



## Production and In Vivo Evaluation of Bread from Wheat and Cassava Flours

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### Abstract

The study was carried out to investigate the effect of supplementation of 10% cassava flour on bread without compromising the nutritive value of wheat based bread. Feeding trial of Wistar-strain rats were used to evaluate the nutritional status of the 10% cassava composite bread. 100% wheat bread was used as control. The results showed that the rats fed on the control diet (6.42g) more than those fed on the 10% cassava composite bread (5.10g). The weight gain of the rat fed on the composite bread was higher (3.06g) than those fed on the control diet (2.84g). Result of the protein efficiency ratio of the rats fed on the composite bread reveals higher values of (12.73) than those fed on the control diet (4.47). The plasma protein of rats fed on the control diet also revealed higher value of (60.40g/ml) than those fed on the composite bread (42.44g/ml). The plasma glucose of the rats fed on the control diet was lower (4.09mmol/l) than those of the composite bread (6.44mmol/l). The plasma glucose of the rats fed on both diets was seen to be above the normal plasma glucose 3.12. The plasma protein of the rats fed on the composite bread was below the normal plasma protein. It can be observed from the data analysis that the rats fed on both the control and composite bread had higher hyperglycemic with continuous feeding of the breads produced from the control and composite flours. It is therefore recommended that bakeries should include protein supplement in the processing of cassava composite bread.

**Keywords:** Wheat Flour; Cassava Flour; Rats

### Introduction

In many developing countries such as Nigeria, bread consumption is continually expanding and there is an increasing dependence on imported wheat which has been found to take up much of the country's foreign exchange earnings per annum. Bread can be described as a fermented confectionary product made mainly from wheat flour, water, yeast, and salt by series of processes involving mixing, kneading, proofing, shaping and baking [1]. It is an important staple food in both developing and developed countries and constitutes one of the major sources of nutrients such as carbohydrates, protein, fibre, vitamins, and minerals in the diet of many people worldwide [2]. The consumption of bread in Nigeria is on a steady increase because it is convenient and ready to eat food normally consumed at breakfast, lunch and sometimes dinner [3]. In Nigeria, bread has become the second most widely consumed non-indigenous food after rice [4]. The rapid urbanization, increasing population and changing food habits have resulted in the preference for convenient foods such as bread, biscuits and other baked products [5,6]. Different flours from cereals (maize, rice,

sorghum and millet) and tubers rich in starch (cassava, cocoyam, sweet potato, yam) and protein rich flours (cowpea, soybean) have been used in bread making to partially substitute wheat flour [7,8]. Eriksson, *et al.* [9] in his work, determined the maximum inclusion level of cassava flour with three different ratios of 10, 20 and 30% composite of wheat and cassava flour for baking bread. Physical analysis of the bread from composite flour revealed decreased in the specific volume of the bread made from wheat and cassava flours with increasing proportion of cassava flour with 10% cassava flour substitution having the highest specific volume similar to the one from 100% wheat bread. Ochelle, *et al.* [10] evaluated the effect of water yam and soybean flours addition on the quality of wheat based bread, chemical analysis revealed increase in all the nutritional parameters of the bread. Nigeria grows staples other than wheat that can be used for bread making. One of such staples is the starchy tuber cassava.

Cassava is one of the most important crops in Africa. Nigeria is the leading producer globally. It's addition in making bread has been suggested as 10% or 20% by some researchers and 40% by

Nigeria government. However, there is evidence in favour of producing bread from composite flour of 90:10 wheat to cassava flour as well as 80:20 due to some physical parameters which compared favorably with that of 100% wheat flour. Although, bread made from different composite flour wheat and cassava has been advocated, their health, especially nutritional implication, has not been investigated. Cassavas are high in cyanogenic glycosides content as well as antinutritional factors that could be detrimental to health. The bitter and sweet cassavas are the two varieties of cassava based on the cyanogenic glycosides of their root [11]. Those with high cyanide content are called bitter cassava while those with little amount of cyanide are called sweet cassava [12]. Nutritionally, cassava is the major source of dietary energy for low income consumers in many parts of tropical Africa [13]. One hundred grams of cassava roots contains 62.5g of water, 34.7g of starch, 1.2g of protein, 0.3g of fat, 33g of calcium and 36mg of vitamin C [14].

The objective of this study was to evaluate the nutritional implication of 10% cassava supplementation to wheat based bread using *in vivo* assay.

## Materials and Method

The raw materials for the bread production are wheat (*Triticum aestivum*) and cassava (*Manihot esculenta* Crantz). The equipments used for this study was from the Department of Food Science and Technology and the Department of Biochemistry, University of Maiduguri. The Randox reagent was purchased from a Distributor at Onitsha, Anambra State Nigeria.

## Animal Experimentation

A total of 20 male and female weanling albino rats, 21-28 days old were obtained from the Animal House of the Biochemistry Department. The rats were distributed randomly into two different groups of ten animals each and transferred into a wooden cage with wire gauze at the top [15]. They were now allowed to acclimatize with growers mash for six days under normal temperature before the commencement of the experiment. The rats in the first group were fed on bread made from 90% wheat and 10% cassava flour. Water was provided ad libitum, 8g of the sample was initially provided to each rat daily but was later increased to 10g, 12g as feeding increases. The residue was weighed and removed before the next feeding. Daily records of food intake and weekly records of weight gained were maintained. The experiment was terminated at the end of two weeks.

## Collection of blood sample for determination of total protein and glucose concentration

The rats were cut by the tails. Blood samples were collected into vials containing fluoride oxalate by pressing the tail. The blood was later centrifuged for 15 minutes at 4000rpm using an 80-2 Electronic Centrifuge. The clear supernatant (plasma) was used for the estimation of total protein and glucose concentrations.

## Determination of total protein

0.5ml of the samples were taken in labeled test tubes, and 2ml of colour reagent was added to it. In preparing the blank, 2ml of colour reagent was taken in another test tube labeled blank and 50ul of distilled water was added to it. These test tubes were incubated for 10minutes at a temperature of 37°C. The samples were transferred from the test tubes into the cuvette and placed in auto colorimeter set at 540nm and their absorbance readings were taken against the reagent blank.

## Calculation

$$\text{Total Protein (g/l)} = \frac{\text{Absorbance X value of standard}}{\text{Absorbance (Standard)}}$$

## Determination of glucose concentration

The content of one vial of reagent R1b was reconstituted with a portion of Buffer R1a and the entire contents was transferred to bottle R1, rinsing bottle R1b several times. 500ul of working reagent (R1) was taken in the test tubes labeled as blank. 5ul of the sample was added to 500ul of the working reagent in the test tubes. The content of the test tubes were mixed and incubated for 10 minutes at 37°C after which 2ml of the distilled water was added. The samples were transferred from the test tubes into the cuvette and placed in the lab tech auto colorimeter set at 500nm and the absorbance readings were taken against the reagent blank.

## Calculation

$$\text{Glucose Concentration (mg/dl)} = \frac{\text{Absorbance of sample X Standard Concentration}}{\text{Absorbance of Standard}}$$

## Assessment of nutritional status

The Nutritional Status of the bread was determined using the protein Efficiency Ratio (PER). This was mathematically calculated using the recommendation of [16].

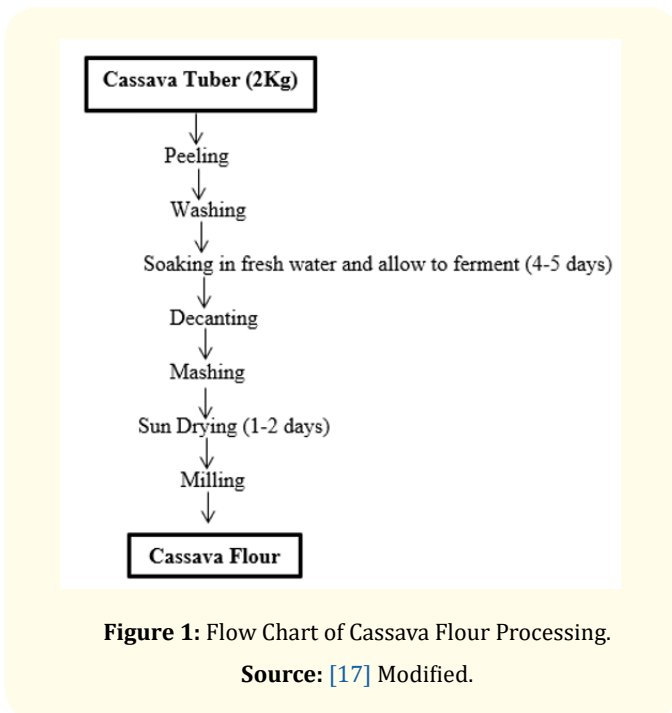
$$PER = \frac{\text{Weight gain}}{\text{Total amount of protein (g) in total food intake (g)}}$$

**Blend formulation of wheat and cassava flours**

Two flour blends, each containing wheat and cassava flour were prepared by mixing flours in the proportion of 90:10 (B). The control sample was 100% wheat flour (A). The two samples were packaged in black low density polyethylene bags and stored at room temperature until use for analyses and bread production.

**Cassava flour preparation**

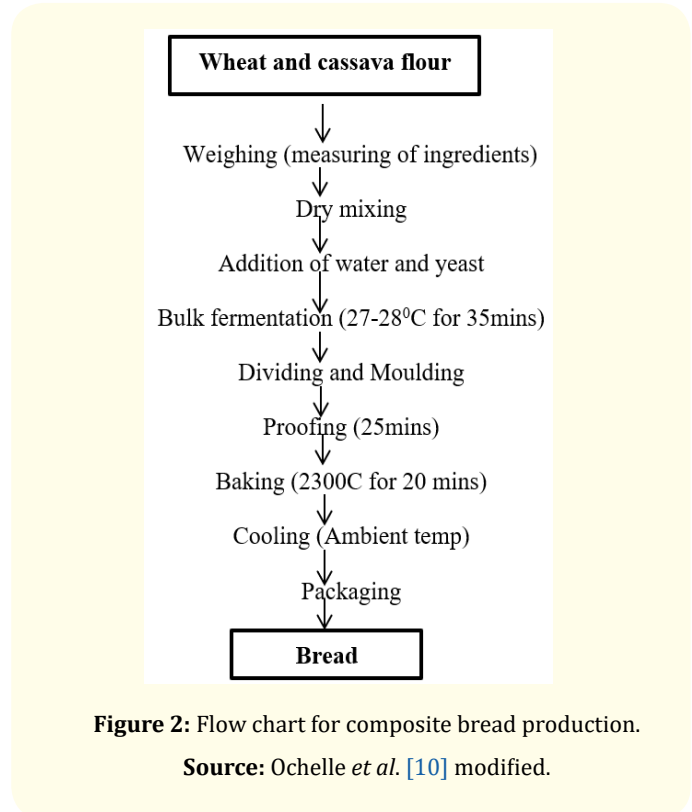
Cassava flour was prepared according to the method described by [17] as shown in Figure 1.



**Baking process**

The two blends of composite flour were baked into bread using the modified method of ochelle, *et al.* [10] The wheat flour and composite flour were mixed with 5 g salt, 10 g yeast, 7 g sugar in 250 ml water followed by manual mixing for 5 min to obtain a dough. The dough was kneaded for some minutes. The kneaded dough was transferred into the baking pans greased with plasticized fat and covered with basins. The dough was allowed to ferment for 35 mins at room temperature in the baking pans. The fer-

mented dough was then allowed to undergo proofing for 25 mins at relative humidity. The bread were cooled to room temperature and used for analyses.



Study period (days)	Food consumed (g)	
	A-Group	B-Group
Exp1	6.73 ± 1.50 <sup>a</sup>	4.88 ± 0.66 <sup>a</sup>
Exp2	6.10 ± 1.28 <sup>a</sup>	5.32 ± 0.78 <sup>a</sup>
ExpT	6.42 ± 1.39 <sup>a</sup>	5.10 ± 0.72 <sup>a</sup>

**Table 1:** Consumption pattern of Rats Fed on 100% wheat bread and 90:10% wheat and cassava bread.

**Keys:**

Exp1: Indicate 1st experiment, Exp 2 indicate 2nd experiment and ExpT indicate total experiment. The numbers of days in each experiment were 7 and 14 respectively

Values are means ± standard deviation of duplicate determinations; Means with same superscript down the column are not significantly (P≥0.05) different Keys: A = Rats fed on (100 % wheat flour control), B = Rat fed on (90% wheat flour and 10% cassava flour).

**Results and Discussion**

Study period (days)	Weight gained (g)	
	A-Group	B-Group
Exp1	3.52 ± 1.50 <sup>a</sup>	2.01 ± 0.94 <sup>a</sup>
Exp2	2.16 ± 1.28 <sup>a</sup>	4.11 ± 1.05 <sup>a</sup>
ExpT	2.84 ± 1.39 <sup>a</sup>	3.06 ± 1.00 <sup>a</sup>

**Table 2:** Weight gain of Rats Fed on 100% wheat bread and 90:10 wheat-cassava bread.

Exp1: Indicate 1<sup>st</sup> experiment, Exp 2 indicate 2<sup>nd</sup> experiment and ExpT indicate total experiment. The numbers of days in each experiment were 7 and 14 respectively

A = Rat weight on (100 % wheat flour control), B = Rat weight on (90% wheat flour and 10% cassava flour).

Values are means ± standard deviation of duplicate determinations; Means with same superscript down the column are not significantly (P≥0.05) different

Study period (days)	Protein Efficiency Ratio (PER)	
	A-Group	B-Group
Exp1	5.62 ± 2.86 <sup>a</sup>	8.68 ± 3.55 <sup>a</sup>
Exp2	2.62 ± 9.09 <sup>b</sup>	16.36 ± 3.85 <sup>a</sup>
ExpT	4.47 ± 4.25 <sup>a</sup>	12.73 ± 3.28 <sup>a</sup>

**Table 3:** Nutritional Quality of 100% wheat bread and 90:10 wheat-cassava bread

**Keys:**

Exp1: Indicate 1<sup>st</sup> experiment, Exp 2 indicate 2<sup>nd</sup> experiment and ExpT indicate total experiment. The numbers of days in each experiment were 7 and 14 respectively

A = PER of Rat on (100 % wheat flour control), B =PER of Rat fed on (90% wheat flour and 10% cassava flour).

Values are means ± standard deviation of duplicate determinations.

Means with same superscript down the column are not significantly (P≥0.05) different

Study period (days)	Total Protein (g/l)	
	A-Group	B-Group
Accl	18.60 ± 9.24 <sup>b</sup>	23.50 ± 9.08 <sup>b</sup>
Exp1	93.83 ± 31.32 <sup>b</sup>	25.91 ± 5.89 <sup>a</sup>
Exp2	68.77 ± 44.41 <sup>b</sup>	77.90 ± 35.39 <sup>b</sup>
ExpT	60.40 ± 28.32 <sup>b</sup>	42.44 ± 16.79 <sup>b</sup>

**Table 4:** Total Protein in Blood of Rats fed with 100% wheat bread and 90:10% wheat-cassava composite bread.

**Keys:**

Accl: Acclimatization

Exp1: Indicate 1<sup>st</sup> experiment, Exp 2 indicate 2<sup>nd</sup> experiment and ExpT indicate total experiment. The numbers of days in each experiment were 7 and 14 respectively

A = (100 % wheat flour control), B = (90% wheat flour and 10% cassava flour).

Values are means ± standard deviation of duplicate determinations.

Means with same superscript down the column are not significantly (P≥0.05) different

Study period (days)	Plasma Glucose (g/l)	
	A-Group	B-Group
Accl	0.82 ± 0.40 <sup>a</sup>	0.58 ± 0.15 <sup>a</sup>
Exp1	1.72 ± 0.43 <sup>a</sup>	1.54 ± 0.41 <sup>a</sup>
Exp2	4.09 ± 2.27 <sup>a</sup>	6.44 ± 2.02 <sup>a</sup>
ExpT	2.21 ± 1.03 <sup>a</sup>	2.85 ± 0.86 <sup>a</sup>

**Table 5:** Plasma Glucose Concentration of Rats fed with 100% wheat bread and 90:10% wheat-cassava composite bread.

**Key:**

Accl: Acclimatization; A = (100 % wheat flour control), B = (90% wheat flour and 10% cassava flour).

Exp1: Indicate 1<sup>st</sup> experiment, Exp 2 indicate 2<sup>nd</sup> experiment and ExpT indicate total experiment. The numbers of days in each experiment were 7 and 14 respectively

Values are means ± standard deviation of duplicate determinations.

Means with same superscript down the column are not significantly (P≥0.05) different:

Results of the consumption pattern of Rats Fed on 100% wheat bread and 90:10% wheat and cassava bread is presented in Table 1. Results indicate that rats in the A-group consumed more of the food (6.42) than those in B-group (5.10). This could be due to the fact that the bread was processed from 100% wheat flour which had pleasant flavour and texture [6].

Weight gain of Rats Fed on 100% wheat bread and 90:10 wheat-cassava bread is presented in Table 2, although the rats in the A-group consumed more, those in the B-group had higher mean weight gain (3.06) than those in the A-group (2.84). In the course of the experiment, it was observed that the rats in A-group excreted and passed urine very often. The rats in the A-group ate more, retained less water and excreted more faces whereas those in the B-group ate moderately, retained more and excreted less. This could be reason behind the increasing weight gain of the rats in the B-group when compared to those in the A-group. However, result shows no significant difference.

The Protein efficiency ratio results are presented in Table 3. The rats in the B-group was higher (12.73) than those of the rats in the A-group this was probably due to the increase in weight gain among rats in the B-group. The statistical results indicate that the plasma protein was significantly different among rats in the two groups in the first week of the experimental period.

The mean plasma protein of the rats in group-A was higher (60.40) than those of the B-group (43.44) as presented in Table 4. This could be due to the protein (gluten) present in the wheat flour that was used in processing the 100% wheat bread, this protein was absent in the composite bread. This indicated that the control diet supported growth more than the composite bread diet.

The plasma glucose level of the rats in the B-group was higher (2.85) than those of the rats in the A-group (2.21) as depicted in Table 5. It was observed that the plasma glucose levels in both groups were increasing as the experimental period progressed. The plasma glucose of normal wistar rats is said to be 3.1 [18]. Hence, it can be said that the rats were hyperglycemic with continuous feeding of the foods. There was no significant difference between their mean (2.53). It can be concluded that the rats in the B-group had higher plasma glucose and so could be supporting the effect of overweight as an associated risk for diabetes in an overfed population [19].

## Conclusion

Results revealed that the weight gain, protein efficiency ratio and plasma glucose of the 90:10 wheat and cassava bread were not

significantly different from that of the 100% wheat bread. Result of the plasma glucose also showed that both the 100% wheat bread and the cassava composite bread were above the normal plasma glucose level, continuous feeding on the bread alone could lead to hyperglycemia. It is recommended that, further study be carried out to investigate the rapid weight gain of rats that fed on the cassava composite bread. The plasma protein of the rats fed on the composite bread was seen to be below the normal plasma protein. It is therefore recommended that, bakeries should include protein supplement in the processing of cassava composite bread.

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