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Evaluation of nutrient composition, glyceamic index and anti-diabetic potentials of multi-plant based functional foods in rats

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Nutrition transition, that is, increase in consumption of high energy-dense foods, with low consumption of fruits and vegetables, has been implicated as the major factors responsible for the increase in prevalence of diet-related diseases such as diabetes and cardiovascular diseases worldwide. Evidence has shown that consumption of plant-based foods prevents the risk of these chronic diseases, hence, the present study formulated and evaluated nutrient compositions and antidiabetic potentials of multi-plant based functional foods from locally available food materials. Food materials (popcorn, moringa leaves, wonderful kola and defatted soybean) were obtained from reputable farms and markets in Akure, Nigeria. The food materials were processed as raw, blanched and fermented flour samples, and blended to obtain nine samples, i.e., R_1, B_1 and F_1 (popcorn 60%, soybean 10%, moringa 20% and groundnut oil 10%), R_2, B_2 and F_2 (popcorn 60%, soybean 10%, wonderful kola 20% and groundnut oil 10%) and R_3, B_3 and F_3 (popcorn 60%, soybean 10%, moringa 10%, wonderful kola 10% and groundnut oil 10%) using Nutri-Survey software. Proximate and mineral compositions of the samples were determined using standard methods. Glycemic index and anti-diabetic potentials were determined using rat models. Proximate compositions (g/100g) of the formulated multi-plant based functional foods were as follows: moisture contents ranged from 6.29 in F_3 to 8.27 in R_3, crude fiber contents from 2.79 in F_2 to 4.68 in B_3 and crude protein contents from 23.22 in B_1 to 30.39 in F_3, while carbohydrate content of the formulations ranged from 52.10 in F_3 sample to 56.94 in B_2 sample, while energy values were between 421.1 in R_1 and 433.7 kcal in B_1. The food samples contain appreciable amount of mineral concentrations require for biochemical activities. Glycaemic index (GI) of the formulated functional foods ranged from 36.98% in R_3 to 44.9.9% in R_1 sample, and were significantly (<0.05) lower than in glucose (a reference sample). The glycaemic load (GL) of F_1 sample had the lowest value (20.3), while B_2 sample had the highest value (25.6). The percentage blood glucose reduction of diabetic-induced rats fed with R_1 samples (63.8%) had the highest values; while those rats fed with B_2 sample (24.1%) had the lowest blood glucose reduction. Statistically, the percentage blood glucose reduction of the formulated functional foods, particularly R_1 and F_1, were comparable to metformin (a synthetic anti-diabetic drug) in terms of antidiabetic activities. The study reported on the nutritional profile of multi-plant based functional foods from popcorn, soybean, wonderful kola and moringa leaves. Findings showed that these functional foods contain appreciable amount of protein, fiber, carbohydrate content within the recommended value for diabetic patients, low glycaemic index and glycaemic load properties and with antidiabetic activities. Hence, the formulated functional foods may be suitable for individuals at risk of diabetes or diabetic patients.

Key words: Multi-plant based functional foods, nutrient compositions and antidiabetic activities.

INTRODUCTION

Diet and nutrition are important factors in the promotion and maintenance of good health at every stage of human life cycles. Recently, rapid socioeconomic changes, along with declines in food prices, increased access to foods and urbanization have resulted in a ‘‘nutritional transition’’ which characterized by a shift from primitive mode of

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nutrition to more energy-dense diets and reduction in physical activity (Vorster *et al*. 1999; Popkin, 2004, 2006, 2009; Astrup et al., 2008). Nutrition transition is characterized by increases in consumption of high fat, energy-dense foods, processed foods high in sugar, animal-based food products, and decrease in the consumption of plant-based food products like fruits, vegetables and whole grains (WHO, 2000; Popkin, 2001; Popkin, 2006, 2009; Astrup et al., 2008).

Evidence has shown that nutritional transition is negatively associated with health status, and it has been implicated as the major factors responsible for the increases in prevalence of diet-related diseases, such as obesity, diabetes and cardiovascular diseases worldwide (WHO/FAO, 2002; Popkin, 2004, 2006, 2009; Astrup et al., 2008). Epidemiological study established that increase in refined sugar intakes, which is characterized with high glycaemic index, has replaced traditional plant-based foods (Brand-Miller, 2004). Refined sugar intakes have been implicated in the etiology of many chronic diseases like diabetes and cardiovascular diseases (Brand-Miller, 2004; Ludwig, 2000; Jenkins et al., 2006; Ma et al., 2006).

The glycaemic index (GI) (a dietary measuring system relating the rate at which carbohydrate-containing foods raises blood sugar after two (2) hours or more of food consumption (post-prandial glycaemia), has clinically important benefits for preventing, managing, and treating a number of chronic diseases such as diabetes, cardiovascular disease (CVD), and some forms of cancer and obesity (Jenkins et al., 2002; Brand-Miller et al., 2003b). Carbohydrate-containing foods are graded as either having a high, intermediate (medium) or low GI depending on the rate at which blood sugar level rises (Mendosa, 2000), which in turn is related to the rate of digestion and absorption of sugars and starches available in that food (FAO/UN, 1998). This, high GI foods (GI range: >70) will break down rapidly during digestion and cause a rapid, but short-lived rise in the blood sugar level during absorption while low-GI foods (GI-range: <55) undergoes slower but gradual release of glucose into the blood stream, while intermediate (medium) glycaemic foods are those ranging between 56 and 69 on the GI scale. Foods that are classified as low GI provide a better response to postprandial glucose, causing a slight increase in circulating levels of insulin and gastrointestinal hormones; therefore, satiety is increased and voluntary food intake is reduced (Bornet et al., 2007; Jenkins et al., 2002). However, increased insulin secretion, caused by foods with high GI, leads to postprandial hyperinsulinemia along with an increase in both hunger and voluntary food intake (Aller et al., 2011). This suggests that a low-GI diet may provide some level of prevention against developing diabetes and obesity and for managing existing cardiovascular diseases (CVD).

Recently, evidence has shown that food intakes containing high glycaemic index and glycaemic load have increased, because of increases in carbohydrate intake and changes in food processing (Ludwig, 2002). Dietary glycaemic index is an indicator of carbohydrate quality that reflects the effect on blood glucose, and the dietary glycaemic load is an indicator of both carbohydrate quality and quantity food (Wolever et al., 1994; Salmeron et al., 1997). Epidemiologic evidence suggests that a diet with a high glycaemic load or glycaemic index may increase the risk of coronary heart disease (Liu et al., 2000; Ford and Liu, 2001; Liu and Manson, 2001) and type 2 diabetes (Salmeron et al., 1997). Glycaemic index is an important tool used in treating people with diabetes and in weight loss programs. Low glycaemic index foods, by virtue of the slow digestion and absorption of their carbohydrates, produce a more gradual rise in blood sugar and insulin levels and are increasingly associated with health benefits. Low glycaemic index foods have thus been shown to improve the glucose tolerance in both healthy and diabetic subjects (Jenkins et al., 1988). Hence, the present study aimed at formulating multi-plant based functional foods with low glycaemic index and antidiabetic activities.

MATERIALS AND METHODS

Sources of food materials and wistar rats

Freshly harvested *Buchholzia coriacea* seeds were bought from Ojee market in Ibadan, popcorn kernels were purchased from Erekensan market, Akure and moringa leaves were freshly harvested from Federal University of Technology, Akure Community, Nigeria. The leaves and seeds were identified and authenticated at Herbarium Unit of Department of Crop Production and Pest, Federal University of Technology, Akure, Nigeria. The wistar rats were purchased from Central Animal House, College of Medicine, University of Ibadan, Ibadan, Nigeria. The study protocol was approved by the Ethical Committee for Laboratory Animals of School of Agriculture and Agricultural Technology, Akure, Nigeria.

Processing of food materials into flour

The food materials were processed into flour using home-based processing methods, that is, blanching and fermentation. These methods were used in order to improve nutritional quality of the food products.

Popcorn flour

Raw popcorn kernels were sorted, winnowed, manually

Table 1. Percentage of raw (R_{1-3}), blanched (B_{1-3}) and fermented (F_{1-3}) popcorn, soybean, *Moringa oleifera* leaves and *Buccholzia coriacea* seeds flour samples in the formulated food samples.

Samples	Popcorn (PC)	Soybean cake	Groundnut oil	*M. oleifera* leaf	*B. coriacea* seeds
R_1	60	10	10	20	0
R_2	60	10	10	0	20
R_3	60	10	10	10	10
B_1	60	10	10	20	0
B_2	60	10	10	0	20
B_3	60	10	10	10	10
F_1	60	10	10	20	0
F_2	60	10	10	0	20
F_3	60	10	10	10	10

washed with distilled water and drained. The drained popcorn kernels were subjected into three processing, i.e., raw, blanching (100°C for 40 min.) and fermentation, using traditional methods. The raw, blanched and fermented kernels were oven dried at 60°C (Plus11 Sanyo Gallenkamp PLC, UK) for 20 h, milled using Philips laboratory blender (HR2811 model) and sieved using a 60 mm mesh sieve (British Standard) to obtain popcorn flour samples. The flour samples were packed in plastic containers sealed with an aluminum foil and stored at room temperature (~27°C) until required for use

Wonderful kola (*B. coriacea*) flour

The fresh wonderful kola was cleaned by the double disinfection method. They were washed thoroughly with distilled water to remove adhering particles after which they were soaked in 80% ethanol for 30 min. The seeds were rinsed with distilled water and then washed with aqueous sodium hypochlorite (NaClO) to reduce surface contamination and rinsed again with distilled water. The wonderful kola seeds were divided into three parts, and processed as raw, blanched and fermented (for three days) using local methods. The raw, blanched and fermented seeds were oven dried at 60°C (Plus11 Sanyo Gallenkamp PLC, UK) for 8 h, milled using Philips laboratory blender (HR2811 model) and sieved using a 60 mm mesh sieve (British Standard) to obtain raw, blanched and fermented wonderful kola seed flour. The flour was packed in a plastic container sealed with an aluminum foil and stored at room temperature (~27°C) until required for use.

Moringa Leaves Flour

Green healthy moringa leaves were trimmed off the stem, rinsed with clean water, and drained to remove dirt. The drained moringa leaves were divided into three parts, one of the parts was treated as raw, while the second and third portions were blanched and fermented (for two days) using local methods. The raw, blanched and fermented moringa leaves were oven dried at 40°C (Plus11 Sanyo Gallenkamp PLC, UK) for 8 h, milled using Philips laboratory blender (HR2811 model) and sieved using a 60 mm mesh sieve (British Standard) to obtain raw moringa leaves flour. The flour samples were packed in a plastic container sealed with aluminum foil and stored at room temperature (~27°C) until required for use.

Defatted soybean cake flour

The defatted soybean cake was milled using Philips laboratory blender (HR2811 model) and sieved using a 60 mm mesh sieve (British Standard) to obtain defatted cake flour. The flour sample was packed in a plastic container sealed with aluminum foil and stored at room temperature (~27°C) until required for use.

Formulations of multi-plant based functional foods

The popcorn, defatted soybean cake, moringa and wonderful cola nut flour were mixed in different proportions using NutriSurvey Linear Programming software to obtain nine formulations as shown in Table 1.

Proximate composition determination

Proximate compositions of the multi-plant-based food sample were determined using the standard procedures of Association of Official Analytical Chemists (AOAC) (2005). Moisture content was determined in a hot-air circulating oven (Galenkamp). Ash was determined by incineration (550°C) of known weights of the samples in a muffle furnace (Hotbox oven, Gallencamp, UK, size 3) (AOAC, 2005). Crude fat was determined by exhaustively extracting a known weight of sample in petroleum ether (boiling point, 40 to 60°C) using TecatorSoxtec (Model 2043(20430001), 69, Slandegarupgade, DK-3400, Hilleroed, Denmark) (AOAC, 2005). Protein content (N × 6.25) was determined by the micro-Kjeldahl method (Method No 978.04) (AOAC, 2005). Crude fiber was determined after digesting a known weight of fat-free sample in refluxing 1.25% sulfuric acid and 1.25% sodium hydroxide (AOAC, 2005). Carbohydrate content was determined by difference, that is, addition of all the percentages of moisture, fat, crude protein, ash and crude fibre and subtracted from 100%. This gave the amount of nitrogen free extract otherwise known as carbohydrate.

% carbohydrate = 100-(%Moisture + %Fat + %Ash + % Crude fibre + %Crude protein)

The energy value of the samples were estimated [in kcal/g] by multiplying the percentages of crude protein, crude lipid and carbohydrate with the recommended factors 4.0, 9.0 and 4.0 respectively as described by Iombor et al. (2009).

Mineral compositions determination

Calcium (Ca), magnesium (Mg), iron (Fe), copper (Cu), zinc (Zn) were determined using Atomic Absorption Spectrophotometer (AAS Model SP9). Sodium (Na) and potassium (K) were determined using flame emission photometer (Sherwood Flame Photometer 410, Sherwood Scientific Ltd. Cambridge, UK) with NaCl and KCl as the standards (AOAC 2005). Phosphorus was determined using Vanodo-molybdate method.

Determination of glycemic index and anti-diabetic activities of formulated food samples in Wistar Albino rats

Experimental animals: Seven-seven male Wistar Albino rats of body weights between 140 – 150 g were purchased from Central Animal House, college of Medicine, University of Ibadan, Ibadan, Nigeria. The rats were divided into eleven groups (7 rats per group), and were housed individually in metabolic cages in a climate-controlled environment with free access to feed and water. The rats were allowed to acclimatize to the new environment for 4 days. After four days of adaptation period, the animals were reweighed and fasted for 12 h (overnight fasting). The blood glucose of the animals were taken at zero time from the tail vein before fed with 2.0 g of the formulated food samples and glucose (control), which were consumed within 25 min. After the consumption, the serum glucose levels of the animals were measured using an automatic glucose analyzer (‘Accu-chek Active’ Diabetes monitoring kit; Roche Diagnostic, Indianapolis, USA) at 0, 30, 60, 90 and 120 min. The glycemic response was determined as the Incremental Area under the Blood Glucose Curve (IAUC) measured geometrically from the blood glucose concentration-time graph ignoring area beneath the fasting level (Wolever, 1993).

Measurement of blood glucose response

Blood glucose curves were constructed from blood glucose values of animals at time 0, after 15, 30, 60 and 120 min. intervals after consumption of the glucose (control) and formulated food samples of each group. The Incremental Area Under the Curve (IAUC) was calculated for reference food (glucose) by the trapezoidal rule (Gibaldi and Perrier, 1982) in every rats in each group separately as the sum of the surface of trapezoids between the blood glucose curve and horizontal baseline going parallel to x-axis from the beginning of blood glucose curve at time 0 to the point at time 120 min to reflect the total rise in blood glucose concentration after eating the reference food (glucose). The Incremental Area Under the Curve (IAUC) from the animals fed with the formulated food samples was similarly obtained. The glycemic Index (GI) for each diet was calculated by ratio of Incremental Area Under two hours of blood glucose response or Curve (IAUC) for each diet to the IAUC for glucose solution standard according to the method of Jenkins *et al*. (1981) and Wolever *et al*. (1991) which also reported by FAO/WHO (1997) using the following equation:

$$GI = \frac{\text{Incremental area under 2h blood glucose curve for food samples (2.0g)}}{\text{Incremental area under 2h blood glucose curve for glucose (2.0g)}} X\ 100$$

Calculation of glycemic load (GL)

Glycemic Load (GL) for each food sample was determined by the method of Salmeron *et al*. (1997). In each individual glycemic load was calculated by taking the percentage of the food’s carbohydrate content in a typical serving food and multiplying it by its glycemic index value. The following formula was used:

$$GL = \frac{Net\ Carbohydrate\ (g) x\ GI}{100}$$

Net Carbs = Total Carbohydrates in the food sample served.

Induction of diabetes mellitus

The baseline blood glucose levels of the animals were measured before induced with aloxan drug. Diabetes

Table 2. Proximate compositions (g/100g) of multi-plant based functional foods.

Parameters		Moisture	Fiber	Fat	Ash	Protein	Carbohydrate	Energy(Kcal)
R1	Mean	7.05	4.53	10.83	4.11	23.56	49.92	391.39
	±SEM	0.03	0.51	0.08	0.10	0.88	1.53	2.14
R2	Mean	7.68	3.13	10.04	3.13	25.76	50.26	394.39
	±SEM	0.08	0.08	0.24	0.14	0.19	0.22	1.58
R3	Mean	8.28	2.77	10.92	3.88	21.58	52.57	394.88
	±SEM	0.47	0.29	0.44	0.13	0.74	0.98	1.91
B1	Mean	7.04	3.78	11.92	3.29	21.60	52.36	403.14
	±SEM	0.26	0.26	0.30	0.14	1.03	0.71	2.48
B2	Mean	7.72	3.53	11.02	3.15	22.03	52.56	397.52
	±SEM	0.72	0.36	0.57	0.11	0.89	0.79	1.64
B3	Mean	7.57	4.55	10.53	3.55	24.43	49.58	390.76
	±SEM	0.72	1.13	0.28	0.07	0.65	0.88	2.08
F1	Mean	7.17	2.62	10.43	2.99	27.91	48.90	401.03
	±SEM	0.04	0.04	0.48	0.09	0.01	0.32	3.04
F2	Mean	7.41	2.58	9.50	3.01	27.00	50.51	395.50
	±SEM	0.04	0.06	0.38	0.13	0.87	1.22	1.98
F3	Mean	6.29	3.01	9.67	3.73	28.48	48.83	396.23
	±SEM	0.04	0.04	0.17	0.01	0.03	0.15	0.81
Popcorn	Mean	7.20	0.85	5.1	1.13	6.67	86.26	417.5
	±SEM	0.20	0.01	0.12	0.05	0.48	0.30	0.37

mellitus was induced by single intraperitoneal injection of freshly prepared solution of alloxan monohydrate (150 mg/kg. body weight) dissolved in physiological saline in overnight fasted Wistar Albino rats (Al-Shamaony et al., 1994). The rats were allowed to drink 5% glucose solution to avoid hypoglycaemic effects of the drug. The blood glucose levels in the animals were measured 72 h after the drug administration (alloxan treatment) through tail tipping using glucometer (Accu-Chek, Active, Roche Diagnostic's 9115 Hague road, Indianapolis, 46256 Lot No 115764) and those found to be diabetic (serum glucose ≥ 250mg/dl) were selected for the study (Ruxue et al., 2004). The induced diabetic rats were divided into eleven groups per 7 rats. Nine of the groups were fed with experimental diets (i.e., R_{1-3} B_{1-3} and F_{1-3}), and the remaining two groups were treated with saline solution and metformin hydrochloride (antidiabetic drug) (Scheen, 1996) and commercial animal feeds respectively. The animals were fed with the diets for 14 days and blood glucose levels were measured in the morning at two days interval by drawing blood from each rat through tail tipping and the blood glucose level was measured using Accu check® glucometer kit (Meiton, 2006).

Statistical analysis

The data were analysed using SPSS version 16.0. The mean and standard error of means (SEM) of the triplicate analyses were calculated. The analysis of variance (ANOVA) was performed to determine significant differences between the means, while the means were separated using the Duncan multiple range test at $p < 0.05$.

RESULTS AND DISCUSSION

Proximate compositions of formulated multi-plant based functional foods

The proximate compositions of multi-plant based functional foods from popcorn, soybean, *Moringa oleifera* leaves and *Buccholzia coriacea* seeds flour are presented in Table 2 and Figure 1. The moisture contents of the formulated diets ranged from 6.29 g/100g in F_3 to 8.27 g/100g in R_3. The lower moisture content values observed in this study imply that the formulated food samples could be stored for a reasonable long period. Moisture content of food samples is the main determinant of food spoilage. It is well established that low moisture content of food samples reduce the activities of microorganisms, and thereby increase the shelf life of the food products. In contrary, high moisture contents in food products facilitate the activities of microorganisms, and thereby reduce the nutritional quality and shelf life of the food products (Alozie et al., 2009). The values of moisture contents that were observed in this present study agreed with the findings of Olitino et al. (2007) and Adeoti et al. (2013). Crude fiber contents of the formulated diets ranged from 2.58 g/100g in F_2 to 4.55 g/100g in B_3 sample. The fiber contents of fermented food samples (F_1-F_3) were lower when compared with raw and blanched formulated food samples. This observation could be attributed to the effects of

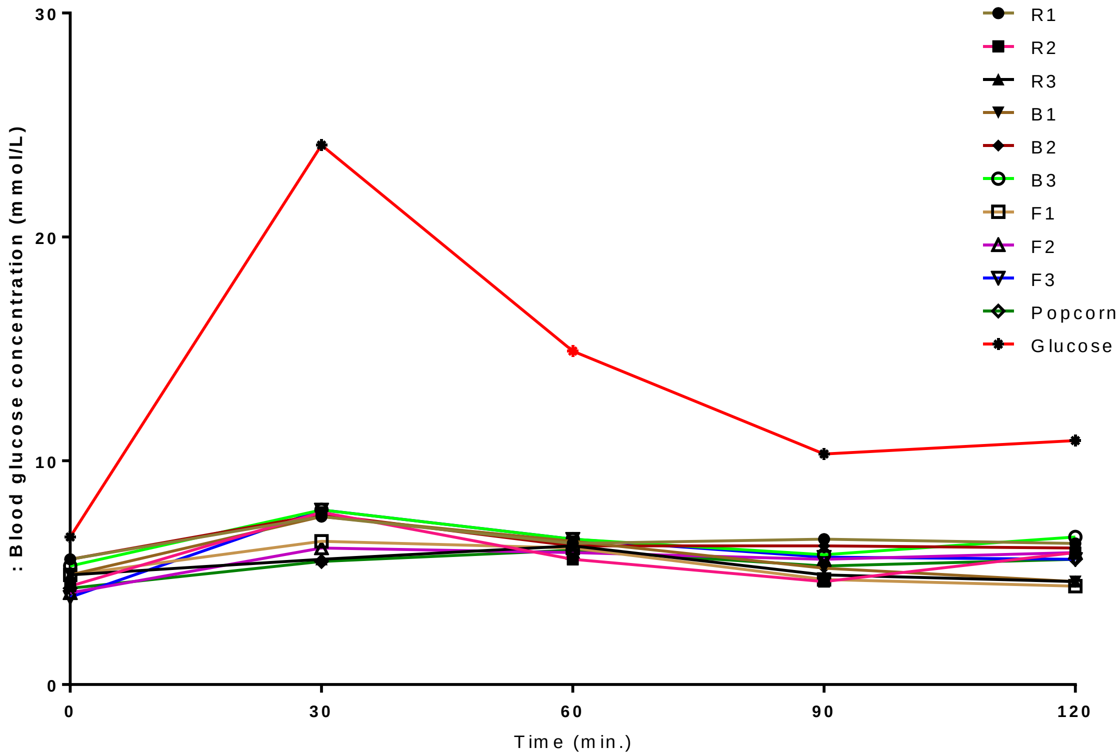

Figure 1. Blood glucose of Wistar rats fed with formulated multi-plant based functional foods from popcorn, soybean, *Moringa oleifera* leaves and *Buccholzia coriacea* seeds flour.

fermentation on the food materials. However, this observation was contrary to the report of Jood et al. (2012), who reported an increased in the insoluble and total dietary fiber content of fermented sorghum based food products. Generally, the fiber contents in the formulated diets of the present study were high, and comparable to the report of Okoye and Maze (2011). Nutritional study has established that adequate fiber intake render some health benefits like preventing coronary heart diseases, constipation and diabetes (Ishid et al., 2000). Therefore, these formulations could serve as good sources of fiber to the consumers. Fermented food samples ($F_{1-3)}$ had the highest protein content (27.00 - 28.48 g/100g) when compared with the raw (R_{1-3}) (21.58 – 25.76g/100g) and blanched (B_{1-3}) (21.60 – 24.42 g/100g) food samples. This finding agreed with the reports of other researchers (Azokpota et al., 2006; Ochanda et al., 2010). Quite a number of scientific studies have reported that fermentation increased the protein contents of food products (Adams, 1990; Nout and Ngoddy, 1997; Zlatica and Jolana, 2007), due to the activities of microorganisms that utilize other nutrients like carbohydrate and fats in the food samples to synthesized protein for their growth and development (Adams, 1990; Nout and Ngoddy, 1997; Zlatica and Jolana, 2007; Azokpota et al., 2006). The value of crude protein in this study was comparable to the values of blends of maize-tilapia fish flour (16%) (Fasasi et al., 2005), maize-bambara groundnut flour (10%) (Wang and Daun, 2000) and maize-soyabean flour (18%) (Sefa-Dedeh et al., 2002). The carbohydrate content of the formulations ranged from 48.83 g/100g in F_3 sample to 52.56 g/100g in B_2 sample, while energy values were between 421.1 kcal in R_1 and 433.7 kcal in B_1. The carbohydrate content of the formulations were low, and were within the daily recommended range values (50 - 60%) for diabetic patients (FAO, 1997; Krauss et al., 2000; Franz et al., 2002; Liu et al., 2000).

Mineral compositions of functional complementary foods

The mineral composition of functional foods is presented in Table 3. The mineral compositions and of the functional foods were as follows: Phosphorous ranged was 66.025 – 334.50 mg/100g, calcium 38.75 – 68.895 mg/100g, iron 1.235 – 4.35 mg/100g, magnesium 25.75 - 101.25 mg/100g and sodium was 38.045 -79.790 mg/100g, while zinc, potassium, copper and manganese values were 0.123 – 0.925, 33.28 -82.890, 6.950 -11.725 and 1.075 -1.625 mg/100 g respectively. The mineral

Table 3. Mineral compositions (mg/100g) of multi-plant based functional foods

Parameters		R1	R2	R3	B1	B2	B3	F1	F2	F3	Popcorn
P	Mean	152.25	176.50	334.50	234.50	192.75	92.75	266.25	224.25	166.25	142.51
	±SEM	0.75	0.50	0.50	0.50	0.25	0.25	0.25	0.25	0.25	0.91
Ca	Mean	62.75	42.75	56.75	68.75	38.75	52.75	57.50	39.75	49.25	134.80
	±SEM	2.20	3.25	2.24	2.23	0.25	1.25	0.50	0.55	0.15	0.07
Fe	Mean	4.30	1.24	2.13	2.49	1.68	2.65	1.03	1.65	2.44	4.12
	±SEM	0.00	0.01	0.00	0.01	0.03	0.05	0.01	0.05	0.01	0.04
Mg	Mean	101.20	25.75	76.50	27.93	96.50	102.75	57.28	12.13	35.78	31.44
	±SEM	0.25	0.25	0.50	4.93	0.50	0.25	0.03	0.03	0.03	0.96
Na	Mean	73.20	79.79	55.55	73.35	71.99	56.55	38.05	62.45	65.58	140.71
	±SEM	0.07	0.23	0.23	0.25	0.45	0.07	0.46	0.07	0.23	0.45
Zn	Mean	0.93	0.42	0.45	0.23	0.18	0.29	0.20	0.12	0.35	1.84
	±SEM	0.07	0.08	0.02	0.01	0.03	0.03	0.03	0.01	0.02	0.08
K	Mean	82.89	70.57	56.81	74.39	68.96	60.51	33.28	60.03	45.19	122.59
	±SEM	0.23	0.53	0.03	0.22	0.02	0.51	0.09	0.52	0.23	0.76
Cu	Mean	8.85	6.95	7.30	9.31	11.73	8.36	6.69	10.98	8.37	1.35
	±SEM	0.05	0.05	0.03	0.03	0.58	0.05	0.06	0.33	0.04	0.04
Mn	Mean	0.45	0.45	0.83	0.65	1.63	1.08	1.53	0.40	0.38	1.96
	±SEM	0.05	0.05	0.07	0.05	0.08	0.13	0.38	0.10	0.08	0.11
Na/K	Mean	0.89	1.14	0.98	0.99	1.05	0.94	1.14	1.04	1.45	1.14
Ca/P	Mean	0.41	0.24	0.17	0.29	0.20	0.57	0.22	0.18	0.74	0.95
Ca/K	Mean	0.76	0.61	1.00	0.93	0.57	0.88	1.73	0.66	1.09	1.10
Zn/Cu	Mean	0.11	0.06	0.06	0.03	0.02	0.04	0.03	0.01	0.04	1.36
Fe/Cu	Mean	0.49	0.18	0.29	0.27	0.15	1.04	0.15	0.15	0.29	3.05

concentrations of these formulations in the present study were similar to the report of Oyarekua and Eleyinmi (2004) for the ogi, a local breakfast diet, produced from corn, sorghum and millet. It is well established that minerals have critical functions in the human body, for instance, calcium, phosphorus, and magnesium are essential in forming and maintaining bones (Power et al., 1999), and in addition, calcium plays a role in blood clotting, while phosphorus and magnesium play essential roles in energy metabolism (Institute of Medicine, 1997). Zinc and iron play important roles in children, and zinc is needed for cognitive development, while iron is required for hemoglobin and myoglobin formation (Adeyeye and Otokiti, 1999; Shahbazi et al., 2009), and both zinc and iron are needed for proper immune functioning (Walsh et al., 1994; Zalewski 1996).

The ratios of sodium to potassium (Na/K) in the functional foods ranged from 0.885 in R_1 to 1.45 in F_3 sample, and the values were higher than in control samples (0.10). Nutritionally, the recommended Na/K molar ratio for human is between 1.4 and 3.4 (FAO/WHO, 1991); hence, the Na/K molar ratio of the present study formulations were suitable for people with cardiovascular diseases (Ogbuagu et al., 2011). Evidence has shown that high intake of potassium against sodium usually prevent cardiovascular diseases (Ogbuagu et al., 2011). Potassium is an essential nutrient and has an important role in the synthesis of amino acid and protein in man (Malik and Scivastava, 1982). The Ca/P molar ratios of the formulations ranged between 0.180 and 0.74, and were comparatively lower than FAO/WHO (1991) recommended value (1.6-3.6). From a nutritional point of view, the importance of adequate vitamin D, phosphorous (P) and calcium (Ca) intake for bone formation and health is well established (Bischoff-Ferrari et al., 2005, Tang et al., 2007; Pettifor, 2008).

Glycaemic index (GI) and Glycaemic load (GL) of the formulated multi-plant based functional foods

The determination of glycaemic index of foods is very important in the management of diabetes, because it helps to characterize foods according to their postprandial glycaemic response rather than their chemical composition (Jenkins et al., 1981). In this study, the glycaemic index of the formulated functional foods ranged from 36.9% in F_1 to 44.9% in B_3 sample (Figure 2), and the values were lower when compared with the reference value (i.e., low-GI<55%). The glycaemic load (GL) of the formulated food samples showed that sample F_1 had the lowest GL value (20.3), while B_2 sample had the highest GL value (25.6) (Figure 3); however, GL of the food samples were slightly above the recommended values (Low GL = <10 and high GL = >20). The low GI observed in this study, particularly for F_1 (a fermented food sample) agreed with the report of other findings that the lactic acid produced during fermentation by microbial

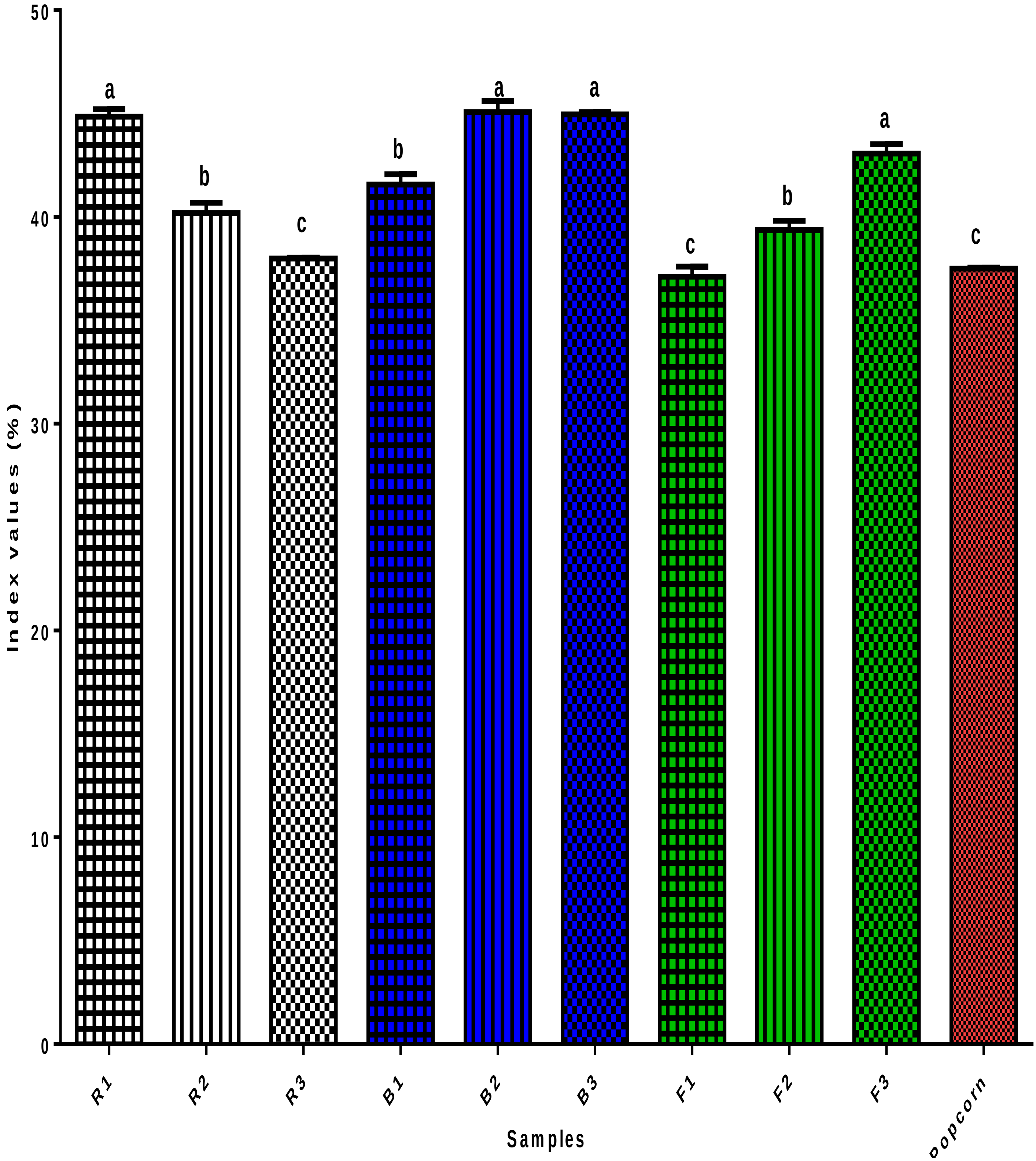

Figure 2. Glycaemic index (GI) of formulated functional foods from popcorn, soybean, *Moringa oleifera* leaves and *Buccholzia coriacea* seeds flour.
Note-GI classification: Low-GI = < 55%; Medium-GI = 56-69%; High-GI = >70%.

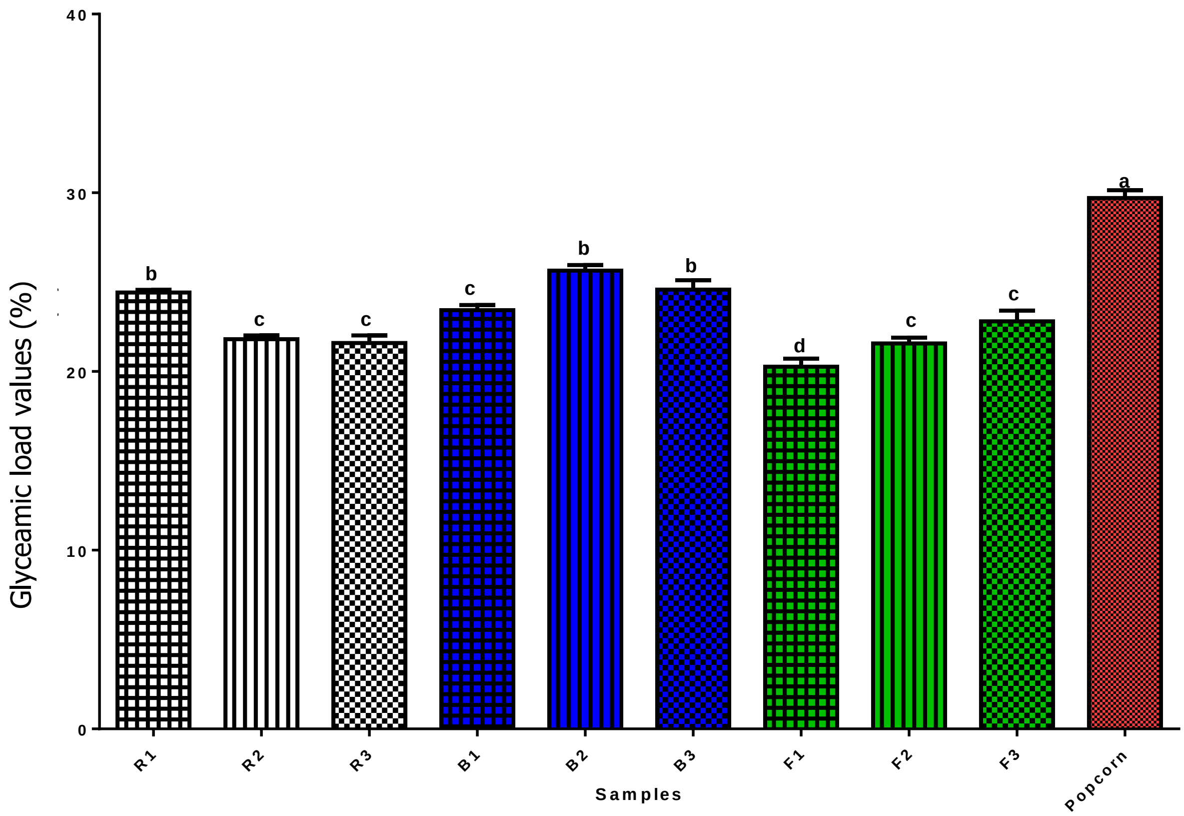

Figure 3. Glycaemic load (GL) of formulated functional foods from popcorn, soybean, *Moringa oleifera* leaves and *Buccholzia coriacea* seeds flour.
Note- GL classification Low-GL = < 10; Medium-GL = 11-19; High-GL = >20.

activities usually delay gastric emptying of starch and thereby reduced the GI of fermented food products (Scazzina et al., 2009; Liljeberg and Bjorck, 1998).

Scientific studies have established that glycaemic index of a food in human is influenced by many factors like rate of digestion/gastric emptying/absorption (FAO/WHO report, 1997; Liljeberg and Bjorck, 1998; Jenkins et al., 2002), nature of the starch/carbohydrates granules and food processing (John and Vladimir, 2004). The GI of foods (%) can be classified into three, that is, high (>70%), medium (56-69%) and low GI (<55) depending on the rate at which blood sugar level rises (Mendosa, 2000), and the rate of digestion, absorption of sugars and starches available in that food (FAO/UN, 1998). The high GI foods rapidly digest and increase the blood glucose level, while low-GI foods undergoes slower but gradual release of glucose into the blood stream. The health benefits of low glycaemic index and glycaemic load foods are their ability to reduce plasma glucose concentration as a result of slower rates of gastric emptying and digestion of carbohydrate in the intestinal lumen and subsequently, a slower rate of absorption of glucose into the portal and systemic circulation (Jenkins et al., 1981; Wolever et al., 1991). Recent studies have implicated high glycaemic load or glycaemic index of foods with increase in the risk of coronary heart diseases (Liu et al., 2000; Ford and Liu, 2001; Liu and Manson, 2001) and type 2 diabetes (Salmeron et al., 1997), hence, information on diets GI may be useful in planning carbohydrate-based foods for individuals with diabetes (FAO/UN, 1998).

Anti-diabetics potentials of the formulated multi-plant based functional foods

Blood glucose concentrations (mmol/L) of diabetic induced rats fed with multi-plant based functional food samples are presented in Figures 4. Percentage reduction of blood glucose of diabetic-induced rats fed with R_1 samples (63.8%) had the highest blood glucose reduction; while those rats fed with B_2 sample (24.1%) had the lowest blood glucose reduction. The finding also showed that among the raw, blanched and fermented formulations, samples R_1, B_1 and F_1, which contained 20% moringa leaves flour, had the highest anti-diabetic

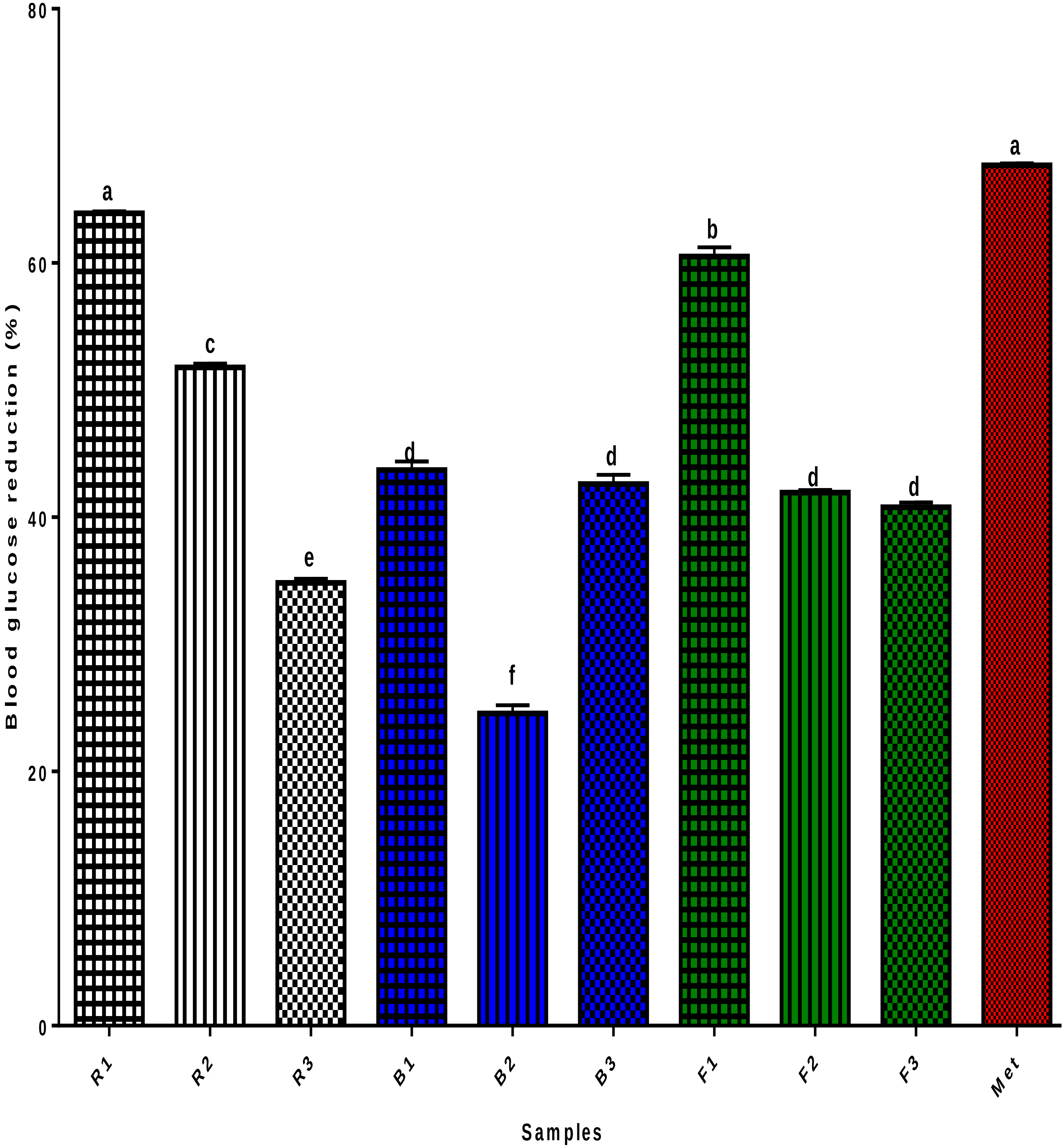

Figure 4. Percentage of blood glucose reduction (%) by the formulated multi-plant based functional foods compared with Metformin (MET) (a synthetic drug).

potentials when compared with other formulations. The antidiabetic activities of the formulated functional foods were significantly ($p < 0.05$) lower when compared with the metformin (a synthetic anti-diabetic drug). This observation could be due to interactions between the bioactive components of moringa leaves or wonderful kola with other food components, which may reduce the antidiabetic activities of the formulated functional foods. However, it is well established that *Moringa oleifera* leaves (Desoky and Youssef, 1997; Kar et al., 2003; Ndong et al., 2007; Dieye et al., 2008) and B. coriacea seeds (Ezeigbo, 2011) have antidiabetic activities; and that their antidiabetic activities may be low when compared with the pharmaceutical drugs, however, their regular intakes as food or herbal drug may have a noticeable long-term physiological effect (Espın et al., 2007).

Scientific data has shown that prevalence of diabetes mellitus is increasing throughout the world, particularly in many parts of developing countries (Shaw et al., 2010). Epidemiological study established that diet related diseases like diabetes mellitus, cardiovascular diseases, etc., are formerly “disease of affluence” in developed countries, but now the disease are increasing in many parts of developing countries (WHO/FAO 2003). This is showing a worrying trend as it is affecting a large proportion of the population and is appearing earlier in life (WHO/FAO 2003). Diabetes is a global epidemic that poses a great public health challenge, hence, the need for widely applicable strategies to reduce the incidence of diabetes is required by focusing on diet modification such as consumption of plant-based foods and increase in physical activity (Pan et al., 1997; Tuomilehto et al., 2001; Shaw and Chisholm, 2003).

Conclusion

The present study reported on the nutritional profile and antidiabetic potentials of multi-plant based functional foods. The findings established that the functional foods contain appreciable amount of protein, fiber, minerals, low glycaemic index and antidiabetic potentials. Hence, the formulated functional foods may be suitable for individuals at risk of diabetes or diabetic patients.

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