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Korrespondenzadresse:
austinelonwa@gmail.com

[1]) Department of Biochemistry, University of Nigeria, Nsukka, Enugu State, Nigeria; [2] Department of Biochemistry, University of Nigeria, Nsukka, Enugu State, Nigeria; [3]) Department of Plant Science and Biotechnology, University of Nigeria, Nsukka, Enugu State, Nigeria

Chemical composition of raw cashew *(anarcardium occidentale)* nuts sourced from Enugu State, South Eastern Nigeria

Chemische Zusammensetzung von rohen Cashewnüssen (Anarcardium occidentale) aus dem Bundesstaat Enugu, Südost-Nigeria

Chinelo C. Nkwocha[1]), Nene O. Uchendu[2]), Angela N. Amujiri[3])

Summary

This work aimed to determine some chemical compositions of cashew nut *(anacardium occidentale).* The concentration of heavy metals, antinutrients, physicochemical properties and polycyclic aromatic hydrocarbons (PAHs) were done using standard methods. Fresh cashew nut samples were collected in bags from Obukpa-Lejja in Nsukka, Enugu State, Nigeria. The cashew nut was fried in a litre of vegetable oil for 10 minutes for easy removal of the seeds from the pods. The data obtained shows that cashew nut contains heavy metals (Cadmium (0.0703 ± 0.0096 ppm), Lead (0.0773 ± 0.0047 ppm) and Mercury (0.8013 ± 0.0785 ppm)); Antinutrients such as Phytate (0.4307 ± 0.0720 mg/kg), Haemagglutinin (10.9522 ± 0.0751 mg/kg), Cardiac glycoside (33.9200 ± 0.1562 %), Oxalate (1.2998 ± 0.1110 mg/kg), Trypsin inhibitor (462.3670 ± 0.1528 mg/kg), Saponin (1.5300 ± 0.0173 %), and Tannin (0.7660 ± 0.0026 %) were present. PAHs present include Acenaphthylene (0.0017 ± 0.0003 µg/g), Fluoranthene (0.0033± 0.0058 µg/g), Phenanthrene (0.0005 ± 0.0002 µg/g), Xylene (0.0008 ± 0.0001 µg/g) and Benzo (b) fluoranthene (0.0013 ± 0.0002µg/g). The physicochemical properties include Acid value (3.4133 ± 0.0777 %), FFA (1.6763 ± 0.0021 %), Viscosity (1.7060 ± 0.0314 Pa.s), Density (0.9363± 0.0012 kg/m3), Saponification (372.4113 ± 0.0015 mg/kgKOH), Iodine value (97.3010 ± 1.1316 mg iodine/g), Peroxide value (31.3330 ± 0.5774Moleq/kg), Refractive index (1.4624 ± 0.0001), Smoke point (83.6670 ± 0.5774°C), Cloud point (104.3330 ± 0.5774°C) and Flashpoint (144.3330 ± 0.5774°C). When the levels of parameters like the antinutrients, heavy metals, physicochemical properties and PAHs exceed a certain level in the system they possess a deleterious effect on the system.

Keywords: antinutrients, cashew nuts, heavy metals, PAHS, physicochemical properties

Introduction

Nuts can be said to be botanical fruits with palatable seeds. They are nutrient-dense with complex matrices abundant in unsaturated fatty acids and other bioactive compounds possessing vitamins, complete protein, phytosterols, minerals, tocopherols, fibres, vitamins, and phenolic compounds. Nuts comprise nearly 4–16% of saturated fatty acids and nearly partial of the total fat comprises of polyunsaturated fatty acids, monounsaturated fatty acids and unsaturated fatty acids, (Desai et al., 2017; Ros, 2010). The cashew tree is a subtropical and tropical tree owned by the family Anacardiaceae, the genus *anacardium* Linn, and the species *anacardium occidentale* Linn var. *annum*. The nut and apple of cashew are the two structural spots of attention in the tree. Nuts make up a significant part of an everyday diet for most consumers and many public health organisations recommend a daily intake of nut as part of an overall healthy diet. Nuts contain a wide variety of health benefits; they improve heart, blood, mental, eye, skin, bone and oral health; They also improve digestive function, memory and metabolism, help manage diabetes, aid in weight loss, boost the immune system, prevent a different type of cancer, lower the risk of formation of gallstones, protect against viral and fungal infections (Jiang et al., 2002). Despite several health benefits, the overindulging of nuts also can have adverse effects (Seeram et al., 2006). Cashew trees are universal across Nigeria and other tropical regions near the equator, therefore the nutritional composition may vary based on origin and geographical location. This study focused on ascertaining the chemical formation of raw cashew *(anarcardium occidentale)* nuts sourced from Enugu State, South Eastern Nigeria.

Materials and methods

Materials

Sample collection

Raw cashew nuts were sourced in bags from Obukpa and Lejja, Nsukka, Enugu State, Nigeria. The cashew nuts were collected on 2nd April 2019. The cashew nut was identified and authenticated by Professor A.O. Nwadinigwe, a botanist, Plant Science and Biotechnology department in the University of Nigeria Nsukka, Enugu State, Nigeria.

Chemicals and reagents

Reagents and chemicals used were procured from Sigma Aldrich, St Louis, USA.

Equipment and instruments

The equipment and instrument used were obtained from Spring Board Research Laboratories, Awka, Anambra State, Nigeria. They include AA240 Absorption spectrophotometer (Agilent Technologies, USA), Beakers (Pyrex, England), Measuring cylinder (Gents products, India), Hand gloves (Supermax Ltd, England), Abbes refractometer (Bellingham, India), Gas chromatography (Accumax, India), Weighing balance (Vikas Ltd, England), Pycnometer (Jianco, India), Thermometer (Branan, Europe), Pipette (Essex, USA), Water bath (Gallenkamp, England), Reflux condenser (Asgi, India), Conical flask (N.r.d. Enterprises, India), Desiccator (Ambala Cantt, India), Test tube (Pyrex, England), GCMS and Oven (Gallenkamp, England).

Methods

Preparation of plant material

Cashew nuts were fried in a litre of vegetable oil for 10 minutes for easy removal of the seeds from the pods. Afterwards, the seeds were oven-dried at a regulated temperature of 40–60 . The oven-dried seeds were sorted to eliminate unpleasant seeds. Pleasant seeds were dehulled and ground into a powder. Powdered samples were kept in an airtight sample container until ready for analysis.

Heavy metal analysis

Heavy metals (mercury (Hg), lead (Pb) and cadmium (Cd)) analysis were carried out with variant AA240 Atomic Absorption Spectrophotometer employing the method of APHA, 1995

Determination of antinutrients

Determination of Saponin and Hemagglutinin
This was done using the method of Obadoni and Ochuko (2001).

Determination of cardiac glycoside

This was done using the method of Wang (2007).

Determination of tannin

This was done applying the Follins Dennis technique as outlined by Pearson (1974).

Determination of phytate

This was carried out applying the technique of Young and Greaves (1940) as endorsed by Lucas Markakes (1975).

Determination of oxalate

This was done using the method of Harborne (1993).

Determination of trypsin inhibitor

This was determined using the method of Osagie (1993).

Physicochemical properties

This was evaluated following the method of AOAC (1990).

Determination of polycyclic aromatic hydrocarbons (PAHs)

AOAC (1990) method was used for the preparation of samples for gas chromatography (GC) analysis.

Statistical analysis

Data obtained were examined employing computer software known as Statistical Product and Solution Service (SPSS) version 23.

Results and discussion

Table 1 showed that the concentration of Pb obtained in the sample (0.0773±0.0047ppm) is lower than those from pineapple 0.37mg/kg, orange 0.32 mg/kg, avocado 0.09 mg/kg (Tegegne, 2015). The content of Pb reported in this study was generally lower than the permissible levels set by FAO/WHO (FAO/WHO, 2001). Thus this study showed that Pb contents are within the permissible limit. Typical symptoms of lead poisoning are brain damage, central nervous disorder, colic, convulsion, anemia and chronic nephritis of the kidneys (Khan et al., 2008). Similarly, results showed that the concentration of Cd obtained in the

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TABLE 1: Heavy metal composition of fresh cashew nut.

Heavy Metals	Concentration (ppm)	Reference values
Cadmium (Cd)	0.0703 ± 0.0096	0.05 mg/kg
Lead (Pb)	0.0773 ± 0.0047	0.1 mmg/kg
Mercury (Hg)	0.8013 ± 0.0785	0.05 mg/kg (CODEX, 1995)

Values are mean ± standard deviation of triplicate determinations

sample (0.0703±0.0096ppm) is lower than that from *arachis hypogeal* seeds 0.290±0.11ppm (Damera et al., 2014). The effects of cadmium on humans are nephrotoxicity, osteotoxicity, cardiovascular toxicity and genotoxicity. Occasional peaks in cadmium intake may cause a drastic increase in fractional absorption of cadmium. About 5% ingested cadmium is absorbed (GMACE, 2001). Mercury was found in the cashew nuts (0.8013±0.0785ppm). The average daily intake of mercury is reported to be between 0.002–0.02mg (GMACE, 2001). Mercury is one of the most toxic elements among studied heavy metals and exposure to high level of this element could permanently damage the brain, kidney and developing foetus (Castro-Gonalez and Mendez-Armenta, 2008). It is among the metals of most concern in human health especially organic mercury. Much has been done in the last decade to limit or remove the sources of mercury contamination of foodstuff.

The antinutritional factors of cashew nut such as tannin, saponin, oxalate, phytate, haemagglutinin, cardiac glycoside and trypsin inhibitor in this study as seen in table 2 were: 0.7660 %, 1.5300 %, 1.2998 mg/kg, 0.4307 mg/kg, 10.9522 mg/kg, 33.9200 %, and 462.3670 mg/kg respectively. The values of saponin (1.5300 %) and tannin (0.76600%) in this work is bigger than (0.1568%, 0.0701%) saponins and (0.0621%, 0.08775%) tannins for sun-dried yellow and red pulps respectively as reported by Okpanachi et al. (2016). Similarly, the values of oxalate in this study (1.29980mg/kg) and phytate (0.43067mg/kg) is also higher than the value of oxalate (0.0287 %, 0.0327 %) and phytate (0.3159 mg/kg, 0.3661 mg/kg) for the sundried yellow and red cashew pulp respectively as reported by Okpanachi et al. (2016).

Food abundant in saponins is significant in individuals' diets to averting peptic ulcer, osteoporosis, check plasma cholesterol, and to truncate heart disease (Loewus, 2002). A diet rich in saponin can be utilized in the therapy of hypercalciuria in humans, suppression of platelet aggregation and dental caries, and as an antidote against acute lead poisoning. Saponin when high in concentrations, impart a bitter taste and may influence nutrient absorption by impeding enzymes (digestive and metabolic) including attaching with nutrients notably zinc (Liener, 2003). Tannin rich foods have a beneficial effect on the therapy and protection of cancer and cardiovascular disease (Habauzit and Morand, 2011). In the diet, if tannin concentration becomes excessive, microbial enzyme roles such as intestinal digestion and cellulose possibly decreased. Tannins form insoluble complexes with proteins and the tannin-protein complexes possibly accountable for the antinutritional properties of tannin-comprising foods (Habauzit and Morand, 2011). Phytate functions in a wide pH-region as an extremely negatively charged ion, consequently, its availability in the diet hurts the bioavailability of trivalent and divalent mineral ions, and such as Cu2+, Zn2+, Ca2+, Fe2+/3+, Mn2+ and Mg2+. In the absence of phytase, phytic acid can obstruct the absorption of these minerals following attaching to them. This leads to enormously insoluble salts that are badly absorbed by the gastrointestinal tract resulting in lesser bioavailability of minerals (Masum et al., 2011). Oxalate can bind to both insoluble (iron, magnesium and calcium) and soluble (sodium and potassium) salts forming oxalate salts. When the level of oxalate is high, it binds to calcium forming insoluble calcium oxalate salt which tends to precipitate in the urinary tract or kidneys, establishing sharp-edged calcium oxalate crystals (Nachbar et al., 2011). Trypsin inhibitor inhibits the activity of the enzyme trypsin in the gut, thereby hindering protein digestion. Excess trypsin synthesis and load on sulfur-containing amino acids in the body are on account of the discharge of cholecystokinin prompted by trypsin inhibitors (Tibe et al., 2007). The difference observed in the values can be a result of the difference in geographical location or different methods of processing.

TABLE 2: The antinutrient constituent of cashew nut.

Antinutrient	Concentration
Phytate (mg/kg)	0.4307 ± 0.0720
Heamaglutinin (mg/kg)	10.9522 ± 0.0751
Cardiac glycoside (%)	33.9200 ± 0.1562
Oxalate (mg/kg)	1.2998 ± 0.1110
Trypsin inhibitor (mg/kg)	462.3670 ± 0.1528
Saponin (%)	1.5300 ± 0.0173
Tannin (%)	0.7660 ± 0.0026

Values are mean ± standard deviation of triplicate determinations

From table 3 below, the physicochemical properties of cashew nut oil such as the iodine value gotten in this study (97.3010 mg/kgKOH) were higher than 42.1 mg iodine/100g reported by Ogungbenle and Afolayan, (2015). Iodine value which is less than 100 is considered as non-drying oil while that greater than 100 is considered as a drying oil, therefore the cashew nut may be said to contain non-drying oil. The saponification value gotten in this study was (372.4113 mg/kgKOH), the value is higher than (139 mg KOH/g) reported by Ogungbenle and Afolayan, (2015). Saponification values exceeding 200mg/KOH indicate the availability of small or slightly small molecular weight fatty acids whereas estimates beneath 190 mg/KOH is a sign that high molecular weight fatty acids are available. Therefore, since the saponification value is greater than 200 mg/KOH, it means that the fatty acids present in the cashew nut may have low molecular weight.

TABLE 3: Physicochemical properties of cashew nut.

Physicochemical properties	Concentration values	Reference
Acid value (%)	3.4133 ± 0.0777	4.0mgKOH/g oil
FFA (%)	1.6763 ± 0.0021	
Viscosity (Pa.s)	1.7060 ± 0.0314	
Density (kg/m^3)	0.9363 ± 0.0012	
Saponification (mg/kgKOH)	372.4113 ± 0.0015	184–196mgKOH
Iodine value (mg iodine/g)	97.3010 ± 01.1316	75–94Wijs
Peroxide value (Moleq/kg)	31.3330 ± 0.5774	15 milliequivalents/Kg
Refractive index	1.4624 ± 0.0001	1.4677–1.4705
Smoke point (°C)	83.6670 ± 0.5774	
Cloud point (°C)	104.3330 ± 0.5774	
Flash point (°C)	144.3330 ± 0.577	

Values are mean ± standard deviation of triplicate determinations. Reference values (CODEX, 1999)

The peroxide value gotten in this study was (31.333 Moleq/kg), Ogungbenle and Afolayan (2015), reported peroxide value of (19.1 Moleq/kg). The high peroxide value gotten suggests that the oil may not go rancid (developing an unpleasant smell or taste) when properly stored because it has already undergone hydrolytic oxidation. The free fatty acid (FFA) value gotten in this study was (1.6763 %), Ogungbenle and Afolayan(2015), reported (4.70%). This low value of FFA suggests that the cashew nut oil may possess a reasonable amount of carboxylic acid functional groups with which an alkali may react to generate a cleaning agent. The acid value gotten for cashew nut oil in this study (3.4133 %) is lower than 11.2% gotten by Ogungbenle and Afolayan (2015). The high saponification value, peroxide value, and low value of FFA indicates that the cashew nut oil may be good for soap making. Similarly, low acid value suggests the oil may partially be good for making soap. The difference observed in the saponification value and peroxide value may be a result of variations in the state of oil conservation such as changes in time for performing the assessments, storage time, furthermore particularities in the course of the oil extraction process.

The refractive index of cashew nut oil for this study was gotten to be (1.4624) and incomparable to 1.460 gotten for cashew nut oil (Ogungbenle and Afolayan, 2015). Refractive index value within this range suggests that the oil may not be as thick as most drying oil whose refractive index falls within 1.475 and 1.485. The values for viscosity, smoke point, cloud point, and flash point for cashew nut oil were: 1.706 Pa.s, 83.667°C, 104.333°C, and 144.33°C respectively: These values showed that the oil may have combustion characteristics.

Polycyclic aromatic hydrocarbons (PAHs) detected in cashew nut sample were a total of five (5) as seen in table 4, out of the five PAHs detected, acenaphthylene, xylene, and phenanthrene are the low molecular weight PAHs (LPAHs) while fluoranthene, benzo(b)fluoranthene are the high molecular weight PAHs (HPAHs) detected. Out of the five number of PAHs detected, fluoranthene contains the highest concentration (0.0033 µg/g) of PAHs while benzo(b)fluoranthene (0.0013µg/g) has the least concentration of PAHs. The total concentration of LPAHs that may have contaminated the cashew soil is (0.003 µg/g) while that of HPAHs is (0.004567 µg/g); from the result gotten, it shows that the cashew nut is majorly contaminated with HPAHs. The total concentration of both the LPAHs and HPAHs that contaminated the cashew nut is (0.0076 µg/g). The total PAH concentration of cashew nut in this work is very much lower than the total PAH reported by Aikpokpodion et al. (2012), on kola nut having HPAHs mean value of 0.8058 µgkg-1 while that of LPAHs has a mean value of 0.05135 µgkg-1. According to the rule proposed by Fernandes et al. (1997), HPAHs contaminate the soil as a result of the high temperature of combustion while LPAHs contaminate the soil through fossil fuel combustion. Therefore, high HPAHs in this study may be a result of the high temperature of combustion. PAHs when contaminated with food show toxic, genotoxic, and immunotoxic effects or are responsible for hormone disruption (Meador, 2003). PAHs such as fluoranthene and benzo (b) fluoranthene when they exceed a certain level in the system tend to be carcinogenic to the system.

TABLE 4: The Polycyclic Aromatic Hydrocarbons (PAHs) present in Cashew Nut.

Polycyclic Aromatic Hydrocarbons (PAHs)	Concentration (µg/g)
Acenaphthylene	0.0017 ± 0.0003
Flouranthene	0.0033 ± 0.0058
Phenanthrene	0.0005 ± 0.0002
Xylene	0.0008 ± 0.0001
Benzo (b) fluoranthene	0.0013 ± 0.0002
Total	0.0076 ± 0.0066

Values are mean ± standard deviation of triplicate determinations. Reference values: Maximum levels of benzo(a)pyrene is 2µg/Kg, sum of benzo(a)pyrene, benzo(a)anthracene, benzo(b)fluoranthene and chrysene is 10µg/Kg

Conclusion

This study revealed that the concentration of lead, cadmium, mercury, antinutrients and PAHs are within safe limits. This study also revealed that the oil extracted from the cashew nut can be used for soap making as revealed through the peroxide, saponification, acid values, and free fatty acid whereas the values of the physical properties showed that the oil may have combustion characteristics. When these parameters exceed an acceptable level they exert a deleterious effect.

Conflict of interest

The authors declare no conflict of interest.

References

Aikpokpodion PE, Oduwole OO, Iloyanomon CI, Adebowale LA (2012): Assessment of polycyclic aromatic hydrocarbon in kola nuts from selected markets in western Nigeria. International Journal of Science and Nature, 3(4): 900–904.

AOAC (1990): Official Methods of Analysis, 15th Edn. Association of Official Analytical Chemists, Washington, D.C., USA.

APHA (1995): Standard method for the examination of water and wastewater, 19th Edn.American Public Health Association, Washington D.C., USA.

Castro-Gonzalez MI, Mendez-Armenta M (2005): Heavy metals: implications associated to fish consumption. Environmental Toxicology and Pharmacology, 26(3): 263–271

CODEX STAN (1995): General standard for contaminants and toxins in food and feed.

CODEX (1999): CODEX standard for named vegetable oils (CODEX-STAN 210-1999).

Damera V, Chinna V, Syeda AU (2014): Effects of heavy metals in *arachis hypogeal* (groundnut) and its treatment. World Journal of Pharmaceutical Research, 3(9): 533–544

Desai D, Raorane C, Patil S, Gadgil R, Patkar D (2017): *Anacardium Occidentale:* Fountain of Phytochemicals; Qualitative Profiling. World Journal of Pharmaceutical Research, 20: 585–592.

FAO/WHO (Codex Alimentarius Commission): Food additives and contaminants. Joint FAO/WHO Food Standard Program, 2001; ALINORM 01/12A: 1–289.

Fernandes MB, Sicre MA, Boireau A, Troczynski J (1997): polyaromatic hydrocarbon (PAH) distributions in the Seine River and its estuary. Marine Pollution Bulletin, 34: 857–859.

Guidelines on metals and alloys-council of Europe (GMACE) (2001): Downloaded from http://www.foodcontactmaterials.com/materials/coe%20metals%20policy%20statement.pdf

Habauzit V, Morand C (2011): Evidence for a protective effect of polyphenols-containing foods on cardiovascular health: an update for clinicians. Therapeutic Advances in Chronic Disease 3: 87–106.

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Harbone JB (1993): Phytochemical methods. London: Chapman and Hall, pp. 11–21.

Jiang R, Manson JE, Stampfer MJ, Liu S, Willett WC, Hu FB (2002): Nut and peanut butter consumption and risk of type 2 diabetes in women. Journal of the American Medical Association, 288 (20): 2554–2560.

Khan SA, Khan L, Hussain L (2008): Profile of heavy metal in selected medicinal plants. Pakistan Journal of Weed Science Research, 14 (1–2): 101–110.

Liener IE (2003): Phytohemagglutinins: Their nutritional significance. Journal of Agricultural and Food Chemistry, 22: 17.

Loewus FA (2002): Biosynthesis of phytate in food grains and seeds. In: Food Phytates. Reddy NR and Sathe SK (Eds). Boca Raton Florida: CRC Press, pp 53–61.

Masum ASMG, Crawford H, Berthold J, Talukder ZI, Hossain K (2011): Minerals (Zn, Fe, Ca and Mg) and antinutrient (Phytic acid) constituents in common bean. American Journal of Food Technology, 6 (3): 235–43.

Meador, JP (2003): Bioaccumulation of PAHS in marine invertebrates. In: Douben PET (Eds). John Wiley and Sons, pp. 147–171.

Nachbar MS, Oppenheim JD, Thomas JO (2000): Lectins in the US diet: Isolation and characterization of a lectin from the tomato (Lycopersicon). Journal of Biological Chemistry., 255: 2056.

Obadoni BO, Ochuko PO (2011): Phytochemical Studies and Comparative Efficacy of the Crude Extracts of Some Homeostatic Plants in Edo and Delta State of Nigeria. Global Journal of Pure and Applied Sciences, 8: 203–208.

Ogungbenle HN, Afolayan MF (2015): Physical and chemical characterization of roasted cashew nut *(anarcadium occidentale)* flour and oil. International Journal of Food Science and Nutrition Engineering, 5 (1): 1–7.

Okpanachi U, Ayoade JA, Tuleun CD (2016): Composition and anti-nutritional (phytonutrients) factors present in both red and yellow varieties of sun-dried cashew pulp. American Journal of Food Science and Health, 2(4): 45–48.

Osagie AU (1993): Antinutritional factors. In: Nutritional quality of plant food. Benin City, Nigeria: Ambik Press Limited, pp. 221–224.

Pearson D (1974): The chemical analysis of foods, 6th Edn. Churchill Livingstone, Edinburgh. p. 451.

Ros E (2010): Health benefits of nut consumption. Forum Nutrition, 2 (7): 652–682.

Seeram NP, Zhan Y, Henning SM (2006): Pistachio skin phenolics are destroyed by bleaching resulting in reduced antioxidative capacities. Journal of Agriculture and Food Chemistry, 54(19): 7036–7040

Tegegne WA (2015): Analysis of heavy metal levels in some edible fruits from some selected markets in Ethiopia. Journal of Modern Chemistry and Chemical Technology, 6 (1): 1–8.

Tibe O, Amarteifio JO, Njogu RMJ (2007): Trypsin inhibitor activity and condensed tannin content in Bambara groundnut (*vigna subterranean* (L.) verdc) grown in Southern Africa. Applied Sciences and Environmental Management, 11 (2): 159–154.

Wang L, Wible BA, Wan X, Ficker E (2007): Cardiac glycosides as novel inhibitors of human ether-a-go-go related gene channel trafficking. Journal of Pharmacological and Experimental Therapeutics. 320: 525–534.

Young SM, Greaves JS (1940): Influence of Variety and Treatment of Phytic acid Content of Wheat. Food Research Journal. 5: 103–105.

Address of corresponding author:
Chinelo C. Nkwocha
Department of Biochemistry
University of Nigeria
Nsukka, Enugu State
Nigeria
austinelonwa@gmail.com