Content of Minerals and Antinutritional Factors in Akara (Fried Cowpea Food)

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Abstract: The aim of the study was the quantifaction of minerals and antinutritional factors in akara (AK) and its crude mass (CM). Deep-frying was performed on 5 consecutive days. Potassium and phosphorus were the most abundant elements naturally present (545-719 mg 100 g^{-1} and 210-375 mg 100 g^{-1}, respectively), while sodium exhibited the highest contents (699-1,869 mg 100 g^{-1}) because of salt addition to CM. The content of antinutritional factors in AK and CM were determined to be: 11.27 ± 0.17 and 9.9 ± 0.14 µmol g^{-1} ($InsP_5$); 2.92 ± 0.03 and 3.75 ± 0.11 µmol g^{-1} ($InsP_6$); 1.73 ± 0.16 and 1.68 ± 0.02 mg eq. CE g^{-1} (tannins); 6.35 ± 0.03 and 6.27 ± 0.03 mg g^{-1} (polyphenols); 0.50 ± 0.00 and 0.0 HU kg^{-1} (hemagglutinins). Deep frying led to a significant reduction ($p \leq 0.05$) in the content of most of the minerals and antinutritional factors analyzed. Nonetheless, AK was shown to be a good source of K, P, Mg, Mn, Mo, Cr, Cu, Fe and Zn. However, bioavailability of the Fe and Zn was low.

Keywords: Akara, Cowpea, Deep-frying, Minerals, Antinutritional factors.

1. INTRODUCTION

Akara is a cultural and touristic icon in the city of Salvador (Bahia, Brazil) sold in the streets as a finger food by typically clothed women called *baianas de acarajé*. It is made from several cultivars of cowpea beans (*Vigna unguiculata* L. Walp), such as *fradinho, macassar, olho de pombo, costela de vaca, boca preta*. In the akara making process, beans are split, decorticated and macerated into a paste. After being seasoned with grated onions and salt, the crude mass is shaped into balls by a wooden spoon and deep-fried in crude palm oil (CPO) [1].

Functional, sensory and nutritional characteristics of akaras are directly related to the variety of beans and CPO. Cowpea legumes are low-in cost and they represent a significant part of the daily food in the Brazilian population. Akara is a source of proteins, vitamins and minerals such as calcium, iron and zinc [2, 3]. However, its nutritional value is usually reduced by the presence of antinutritional factors such as phytates, fibers, lectins, polyphenols and tannins that affect minerals bioavailability [2, 4, 5].

Phytate (*myo*-inositol 1, 2, 3, 4, 5, 6-hexakisdihydrogenphosphate) is the major storage form of phosphorus (P) in plant seeds, especially in legumes, cereals and oilseed crops [6, 7]. Phytate affect the nutritional value of plant-derived foods due to its ability to form complexes with calcium, iron, zinc, copper and magnesium, thereby reducing the bioavailability of the minerals [1, 5, 8]. Phytate levels are reduced during certain food processing and preparation techniques such as baking, extrusion, fermentation and germination [9, 10] by endogenous phytases resulting in inorganic phosphate and partially phosphorylated *myo*-inositol phosphates [11]. Despite the mineral-chelating properties, phytate has also been investigated for its beneficial effects on diabetes mellitus, renal lithiasis, arteriosclerosis and antioxidant action in biological systems [7].

Hemagglutinins (HG) also called lectins, constitute an ubiquitous class of proteins which are widely distributed in the plant kingdom. They belong to the carbohydrate-binding proteins of non-immune origin, which agglutinate cells or precipitate polysaccharides and glyco-conjugates. HG also have an ability to bind to surface specific receptors sites of intestinal cells, leading to non-specific interference on nutrients absorption [12]. Condensed tannins are reported to form tannin-protein complexes which result in protein insolubility. Furthermore iron and calcium bioavailability

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is impaired due to the chelating ability of condensed tannins [13, 14, 15]. Polyphenolic compounds have also been reported to reduce the bioavailability of some minerals as well as digestion and/or utilization of digested and absorbed proteins [16].

Akara plays an important role in the diet of the population of Salvador. However, elevated levels of antinutrients in this finger food may result in a reduction of the availability of essential nutrients, such as iron, zinc and calcium [1, 3]. Nonetheless, there are only a few studies regarding akara and its content of minerals and antinutritional factors. Therefore, the aim of this study was to quantify minerals and antinutritional factors in akara samples and their crude mass.

2. MATERIAL AND METHODS

2.1. Samples Collection and Frying Experiment

Preparation of akara was performed by a *baiana de acarajé* according to her traditional practice [1]. 5 kg of crude mass (CM) of akara (AK) were bought at São Joaquim Fair, in Salvador city (Brazil). 30 L of integral (a mixture of liquid and solid phases) CPO industrialized (conditioned in tinplate cans) were purchased at the same fair. This oil was placed in a stainless recipient and heated up to 45 ºC, allowing oil fusion and homogenization, to be used in frying experiment.

CM was placed in an aluminum pan, prepared and shaped into balls (80-110 g each). Then, 5 L of CPO in an enameled pan was heated for 12 minutes in the presence of an onion [1]. Subsequently, 5 of those balls were successively added and deep-fried in CPO. Thereafter, the fried balls (AK) were removed and drained on absorbent paper until reaching ambient temperature. Total frying time a day was 5 hours and the frying process was performed on 5 consecutive days. The maximum daily oil turnover was 2 L. A total of 110 AK/day was obtained – simulating akara average quantity market in *baiana's* point of sale [1].

Each day, 10 CM samples were randomly collected as well as 10 AK close to the end of the daily frying process. All samples were stored in *Ziploc* bags at –20 ºC. During sample preparation, the 10 CM and the 10 AK samples were joined and the quarter method was applied until 50 g of each sample was obtained [17]. Then, samples were identified in accordance with frying time of each day: CM 5h, AK 5h (day 1); CM 10h, AK 10h (day 2); CM 15h, AK 15h (day 3); CM 20h, AK 20h (day 4); CM 25h, AK 25h (day 5).

2.2. Sample Preparation

Akara and its crude mass were stored for 24 hours at-80 ºC before freeze-drying (Freeze-dryer LS 3000 D, Terroni Equipamentos Científicos Ltda., Brazil). Then, they were ground in a domestic stainless steel food processor (Cuisinart Coffee Grinder, Model DCG-20). The ground material was stored in amber bottles at 25 ºC for subsequent analysis.

2.3. Antinutritional Factors

2.3.1. Condensed Tannins

Condensed tannins were extracted with HCl: methanol (1:100 v/v) for 2 h with mechanical shaking at 25 °C and centrifuged at 5000 *g* at 15 °C for 15 min. Aliquots were immediately analyzed for tannins using the 0.5 % vanillin assay [18].

2.3.2. Polyphenols

Total phenols were extracted with water. An internal standard curve was prepared by adding 10 mL of 0–0.01 % tannic acid to the flasks. The flasks were heated for 30 min at 70 °C with constant shaking. Clear supernatants were collected after centrifugation at 2500 *g* for 15 min followed by filtration. Polyphenols were determined using the Folin–Denis reagent [19].

2.3.3. Haemagglutinating Activity

Haemagglutination assays, using trypsin-treated rabbit erythrocytes, were carried out by a serial dilution method as described by Grant *et al*. [20]. One unit of haemagglutinating activity (HU) was defined as that contained in the amount of sample in the last dilution which caused 50 % agglutination of the red blood cells.

2.3.4. Myo-Inositol Phosphate Analysis

Quantification of *myo*-inositol phosphates was performed in freeze-dried samples, by HPLC ion-pair chromatography using an Ultrasep ES 100 RP18 column (2 x 250 mm) as described by Greiner & Konietzny [21]. A mixture of the individual *myo*-inositol phosphate esters ($InsP_3$–$InsP_6$) was used as the standard.

2.4. Minerals

High purity analytical stock solutions (Titrisol®, Merck) of each element were used to prepare multi-element reference solutions and calibration curves. The quantification of the minerals was performed by ICP OES and ICP-MS using external calibration. The relative standard deviations were less than 10 % for all

minerals investigated. The paired t test at a 95 % confidence level showed that there was no significant difference between the means of the certified and determined values for most analytes under investigation.

2.4.1. Digestion Procedure

Approximately 500 mg of each sample was digested using a microwave-assisted procedure. 7 mL of concentrated HNO_3 (65 % (w/w)) and 1 mL of H_2O_2 (30 % (v/v)) in closed TFM vessels were used for digestion. Heating was performed in four successive steps: linear temperature increase from room temperature to 90 °C in 4 min (maximum power of 500 W); 3 min at 90 °C (maximum power of 500 W); linear temperature increase from 90 °C to 180 °C in 10 min (maximum power of 1,000 W); 15 min at 180 °C (maximum power of 1,000 W) [22]. All samples were analyzed in triplicate, and a set of digestion blanks were prepared together with each sample batch.

2.4.2. ICP OES and ICP-MS Analysis

Quantification of minerals (Ca, Cu, Fe, K, Mg, Mn, Mo, Na, P and Zn) was performed by ICP OES with axially viewed configuration (VISTAPRO, Varian, Mulgrave, Australia). The following parameters were used: RF generator (40 MHz), power (1.2 kW), plasma flow (15.0 L min^{-1}), auxiliary flow (1.5 L min^{-1}), nebulizer flow (0.7 L min^{-1}), sample flow (0.7 L min^{-1}) and dwell time (1 min). V-groove nebulizer and Sturman–Masters chamber were used to put system solutions in the spectrometer, while Ca, 396.847 nm; Cu II, 324.754 nm; Fe II, 238.204 nm; K, 766.491 nm; Mg II, 280.270 nm; Mn II, 257.610 nm; Mo II, 202.032 nm; Na, 568.821 nm; P, 213.618 nm; and Zn II, 213.857 nm were used as atomic emission line (I) and ionic emission line (II). ICP-MS (Thermo Scientific, 2008) has been applied to the quantification of Al, As, Cd, Co, Cr, Ni and Se. The following parameters were used: power (1.35 kW), plasma argon flow (13.0 L min^{-1}), auxiliary argon flow (0.7 L min^{-1}), nebulizer argon flow (0.87 L min^{-1}), sweeps (100), dwell time (10 ms) and CCT gas flow (6.5 L min^{-1}). The following isotopes were determined: ^{27}Al, ^{75}As, ^{111}Cd, ^{59}Co, ^{53}Cr, ^{60}Ni and ^{82}Se; ^{72}Ge (internal standard) in the peak jump analysis mode [22].

2.5. Phytate to Zinc, Iron and Calcium Molar Ratios

To calculate the phytate to mineral ratios the following molecular masses were used: phytate: 660 g mol^{-1}; Fe: 56 g mol^{-1}; Zn: 65 g mol^{-1}; Ca: 40 g mol^{-1} [23].

2.6. Statistical Analysis

Data analysis was performed using Statistica 6.0 (*Statistica for Windows*, 2006). Each sample was analyzed in triplicate (and *myo*-inositol phosphates in duplicate). All values were given as mean ± standard error (SE). Relative standard error (RSD %) were calculated, and results were expressed as 95 % confidence intervals. Correlations between minerals and antinutritional factors were assessed by Spearman's rank correlation test. Data were subjected to multivariate analysis by Principal Components Analyze (PCA) and Hierarchical Cluster Analysis (HCA). PCA and HCA were applied in a data matrix (10 x 15) generated with mean values of mineral content. However, the Cd and Ni were not included because these elements were not detectable in all samples (Table **1**). All data were processed by standardization, and Varimax normalized method was applied.

3. RESULTS AND DISCUSSION

3.1. Minerals

Mineral were classified into macroelements (Ca, K, Mg, Na and P), microelements (Cu, Fe, Mn and Zn), trace elements (Al, Co, Cr and Se) and ultra-trace elements (As, Cd, Ni and Mo) (Table **1**). The content of the different minerals in the CM samples showed considerable differences. Especially the mineral content of the CM 5h sample differed significantly from those of the other CMs samples (Table **1**). The only elements that did not present significant difference ($p > 0.05$) among the different CM samples (10, 15, 20, 25 hours) were As, Co, K, Fe (Table **1**). The observed differences could be explained by different methods used for preparation, with variations in time and temperature of seed maceration and a mixing of different beans varieties [1], as well as a mixture with others legumes [24].

In general, deep frying resulted in a significant reduction ($p \leq 0.05$) in the content of most of the minerals studied, especially in the content of Ca, K, Mg, P, Zn, Mo and Mn (Table **1**). Nevertheless, Cr, Al and Co values were increased ($p \leq 0.05$) in almost all AK samples compared to the corresponding CM samples (Table **1**). Minerals are heat-stable and are therefore not affected by deep frying. However, if wet heating is performed, leaching of minerals can occur into the cooking liquid, and thus indirectly affect the mineral content. According to Vaquero [25], the cooking process (especially frying) can result in an increase in cell separation of the cooked tissues and in

Table 1: Minerals Content (Macro, Micro, Trace and Ultra-Trace Element) In Crude Mass (CM) And Akara (AK) (Cowpea Food) from 25h Deep-Frying (5h/Day) in Crude Palm Oil

Mineral	Sample	5h	10h	15h	20h	25h
MACROELEMENTS (mg 100 g^{-1})						
Ca	CM	44.31 ± 1.06^{a,z}	32.64 ± 1.68^{a,y}	36.61 ± 0.64^{a,x}	46.13 ± 2.19^{a,z}	38.45 ± 1.89^{a,w}
	AK	38.30 ± 0.88^{b}	27.40 ± 0.33^{b}	25.05 ± 0.33^{b}	39.15 ± 1.24^{b}	28.10 ± 0.49^{b}
K	CM	719.91 ± 45.40^{a,z}	708.26 ± 58.99^{a,y,z}	652.68 ± 7.62^{a,y}	649.47 ± 17.60^{a,w,y}	659.55 ± 20.73^{a,w,y,z}
	AK	613.01 ± 3.85^{b}	602.87 ± 26.11^{b}	545.31 ± 6.34^{b}	612.64 ± 12.21^{b}	566.94 ± 2.10^{b}
Mg	CM	115.27 ± 3.95^{a,z}	145.72 ± 8.59^{a,y}	136.13 ± 2.17^{a,x}	145.76 ± 6.14^{a,y}	154.59 ± 1.74^{a,y}
	AK	106.98 ± 2.01^{b}	125.74 ± 4.09^{b}	116.16 ± 1.51^{b}	125.71 ± 1.51^{b}	117.66 ± 0.61^{b}
Na	CM	1,869.56 ± 71.82^{a,z}	924.83 ± 51.23^{a,y}	1,187.05 ± 17.81^{a,x}	1,043.52 ± 37.56^{a,w}	699.16 ± 27.58^{a,v}
	AK	1,019.69 ± 12.72^{b}	703.63 ± 25.61^{b}	818.98 ± 6.74^{b}	1,180.03 ± 24.68^{b}	1,517.00 ± 3.09^{b}
P	CM	225.20 ± 6.90^{a,z}	334.66 ± 15.40^{a,y}	323.59 ± 6.23^{a,y}	375.40 ± 15.88^{a,x}	366.91 ± 4.35^{a,x}
	AK	210.55 ± 5.28^{b}	285.12 ± 5.17^{b}	269.00 ± 4.56^{b}	353.90 ± 4.33^{b}	271.19 ± 2.49^{b}
MICROELEMENTS (µg g^{-1})						
Cu	CM	2.05 ± 0.03^{a,z}	2.49 ± 0.04^{a,y}	2.44 ± 0.08^{a,y}	2.99 ± 0.19^{a,x}	2.39 ± 0.03^{a,w}
	AK	1.82 ± 0.03^{b}	1.99 ± 0.02^{b}	1.90 ± 0.10^{b}	3.21 ± 0.09^{a}	1.79 ± 0.16^{b}
Fe	CM	38.88 ± 1.20^{a,z}	43.83 ± 2.46^{a,y}	41.96 ± 0.59^{a,y}	48.75 ± 2.93^{a,x,y}	46.09 ± 1.27^{a,x,y}
	AK	39.51 ± 1.61^{a}	39.66 ± 0.13^{b}	38.28 ± 0.07^{b}	50.64 ± 2.00^{a}	39.61 ± 1.28^{b}
Mn	CM	15.89 ± 0.48^{a,z}	13.97 ± 0.65^{a,y}	12.60 ± 0.21^{a,x}	12.92 ± 0.67^{a,y}	14.20 ± 0.26^{a,y}
	AK	14.84 ± 0.39^{b}	11.46 ± 0.23^{b}	10.72 ± 0.26^{b}	11.20 ± 0.15^{b}	14.32 ± 0.14^{b}
Zn	CM	29.12 ± 0.93^{a,z}	30.70 ± 1.57^{a,y,z}	29.14 ± 0.72^{a,y,z}	31.90 ± 1.59^{a,y}	33.90 ± 0.67^{a,x}
	AK	26.83 ± 0.56^{b}	25.28 ± 0.51^{b}	23.79 ± 0.32^{b}	29.82 ± 0.59^{b}	26.19 ± 0.13^{b}
TRACE ELEMENTS (µg g^{-1})						
Al	CM	66.36 ± 2.66^{a,z}	26.36 ± 0.85^{a,y}	53.06 ± 4.41^{a,x}	45.53 ± 10.76^{a,w}	32.38 ± 5.59^{a,w}
	AK	62.84 ± 1.51^{a}	23.47 ± 5.01^{a}	42.74 ± 0.41^{b}	49.92 ± 0.12^{a}	46.00 ± 9.30^{b}
Co	CM	0.09 ± 0.01^{a,z}	0.03 ± 0.00^{a,y}	0.03 ± 0.00^{a,y}	0.03 ± 0.00^{a,x,y}	0.03 ± 0.00^{a,x,y}
	AK	0.08 ± 0.00^{a}	0.03 ± 0.00^{a}	0.03 ± 0.00^{a}	0.07 ± 0.00^{b}	0.08 ± 0.00^{a}
Cr	CM	2.64 ± 0.10^{a,z}	0.73 ± 0.03^{a,y}	0.98 ± 0.07^{a,x}	1.03 ± 0.05^{a,w}	0.90 ± 0.12^{a,y}
	AK	1.27 ± 0.03^{b}	0.99 ± 0.06^{b}	1.18 ± 0.03^{b}	1.71 ± 0.03^{b}	1.85 ± 0.11^{b}
Se	CM	0.12 ± 0.02^{a,z}	0.04 ± 0.00^{a,y}	0.07 ± 0.01^{a,x}	0.04 ± 0.01^{a,w,y}	0.03 ± 0.01^{a,w,y}
	AK	0.06 ± 0.00^{b}	0.03 ± 0.00^{b}	0.03 ± 0.00^{b}	0.04 ± 0.01^{a}	0.04 ± 0.01^{a}
ULTRA-TRACE ELEMENTS* (µg g^{-1})						
As	CM	0.03 ± 0.00^{a,z}	0.01 ± 0.00^{a,y}	0.01 ± 0.00^{a,y,z}	0.01 ± 0.00^{a,x,y}	0.01 ± 0.00^{a,x,y}
	AK	0.01 ± 0.00^{b}	0.00 ± 0.00^{a}	0.01 ± 0.00^{a}	0.00 ± 0.00^{a}	0.01 ± 0.00^{a}
Mo	CM	1.12 ± 0.05^{a,z}	0.90 ± 0.08^{a,y}	1.31 ± 0.13^{a,x}	1.98 ± 0.14^{a,w}	1.06 ± 0.03^{a,y,z}
	AK	0.85 ± 0.06^{b}	0.83 ± 0.05^{b}	0.91 ± 0.05^{b}	1.50 ± 0.07^{b}	0.83 ± 0.01^{b}

* Ultra-trace elements: Cadmium (Cd) < 0.0021 µg g^{-1}, for all samples; Nickel (Ni) < 0.0030 µg g^{-1}. Different letters (a−b) within the same column indicate significant differences between mineral contents determined in CM and AK; Different letters (v−z) within the same row for each mineral content in CM – Mann Whitney ($p \leq 0.05$)

leaching of minerals into fat medium. Moreover, it seems that food with low fat content and high water content (as CMs) are more susceptible mineral loss than fatty food, which may lead to a variation in their mineral composition.

Potassium (K) was the most abundant element naturally present in the CM as well as AK samples (Table **1**) and it is also the most abundant mineral in raw beans (957 to 2,899 mg 100 g^{-1}), [22, 24, 26]. On the other hand, sodium (Na) was determined to be the

most abundant mineral in CM and AK (Table **1**) showing a very wide range in the different samples.This reflects the random addition of salt to the CM (*baiana de acarajé* practice) [1]. It greatly exceeds literature values for cowpea beans (6.1 to 102.0 mg 100 g^{-1}) [3, 22] and akara (305 mg 100 g^{-1}) [26]. Therefore, an average sized akara (80 g) contains 37.5-80.9 % and 9.3-10.4 % of the Dietary Reference Intakes (DRIs) [27] for Na and K (Table **2**), respectively. It is worth highlighting that the akara Na content could be very high, and so it might be harmful to health.

Phosphorus (P) was the second most abundant mineral naturally present in the CM and AK samples (Table **1**). These contents were within the range commonly found in beans (266 mg 100 g^{-1}) [22, 28]. In addition, magnesium (Mg) content in the CM and AK samples was remarkable (Table **1**) and the values were in accordance with those reported in the literature for Brazilian cowpea [3]. A medium sized akara (80 g) contains 26.7–31.4 % (for female) and 20.4–24.0 % (for male) of recommendation fro Mg and 24.0-40.4 % of the recommendation for phosphorus [29] (Table **2**).

Calcium (Ca) levels were relatively low in the CM and AK samples (Table **1**) compared to those already reported for cowpea varieties (91-161 mg 100 g^{-1}) [24]. By removing the seed coat (testa), the dehulling of cowpea grains at the beginning of the akara preparation process may significantly reduce the calcium content. Mamiro *et al.* [30] showed that cowpea grains had higher levels of calcium varying between 958.1 and 992.4 mg kg^{-1} than dehulled cowpea and cowpea flour (303 to 311 mg kg^{-1}). Consumption of a medium size akara do not significantly contribute to the daily recommendation [31] for Ca (Table **2**).

Iron (Fe) was detected in concentrations between 38.28 and 50.64 µg g^{-1} in akara (Table **1**). Iron concentrations of cowpeas have been reported to vary between 61 and 75 µg g^{-1} seed flour [3] slightly higher than the values determined in this study. The composition of legumes depends not only on the species or variety, but also on the growing conditions such as soil, rain fall and other conditions [28]. A medium sized akara (80 g) contained 38.0-50.6 % (male) and 16.9-24.5 % (female) (Table **2**) of the daily needs of iron [32].

Zn and Mn contents obtained in this study (Table **1**) are comparable to those already reported for cowpeas [24, 30, 33]. According to the DRIs [31], 80 g of akara contained 24.0-30.0 % and 17.5-21.8 % (Table **2**) of the Zn requirement for the average female and male, respectively, and 48.9-66.7 % of the DRIs [34] of Mn for female and 38.3-52.2 % for male (Table **2**).

Table 2: Contribution (%) of Akara (Fried Cowpea Food) to the Dietary Reference Intakes (Dris) Of Minerals in Adults

Akara[a] mineral	Range Content[b]	DRIs[c]		% DRIs[d]	
		Male	Female	Male	Female
Macroelements (mg 100 g^{-1})					
Ca	25 – 39	1,000		2.0 – 3.12 %	
K	545 – 613	4,700		9.3 – 10.4 %	
Mg	107 – 126	420	320	20.4 – 24.0 %	26.75 – 31.5 %
Na	704 – 1,517	1,500		37.5 – 80.9 %	
P	210 – 354	700		24.0 – 40.4 %	
Micro, Trace And Ultra-Trace Elements (µg g^{-1})					
Cu	1.8 – 3.2	900		16.0 – 28.4 %	
Cr	1.0 – 1.9	35	25	228.6 – 434.3[e] %	320.0 – 608.0[e] %
Fe	38.3 – 50.6	8,000	18,000	39.3 – 50.6 %	17.0 – 22.5 %
Mn	10.7 – 14.8	2,300	1,800	37.2 – 51.5 %	47.6 – 64.8 %
Mo	0.8 – 1.5	45		142.2 – 266.7[f] %	
Se	0.0 – 0.1	55		14.5 %	
Zn	23.8 – 29.8	11,000	8,000	17.3 – 21.7 %	23.8 – 29.8 %

a: one average sized akara (80 g); b: minimum – maximum; c: DRIs [27, 29, 31, 32, 34], requirements to adults (19 to 50 years); d: DRIs percentage achieved with one average sized akara; e: < UL (Tolerable Upper Intake Level) [39]: 1 mg/day: Cr (0.152 mg/akara); f: < UL [40]: 2,000 µg/day: Mo (120 µg/akara).

Chromium (Cr) levels in foods are generally estimated to be low, about 10−1,300 μg kg^{-1} [35]. Aremu *et al.* [33] found chromium levels below the threshold of the detection system used. Nonetheless, in CM and AK samples chromium levels of 2.64 and 0.73 μg g^{-1}, and 0.99-1.85 μg g^{-1} respectively, have been determined (Table **1**). Thus a medium sized akara contained more than 100 % (Table **2**) of the daily Cr requirements of an adult [32]. This might be due to the high chromium content in Brazilian soils [35]. Furthermore, the Mo content of a medium sized akara (Table **2**) exceeds the human need too [32].

Aluminum (Al) levels in the samples under investigation ranged from 26 to 66 μg g^{-1} in the CM and from 23 to 63 μg g^{-1} in the AK samples (Table **1**). Since the 50^{ths}, the interaction between aluminum household utensils and foods prepared in them is well known [36]. Therefore, this variation may be explained by the addition of water to soak bean grains, chopping in aluminum processors and mass whipping in aluminum pan.

Furthermore, high levels of aluminum exposure may have the potential to produce neurotoxicity and to affect the male reproductive system [37]. The European Food Safety Agency [37] defined a tolerable weekly intake of Al as 1 mg Al kg^{-1} of body weight a week If a 70 kg person consumes one medium sized akara (80 g) every day for one week, his Al intakes would be close to the tolerable weekly intake of 37 mg.

3.2. Minerals and Antinutritional Factors

Di-and trivalent cations, as Ca^{2+}, Cu^{2+}, Fe^{2+}, Fe^{3+}, Zn^{2+}, form water insoluble chelates with $InsP_6$ and $InsP_5$. Zn or Fe could also tightly bind to phytate-calcium complexes with the result of an even lower solubility [7, 24]. In this regard, some critical values of the phytate/mineral molar ratio, as a measure of potential mineral availability for iron, calcium and zinc have been proposed [23]. In all akara samples analyzed, the phytate/Fe molar ratio was above the proposed critical value of 1.0, with a mean value of 18.83. Thus, iron bioavailability in the akara under investigation might be pretty low. The molar ratio of phytate/Ca was determined to be below the critical value of 0.24 [23] – mean value of 1.79. Thus calcium absorption from akara should not be impaired by its phyate content. Based on absorption studies in humans, phytate/Zn molar ratios <5, between 5 and 15 and >15 have been associated with high, moderate and low zinc bioavailability, corresponding to approximately 50 %, 30 % and 15 % of total zinc, respectively [38]. In the akara samples, this calculated mean molar ratio was determined to be 29.68-36.20. This suggests that the phytate present in akara will also significantly reduce the absorption of zinc.

3.3. Antinutritional Factors

No significant levels of $InsP_3$ or $InsP_4$ were detected in CM or AK samples. Perparation of akara resulted in an 30 % decrease in $InsP_6$ content and an corresponding increase in $InsP_5$ (Table **3**). The concentration of the different *myo*-inositol phosphates is in accordance with those reported in the literature [2, 3, 4]. The decrease in phytate content is very likely due to a partial dephosphorylation of the hexaphosphate ($InsP_6$) to pentaphosphate ($InsP_5$) (r= -0.947). Phytate

Table 3: Antinutritional Factors Content of Crude Mass (CM) and Akara (AK) (Cowpea Food) from 25h Deep-Frying (5h/Day) in Crude Palm Oil

Antinutritional Factor *	Sample	5h	10h	15h	20h	25h
$InsP_6$ (μmol g^{-1})	CM	11.45 ± 0.04	11.39 ± 0.02	11.34 ± 0.02	11.17 ± 0.02	10.99 ± 0.04
	AK	10.15 ± 0.02	9.95 ± 0.04	9.86 ± 0.01	9.79 ± 0.04	9.79 ± 0.08
$InsP_5$ (μmol g^{-1})	CM	2.90 ± 0.01	2.89 ± 0.02	2.90 ± 0.01	2.93 ± 0.01	2.97 ± 0.01
	AK	3.57 ± 0.03	3.73 ± 0.05	3.77 ± 0.07	3.83 ± 0.03	3.87 ± 0.01
Condensed tannins (mg eq. CE g^{-1})	CM	1.74 ± 0.01	1.73 ± 0.01	1.74 ± 0.01	1.73 ± 0.04	1.72 ± 0.00
	AK	1.69 ± 0.01	1.69 ± 0.01	1.71 ± 0.01	1.67 ± 0.03	1.67 ± 0.05
Polyphenols (mg g^{-1})	CM	6.38 ± 0.04	6.34 ± 0.06	6.36 ± 0.06	6.34 ± 0.01	6.34 ± 0.02
	AK	6.28 ± 0.04	6.30 ± 0.03	6.27 ± 0.02	6.27 ± 0.02	6.26 ± 0.06
Haemagglutinating activity (HU kg^{-1})	CM	0.50 ± 0.00	0.50 ± 0.00	0.50 ± 0.00	0.50 ± 0.00	0.50 ± 0.00
	AK	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00

$InsP_4$ and $InsP_3$ presented 0.00 ± 0.00 μmol g^{-1} for all samples analyzed. * All samples were significantly equal (Mann Whitney, $p \leq 0.05$) within the same column and row between antinutritional factors contents determined in CM and AK.

hydrolysis can occur during food preparation and production, either by phytase from plants or microorganisms [7].

Haemagglutinating activity in the CM samples was determined to be 0.5 HU Kg^{-1}. No haemagglutining activity was determined in the akara samples (Table **3**). The decrease is very likely due a temperature induced denaturation of the lectins [14].

The content of condensed tannins ranged from 1.72 to 1.74 mg eq. CE g^{-1} in the CM samples and from 1.67 to 1.69 mg eq. CE g^{-1} in the AK samples (Table **3**). The observed loss is very likely due to the soaking process involved, because condensed tannins are water soluble [6]. The observed levels of tannin might not affect the nutritional potential of the cultivars since the values represent less than 10 % of the total dry weight of the samples [24]. Furthermore, Ghavidel & Prakash [13] reported acorrelation between tannins and iron and calcium bioavailability. In cowpeas, a reduction in iron and calcium bioavailability occurred with tannin value ranging from 2.5 to 4.7 mg eq. CE g^{-1}. No significant statistic correlation was found between tannins and any mineral in this study.

Polyphenol content ranged between 6.34 and 6.38 mg g^{-1} in the CM samples and 6.26 to 6.30 mg g^{-1} in the AK samples. There were none significant statistical difference ($p \leq 0.05$) between all samples (Table **3**). The values are in accordance with the range reported in dry beans (5.87-6.62 mg g^{-1}) [2]. Polyphenols have been associated with metal chelating activities [16], although Petry *et al.* [15] did not find a reduction in iron absorption with polyphenols concentrations up to 20 mg. Values above 50 mg lowered Fe bioavailability by 14 %.

3.4. Multivariate Analysis

Scores chat biplot (Figure **1b**) showed that CM 10, 15 and 20 h, and AK 20h contained higher levels of Mg, P, Fe, Cu, Zn, Mo and Ca and they are located at positive PC2, whereas CM 5h was separated from the other CM samples because of the higher levels of Al, Na, As, Cr, Co and Se, being located at positive PC1. AK 10, 15 and 25h samples, which had lower minerals contents, were clustered in the negative PC2 (Figures **1a** and **1b**).

The applied HCA (Figure **2**) confirms through the cluster, the separation of the following groups: CM 5h; AK 5h; AK 10, 15 and 25h; AK 20h and CM 10, 15, 20, 25h (Figures **1b** and **2**). PCA and the clustering showed differences among the mineral content of crude masses, reflecting CM and AK 5h separation from other samples and AK 20h clustering with CM samples, whereas its crude mass (CM 20h) still had high levels of Ca, Fe and P (Table **1**). Frying had a significant effect on AK and CM group separation, because of a loss in the content of most of the minerals quantified.

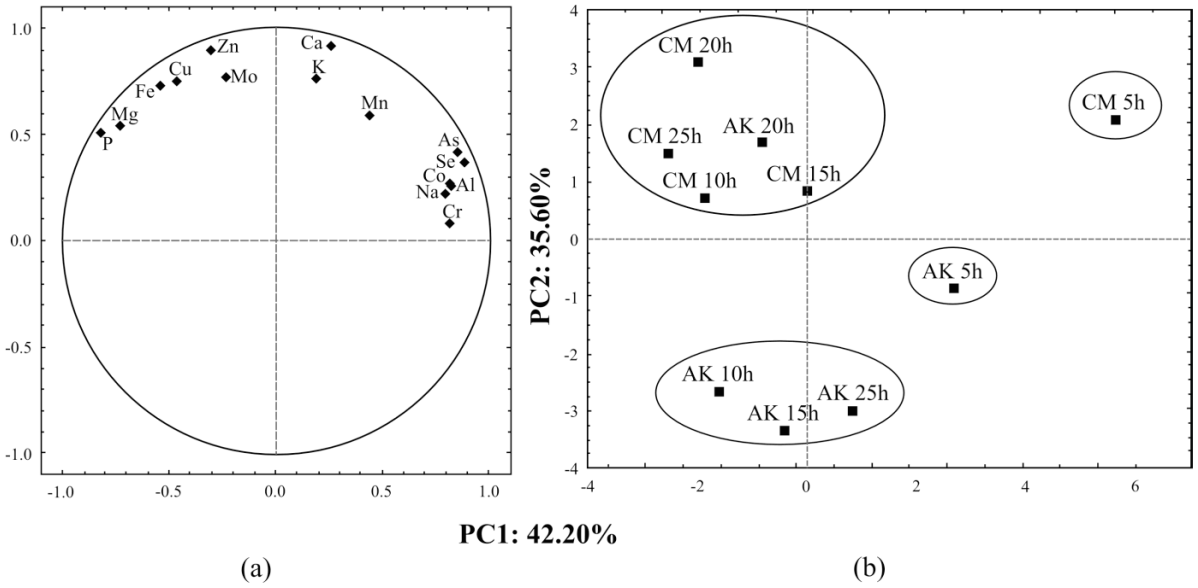

Figure 1: Loadings (**a**) and scores (**b**) plots (PC1 x PC2) of akaras (AK) and its crudes mass (CM) in deep-frying (5h to 25h).

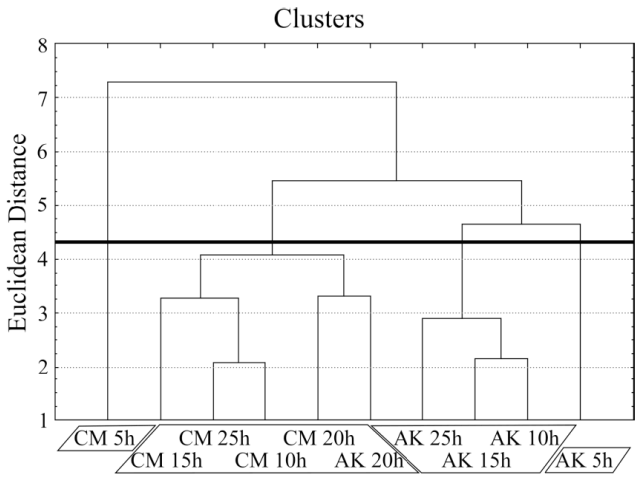

Figure 2: HCA of akara (AK) and its crude masses (CM) in deep-frying (5h to 25h).

4. CONCLUSIONS

Akara, an inexpensive street food, is an integral part of the diet of population of Salvador and it is accessible to economically underprivileged people which often eat it as a main meal. Data obtained from this study showed that akara could partially or totally meet the daily dietary recommendation for chromium, copper, magnesium, manganese, molybdenum, phosphorus and potassium in this population.

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