International Journal of Food Science and Nutrition
ISSN: 2455-4898; Impact Factor: RJIF 5.14
Received: 02-01-2020; Accepted: 03-02-2020; Published: 02-03-2020
www.foodsciencejournal.com
Volume 5; Issue 2; 2020; Page No. 04-08

Determination of potassium bromate and proximate compostion of selected breads sold within Port Harcourt metropolis

Wordu GO[1], Akusu OM[2]
[1, 2] Department of Food Science and Technology, Programme in Food Sciences and Home Science and Management Rivers State University, Port Harcourt, Nigeria

Abstract
The aim of the study was to evaluate the residual bromate and proximate composition of some selected bread samples sold in Port Harcourt metropolis. Samples were collected from Rumuokoro, Diobu town, GRA phase 3 and Trans-Amadi of Port Harcourt metropolis. The result of the analysis showed that the concentration of residual potassium bromated content was minimal in all bread and were within safe limits. The proximate analysis for the bread showed the moisture content ranged from (26-43-42.92%), ash content (0.88 – 188%) fat (7.34 – 18.51), protein (8.97-12.01%), crude fibre (1.47 – 4.26%) and carbohydrate (32.11-47.60%), for local bread, while the high class breads showed moisture (30.33 – 35.23%), ash (0.68-1.69%), fat (5.43 – 20.73%), protein (10.33 -11.91%), crude fibre (2.21 – 4.90%) and carbohydrate (29.69 – 45.97%). Results of the proximate analysis revealed that the bread sample were found to be rich in macro-nutrients, at the same time fat and ash content of the bread samples were higher than the standard limits. The bread contents of residual potassium bromate in various location within Port Harcourt metropolis were observed to be within approval standard. And the need for continuous surveillance is suggested.

Keywords: determination, bromate, proximate, Harcourt, Etropolis

1. Introduction
Bread is a staple foodstuff in Nigeria and is eaten in most countries of the world (Pagewife, 2002). It may be described as a fermented confectionary product produced mainly from wheat flour, water, yeast and salt by series of process involving mixing, kneading, proofing, shaping and baking (Dewettinck *et al*, 2008). It is low in saturated fat, very low in cholesterol and it is also a good source of thiamin and folate (Cranton, 2004). Bread is also rich in protein, vitamins and minerals (Pagewise, 2002) [8, 9]. The major challenge in both flour milling industries and bakeries is the baking quality of flour which is determined, by the capacity of the dough prepared from it to retain gas yeast feed on sugar to produce alcohol and carbon dioxide gas; this gas makes the bread to rise, bread improvers are added to speed up bread making. There are approximately 60 approved chemicals for making flour and bread. Although no single manufacturer uses all 60 additives, eight or more are commonly used. For instance, if carbon dioxide is not used, bleaching agents such as benzogy peroxide and nitrogen peroxide may be added together with maturing agents such as potassium bromate or potassium iodate (McGee, 1997). Potassium bromate is an additive some bakers use to help bread rise rapidly and create a good texture in the finished product. Potassium bromate is a flour improver, it strengthens the dough, allowing higher rise. It is an oxidizing agent and under right conditions will be completely used up in bread making (Akunyili, 2005). The use of potassium bromate has been a common choice among flour millers and bakers throughout the world because it is cheap and probably the most efficient oxidizing agent (Akunyili, 2005). Potassium biomate (K $Br0_3$) is one of the food additive that have been used in limited way and amounts by the baking industry for almost a century with no known health concern. It has been used in baking since 1914 when a potent was issues by the United States Patent Office (American Institute of Banking, 2008). In human, potassium bromated cause cough and sore throat on inhalation, abdominal main, diarrhoea, nausea, vomiting, kidney failure, hearing loss as well as redness and pain in both eye and skin (Akunyili, 2005). Potassium bromate has been banned worldwide except in Japan and the United States (Starr, 2002) [11]. The World Health Organisation in 1994 stated that this ingredient was no longer acceptable for use as it was a possible human carcinogen (WHO, 1994) [12]. In Nigeria, the use of potassium bromate in flour milling and baking has been banned by National Agency for Food and Drug Administration and Control (NAFDNC) since 1993 (Akunyili, 2004) [2]. Following the harmful effects of potassium bromate on the consumers health and the increased consumption of bread within Port-Harcourt, the largest industrialized city in south east Nigeria, it becomes necessary to determine the presence of this carcinogenic chemical in bread sold and retailed in Port Harcourt.

Materials and Method
Materials
Port Harcourt is the capital of Rivers State Nigeria. It is the largest city in the South-Eastern part of Nigeria. It lies along the Bonny River and is located in the Niger Delta. Some of Port Harcourt's most popular and well known residential areas are, Port Harcourt township, government reserved Area (GRA), phase 1-5, Elekahia, Rumuomasi, D-line, Elelenwo, Diobu, Ogbunabali, Rumuola, Rumuokoro, Trans-Amadi, Rumuokuta and Borokiri (Okafor, 1973).

Sample Collection
A total of twenty (20) different brands of bread sample (two

from each location) were collected from the Port Harcourt metropolis using a random sampling technique. These areas are Rumuokoro, Diobu, Township, GRA phase 3 and Trans-Amadi areas.

Sample Preparation

A circular samle of 2cm in diameter from the centre of each bread sample was taken and dried to a constant weigth in an oven at 105^0C The crust was ground to a fine powder and stored in air tight container for the various analysis.

Determination of Potassium Bromate

Potassium bromate in the bread samples were qualitatively and quantitatively analyzed using the method is reported by Emeje *et al*., (2010). One gramme of the bread samples were weighted out from each bread sample using an analytical weighting balance. This was transferred into a test tube. Ten milliliter (10ml) of distilled water added; the mixture was shaken and allowed to stand for 20 min. at 28 $\pm$ 10^0C. Heating was done using a water both and the temperature of the mixture was controlled using a thermometer after cooling, a 5.0ml volume was decanted from the test tube. A 50ml quantity of freshly prepared 0.5^0C potassium iodie solution in 0.1N hydrochloric acid was added. Any colour change was noted the presence of potassium bromate was indicated by a chance in coloour from light yellow to purple. The absorbance of the samples was taken at 620nm using a uv-vis spectrophotometer. Analysis was done on three replicates for each of the samples. The mean absorbance produced by the standard of potassium bromate (corrected for the standard blank) was plotted versus the concentration of the analyze in the sample to produce a calibration curve. The concentration of potassium bromated in each bread sample were obtained by extra-polation from a potassium bromated standard curve. Proximate analysis were determined acceding to the method of A.O.A.C (2012).

Moisture Content Determination

Moisture crucibles were dried for 15 minutes at 105^0C. They were allowed to cool in the desecrator for 15 minutes and the weights of the crucibles were taken. Two (2g) of the of each bread sample was weighed and and transferred into each crucible and weight of the crucible with sample taken. The crucibles with sample were then placed in the oven at 105^0C and allowed to dry for 2 hours after which the sample was removed and allowed to cool in the desiccators for 15 minutes. The crucibles with sample were recorded. The moisture contents of the samples were then calculated using the formular below-

$$\% \text{ moisture content} = \frac{Moisture\ loss}{Sample\ weight} \times \frac{100}{1}$$

Moisture loss = (crucible weight + sample before drying)
(Crucible weight + sample after drying)
Sample weight = (curable weight + sample before drying)-Crucible weight

Ash content determination

The crucibles were thoroughly washed cleaned and placed in the oven to dry for 30 minutes. They were cooled in the desiccators for 15 minutes and the weight taken. Two (2g) of the bread sample was weighted into the crucibles after which they were taken to the muffle furnace for ashing at 550^0C for four hours. The samples were the removed and allowed to cool to room temperature in the desiccators and the weight recorded as follows:

$$\%\ of\ Ash = \frac{weight\ of\ ash}{sample\ weight} \times \frac{100}{1}$$

Ash = weight of crucible after drying–weight of empty curable
Sample weight–weight of curable+sample–weight of empty crucible

Crude protein content determination

Bread sample (2.0g) was weighed and transferred into a digestion flask. And 0.3g of catalyst (copper sulphate) was weighed into the flask. Three (3g) of sodium sulphate was also weighed into the flask. Tweleve (12ml) of concentrate H_2SO_4 was added to the flask. The flask was then transferred to the digestion rack and taken to the fume cupboard and heated for a minum of 3 hours at 300^0C – 420^0C. When a clear solution was formed water was carefully added and poured into a flat bottom flask. The solution was then made up to 100ml. Ten (10ml) of 2% boric acid was measured into 100ml conical flask (as receiving flask) Twenty (20ml) of the digest was transferred into a kjeldahl flas and 20ml of 45% NaoH added steam was passed into the digest and collected into the boric acid indicator until it gets to the 50ml mark, $0.05NHSO_4$ was poured into the burette and titrated against the condensed sample plus indicator until it slightly turns pink. The reading was taken as the final titre and the percentage nitrogen and crude protein were calculated. A blank solution was also prepared.

Calculations

$$\%\ N = \frac{Sample\ titre - blank\ tibre\ x\ nomality\ of\ acid\ x\ 1.4}{sample\ weight\ (gm)}$$

Protein content = % Nitrogen x protein factor (6.25)

Fat Content Determination (AOAC, 2012)

A clean, dried 500ml round bottom flask was weighed (WI) and 300ml of petroleum ether for the extraction was poured into the flask fitted with soxhlet extraction unit. The extractor thumble containing 2.0g of the bread sample was fixed into the soxhlet extract unit. A round bottom flask and a condenser was connected to the Soxhlet exactors and coldwater circulation put on. The heating mentle was switched on and the heating rate adjusted until the solvent is refluxing at a steady rate. Extraction was carried out for six hours. The solvent was recovered and the oil dried in the oven at 70^0C for one hour. The round bottom flask and oil were cooled than weighted (W2). The lipid content was

Calculated thus

$$\%\ crude\ lipid\ content = \frac{W_2 - W_1}{sample\ weight} \times \frac{100}{1}$$

Crude Fiber Determination (AOAC, 2012)

The defatted sample was placed in a beaker and 25ml of

1.25% H_2So_4 was added to the sample. The mixture was then heated and allowed to boil for 4-5 minutes in a heating mantle and the beaker covered with a wash glass. A funnel with filter paper was used to filter the boiled sample using boiling water torinse the remains in the beaker. This process was continued until the sample washing was free from acid. Twenty-five (25ml) of NaoH was measured and used to wash the residue back into a beaker and brought to boil. It was then boiled for 4-5 minutes and then allow to stand for 1 minute. The sample was filtered through an ashless filter paper that has been dried and weighted. It was washed thoroughly using boiling water until the washing was free from the base. The filter papers were transferred to the oven and allowed to dry for one hour cat 105^0C after which it was cooled and weighed. The filter paper was transferred to an already dried and weighed crucible and then ashed for 2 hours at 550^0C. The weights were taken and the result calculated,

Calculates

$$\% \text{ fiber} = \frac{\text{Crude fiber}}{\text{sample weight}} \times \frac{100}{1}$$

Residue weight = (weight of filter paper + residue)-filter paper weight
Ash = (weight of crucible + ash) - weight of crucible
Actual ash – weight of ash – weight of residue of the blank
Crude fiber = Residue weight - Actual weight
Carbohydrate determination by difference carbohydrate were determined by the calculation,
Carbohydrate = 100 - (moisture + fat protein + crude fibre)

Statistical analysis
Analysis were done intripliation. All values obtained were subjected to analysis of variance (ANOVA) and the differences in mean was subjected to LSD.

Results
Table 1 shows the proximate composition of high quality breads sold within Port Harcourt Metropolis Moisture content of the bread samples ranged from 30.33 – 35.23% with bread samples from government reserve areas (GRA) having high moisture content and bread samples from trans Amadi (TR) recording the lowest. It also showed that breads from Trans Amadi (TB1 and TR2) and Rumuokoro (RA1) did not differ significantly (P >0.05) from each other whereas breads collected from Diobu (DA2) (DA2), TR, RA and TA differed significantly (P < 0.05).
Ash content of the high quality breads ranged from 0.68 - 1.69% with bread collected from DA2 recording the highest and GRA1 lowest. Results showed that breads from GRA2, TR, RA and TA did not differ significantly (P >0.05) from each other.
Fat content ranged from 5.43-20.73% with DA2 bread recoring the lowest and RA2 bread the highest. Results showed that breads collected from GRA2, RA1 and DA1 did not differ significantly (P > 0.05) in fat content while breads from TA1, TA2, DA1, DA2 and TR2 differed significantly (P < 0.05).
Protein content of the bread samples collected ranged form 10.33 – 11.91% with bread from TR1 recording the lowest and RA2 the highest. Results showed no significant (P > 0.05) difference in the protein content of the bread samples, except for TR1 which differed significantly (P < 0.05) from RA1 and 2 and TR2.
Crude fibre content ranged from 1.47 -4.90% with bread samples and DA2 recording the lowest and TR2 the highest. The results showed that bread samples from GRA, TR1, RA2, and DA1 showed no significant (P >0.05) differences while TRD, DA2 and TA1 different significantly (P > 0.05) in their crude fibre values. Carbohydrate content of the bread sample ranged from 29-45.97% with bread samples from TA2 recording the highest and RA2 the lowest.

Table 1: Proximate composition of selected high quality bread sold with in Port Harcourt metropolis

Samples	% moisture	% Ash	% Fat	% Protein	% crude fibre	% C+10
TA1	34.32±0.36ab	1.12±0.09ab	10.17±0.33c	11.18±1.21ab	3.40±27bc	38.31±0.8bc
TA2	34.11±0.98ab	1.08±0.06ab	5.85±0.02ab	10.71±0.42ab	2.28±0.89bcd	38.31±0.8bc
DA1	33.47±0.00bc	1.59±0.19a	17.21±0.02b	11.19±0.81ab	2.85±1.07bcd	33.70±1.71a
DA2	32.41±34c	1.69±0.16a	15.43±1.30c	10.79±0.21ab	1,47±0.64d	38.23±67bc
RA1	30.33±0.64d	1.02±0.34a	17.8±0.90b	11.72±0.18a	3.75±0.04ab	3.87±1.40cd
RA2	32.58±0.58a	1.50±0.17a	20.73±1.17a	11.91±0.10a	3.56±0.54bc	29.69±1.45
TR1	30.34±08d	1.39±0.19ab	11.11±0.1dc	10.33±0.59b	2.54±18bcd	44.30±0.4a
TR2	28.53±0.54d	1.14±0.11ab	12.35±0.78dc	11.78±0.81b	4.90±0.78a	38.77±2.05b
GRA1	35.23±0.54+a	0.68±0.13b	5.49±.15f	10.89±0.69b	3.10±0.20bc	44.62±1.49a
GRA2	33.47±0.00bc	1.13±0.00ab	17.21±03b	11.00±0.00ab	2.21±0.77cd	35.00±0.76a
LSD	1.47	0.69	1.53	1.29	1.47	2.71

Results are expressed as mean ±SD, n= 3 values in the same column having different superscript are significantly different ate (P< 0.05)

Key: TA1 and 2 = Bred collected from town
DA1 and 2 = Bread collected from Diobu
RA 1 and 2 = Bread collected from Rumuokoro
TR1 and 2 = Breads collected from Trans-Amadi
GRAI and 2 = Breads collected from GRA.

Table 2 shows the proximate analysis of local breads sold within Port Harcourt metropolis. Moisture content of the bread samples ranged from 26.43 – 42.92% with GRA 1 recording the lowest and GRA 1 the highest GRA was significantly (P < 0.05) different from all the bread samples while other bread samples showed no significant (P > 0.05) difference. Ash content ranged from 0.76 – 1.88% with bread sample from D4 (DA2) and Rumuokoro 1 (RA) recording the highest which GRA lowest. Bread sample from Town 2 (TA2), DA, GA and Trans Aamdi 1 (TRI) showed no significant (P > 0.05) difference from each other while TA1, GRA and TR2 followed this same trend. Fat content of the bread sample allowed range from 3.34 – 18.51% with brand sample from TA2 recording the highest and this was significant (P <0.05) different from all other DA2 had the lowest fat content significantly (P < 05)

different from others while breads from RA2, TRI and DA1 showed no significant (P < 0.05) difference from each other. Protein content of the bread principles ranged from 8.57 – 12.01% with DA2 bread recording the highest and DA1 the lowest. Results shows that bread samples collected from GRA2, TR2, RA2, TA1 showed no significant (P < 0.05 difference from cash other while bread sample from DA1, DA2 and TA1 different significantly (P < 0.05) in their protein content. Crude fibre content of the bread collected ranged form 1.47 – 4.26% with DA2 bread recording the highest and GRA1 the lowest. Result show that bread sample collected from GRA, RA1,TR1, TA and DA1 showed no significance (P <0.05) difference from each other while bread sample from DA2, RA1 and TR2 differed significantly (P < 0.05) in terms of their crude fibre content. Carbohydrate content of the breast samples ranged from 32.11 – 47.60% with bread samples form TA2 recording the lowest and GRA1 the highest. Bread sample collected fro GRA1, DA1, RA2 and TA2 had carbohydrate contents significantly (P < 0.05) different from each other while sample from GRA 2, TR1 and RA2 showed no significant (P < 0.05) difference from each other.

Table 2: Proximate composition of selected local breads sold within Port Harcourt metropolis

Samples	% moisture	% Ash	% Fat	% Protein	% crude fibre	% C+10
TA1	31.98±1.54^{b}	0.99±0.19^{b}	12.37±0.16^{d}	9.4±0.35^{acd}	3.58±0.76-^{ab}	41.61±32.8^{bc}
TA2	33.90±0.08^{b}	1.58±0.00^{a}	18.51±0.33^{a}	10.37±0.01^{bc}	3.55±0.00^{ab}	32.11±0.41^{c}
DA1	27.51±3.17+c	1.49±0.11^{a}	15.42±0.59^{b}	12.01±0.23^{a}	3.53±0.06^{ab}	42.55±0.78^{b}
DA2	32.92±0.86^{b}	1.88±0.17^{a}	7.34±0.11^{f}	8.57±0.44^{c}	4.26±1.13^{a}	45.03±2.72^{ab}
RA1	34.31±.11^{b}	1.88±0.73^{a}	9.82±0.89^{c}	8.78±0.01^{c}	2.50±0.91^{c}	42.73±1.67^{b}
RA2	34.50±0.30^{b}	1.52±0.39^{a}	14.58±0.28^{b}	9.89±0.50^{bcd}	3.48±0.11^{ab}	36.04±0.85^{d}
TR1	32.66±1.52^{b}	0.88±0.02^{a}	14.74±0.73^{b}	10.57±0.80^{bc}	2.44±0.42^{bc}	38.73±1.86^{cd}
TR2	34.82±2.02^{b}	1.11±0.04^{b}	13.46±0.05^{c}	11.09±0.36^{bcd}	2.91±1.06^{c}	37.8±0.13^{d}
GRA1	26.43±1.47^{c}	0.99±0.01^{b}	13.43±0.11^{c}	9.12±0.00^{ab}	1.47±0.55^{b}	47.60±0.09^{a}
GRA2	42.92±1.62^{a}	0.76±0.00^{b}	9.24±0.00c	9.85±0.83^{dc}	2.26±0.41^{bc}	38.60±2.00^{cd}
LSD	3.69	0.39	0.84	1.24	1.64	3.47

Result are expressed as mean ISD, n =3. Values in the same column having different superscript are significantly different at P < 0.05.

Key
TA1 and 2 = Bread collected from Town;
DA1 and 2 = Breads collect from Diobu
RA1 and 2 = Breads collected from Rumuokoro
TR1 and 2 = Bread collected from Trans-Amadi
GRA 1 and 2 = Breads collected from GRA

Table 3 shows the concentration (mg/l) of potassium bromated selected in high quality breads sold within Port Harcourt metropolis. Bromate concentration ranged from 0.022–0.03mg/l, with breads purchased from Town 1 recording the highest and breads from GRA 2 the lowest.

Table 3: concentration of potassium brough (Mg/l) in selected high quality breads sold within Port Harcourt metropolis

Bread Samples	Bromate content of High quality breads (mg/l)
TA1	0.038
TA2	0.026
DA1	0.031
DA2	0.037
RA1	0.027
RA2	0.026
TR1	0.029
TR2	0.026
GRA1	0.025
GRA2	0.022

Key
TA1 and 2 = Breads collected from Town
RA1 and 2 =- Breads collected from Rumuokoro
TR1 and 2 = Breads collected from Trans-Amadi
GRA1 and 2 = Breads collected from GRA

Table 4 shows the concentration of (mg/l) of potassium bromate in selected local breads sold within Port Harcourt metropolis. bomate content for local breads ranged from 0.022–0.034mg/l with bread purchased from Town 1 recording the lowest and breads from Trans-Amadi the highest.

Table 4: Concentration of potassium promate (mg/l) in selected local class breads sold within Port Harcourt metropolis.

Bread Samples	Bromate content of local breads (mg/l)
TA1	0.022
TA2	0.025
DA1	0.027
DA2	0.025
RA1	0.022
RA2	0.025
RA1	0.031
TR1	0.034
GRA1	0.025
GRA2	0.029

Key
TA1 and 2 = Breads collected from Town
DA1 and 2 =- Breads collected from Diobu
RA1 and 2 = Breads collected from Rumuokoro
TR1 and 2 = Breads collected from Trans-Amadi
GRA1 and 2 = Breads collected form GRA

Discussion
Moisture content the local class of bread samples ranged from 26.43 – 42.92% while that of the high quality ranged from 30.33 – 35.23%. The values of the moisture content of the local and high quality breads fell within the ranges (40.00%) permitted by SON (2004) [10] except for local bread purchased from GRA2 which had a mixture content of 42.92%. The moisture content of foods is usually used as an indicator of food quality. It is important to measure the moisture content in breads beause of its potential impact on the microbial and physical properties of bread (Olaoye *et al*., 2006). Ash content of the high quality class bread ranged from 0.68 – 1.69% while the class bread ranged from 0.88 – 1.88%. Ash content is an indicator of minerals present. The ash content of the local and high quality breads

were higher than limits (0.60%) promitted by SON (2004) [10] except for high quality bread purchased from GRA1 which had ash content of 0.68%, similar finds were reported by Eke *et al* (2013) [4] which observed high ash content (4.06 – 7.93%) in bread brands sold in Benue and Nassarawa State in Nigeria.

Protein content of the local class bread ranged from 8.57 – 12.01% which the high quality class bread ranged from 10.33 – 11.91% protein content of the local and high quality breads were within limits permitted by SON (2004) [10] excpt for local breads bread purchased from Diobu 2, Town 1, Rumuokoro 1and GRA 1 with protein content of 8.57%, 9.445%, 8.78% and 9.12% respectively. Fat content of the high quality bread ranged from 5.43 – 20.73% while that of the local breads were between 7.34 – 18.51%. The fat content of the local and high quality breads were all higher than the limts of 2.0% permitted by SON (2004) [10]. Crude fiber content of the local class and high quality bread ranged from 1.47 – 4.26% and 1.47 – 4.9% respectively. The crude fiber content were within the limits of 5.0% permitted by SON (2004) [10]. The high values for protein, fat, crude fiber and ash could be as a result of the flours used in composite with wheat, as well as other additives used in producing these bread samples. The concentrations of residual potassium bromated solid in various location within Port Harcourt metropolis were observed to be lower than approved FAD limit. The low concentration recorded in all the locations might be due to high degree of population of people from different part of the world and Rivers State covernment addresses to backers in Port Harcourt metropolis.

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