6.53a ± 1.91	7.07a ± 1.14	6.07a ± 1.34	6.20ab ± 1.35	5.67ab ± 1.45	6.77a ± 1.10
C(80:20)	6.80ab ± 1.64	6.93a ± 1.53	6.47a ± 1.38	6.23abc ± 1.33	6.20b ± 1.16	6.70a ± 1.02
D(70:30)	7.43bc ± 0.90	7.10a ± 1.12	7.20b ± 1.03	6.77bc ± 1.07	7.03c ± 1.22	7.70b ± 0.65
E(60:40)	7.67c ± 1.06	7.13a ± 1.28	7.20b ± 1.54	6.77bc ± 1.70	7.47c ± 1.63	7.60b ± 1.45
F(50:50)	7.57bc ± 1.10	7.40a ± 1.19	7.37b ± 1.13	7.00c ± 1.29	7.57c ± 1.28	8.17b ± 0.91

* Values are means of duplicate samples ± standard deviation. Means with the same superscript in the same column are not significantly ($p>0.05$) different. (YCF: TNF) - Yellow cassava flour: Tiger-nut flour.

Following the patients evaluation, computer software was written that linked the patient's responses with a feedback source in form a message for each message. The computer program generates message tailored to individual patients based on his/her survey results. However, in this study, the patients were surveyed with a baseline questionnaire by evaluating and scoring the flour blends based on the sensory evaluation of six parameters which are color, flavor, taste, aftertaste, mouthfeel/texture and overall acceptability. It was indicated that sample F (YCF: TNF - 50: 50) has the highest preference as indicated by the patients. Therefore, personalized dietary and psychosocial messages sent to patients are based on patients' sensory evaluations on the different sample of the diet. The process of the computer-tailored message is summarized in Figure 4 similar to the other studies [46,58-63].

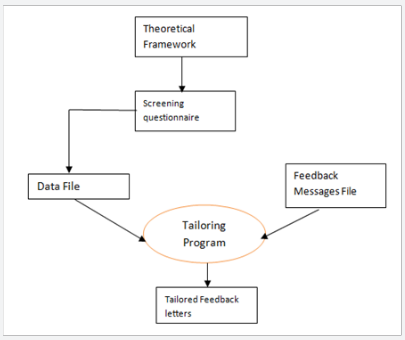

Figure 4: Process of Computer Tailored Message Brug et al [46].

Results and Discussion

Table 1 show that the mean bulk density decreased from 0.94 - 0.71g/cm3. There is a statistically significant ($p<0.05$) difference between the bulk density group means of the flour samples. This means that they have different packaging characteristics. WAC and OAC of the composite flours ranged

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from 3.50 - 2.65ml and 1.15 - 0.85ml respectively. The rate at which each sample absorbed water and oil decreased with increased tiger-nut substitution, however, there is no significant ($p>0.05$) difference in the WAC and OAC of the flours. The LGC of the flour samples decreased from 18.00 - 16.00% as the tiger-nut content increased.

The proximate composition of the flour samples as represented in Table 2 shows that there is a statistically significant ($p<0.05$) difference between the group means' proximate composition of the flour samples as determined by one-way ANOVA in Table 6 below. For protein content of the flour sample, (F (5, 6)= 1.408E3, p = 0.000); for ash content, (F (5, 6)= 584.507, p = 0.000); for moisture content, (F (5, 6)= 162.154, p = 0.000); for fibre content, (F (5, 6)= 724.444, p = 0.000); for fat content, (F (5, 6)= 4.203E3, p = 0.000); and carbohydrate content, (F (5, 6)= 2.576E3, p = 0.000).

Table 6: Summary statistics from One-Way ANOVA on proximate composition of the flour samples.

ANOVA						
		Sum of Squares	**df**	**Mean Square**	**F**	**Sig.**
PROTEIN	Between Groups	58.859	5	11.772	1.408E3	0.000
	Within Groups	0.05	6	0.008		
	Total	58.909	11			
ASH	Between Groups	5.967	5	1.193	584.507	0.000
	Within Groups	0.012	6	0.002		
	Total	5.979	11			
MOISTURE	Between Groups	5.506	5	1.101	162.154	0.000
	Within Groups	0.041	6	0.007		
	Total	5.547	11			
FIBRE	Between Groups	7.184	5	1.437	724.444	0.000
	Within Groups	0.012	6	0.002		
	Total	7.196	11			
FAT	Between Groups	31.348	5	6.270	4.203E3	0.000
	Within Groups	0.009	6	0.001		
	Total	31.356	11			
CHO	Between Groups	227.869	5	45.574	2.576E3	0.000
	Within Groups	0.106	6	0.018		
	Total	227.975	11			

Moreover, the researchers sought to find out the level of the significant that exists between each content of the compositions of the flour samples (A-F) of yellow cassava flour (YCF) and tiger nuts flour (TNF), using a Duncan post hoc test. It revealed that the protein (10.10e±0.09 in Table 2, p = 0.00) and fibre (6.00d±0.01 in Table 2, p = 0.00) contents of the flour samples were statistically higher after adding the tiger nuts in each sample of flour blends compared to the protein (4.50a±0.16 in Table 2, p = 0.00) and fibre (3.95a±0.05 in Table 2, p = 0.00), when tiger nuts were not added. Conversely, it was also revealed that the carbohydrate content (71.06b±0.16) of the flour sample were statistically lower after adding the tiger nuts compared to its contents (82.51f±0.21) when tiger nuts were not added.

The protein content of the flour samples increased from 4.50 - 10.10% with increased tiger-nut substitution. Moisture and carbohydrate contents of the composite flours reduced while the ash content increased. It is worthy of note that increase in tiger-nut substitution enhanced the fibre content of the flour blends. Increase in fibre and fat content can be attributed to high fibre contents of tiger-nut flour [64]. However, the intake of fibre among people living in Nigeria remains low.

Similarly, dietary fibre intake among people living in Western countries, also, remains low, and according to the Third National Health and Nutrition Examination Survey (1998), fibre intakes averages 17g per day in the United States. Although patients with diabetes are advised to increase their intake of dietary fibre, in the NHANES study [65], their average daily intake was found to be only 16g. Chandalia et al. [66] found that the high-fibre diet improved glycaemic control, as evidenced by decreases in the mean daily pre-prandial and 24-hour plasma glucose concentrations. Urinary glucose excretion was also lowered by the high-fibre diet. The high-fibre diet lowered glycosylated haemoglobin values slightly but not significantly. The high-fibre diet also lowered 24-hour plasma insulin concentrations. Other intervention details involving fibre supplementation for type 2 diabetes mellitus can reduce fasting blood glucose and blood glucose glycosylated haemoglobin (HbA1c) and suggested that increasing dietary fibre in the diet of patients with type 2 diabetes is beneficial

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and should be encouraged as a disease management strategy is found in these studies [67-71].

Why the intake of dietary fibre in patients with diabetes mellitus remains low, despite its well-documented effect of lowering plasma glucose concentrations, remains unexplained. As a result, we sought to use persuasive technology to persuade the diabetic mellitus patients about the intake of dietary fibre. Firstly, the patients from the five hospitals were provided with samples (A-F) of improved cassava fufu. They took a sip of water and rinsed their mouth between sample tests. Sensory evaluations were carried out on the improved cassava fufu produced from the flour blends (A-F) by the patients as a follow up intervention and lasted for five weeks.

The patients were surveyed with a baseline questionnaire by evaluating and scoring the flour blends based on the sensory evaluation of six parameters: color, flavour, taste, aftertaste, mouthfeel/texture and overall acceptability using 9- likert scale (9= Like extremely, 8= Like very much, 7= Like moderately, 6= Like slightly, 5= Neither like nor dislike, 4= Dislike slightly, 3= Dislike moderately, 2= Dislike very much, 1= Dislike extremely). The patients also indicated the sample they liked best, reasons, and other comments on the samples. The summary statistics from one-way analysis of variance (ANOVA) on the sensory evaluation results of the sensory attributes of the flour samples (A-F) by the patients are represented in Table 7 below.

Table 7: Summary statistics from one-way analysis of variance (ANOVA) on the sensory evaluation of the flour samples

ANOVA						
		Sum of Squares	df	Mean Square	F	Sig.
COLOUR	Between Groups	40.361	5	8.072	3.468	0.005
	Within Groups	405.033	174	2.328		
	Total	445.394	179			
FLAVOUR	Between Groups	3.894	5	0.779	0.507	0.77
	Within Groups	267.1	174	1.535		
	Total	270.994	179			
TASTE	Between Groups	68.044	5	13.609	7.371	0.000
	Within Groups	321.267	174	1.846		
	Total	389.311	179			
AFTERTASTE	Between Groups	29.733	5	5.947	3.052	0.011
	Within Groups	339.067	174	1.949		
	Total	368.8	179			
MOUTHFEEL	Between Groups	164.717	5	32.943	16.367	0.000
	Within Groups	350.233	174	2.013		
	Total	514.95	179			
OVERALL	Between Groups	79.6	5	15.920	11.879	0.00
ACCEPTANCE	Within Groups	233.2	174	1.340		
	Total	312.8	179			

It shows that there is a statistically significant difference ($p<0.05$) between the group means sensory attributes of the flour samples as determined by one-way ANOVA. For color content of the flour sample ($F(5, 174)= 3.468$, $p = 0.005$); for flavor content ($F(5, 174)= 0.507$, $p = 0.770$); for taste content ($F(5, 174)= 7.371$, $p = 0.000$); for after test content ($F(5, 174)= 3.052$, $p = 0.011$); for mouth feel ($F(5, 174)= 16.367$, $p = 0.000$); and overall acceptability ($F(5, 174)= 11.879$, $p = 0.000$).

To examine the flour samples ((A-F) of yellow cassava flour (YCF) and tiger nuts flour (TNF)) that significantly differ from each other; we used the Duncan post hoc test. The test revealed that the color (7.57bc±1. p = 0.005), after test (7.00c±1.29, p=0.011) from Table 5, and overall acceptability (8.17b±0.91, p = 0.00) from Table 4. 47, attributes of the flour samples were statistically higher after adding the tiger nuts in each sample of flour blends compared to the color (6.57a±2.11 in Table 5, p = 0.005), after test (5.83a±1.56 in Table 5, p=0.011) and overall acceptability (6.27a±1.55 in Table 5, p= 0.000) when tiger nuts were not added. Conversely, it was also revealed that the carbohydrate content (71.06b±0.16) of the flour samples were statistically lower after adding the tiger nuts compared to its contents (82.51f±0.21) when tiger nuts were not added. There was no statistically significant difference between the flavor of the flour samples (P = 0.770).

Figure 5 shows the comparison chart for sensory evaluation of the mean scores of the improved cassava fufu samples. The color for sample E (YCF: TNF - 60:40) was most preferred followed by sample F (YCF: TNF - 50:50), however, it was also reported that higher protein content is associated with darker products Fiszman et al. [72] and from the flour samples,

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sample F is darker. There is no significant difference (p<0.05) in the flavor of all the samples and in all the parameters tested, excluding color, sample F (YCF: TNF – 50: 50) received the highest rating.

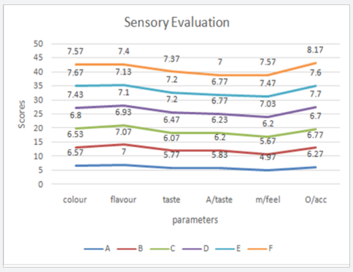

Figure 5: Comparison Chart for Sensory Evaluation Scores.

The survey results were entered into a data file and tailored messages were sent to each individual patients based on his/her survey results to motivate them to maintain a regular intake of the new fufu dietary following similar procedures highlighted by Brug et al. [46] on the application and impact of computer-generated personalized nutrition education. Similarly, it is in line with research conducted by Cialdini [58] that we showed the tailored messages sent to diabetic patients. The tailored messages sent to the patients as shown in Table 8 were derived from the six persuasive principles.

In this study, diabetic patients sampled beliefs that all the parameters tested, sample F (YCF: TNF - 50: 50) has the highest preference. Therefore, personalized dietary and psychosocial messages as shown in Table 8 sent to these patients are more likely to be read, remembered, and seen as personally relevant and above all, have greater effects in motivating these patients to change their dietary intake.

Conclusion

In this work, improved cassava fufu was produced with yellow cassava and tiger-nut flour blends at various levels of substitution. The flours (A-F) were analyzed for their functional, nutrient/proximate composition, anti-nutrient and microbial properties. Results obtained from these analyses showed notable changes in the functional properties and increase in protein, ash, fibre and fat. The samples (F) that contain the highest percentage of tiger-nut substitution were preferred most in all the parameters tested. Thus, fortifying of yellow cassava with this 'high-fibre' tiger-nut would increase their nutritional value. The microbial analysis carried out showed no trace of fecal contamination and that the product can be kept for a long. The increase in tiger nut fat should not raise any alarm because tiger-nut oil is unsaturated oil which lowers the low density lipo-protein (LDLP); the bad cholesterol and increase high density lipo-protein (HDLP); good cholesterol. Hence, it reduces the levels of triglycerides in blood and the risk of forming bloody clots, thereby preventing arteriosclerosis.

Thus, tiger-nut fat is 'heart-friendly'. In addition, the reduction in LGC (least gelation concentration) of the flour samples with increased tiger nuts substitution indicates that the samples that have higher content of tiger nuts will take lesser time to gel or get done. The results also showed that inclusion of tiger nuts to yellow cassava fufu at levels 10-50% brought about increase in the protein, fat, ash and fibre. Increase in dietary fibre and decrease in carbohydrate contents of the flour blends are of utmost importance in this work. 30 diabetic patients were sampled from four hospitals in Nigeria and the patients evaluated and scored the product based on color, flavor, taste, aftertaste, mouth feel/texture and overall acceptability.

The patients were surveyed with a baseline questionnaire, and the survey results were entered into a data file. Tailored messages were sent to each individual patients based on his/her survey results in order to persuade them to maintain the regular intake of the dietary fibre. Therefore, the intended aim of this study; to produce and evaluate improved cassava fufu from yellow cassava tiger-nut composite flour that will add to the diet of the diabetic patients and persuade the patients to maintain the regular intakes of the improved product with high fibre using persuasive technology were actualized.

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