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Comparative Mineral Profiles and Yields of Food Grade Ash from Cow, Goat and Pig Femur Bones

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Abstract: Bones are by-products of abattoir and home consumption of carcases which constitute environmental pollution that attracts houseflies and birds especially vultures in large numbers. Bone dumping is of public health concern because of the health implications but can be harnessed into food grade ash for food fortification because of their high mineral content. This work quantified and compared the mineral content of food grade ash from cow, goat and pig femur bones for uses. The bone samples were procured, sun-dried cleaned, incinerated, dry ashed and analysed for their micro and macro mineral contents using atomic absorption spectrophotometer (AAS) method. The results showed that the femur bones of cow yielded 38.02% raw ash and 10.60% dry ash, goat yielded 40.57% raw ash and 5.86% dry ash while pig yielded 35.60% raw ash and 8.99% dry ash. Results of macro minerals revealed that calcium content range of 610.63-723.16 mg/100g, sodium 2.15-4.07mg/100g, magnesium 7.18-11.23mg/100g, phosphorus 93.11-280.62 mg/100g, while potassium ranged from 2.26 to 3.47 mg/100g. Micro mineral composition showed that copper ranged from 0.001-0.004 mg/kg, iron from 0.022 -1.93 mg/kg, zinc from 0.016-0.144 mg/kg, manganese from 0.007-0.108 mg/kg and sulphur from0.078-0.311 mg/kg. All the none essential heavy metals (toxic minerals) content of the femur bone samples were lower than and recommended safe limit for human consumption and therefore safe. Cow femur bone had the best mineral composition followed by goat and pig femur bones.

Keywords: Cow, Goat and Pig Femur Bones, Raw and Dry Ash, Mineral Contents

1. Introduction

Bone is defined as a dynamic tissue that performs mechanical, biological, and chemical functions [1]. It is a lightweight, strong, hard and rigid organ that constitutes part of the vertebrate skeleton. Bones which are by-product of Abattoir and domestic consumption of carcases consist largely of calcium phosphate and collagen. Animal bones consist of skeleton of most vertebrates which supports the body and protects delicate organs like brain, spinal cord among others. They produce red and white blood cells, stores minerals and enable movement of the vertebrate animals. The Latin word for bone is *os* and the use as a prefix describes bones as in osseous and osteopathy. The *osseous* (bone tissue) is a hard tissue with dense connective tissue and internal honey comb-like matrix that contributed to bone rigidity. Bone tissue is made up of different types of bone cells like osteoblasts and osteocytes that are involved in the formation and mineralization of bone. The mineralized matrix of bone tissue has an organic component of mainly collagen called ossein and an inorganic component made up salts of calcium and phosphate.

Animal femur bones are the largest bone in the body or thigh-bone which is known to contain trace amounts of toxic metals in addition to minerals [2]. Femur bone is a hallow-like structure with a covering membrane known as periosteum beneath which is the cortical (hard) bone layer [3]. The central hollow houses the bone marrow in the form of a

capillary fluid. Each femur bone has two diverging globular structures at its ending known as the articular cartilage that are filled with spongy bone known as trabecular [3]. About 70 to 80% of bone mass is determined genetically while the resulting 20 to 30% is by external factors like diet [4] which has a significant effect on bone mineralization [5].

Minerals are inorganic chemical substances present in all body tissues, bones and fluids which presence is necessary for the maintenance of certain physicochemical processes that are essential to life. They are available as macro, micro and toxic metals to the body. Macro and micro are beneficial to the body while the toxic minerals are health threats. Minerals assist the body in energy production and other biochemical process like maintenance of human health because the human body cannot manufacture minerals. To remedy their deficiency threats to human health, minerals should be incorporated in the diet to increase the nutritional values and restore minerals and organic materials [6]. Bone meal (finely crushed bone) is a good calcium supplement [7, 2]. Biscuit bones can as well be ground and used to fortify human food for stronger bones and proper bone formation. Calcium also plays a vital role in nerve and muscle function, blood clothing, lowering of blood pressure, normal functioning of heart muscles cells and activation of certain enzymes hormones secretion and prevents bowel cancer by binding bile salts in the colon. Excess bile salts in the colon have been associated with increased risk of colon cancer [8].

Calcium and phosphorus are the two major minerals found in bones which form deposits of calcium phosphate held within skeleton by soft, fibrous, organic materials. According to Ebeledike *et al*. [9], calcium content of bones varies among animals. Calcium content of chicken bone unlike phosphorus is higher than those of cattle and goat. Also, species [10] and nutrition affect calcium-phosphate ratio which varies between 1:3 and 2:0 (per weight) and race minerals such as magnesium, sodium, potassium and carbonate. Calcium-phosphorus ratios of animal bones (cattle, goat pig) also vary significantly with animal parts depending on their diet. In their work on animal bones, Ebeledike *et al.*[9] reported calcium content of 730.33 mg/100 g for cow, 655.60 mg/100 g for goat and623.93 mg /100 g for pig bone. Also phosphorus content of 293.05 mg/100 g for goat, 278.87 mg/100 g for cow and 98.18 mg/100 g for pig bone. Bones are constantly being reformed depending on the amount of calcium entering and leaving the bones each day. Therefore, constant supply of calcium from the body stores and the diet is necessary throughout our life time more important during growth phases and in deficiencies [11]. The turnover of bone calcium varies with age which is more rapid in infants, by age one, they have entirely new bones. As adults get older, the bones begin to lose calcium and bone mass leading to osteoporosis in certain individuals. Due to beneficial and some toxic mineral content of femur bones which could limit their use as food fortificants, this study therefore aimed at assessing the mineral profiles of cow, goat and pig femur bones for their essential and non-essential (toxic) mineral contents.

2. Materials and Methods

2.1. Collection of Femoral Bone Sample

Cow femur bones (Figure 1) were procured from Ubakala Abattoir, goat femur bone (Fugure 2) was procured from Ndume Goat market and pig femur bone (Figure 3) was from Umuariaga, Umudike road side market all in Umuahia North Local Government in Abia State. Nigeria.

Figure 1. *Cow femur bone.*

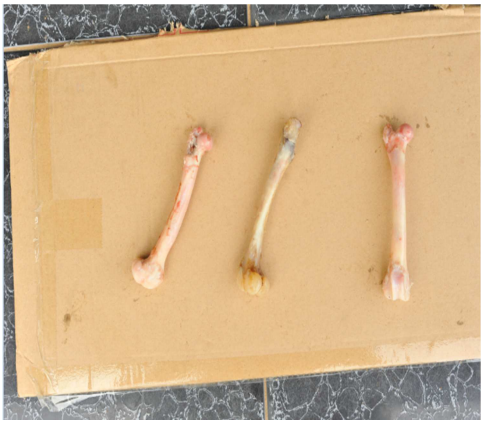

Figure 2. *Goat femur bone.*

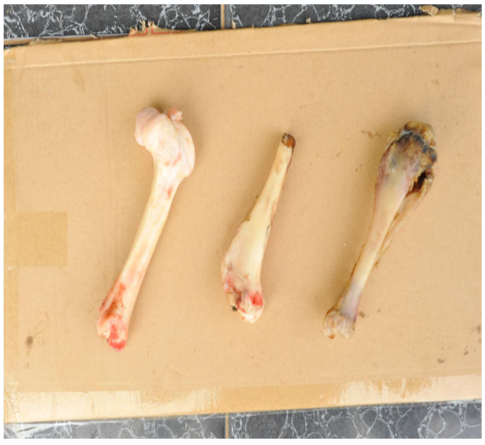

Figure 3. *Pig femur bone.*

2.2. Preparation of the Bone Samples for Analysis

The ash samples were prepared (Figure 4) according to

Okwunodulu *et al*. [12] with slight modifications. The bones were separately cleaned by scrapping with knife to remove some adhering flesh, sun dried to remove adhering moisture to enable their open air incineration and stored in moisture free containers for incineration.

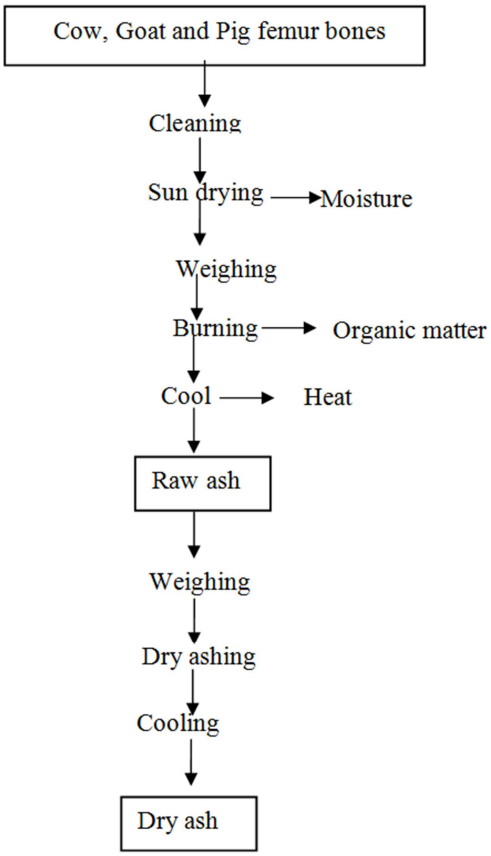

Figure 4. *Flow chart for production of raw ash from cow, goat and pig femur bones.[12].*

2.3. Analysis

2.3.1. Raw Ash Yield

The dried bones were separately weighed and incinerated in open air on a cleaned sheet of metal to obtain raw ash, allowed to cool ground with mortar and pestle, sieved and stored in air-tight container for dry muffle furnace ashing.

The raw ash yield of individual bone sample was determined using the formula:

$$\text{Raw ash yield}=\frac{W_2}{W_1} X \frac{100}{1}$$

Where;

W_1=weight of bones before open air incineration

W_2=weight of ash after incineration

2.3.2. Dry Ash Yield

Three crucibles were washed; oven dried, marked CB for cow bone, GB for goat bone and PB for pig bone samples) and weighed. Exactly 1.004 g of each individual raw ash sample was measured into each crucible according to labels, place into the muffle furnace weighed and ashed at 750^{o}C for 8 hours. After ashing, the crucibles and the greyish white ash were brought out from the furnace using the crucible tongs and were placed in the desiccator to cool before reweighing. The percentage ash yield was determined using the formula:

$$\text{Dry ash yield (\%)}=\frac{W_3-W_1}{W_2-W_1} \text{ x } 100$$

Where;

W_1=weight of empty crucible

W_2=initial weight of empty crucible + raw ash sample before ashing

W_3=Final weight of crucible + ash after muffle ashing

2.3.3. Preparation of Ash Samples for Mineral Analysis

The ash samples were prepared for mineral analysis according to Carpenter and Henricks [13]. The ash samples were separately dissolved in 10 ml of 2M HCL solution and diluted to 100 ml in a volumetric flask using distilled water and filtered. Their filtrates (extract) were used for the analysis

2.3.4. Phosphorus Determination

This was determined by the molybdo Vandate method of Onwuka [14]. A 2 mg of dry ash digest of each sample was separately measured each into a 50ml volumetric flask. The same volume of distilled water and standard phosphorus solution were measured into two 50 ml flasks to serve as reagent blank and standard, respectively. Two ml of phosphorus colour reagent blank (Molybdo Vandate solution) was added into each of the flask and allowed to stand at a room temperature for 15 min thereafter made upto 20 ml mark with distilled water and the absorbance was taken in a spectrophotometer at a wave length of 540 nm with the reagent blank at zero. The phosphorus content was calculated using the formula.

$$\text{P (mg/100g)}=100 \text{ x}\frac{A_u}{w} \text{ x } \frac{C}{A_s}\text{x } \frac{V_t}{V_a}$$

Where;

W=weight of ash sample

A_u=absorbance of test sample

A_s=absorbance of standard phosphorus solution

c=concentration of standard phosphorus solution

V_t=total volume of extract (filtrate).

V_a=Volume of extract analysed

2.3.5. Calcium and Magnesium Determination

Calcium and magnesium contents of the sample extract were carried out using versanate EDTA complexiometric titration described by Capenter and Hendricks [13]. A 20 ml of each extract was dispersed into a conical flask, pinch doses of the masking agents (potassium cyanide, potassium, ferocyanide, hydroxylamine hydrochloride) were added into each. Then 20 ml of ammonia buffer was added to adjust the pH to 10.0. A pinch of the indicator Eriochrom black T was added, shaken very well and titrated against 0.02 NEDTA solution until the colour changed from mauve to a permanent deep blue colour. The result of the titration gave an indication of the amount of a combined concentration of calcium and

magnesium. This is as a result of Ca^{2+} and Mg^{2+} forming complexes at pH 10.0 with EDTA. Same titration was repeated except that the pH was adjustedto 12.0 with 10% HaOH solution before titrating with 0.02N EDTA using selechrome dark blue (Calcon) as an indicator insteadof Eriochrome black T at pH 12.0. A reagent blank was titrated to serve as a control. The experiment was repeated two more times. Calcium and magnesium contents were calculated separately using the formula;

$$\% \text{ calcium or Magnesium} = \frac{100}{w} \times E_W \times N \times \frac{V_f}{V_a} \times T - B$$

Where;
W=weight of sample analysed
EW=equivalent weight
N=Normality of EDTA
V_f=total volume of extract
V_a=volume of extract titrated
T=titre valve of the sample
B=titre valve of blank

2.3.6. Potassium and Sodium Determination

Flame photometry was used to determine the concentrations of potassium and sodium as described by Carpenter and Hendricks [13]. The instrument (Digital flame photometer ME-881) was set up according to the manufacturer's instruction. The equipment was switched on and allowed to stay for about 10 minutes. The gas and air inlet were opened and the start knob was turned on. The equipment was ignited and the flame was adjusted to a non-luminous (blue) flame. Standard potassium and sodium solutions were prepared separately and each was diluted to concentrations of 2, 4, 6, 8 and 10 ppm. Then appropriate filter for potassium and sodium was selected. The highest concentrated standard solution (10 ppm) was aspirated and its emission intensity adjusted to 100 units. Thereafter, starting with the least concentrated (2 ppm), each standard solution was aspirated and caused to spread over the non-luminous butane gas flame. The emission intensity was read directly on the instrument and recorded. Then the sample digest were also aspirated and their readings recorded, the emission intensities of the standard were plotted against their concentrations to obtain a standard curve, (calibration graph) for each element. Subsequently, the optical density emissions recorded from each of the samples were matched against those in the curve by using the curve to extrapolate the quantity of each potassium and sodium ions in the samples. The experiment was repeated two more times to get a mean concentration. The concentration of the test minerals were calculated as follows

$$K \text{ (mg/100 g) or Na (Mg/100g)} = \frac{100}{w} \times \frac{1}{1000} \times X \times \frac{V_f}{V_a} \times D$$

Where;
W=weight of sample used
X=concentration (in ppm) from curve
V_f=total volume of the extract (digest) flamed
V_a=volume of the extract (digest) flamed.
D=dilution factor where applicable.

2.3.7. Determination of Macro Minerals

These were determined using atomic absorption spectrophotometer (AAS), model Buck scientific 205 as described by AOAC [15]. An aliquot of filtrate was aspirated into the AAS and their absorption values corresponding to the different minerals were recorded. Standard solutions of the minerals (Zinc, chromium, iron, copper, manganese, sulphur, silicon, cadmium, lead, aluminium and argon) were also prepared and aspirated into AAS at different wavelengths. A calibration graph was then prepared for the elements from where the amount of the element present in each sample was read. The general formula for the determination of their concentration is shown as:

$$\text{Heavy metal (mg/kg)} = \frac{\text{concetration} + \text{sample solution} \frac{\mu g}{L} \times \text{ML of samples}}{\text{sample weight} \times 1000}$$

3. Statistical Analyses

Data obtained from all the analyses were subjected to analysis of variance (ANOVA) of a completely randomized design (CRD) using the SPSS version 22.0. The treatment means were separated using Duncan multiple range test at 95% confidence level ($p<0.05$).

4. Results and Discussion

4.1. Ash Content Yield of the Bones Samples

The results of raw and dry ash contents are presented in Table 1.

Table 1. *Raw and dry ash yields of the femur bone samples (%).*

Samples	Raw ash	Dry ash
Cow bones	38.02^{b}±0.03	10.76^{a}±0.04
Goat bones	40.57^{a}± 0.04	5.86^{c}±0.04
Pig bones	35.6^{c} ±0.07	8.99^{b}±0.13

Values are means of triplicate determinations ± standard different superscripts are significantly different (P<0.05) deviation. Mean values in the same column with

4.2. Raw Ash Yield

There was significant ($p<0.05$) variations in raw ash yield among the bone samples. Raw ashyield of GB (40.57%) was the highest followed by the CB (38.02%) and CB (35.6%). The difference could be due nutrition, particle size of the bone samples, organic matter composition, animal species and their age [1, 9]. This result signified that GB may likely yield more mineral than the rest bone samples if the dry ash is equally the highest as ash is a measure of mineral composition [14].

4.3. Dry Ash Yield

Dry ash yield of GB had the highest value (40.57%)

followed by CB (38.02%) while PB (35.6%) was the least. There is significant (P<0.05) variations in the dry ash yield of the bone samples. This could be attributed in part to the high ashing temperature in the muffle furnace, specie of the bones, levels of their organic matter content and nutrient [9, 16]. The amount of mineral present in the bones, age, bone density [10] and age [1] are also sources of mineral variations. Levels volatilization of some minerals like iron, copper, lead, phosphorous and zinc due to high muffle furnace temperature [16] had been reported as well. These may have been the reason for the interesting observation in CB with the highest raw ash yield (40.57%) and least dry ash value (5.86%). Besides, the general low dry ash values than the raw ash also attested to these sources of variations.

4.4. Macro Mineral Composition of the Ash Samples

The results are presented in Table 2.

4.4.1. Calcium

There was significant (p<0.05) calcium variation among all the bone samples traceable to calcium-phosphorous ratio, species, animal type and nutrition [10, 9]. Also, genetic constitution affects calcium content of animal bone [5, 4]. Their higher calcium values than other minerals validated the report that calcium is the most abundant mineral in animals of both sexes [17, 18]. High calcium content also indicated that the ashes are of food grade with high density calcium [19, 14] liable to meet adults' calcium RDI of 800-1200 mg/day [18]. Consumption of111-166 g, 131.03-196.53 g and 123.53-185.29 g of foods containing respective CB, GB and PB ashes will meet the calcium RDI. They are therefore good calcium source with cow bone (CB) having an edge followed by pig bone (PB). Calcium content of CB was the highest (723.16 mg/100g) followed by PB with 647.64 mg/100gand GB (610.53 mg/100g). Calcium content of CB and GB obtained in this study were lower than the respective values of 730.33 mg/100g and 655.60 mg/100g but PB was higher than 623.93 mg/100 g reported by Ebeledike *et al*. [9]. Calcium is responsible for good teeth and bones as well as some important biochemical reactions in the body. They include among others nerve and muscle function, blood clothing, lowering of blood pressure, normal functioning of heart muscles cells and prevention of bowel cancer by binding bile salts in the colon [20, 8].

Table 2. *Essential macro minerals content of femur bones samples (mg/100g).*

Sample	Ca	Na	Mg	P	K
Cow bones	723.16^{a}±0.04	4.07^{a}±0.02	8.42^{b}±0.02	216.87^{b}±0.03	3.12^{b}±0.02
Goat bones	610.53^{c}±0.04	2.15^{c}±0.02	7.18^{c}±0.02	280.62^{a}±0.03	3.47^{a}±0.02
Pig bones	647.64^{b}±0.05	2.81^{b}±0.00	11.23^{a}±0.01	93.11^{c}±0.01	2.26^{c}±0.01

Values are means of triplicate determinations ± standard deviation. Mean values in the same column with different superscripts are significantly different (P<0.05). Ca=calcium, Na=sodium, Mg=magnesium, P=phosphorous and K=potassium.

4.4.2. Sodium

Like calcium, there was significant (p<0.05) sodium variation among the samples with same trend of increase. Sodium content of the CB (4.07 mg/100g) was the highest followed by PB with 2.81 mg/100g and GB (2.15 mg/100g). Therefore, CB is comparatively a richer sodium source than the rest bones. With the sodium RDI of 2400mg/100g [21], consumption of 58.97 kg, 111.63 kg and 85.41 kg of foods containing respective CB, GB and PB ashes will meet sodium RDI. Therefore, either of the bone ash is a good sodium source. Low vales of sodium in this study could be traced to their volatilization due to high temperature of muffle furnace ashimg [16]. Sodium helps among others to maintain body fluid balance, blood pressure, muscle contraction, nerve transmission [22] and water balance inside and outside the cells [20].

4.4.3. Magnesium

There was significant (p<0.05) magnesium content among the bone ashes with PB having the highest value (11.23 mg/100g) followed by CB (8.42 mg/100g) and GB (7.18 mg/100g). The variations could be traced to variety, diet [5, 4], 2002) and age of the animals [1]. Magnesium content of all the bone ashes recorded in this study was higher than 6.1% reported by Keene *et al*. [1] from metacarpal bone. The difference could be attributed to bone type, age and nutrition of the animals [23]. The RDI of magnesium is 310 to 420 mg/day [24] which requires consumption of3.7-5 kg, 4.3-5.8 kg and 2.8-3.7 kg of foods containing respective CB, GB and PB ashes to meet the RDI. The bone ashes are therefore poor magnesium sources. Magnesium is essential for more than 300 different body enzyme systems responsible for energy generation. It is needed for healthy bones and blood vessels, and essential in nerve and muscle activities [8].

4.4.4. Phosphorous

The phosphorus content of GB (280.62 mg/`00g) was significantly (p<0.05) higher than CB (216.87 mg/100g) and PB (93.11 mg/100g). Phosphorus content of all the samples was lower than the respective values of278.87 mg/100 g (CB), 293.05 mg/100 g (GB) and 98.18 mg/100 g (PB) reported by Ebeledike *et al*. [9]. The variations could be due to type, genetic constitution, diet [5, 4], age [23] and specie [10]. However, this study revealed that GB is the best phosphorous source followed by CB and PB. Considering adult phosphorous RDI of 700 mg/d [25], consumption of 322.77 g, 249.45 g and 751.80 g of foods containing respective CB, CB and PB ashes will meet the RDI. All the samples are good sources. Phosphorous plays a vital role in nearly all chemical reactions in the body, necessary for healthy bones and the production of energy [10].

4.4.5. Potassium

There was significant (p<0.05) potassium variation

among the bone samples with that of GB (3.47 mg/100g) having a higher value than CB (3.12 mg/100g) and PB (2.26 mg/100g). This means that the goat bone is a better potassium source than the rest bone samples. Meeting the RDI of 3500 mg/d. [21] involves consumption of 112.18 kg, 100.86 kg and 154.87 kg of foods containing respective CB, GB and PB ashes which may not be possible. Therefore, none of the bone sample is a good potassium source. Low potassium values obtained in this study could be due to volatilization of some potassium at high temperature during dry ashing in the muffle furnace [16]. Incorporation of potassium into diets will eenhance muscle strength, metabolism, regulation of body fluid balance, transmission of nerve impulses, muscle contraction and proper metabolism [26].

4.5. Micro Mineral Composition of Femur Bone Samples

The results are presented in Tables 3.

Table 3. *Essential micro minerals of the bone samples (mg/kg).*

Sample	Copper	Iron	Zinc	Manganese	Sulphur
Cow bones	0.001^{a}±0.00	0.193^{a}±0.00	0.144^{a}±0.00	0.108^{a}±0.00	0.311^{a}±0.00
Goat bones	0.003^{a}±0.00	0.080^{b}±0.00	0.016^{c}±0.00	0.010^{b}±0.00	0.078^{c}±0.00
Pig bones	0.004^{a}±0.00	0.022^{c}±0.00	0.035^{b}±0.00	0.007^{c}±0.00	0.109^{b}±0.00

Values are means of triplicate determinations ± standard deviation. Mean values in the same column with different superscripts are significantly different (P<0.05).

4.5.1. Copper

There was no significant (P>0.05) variation in copper content among the bone samples. Despite this, PB had the highest copper content (0.004 mg/kg) followed by GB (0.003 mg/kg) and CB (0.001 mg/kg). This slight variation could be attributed to ionic charge, food, water, air or absorption through the skin [27] which never had any significant (p<0.05) effect. With the RDI of 2 mg/d or 20 mg/kg [21], consumption of2000kg, 666.7kg and 500 kg of foods containing respective CB, GB and PB ashes will meet the copper RDI. As this is not feasible, all the samples may be regarded as poor source. Copper is vital for healthy body development and proper functioning of the brains, nervous systems and cardiovascular systems. Copper also boosts the formation of many enzymes responsible for iron metabolism in the body [28].

4.5.2. Iron

Iron content of CB (0.193 mg/kg) was significantly (p>0.05) higher than the rest with GB (0.080 mg/kg) coming next and PB (0.022 mg/kg) the least. Therefore, CB is a richer source of iron than others. The variations still justify the effects of animal species on their mineral content [9]. Consumption of 51.81-93.26 kg, 125-225 kg and 454.54-818.18 kg of foods containing respective CB, GB and PB ashes williron RDI of 10 to 18mg/d or 100 mg/kg to 180mg/kg [29]. This therefore, projected either of the bone samples as good iron source. Iron works in synergy with protein and copper to produce red blood cells that transport oxygen from lungs to all the tissues where they are needed for maintaining all body's life functions like fuelling the cell division, energy production and growth of a developing body [17].

4.5.3. Zinc

The zinc content of the bone samples ranged with significant (p<0.05) difference from 0.016 in GB through 0.035 mg/kg on PB to 0.144 mg/kg in CB. This indicated that CB is a richer zinc source than the rest bone samples. General lower values of zinc may have been as a result of dry ashing [16]. Meeting the zinc RDI of 15mg/d [21] requires consumption of 104.17 kg, 937.5 kg and 42.86 kg of foods containing respective CB, GB and PB ashes daily. They are therefore poor zinc sources. Zinc enhances healthy immune system, maintain taste and smell and protect the liver from chemical damage [20].

4.5.4. Manganese

Manganese content of the bone samples ranged with significant (p<0.05) variation from 0.007 mg/kg in PB trough 0.010 mg/kg in GB to 0.108 mg/kg in CB. To meet the manganese RDI of 1.6 to 11mg/d or 16 to 110mg/kg/d [24], it will require consumption of 14.81-101.05 kg, 160-11000 kg and 228.57-1571.42 kg of foods containing respectively CB, GB and PB ashes. With this, none of the bone sample is a good source. Manganese among others regulates blood sugar, supports healthy bone, nerves and immune system. It is vital for antioxidant and enzyme function, prevents osteoporosis, inflammation, arthritis and osteoporosis [20, 30].

4.5.5. Sulphur

There were significant (P<0.05) difference among all the bone samples with CB having the highest value (0.311 mg/kg) followed by PB (0.109 mg/kg) and GB (0.078 mg/kg). This indicates that the CB should be a better source than the rest bone samples. Sulphur aids in growth and repairs of worn-out tissues [28].

4.6. The non-essential Heavy Metals

The results are presented in Table 4.

4.6.1. Silicon

The PB had the highest silicon content of 1.342 mg/kg followed by CB (0.422 mg/kg) and GB 0.123 mg/kg. There were significant (P<0.05) silicon variation among the bone samples. Silicon is not a common human poisoning heavy metals and its minute quantities in this study may suggest

that they are within safe limit and therefore safe for human consumption. The GB is safer than all the samples followed by CB and PB.

Table 4. *Non-essential heavy metals of the bone samples (mg/kg).*

Sample	Silicon	Cadmium	Lead	Aluminium	Argon
Cow bones	0.422^{b}±0.00	0.024^{a}±0.00	0.003^{c}±0.00	0.102^{b}±0.00	0.017^{a}±0.00
Goat bones	0.123^{c}±0.00	0.021^{a}±0.00	0.017^{a}±0.00	0.000^{c}±0.00	0.015^{ab}±0.00
Pig bones	1.342^{a}±0.00	0.023^{a}±0.00	0.008^{b}±0.00	0.212^{a}±0.00	0.012^{b}±0.00

Values are means of triplicate determinations ± standard deviation. Mean values in the same column with different superscripts are significantly different ($P<0.05$)

4.6.2. Cadmium

Cadmium content of the bone samples ranged without significant ($p<0.05$) variation from 0.021 mg/kg in GB through 0.023 mg/kg to 0.024 mg/kg in CB. With toxicity level of >0.05 mg/kg [31], cadmium levels in the bone samples (0.021-0.024 mg/kg) were <0.05 mg/kg which implied that all the bone samples are safe food fortificants. This results confirmed the report of Johri *et al*. [32] that <0.05 cadmium level is safe for human health. Beyond the safe limit will result in kidney failure, bone softening (osteoporosis) [33] and prostate cancer [31].

4.6.3. Lead

The GB contained the highest lead content (0.017 mg/kg) which is significantly higher than the rest bone samples followed by PB (0.008 mg/kg) and CB (0.003 mg/kg). Comparing these values with the toxicity level of >3mg per week for adults [34], all the bone samples are safe food fortificants as their daily consumption for a week cannot sum up to >3 mg. Consumption of lead beyond safe limit for long period results in immune dysfunction [35] and carcinogenicity [34].

4.6.4. Aluminium

Aluminium content of PB (0.212 mg/kg) was significantly ($p<0.05$) higher than that of CB (0.102 mg/kg) and GB (0.00 mg/kg). Aluminium is only poisonous to human as aluminium phosphide (Alp) at the level of 0.15 to 0.5 grams [36], therefore the aluminium levels in all the bone samples were not poisonous. Beyond the safe limit for long period, deadly profound shock, myocarditsis and multi-organ failure may result [36].

4.6.5. Argon

There was significant ($p<0.05$) argon variation between the bone samples with CB having the highest value (0.017 mg/kg) followed by PB (0.12 mg/kg) and GB (0.01 mg/kg). Argon is not a common human toxic heavy metals and with its occurrence in minute quantities as obtained in this study, they may not potential health risk.

5. Conclusions

Cow femur bone had higher levels of calcium, sodium, iron, zinc, manganese and sulphur than the rest bones. Goat femur bone contained highest quantity of phosphorus and potassium. Pig bone was highest in magnesium and copper. All their ashes were of food grade and were only good sources of calcium and phosphorous. All the bones had high calcium-phosphorous ratio of 3:1 for cow bone, 2:1 for goat bone and 7:1 for pig bone which implies that they will form a strong bone when used consumed in foods. They had lower levels of non-essential heavy metals, below the recommended toxic levels and therefore are safe for human consumption. They are therefore recommended for use in food industry as an acceptable food fortificants in food processing, at homes and for public health intervention to enhance inadequate intake of essential minerals and prevent hidden hunger.

This study therefore revealed that environmental pollution due to dumped bones from abattoirs and domestic consumption of animal carcases can be transformed into food fortificants. Food forticicants are used to enhance nutrient status of human foods so as to prevent their associated deficiency symptoms.

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