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Comparative Assessment of Proximate Analysis, Minerals Content and Heavy Metal Accumulation in Ripe and Unripe *Capsicum frutescens*, *Capsicum chinenses and Capsicum annuum* using Mile 12 International Market as a Case Study

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Abstract: ***This study investigated the proximate analyses of nutritional and heavy metal content of ripe and unripe in three pepper varieties (Capsicum frutescens, Capsicum annum and Capsicum chinenses) gotten from Mile 12 market, Lagos state. Pepper has been used for thousands of years as spices in food to enhance the flavour, colour and aroma of food. They are added at a substantial quantity to produce a characteristic taste of cuisine in Nigeria and other parts of the world. They are also known for their preservative and medicinal value. Raw data handling were as specified in American Public Health Association (APHA), American Water Works Association (AWWA) and Water Environment Federation (WEF).Means of the samples were determined using Statistical Package for the Social Sciences (SPSS). The result of the analysis showed that the proximate composition of Capsicum annum was significantly (p<0.05) higher than Capsicum chinenses and Capsicum frutescens in moisture, crude protein, crude fat, total carotenoids and ash contents. Capsicum chinenses was significantly (p<0.05) higher than Capsicum annuum and Capsicum frutescens carbohydrate and sugar contents. Capsicum frutescens was significantly (p<0.05) higher than Capsicum annuum and Capsicum chinenses in fibre contents. Mineral composition showed that Capsicum annuum was significantly (p<0.05) higher Capsicum frutescens and Capsicum chinense in calcium, sodium, magnesium, potassium contents. Capsicum chinenses was significantly (p<0.05) higher than Capsicum annuum and Capsicum frutescens in iron and manganese contents. Capsicum frutescens was significantly (p<0.05) higher than Capsicum annuum and Capsicum chinenses in phosphorus and zinc contents. Heavy metal contents showed that Capsicum frutescens and C. chinenses have same values for copper and significantly (p<0.05) higher than Capsicum annuum. Capsicum frutescens was significantly (p<0.05) higher than Capsicum annuum and Capsicum chinenses in selenium contents. And, they all are insignificantly (p>0.05) different in lead, chromium and cobalt. The results indicate that unripe Capsicum spp has better nutritional values than the ripe Capsicum spp. Although, both are used as major condiments in preparation of traditional diets in Nigeria and can be utilized as sources of protein supplement, valuable minerals and dietary antioxidants. They can serve as scavengers of free radicals reported to be associated with occurrence of chronic and degenerative diseases such as cardiovascular diseases. Hence, their cultivation and consumption should be encouraged.***

Keywords: Capsicum frutescens, Capsicum annuum, Capsicum chinenses, ripe and unripe, Heavy Metals, Proximate values, Lagos

1. Introduction

The genus Capsicum comprises a large and diverse group of plants producing fresh fruits varying from sweet to hot. Originating from Latin American tropical regions, spreading from Chile to the southeastern United States, the Capsicum species are cultivated and appreciated around the world due to the unique flavor, spice uses, and presence of hot taste of the fruits. They are consumed fresh and in different forms of processed products [1]

Capsicum comprises about 30 species, of which, five are domesticated. These comprise *Capsicum annuum L*. (Hot and Sweet peppers), *Capsicum chinense Jacq*. (Aromatic chili pepper), *Capsicum frutescens L*. (Bird pepper), *Capsicum baccatum L*. (Aji) and *Capsicum pubescens* Ruiz and Pav. (Rocoto). The first three species are the most cultivated in both tropical and temperate zones. *C. annuum* often forms a complex with *C. frutescens* and *C. chinense*. In Africa, they are generally considered together as *C. annuum L*. [2;3].Three out of the five domesticated species, namely: *Capsicum annuum, Capsicum frutescens* and *Capsicum chinense* grow well in many communities of Nigeria and constitute important spice in most foods. The following varieties are widely grown in Nigeria and commonly consumed in the South West, that is *Capsicum frutescens:* these are cayenne red pepper, they are known as Bird pepper (local name Ata wewe), cayenne pepper (locally

known as Ata Sombo) and Bird eye chilli pepper (known locally as Ata bawa) and *Capsicum annum*: Bell pepper (Tatase) and Sweet pepper (Ata rodo) [4; 5].

In all, these species are the most common and extensively cultivated of the five domesticated species. It is an important economic food crop which is rich in bioactive nutrients and dietary antioxidants [6]. The intake of these compounds in food is an important health-protecting factor. They have been recognized as being beneficial for prevention of widespread human diseases, including cancer and cardiovascular diseases, when taken daily in adequate amounts [7]. Also, *Capsicum frutescens L*. is known for their preservative and medicinal value [8; 9]. They are good sources of essential minerals such as Magnesium, Zinc, Iron, Phosphorous and Potassium [8].

Capsicum chinense is an excellent source of vitamin- A and C. In addition, capsaicinoids exhibit antioxidant activity and have been demonstrated to protect linoleic acid against free radical attack [10].

Traditionally pepper has been used for its stimulant actions, especially with respect to the circulatory and digestive systems. It is said to increase blood flow thus allaying peripheral vascular disorders, decrease blood pressure, tonify the nervous system, increase appetite, relieve indigestion, and act as a carminative (relieves gas and flatulence). It has antiseptic and antibacterial properties and has made for an excellent gargle for sore throats. Consequently, the uses of these spices cannot be over emphasized.

2. Materials and Methods

Preparation of Samples

Three pepper varieties (*Capsicum annuum, Capsicum chinense*, and *Capsicum frutescens*) were purchased fresh from Mile 12 International Market, Lagos State located in South Western Nigeria. They were washed, first with running water and then with distilled water to remove sand particles, microbes, and other substances that could affect the result of the analysis. The samples were then preserved by storage in clean zip locked bags, at 4°C, until analyses. Before performing the analyses, the residual moisture was evaporated at room temperature. Carotenoids and Vitamin C were analyzed immediately. All the experiments were conducted in triplicates.

Proximate Analyses

Moisture content: Moisture content was determined by drying the sample to a constant weight at 105^0C according to [11].

Ash content: Ash content was measured by calcination at 550^0C to a constant weight, according to [11].

Crude Protein content: Nitrogen content was determined according to the Kjeldahl method and nitrogen value was multiplied by 6.25 as a conversion factor [11].

Crude fibre content: Crude fibre was determined by Acid-alkaline-gravimetric method following the AOAC method [11].

Available carbohydrates: Available carbohydrate was estimated by difference using the relation: 100 - (% crude proteins + %crude lipid + % Crude fibre + %ash) [11].

Energy content: Energy content was estimated in kcal/100g by the Atwater general factors system. The percentage available carbohydrate, crude protein and crude lipid were multiplied by 4, 4, and 9 respectively [12].

Mineral Analysis

The minerals were determined after sample wet digestion with a mixture of $HNO_3/HCl_4O/H_2SO_4$ in the ratio 9:2:1 v/v, respectively. Ca, Mg, Fe, Cu, Zn, Cr, Mn, Co and Pb were determined using atomic absorption spectrophotometer. The Na and K contents of the sample were determined using atomic emission spectrometer and phosphorus by colorimetric method [11].

Data Analysis

The data generated were expressed as mean ± standard deviation of triplicate determinations.

3. Results

The results of the proximate analysis as shown in Table 1 , figures 1,2,3 and 4 show ripe *Capsicum annuum* to have the highest moisture content (84.463± 2.365%) which was higher than the moisture contents of all the other pepper and unripe *Capsicum chinenses* (75.663±4.971%) has the lowest moisture content. Ripe *Capsicum frutescens* has the lowest crude protein value (2.098±0.186%) which was lower than those of ripe *Capsicum chinenses* (2.145±0.886%) and Capsicum annuum (2.642±0.348%) while unripe *Capsicum annuum* (3.405±0.325%) has the highest protein value. The fat content of ripe Capsicum annuum (2.108±0.45%) has the highest value while unripe *Capsicum frutescens* (1.118±0.087%) has the lowest fat content. The crude fibre contents of the unripe *Capsicum frutescens* (2.983±0.418%) was the highest and unripe *Capsicum annuum* (1.658±0.059%) has the lowest fibre content. Unripe *Capsicum annuum* have the highest ash contents (3.088±0.107%) when compared with all other varieties but ripe *Capsicum frutescens* (1.940±0.148%) has the lowest value. The carbohydrate content of unripe *Capsicum chinenses* (14.270±5.197) was higher than those of the other two pepper with ripe *Capsicum annuum* having the lowest carbohydrate contents (6.240±2.705%). Ripe *Capsicum chinenses* has the highest sugar content (1.563±0.386%) and unripe *Capsicum frutescens* (0.875±0.189%) has the lowest sugar content. Ascorbic acid has the highest and lowest content in ripe *Capsicum annuum* (172.965±12.799%) and unripe *Capsicum annuum* (82.570±5.726%) respectively. In Phytic acid both unripe *Capsicum frutescens* and *Capsicum annuum*, have same value of (0.047±0.12%) and ripe *Capsicum annuum* has the lowest value of (0.032±0.009). The total carotenoids of unripe *Capsicum annuum* (0.813±0.198%) has the highest content with unripe *Capsicum frutescens* (0.455±0.052) having the lowest value.

The results of mineral composition of *Capsicum annuum*, *Capsicum chinenses*, and *Capsicum frutescens* as shown in Table 2, figure 5 shows that unripe *Capsicum annuum* has the highest calcium value (69.658±3.912%) and unripe *Capsicum frutescens* has the lowest calcium value (48.183±19.433%). Magnesium content shows that ripe *Capsicum annuum* (39.960±2.450%) has the highest Mg content and ripe *Capsicum frutescens* (18.763±1.904%) also have the lowest Mg content. Ripe *Capsicum annuum* (15.645±1.279%) and unripe *Capsicum chinenses* (10.058±1.201%) has the highest and lowest sodium content respectively. Ripe *Capsicum annuum* has the highest potassium content (53.200±1.338%) with unripe *Capsicum frutescens* (35.153±8.501%) having the lowest value. Iron content shows that ripe *Capsicum chinenses* has the highest iron content (4.240±0.594%) and unripe *Capsicum annuum* has the lowest iron content (2.050±0.099%). Manganese content shows the highest and lowest values in unripe *Capsicum chinenses* (4.995±1.117%) and ripe *Capsicum chinenses* (3.338±1.477%) respectively. Ripe *Capsicum frutescens* has the highest phosphorus value (40.323±2.944%) and unripe *Capsicum annuum* having the lowest phosphorus value (16.270±1.259%). Unripe *Capsicum frutescens* (2.960±0.290%) contains the highest amount of Zinc when compared with other pepper varieties while unripe *Capsicum annuum* (1.563±0.560%) has the lowest.

The results of heavy metal contents of *Capsicum annuum*, *Capsicum chinenses*, and *Capsicum frutescens* as shown in Table 3 shows that ripe *Capsicum chinenses* and unripe *Capsicum frutescens* both have the highest copper value (0.063±0.091%) while unripe *Capsicum annuum* (0.011±0.005%) has the lowest copper content. Ripe *Capsicum frutescens* (0.031±0.015%) contains the highest amounts of selenium when compared with other pepper varieties but unripe *Capsicum annuum* (0.008±0.002%) has the lowest content. Lead, Chromium and Cobalt of all the pepper varieties have low values of 0.001±0.001, 0.002±0.002 or 0.003±0.003, table 3, figure 6.

Table 1: Proximate Composition of Capsicum frutescens, Capsicum chinenses and Capsicum annuum

		Pepper					
		Capsicum frutescens (Ripe)	*Capsicum frutescens* (Unripe)	*Capsicum chinenses* (Ripe)	*Capsicum chinenses* (Unripe)	*Capsicum anuum* (Ripe)	*Capsicum anuum* (Unripe)
Moisture (%)	Mean+SD	80.85±6.617	77.353±6.08	79±8.576	75.663±4.971	84.463±2.365	81.223±2.034
Crude Protein (%)	Mean+SD	2.098±0.186	2.295±0.437	2.145±0.886	3.01±0.71	2.642±0.348	3.405±0.325
Crude Fat (%)	Mean+SD	1.825±0.181	1.118±0.087	1.923±0.677	1.463±0.519	2.108±0.45	1.538±0.438
Crude Fibre (%)	Mean+SD	2.54±0.589	2.983±0.418	2.68±0.691	2.56±1.101	2.18±0.103	1.658±0.059
Ash (%)	Mean+SD	1.94±0.148	2.68±0.762	2.22±0.188	3.035±0.228	2.368±0.314	3.088±0.107
Carbohydrate (%)	Mean+SD	10.748±6.238	12.675±5.63	12.01±9.231	14.27±5.197	6.24±2.705	9.09±1.478
Sugar (brix)	Mean+SD	1.428±0.255	0.875±0.189	1.563±0.386	0.92±0.178	1.465±0.472	0.995±0.182
Ascorbic Acid (mg/100g)	Mean+SD	138.348±42.12	99.233±15.528	136.253±54.71	88.048±11.641	172.965±12.799	82.57±5.726
Phytic Acid (mg/100g)	Mean+SD	0.038±0.021	0.047±0.013	0.04±0.003	0.037±0.007	0.032±0.009	0.047±0.012
Total Carotenoid (mg/100g)	Mean+SD	0.615±0.128	0.455±0.052	0.708±0.094	0.638±0.069	0.803±0.068	0.813±0.198

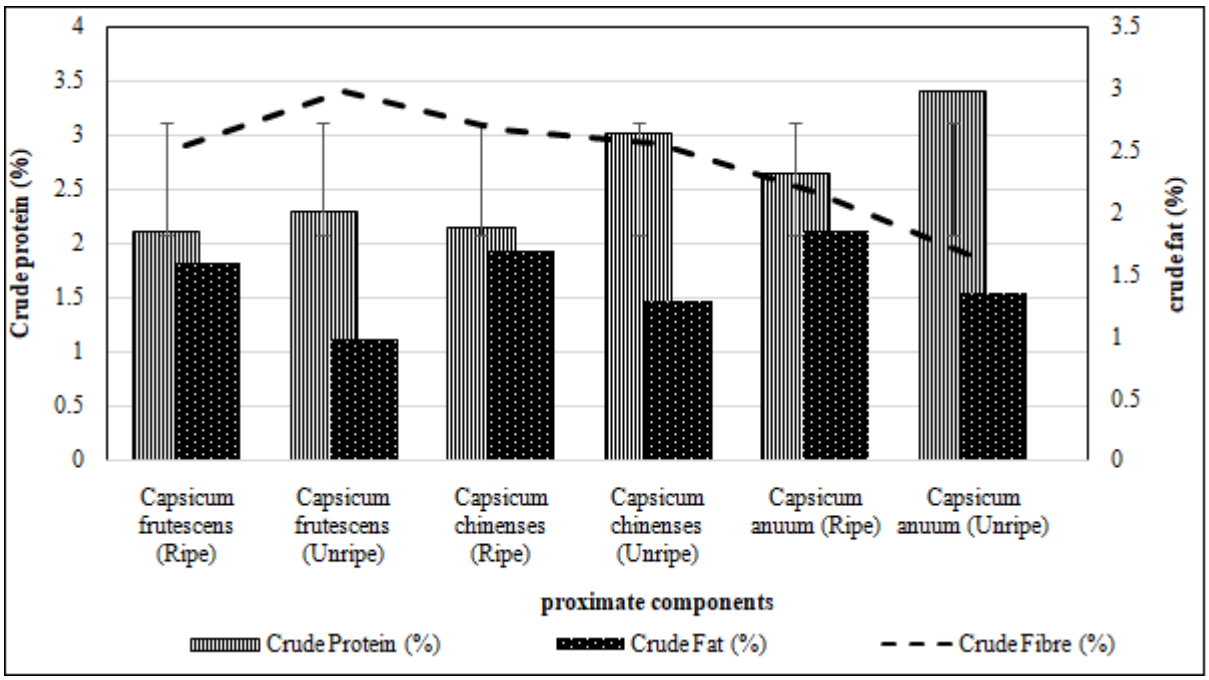

Figure 1: Mean value of Proximate composition of Crude protein, Crude fat and Crude fibre in Capsicum species

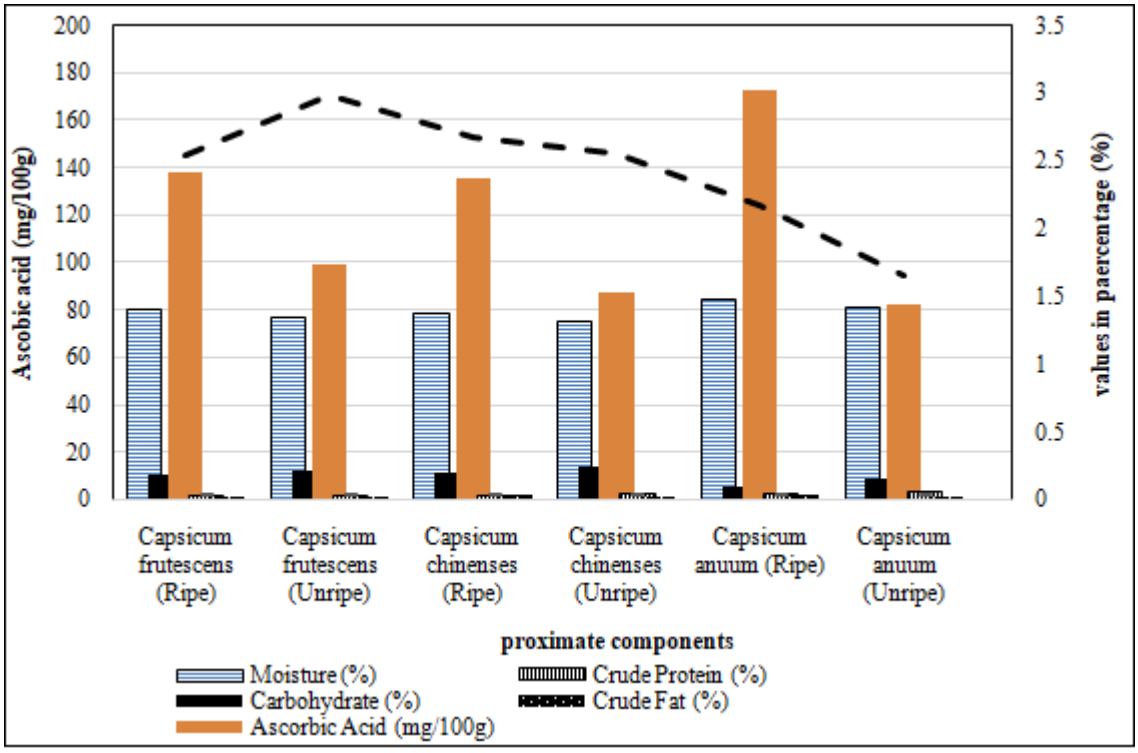

Figure 2: Mean value of Proximate composition of Crude protein, moisture, carbohydrate, and Ascobic acid in Capsicum species

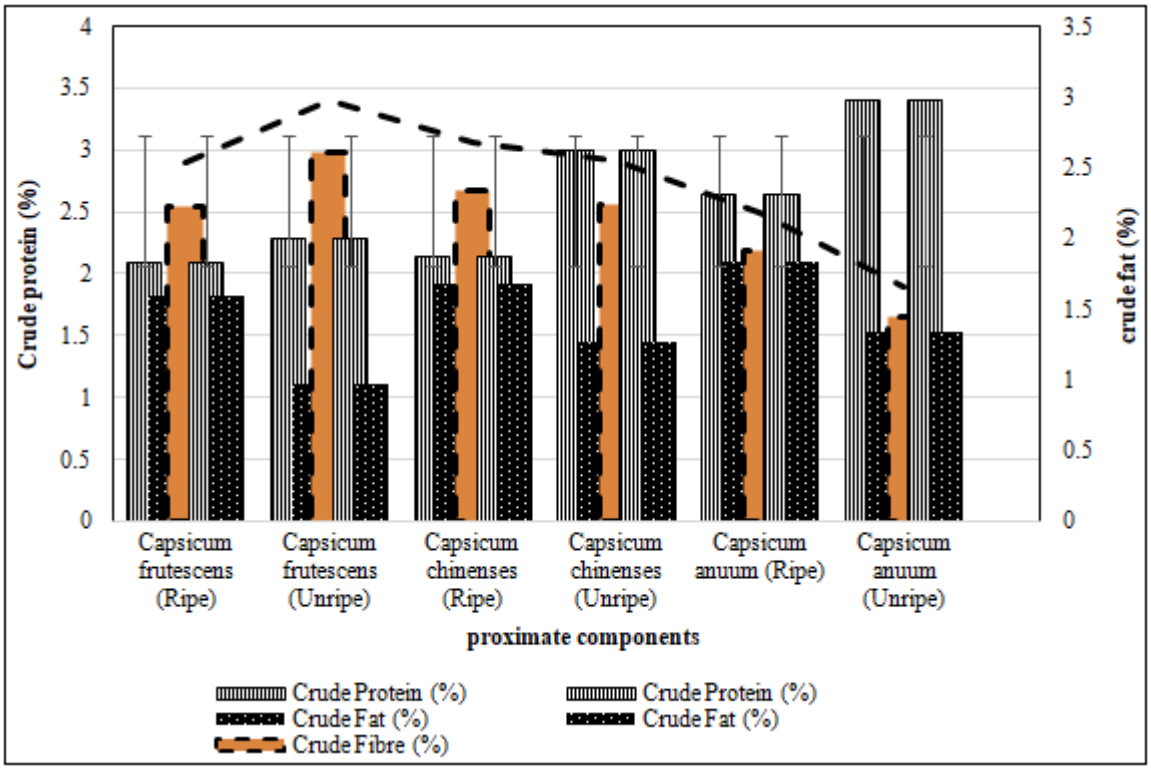

Figure 3: Mean value of Proximate composition of Crude protein, Crude fat and Crude fibre in Capsicum species

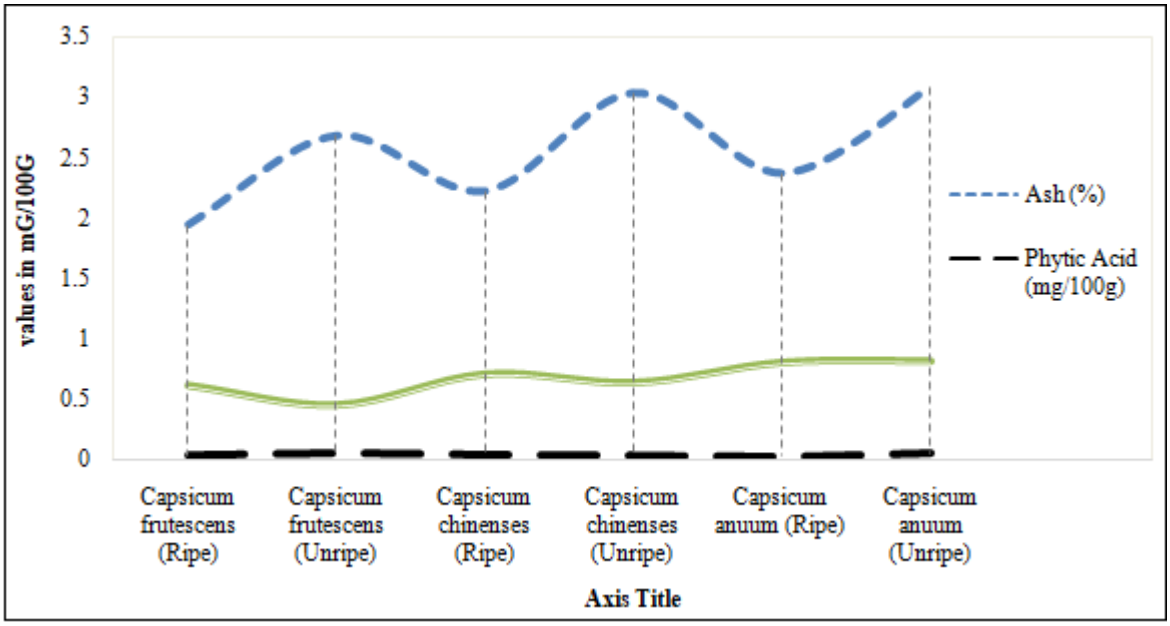

Figure 4: Proximate composition of Ash, phytic acid and total Carotenoid in Capsicum species

Table 2: Minerals Composition of Capsicum frutescens, Capsicum chinenses and Capsicum annuum

Minerals composition		PEPPER					
		Capsicum frutescens (Ripe)	Capsicum frutescens (Unripe)	Capsicum chinenses (Ripe)	Capsicum chinenses (Unripe)	Capsicum anuum (Ripe)	Capsicum anuum (Unripe)
Calcium (mg/100g)	Mean+SD	49.043±5.639	48.183±19.433	59.163±3.37	68.508±5.305	63.12±2.28	69.658±3.912
Magnesium (mg/100g)	Mean+SD	18.7631.904±	23.475±12.121	28.35±15.649	34.603±3.258	39.96±2.45	34.178±3.594
Sodium (mg/100g)	Mean+SD	11.655±0.977	12.913±4.227	13.173±3.678	10.058±1.201	15.645±1.279	10.798±0.673
Potassium (mg/100g)	Mean+SD	47.595±10.507	35.153±8.501	44.073±9.663	37.638±0.963	53.2±1.338	35.287±3.298
Iron (mg/100g)	Mean+SD	3.513±1.469	2.998±2.136	4.24±0.594	4.055±2.223	4.05±0.409	2.05±0.099
Manganese(mg/100g)	Mean+SD	4.253±0.861	3.98±1.084	3.338±1.477	4.995±1.117	4.065±0.741	3.57±0.564
Phosphorus (mg/100g)	Mean+SD	40.323±2.944	37.49±10.687	31.85±6.185	19.285±2.75	23.798±3.395	16.27±1.259
Zinc (mg/100g)	Mean+SD	2.17±0.483	2.96±0.29	2.657±1.055	2.605±1.745	1.858±0.168	1.563±0.56

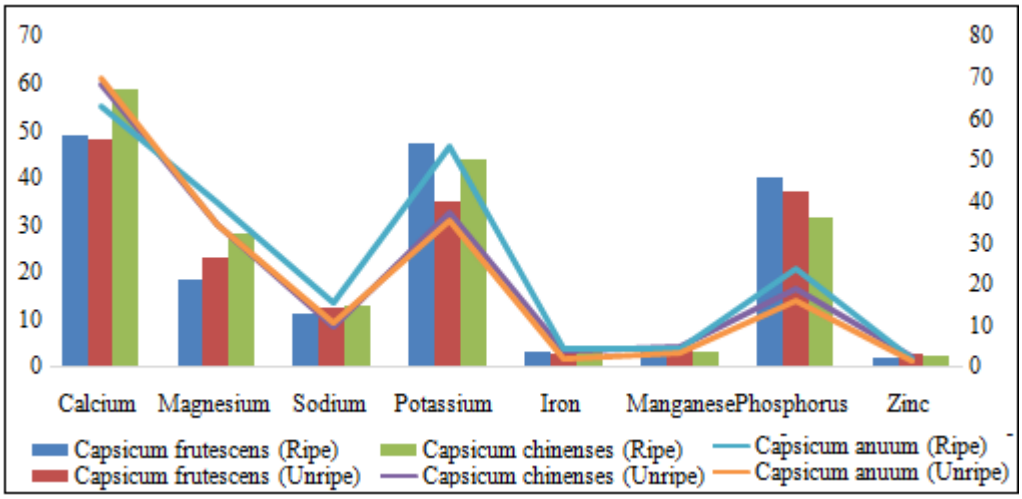

Figure 5: Minerals Composition of *Capsicum frutescens*, *Capsicum chinenses* and *Capsicum annuum*

Table 3: Heavy Metal Content of Capsicum frutescens, Capsicum chinenses and Capsicum annuum

Heavy metals		PEPPER					
		Capsicum frutescens (Ripe)	Capsicum frutescens (Unripe)	Capsicum chinenses (Ripe)	Capsicum chinenses (Unripe)	Capsicum anuum (Ripe)	Capsicum anuum (Unripe)
Copper (mg/100g)	Mean+SD	0.027±0.012	0.063±0.091	0.063±0.091	0.015±0.004	0.013±0.008	0.011±0.005
Selenium (mg/100g)	Mean+SD	0.031±0.015	0.027±0.01	0.026±0.015	0.013±0.005	0.015±0.005	0.008±0.002
Lead (mg/100g)	Mean+SD	0.001±0.001	0.001±0.001	0.001±0.001	0.001±0.001	0.001±0.001	0.001±0.001
Chromium (mg/100g)	Mean+SD	0.001±0.001	0.002±0.001	0.001±0.001	0.003±0.003	0.002±0.002	0.002±0.001
Cobalt (mg/100g)	Mean+SD	0.001±0.001	0.001±0.001	0.001±0.001	0.002±0.001	0.001±0.001	0.002±0.001

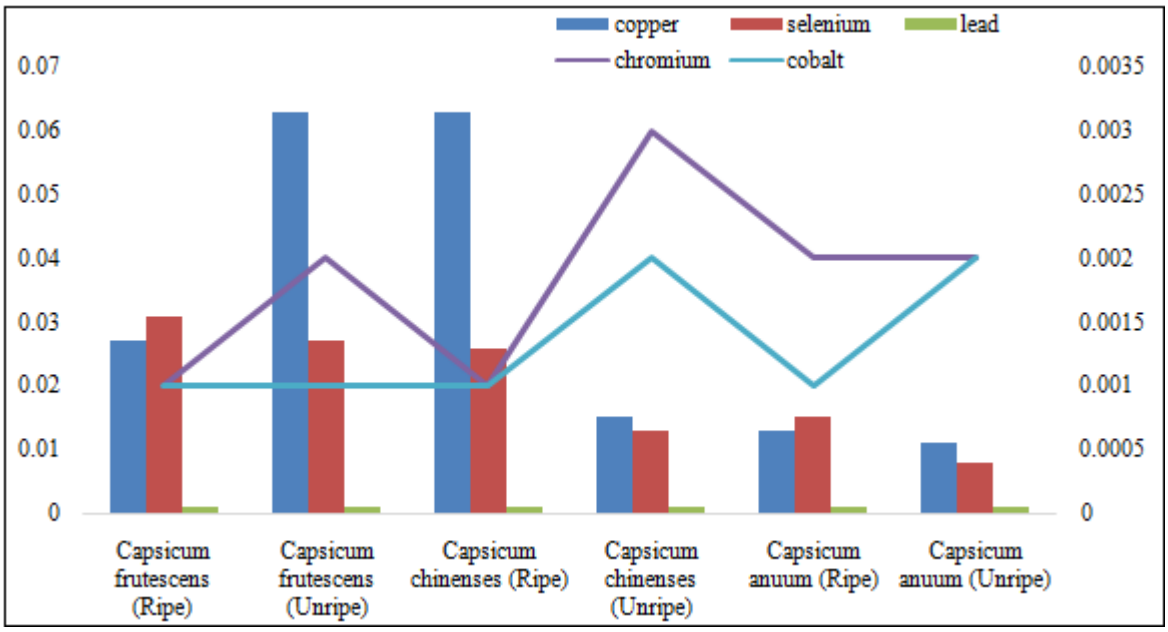

Figure 6: Heavy Metal Content of *Capsicum frutescens*, *Capsicum chinenses* and *Capsicum annuum*

4. Discussion

The result of the analysis revealed that the level of moisture content in all samples were between 77% to 84%. This is higher than the normal percentage value of 50%. The highest % values were gotten from the ripe *Capsicum spp.* The values of the result gotten are still within the permissible limit as recorded by [4], which was 82% to 85%. However, the unripe *Capsicum spp* has lower values which make it advisable for consumption compared to that of ripe. The high moisture content in both samples investigated suggests that they could not be stored for a long period of time without spoilage since water enhances microbial activity leading to food spoilage.

The crude protein content of all samples investigated were relatively low, this could be because protein from plant sources are considered to be of low biological value because an individual plant source does not contain all the essential amino acids. This agrees with the work of [13] and the findings of [14]. They reported that the protein values recorded for the pepper varieties are lower than some commonly consumed plant proteins in Nigeria. The World Health Organization (WHO) sets the safe level of protein intake at 0.83g per kilogram per day, which is expected to meet the protein needs of 97.5% of the world's healthy adult population. In this study, the ripe and unripe *Capsicum spp* have very low values between 2-3.4%. Though, both values are very low in comparison of WHO, but the unripe *Capsicum spp* has a higher protein content therefore advisable for consumption. These low crude protein values of the pepper varieties suggests that pepper need to be combined with other food substances of high protein value in order to meet the protein requirements of individuals. This report agrees with the suggestion of [15].

The ripe *Capsicum spp* have the highest values of crude fibre than those of the unripe, both samples having values between 1.6-2.9% and this agrees with the findings of [16] who reported that percentage insoluble fibre content of pepper varieties ranged from 1.00±0.6 to 7.3±2.2%. Soluble fibre lowers blood cholesterol and glucose level while insoluble fibre is essential in enhancing digestion and bowel movement [17]. FAO sets the level of fibre intake to be 22.5%.

The level of Ash recorded in all samples for this study is between 1.9 and 3.0%. The values are still within the limit FAO recommended for *Capsicum spp* to be 5.7%. The ash content suggests that the pepper varieties could be a good source of valuable minerals [14]. As regard this, unripe *Capsicum spp* is preferable for consumption than the ripe *Capsicum spp* because they have higher values.

Both the 1997 FAO/ WHO expert consultation and the 2002 WHO/FAO experts consultation recommended that total carbohydrate should provide 55%-75% total energy. The carbohydrate content in this result for all samples varies between 6.2-14.2%. Though, the values are very low in comparison of WHO, but unripe *Capsicum spp* has a higher carbohydrate content therefore advisable for consumption. However, these carbohydrates present, even when high in value might not be nutritionally assessable since most of them are bound to remain undigested in the body [18]. Fresh pepper contains a higher amount of ascorbic acid than the other fruits and vegetables [19;20]. In addition, ascorbic acid content of the fresh peppers increases during ripening, but this value decreases during the post-harvesting period [21; 22] WHO recorded that ascorbic acid should provide about 60-200mg/100g. Ascorbic acid content in this result is between 82-172.9%. The values are within the permissible range recorded by WHO.

The Calcium content in this study for both the ripe and unripe *Capsicum spp* is between 48-69mg/100g, this is found to be within the range gotten by [16], which was between 27.2- 54.6 mg/100g. WHO recommended an average of 1000mg to be provided in the body. Therefore, all pepper varieties are safe for consumption.

For magnesium, WHO standard is 30mg/kg depending on the age range. In this study Magnesium content is recorded

between 18.7-39.9mg/100g for both *Capsicum spp.* The unripe samples have higher values than the ripe, which makes unripe *Capsicum spp* more suitable for consumption although both are below the WHO recommendation.

The average amount of Zn in the adult body is about 1.4–2.3g Zn [23;24;25;26]. According to WHO, the zinc value is 0.60mg/kg. In this study, zinc content is recorded between 1.5-2.9mg/100g for both *Capsicum spp.* The unripe samples have higher values than the ripe, which makes ripe *Capsicum spp* more suitable for consumption although both are above the WHO recommendation.

The mean values of Phosphorus in all samples vary between 16-40mg/100g. The values obtained from the samples are within the permissible limit recommended by WHO which is 70mg/kg; making both varieties are suitable for consumption.

According to WHO, copper content should be within the range of 0.05- 0.5mg/100g. Copper in the tested samples has the values between 0.011-0.063mg/100g and are very fit to consume as regards the WHO limit.

According to [27] the normal intake for Lead, Chromium and Cobalt are 2.00mg/kg, 1.30mg/kg and 0.40mg/kg respectively. In this study, they all have mean values of 0.001-0.003mg/100g for all ripe and unripe *Capsicum spp* making them suitable for consumption.

In conclusion, this study showed the proximate mineral nutrient composition and heavy metals of ripe and unripe *C. frutescens, C. chinense and C.annuum*. The study revealed that both varieties contain substantial amount of minerals (Ascorbic acid, Ca, Mg, Na) and permissible levels of heavy metals (Zn, Se, Cr, Pb, Co and Sulphur). Comparatively, the average proximate and mineral values of the unripe sample are higher than those of the ripe samples analyzed. The findings suggest that both varieties contain appreciable amounts of nutrients; however, the consumption of the unripe varieties should be encouraged due to their higher nutritional and mineral quantities inherent in them. It was also observed that no one particular pepper variety is rich in all nutrients; hence the need to consume the pepper as combinations with themselves is imperative.

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