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# *IN-VITRO* NUTRITIONAL, PHYTOCHEMICAL, ANTIOXIDANT AND ANTI-INFLAMMATORY ANALYSES INDICATED DISPARITY IN *Cola nitida L.* and *Garcinia kola* Heckel USED IN NIGERIA

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## ABSTRACT

The research affirmed that *Cola nitida L.* and *Garcinia kola* Heckel used in Nigeria are quite different medicinally and nutritionally. The proportions of all nutrients analyzed quantitatively (lipid, moisture, fibre, ash, and protein except carbohydrate) were greater in *C. nitida* than in *G. kola* Heckel ($p<0.05$). *C. nitida* has a lower energy density than *G. kola Heckel* (332.20 kcal/g vs 349.10 kcal/g sample) ($p<0.05$). Alkaloids, flavonoids, phenolics, and cardiac glycosides were all significantly greater in *G. kola* Heckel than *C. nitida* at $p<0.05$, in addition to saponin, tannin, and steroids. A 72-hour methanol extraction of kola nuts by cold maceration showed that *C. nitida* had significantly lower total phenolic, flavonoid, and tannin contents than *G. kola* Heckel (TPCs: 1815.38±125.90 µg GAE; TFCs: 385.80±37.24 µg QE; and TTCs: 2643.75±36.08 µg TAE/g sample). 2.2-diphenyl-2-picrylhydrazyl (DPPH) and ferric reducing antioxidant power (FRAP) assays showed that *G. kola* Heckel ($IC_{50}$ = 133.70 ± 0.11 µg/mL) was more effective than *C. nitida* ($IC_{50}$ = 226.70 ± 0.21 µg/mL) at the 0.05 level. Studies on anti-inflammation found similar effects for *G. kola* Heckel and *C. nitida* (antiproteinase activity $IC_{50}$ = 162.90 and 223.60 µg/mL; albumin denaturation: $IC_{50}$ = 155.10 and 347.20 µg/mL; and membrane stabilization: $IC_{50}$ = 183.50 and 271.70 µg/mL). Thus, it is proposed here that *G. kola* Heckel, in comparison to *C. nitida*, may be an acceptable source of energy, an antioxidant, and an anti-inflammatory agent in the nutraceutical and pharmaceutical sectors for the benefit of humans.

**Keywords**: Anti-inflammatory, Antioxidant, *Cola nitida*, *Garcinia kola* Heckel, Phytoconstituents

## INTRODUCTION

The availability, affordability, and safety of plant ingredients have led to the shift from mainstream medicine to herbal medicine. Medicine, fibre, food, construction materials, and other necessities come from the aerial parts of plants, such as seeds, stems and leaves among others (Khan *et al.*, 2023; Hussain *et al.*, 2020; Apiamu *et al.*, 2018; Evuen *et al.*, 2016). Plants provide the body with minerals, salts, vitamins, and some hormone precursors in addition to protein and energy (Attah *et al.*, 2022; Evuen *et al.*, 2022). More than 80% of humanity relies on conventional therapies for primary healthcare, according to global report (WHO, 2011). This was necessitated by the fact that "phytochemicals," the chemical compounds found in plants, are responsible for the therapeutic effects of the plants, and is necessary to perform certain physiological functions in humans (Evuen *et al.*, 2022; Himal *et al.*, 2008). Plants contain many phytochemicals, such as alkaloids and phenolic compounds amongst others with unique physiological properties. Also, epidemiological studies have considered the possibility that the eating of some curative plants, particularly nuts, may reduce the risk and severity of human diseases due to the intrinsic phytochemicals with known antioxidant potentials, and other biological functions initiated by their high mineral and vitamin concentrations (Odewade and Odewade, 2023; Fung *et al.*, 2020; Ogunlade *et al.*, 2014; Duraipundiyam *et al.*, 2006). The actions of these phytonutrients were based on their capacities to sequester radicals, chelate transition metals, and enhance antioxidative defence system (Ogunlade *et al.*, 2014). Among the plants of diverse medicinal heritages known in scientific literature (Evuen *et al.*, 2022; Apiamu *et al.*, 2018; Odoemelum, 2005), the kola plants especially their nuts, were comparatively considered as the mainstay of this study.

In the present study, kola nuts were selected above other types of nuts due to their unique properties. The leaves of *C. nitida*, with their distinctive bitter taste, are ideal for refreshing the palate, and the twigs are used as sources of alkaloids in medical compositions (Joshua *et al.*, 2017). Leaves, flowers, twigs, fruit follicles, and bark of *C. nitida* are all used to make a tonic that is said to cure dysentery and other digestive issues associated with characteristics of cough and pain (Okeke *et al.*, 2015; Atawodi *et al.*, 2007). The bacterium, "*Staphylococcus aureus,*" and a host of other pathogenic bacteria have been shown to be susceptible to *C. nitida* nut extracts in an *in vitro* investigation (Ebana *et al.*, 1991). Each extract demonstrated inhibitory activity against the test organisms. In a study, *C. nitida* extract was found to suppress pituitary cell synthesis of Leutinizing hormones (LH), indicating that this extract may control gonadotropin production (Ayebe *et al.*, 2012). Anticarcinogenic, antibacterial, and anti-diabetic properties have all been shown in *C. nitida* (Endrini *et al.*, 2011). However, the species belonging to the genus Garcinia belong to the large, diverse, and tropical family "Clusiaceae," from which they get their fruits and therapeutic uses. Most of these species are still around in bare and semi- domesticated forms with significant regional importance, but they have lately been recognized as so-called neglected or underutilized crops (Manourová *et al.*, 2019). Africans have relied on the medicinal and spiritual properties of the shrub, *G. kola* Heckel (Clusiaceae) for centuries. The indigenous inhabitants in Africa have long picked the fruits from trees in these regions due to the plants' natural homes in the humid tropical woodlands found there.

Farmers in certain regions, away from natural forests, cultivate and manage tree gardens known as agroforests. One of the most significant "non-timber forest products (NTFPs)" in Africa is the seed of this plant (Dranca and Oroian, 2016). The name "wonder plant" is often applied to this plant because of its many potential medical applications (Onasanwo and Rotu, 2016). People in Africa eat the seeds, which are the most valuable part of the plant. This is done for several reasons, including the seeds' astringent taste and their traditional use as a treatment requirement for gastrointestinal issues amongst others (Omeh *et al.*, 2014). The kernel is loaded with beneficial compounds, like tannins and flavonoids. Among these chemicals, the biflavonoid kolaviron complex has garnered the most interest. Neuroprotective, anti-inflammatory, and antibacterial properties, among others, have been hypothesized for this chemical by Manourová *et al.* (2019). Additionally, kolaviron's therapeutic promise extends well beyond its anti-malarial characteristics and capacity to hasten wound healing since it has been shown to reduce symptoms of neurological diseases and Alzheimer's disease (Nwaehujor *et al.*, 2015; Omotoso *et al.*, 2018).

There is a dearth of in-depth comparisons between *C. nitida* L. and *Garcinia kola* Heckel, two extensively utilized plants in Nigeria, regarding their antioxidant, nutritional, phytochemical, and anti-inflammatory profiles. Although the nutritional and therapeutic potentials of these nuts have been studied separately in the past (Omwirhiren *et al.*, 2016; Dah-Nouvlessounon *et al.*, 2015), but a more comprehensive analysis is needed. Without such comparisons, it is difficult to assess the similarities and disparities in the nutritional, phytochemical, antioxidant, and anti-inflammatory markers in the plants thereby preventing their optimal utilization in the study purpose indicated hereafter. Thus, the study's overarching goal was to emphasize the disparity claim between the health benefits of the two kola nuts, and their actual use in dietary supplements, functional foods and medicine respectively.

## MATERIALS AND METHODS

### Chemical reagents

The analytical grade reagents, such as folin-ciocalteu phenol reagent, gallic acid (GA), quercetin ($C_{15}H_{10}O_7$), aspirin ($C_9H_8O_4$), boric acid, tannic acid (TA) and potassium hexacyanoferrate (iii) trihydrate ($K_4Fe(CN)_6.3H_2O$)among others used in this study were procured and supplied by Pyrex Scientific Company, Benin City, Nigeria.

### Plants' nuts of study

*Cola nitida* and *Garcinia kola*, the two most popular types of kola nuts in Nigeria, were purchased from the central market of Abraka Community, and identified by a plant taxonomist at the Department of Botany, Delta State University, Abraka with specimens (ie., voucher specimens: DELSUH105 for *G. kola* & DELSUH106 for *C. nitida*) deposited in the University herbarium. Samples of nuts were washed, air-dried at normal temperature (27 ± 1$^oC$), and pulverized into a fine coarse powdery form, following a 72-hour cold maceration in a pre-determined amount of methanol, and powdered seed samples (i.e., 1250.05 g of *C. nitida* and 1320.4 g of *G. kola*) were sieved by means of Whatman No. 1 filter paper. A rotary evaporator set at 40°C was used to concentrate the filtrate and evaporate the extracting solvent in each case. The yields of the crude extracts, precisely 650.03 g of *C. nitida* and 528.16 g of *G. kola*, were estimated based on the mathematical model shown in Equation (1);

$$\% \text{ Extraction yield } (g/100\ g) = \frac{W_{final}}{W_{initial}} \times 100 \qquad (1)$$

Where; $W_{initial}$ = fresh weight of pulverized plant sample and $W_{final}$ = final weight of extract after evaporation of extracting solvent. The yielded extracts were subjected to various *in vitro* studies.

### Proximate and phytochemical analyses

All nutritional components (i.e., moisture, protein, fat, ash, fibre & carbohydrate contents), and energy were determined using exactly ten grammes (10 g) of freshly gathered kola nuts (*C. nitida* & *G. kola*), in triplicate results as highlighted by the adopted assay protocol (AOAC, 2003). Phytochemical characteristics such alkaloids, tannins, phenolics, glycosides, flavonoids, steroids, saponins, and anthraquinones were also analyzed qualitatively for quantitative analysis of the kola nuts based on the set objective of the existing study (Harbourne, 1984).

### *In-vitro* antioxidant analyses

The two kola samples' total phenolic contents (TPCs) were calculated using a gallic acid (GA) standard calibration curve of y = 0.0013x + 0.174 at 765 nm, with an R-squared value of 0.961, based on the modified folin-ciocalteu procedure stated by McDonald *et al.* (2001), where y and x indicated absorbance and concentration of GA from 50 to 500 g/mL. Using a quercetin (QE) standard calibration curve (y = 0.0011x + 0.3939, R-squared = 0.877, where y is absorbance and x is the graded concentrations of QE from 50 to 500 g/mL), The total flavonoid contents (TFCs) of the samples were determined at 415 nm. This method recognized the colorimetric approach highlighted by Chang *et al.* (2002). Micrograms of QE equivalent per gramme of material were used to express the TFC. Slightly modified from the folin-ciocalteu method described by Singh *et al.* (2016), total tannin content (TTC) was calculated at 725 nm using tannic acid (TA) in a standard calibration curve (y = 0.0008x + 0.2124, R-squared = 0.9718 such that y depicts absorbance units and x depicts graded concentrations of TA from 50 to 500 g/mL).

### Assessment of free radical-scavenging activity

According to the method described by Saeed *et al.* (2012), the ability of the kola nut samples to scavenge DPPH radical was measured at 517 nm by observing the transition of 2,2-diphenyl-2-picrylhydrazyl (DPPH) from a deep violet to a stable yellow-colored complex (diphenyl- picrylhydrazine). In the method described by Yildirim *et al.* (2001), the kola nut samples' ferric reducing power (FRAP) levels were assessed by measuring their capacity to reduce the oxidation state of potassium ferricyanide from $Fe^{3+}$ to $Fe^{2+}$ during its reaction with ferric chloride to yield a ferric-ferrous complex, with the resulting complex's absorbance at 700 nm.

### *In-vitro* anti-inflammatory analyses

The current study evaluated kola nut samples' impacts on albumin denaturation status, antiproteinase activity, and membrane stabilization to assess their anti-inflammatory potentials using *in vitro* assay procedures previously documented in scientific reports (Eshwarappa *et al.*, 2016; Oyedapo and Famurewa, 2008). Dimethyl sulfoxide (DMSO) was progressively used as diluent of samples from 25 g/mL to 500 g/mL. The positive control was the anti-inflammatory drug aspirin (100 g/mL), while the negative control was dimethyl sulfoxide (DMSO).

**Data analyses**

Data of triplicate determinations (n=3) for the respective *in vitro* studies of the kola samples were represented as mean and its standard error, and these were analyzed using GraphPad software v. 8.0 for graphical plots, extrapolation of inhibitory concentration at 50 % ($IC_{50}$) values, and application of unpaired t-test, where mean values with p<0.05 were considered significantly different among samples.

**RESULTS**

**Nutritional disparity**

The nutritional compositions of the kola nut samples are clearly presented in Fig. 1. Among the nutritional contents (lipid, ash, moisture, protein, fibre & carbohydrate) assessed, the percentages of lipid, moisture, fibre and protein contents were statistically higher (p<0.05) in *C. nitida* than that found in *G. kola*. However, the percentages of ash and total carbohydrate contents were significantly higher in *G. kola* than *C. nitida* at p<0.05. In spite of these variations, *G. kola* may be a more promising energy source than *C. nitida* since the former with an energy content of 332.20 ± 2.35 kcal differ significantly (p<0.05) from the later with an energy content of 349.10 ± 1.08 kcal per gram of each sample.

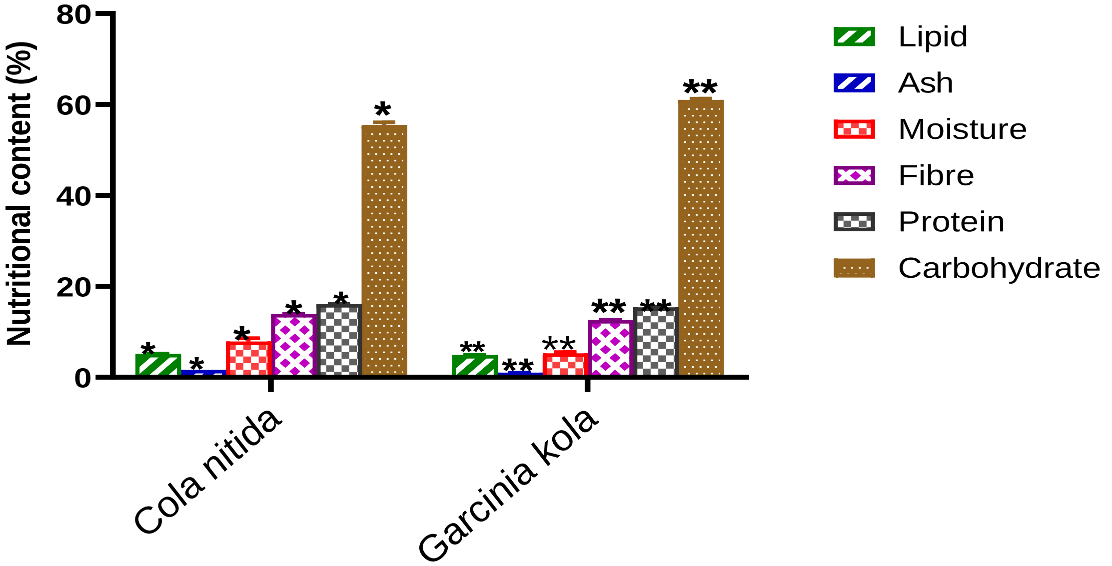


Figure 1**:** Nutritional compositions of *C. nitida L.* and *G. kola*. Each bar represents mean ($\bar{X}$) and standard error (SE) of mean values, expressed as $\bar{X} \pm$ SE of three replicate values. Bars with different asterisks differ significantly (p<0.05).

**Disparity in phytoconstituents**

Anthraquinones were absent from the preliminary phytochemistry, but flavonoids, alkaloids, tannins, saponins, steroids, phenolics, and cardiac glycosides were all present. Fig. 2 illustrates a quantitative comparison of the phytoconstituent levels in two kola nut species. *C. nitida* had a higher overall composition of saponin, tannin, and steroids than *G. kola* did (p<0.05), out of the seven phytoconstituents tested. Contrarily, *G. kola* had significantly larger concentrations of alkaloids, flavonoids, phenolics, and cardiac glycosides than *C. nitida* (p<0.05). This observation provided more evidence that it is a superior antioxidant, anti-inflammatory, and antibacterial source than *C. nitida* in the pharmaceutical industry.

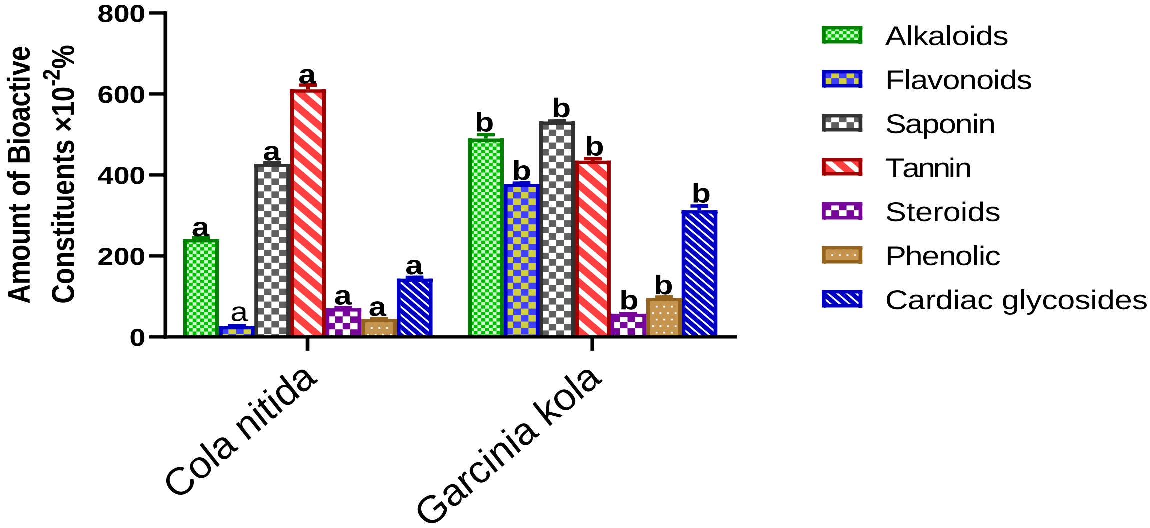


Figure 2**:** Compositions of phytoconstituents in the kola nuts samples. Each bar represents mean ($\bar{X}$) and standard error (SE) of mean values, expressed as $\bar{X} \pm$ SE of three replicate values. Bars with different asterisks differ significantly (p<0.05).

**Disparity in the polyphenolic compositions**

From the data obtained for gallic acid, a standard calibration curve developed was employed for the extrapolation and quantifications of TPCs of the two species of kola. The regression model developed was considered adequate for the estimation of the earlier mentioned endpoint: This was further substantiated with a regression coefficient ($R^2 = 0.961$), which accounted for 98.1% of data generated during the experimentation. On the account of the calibration curve, the TPCs of *C. nitida* (1815.00 ± 21.10 µg GAE/g of sample) and *G. kola* (3926 ± 57.80 µg GAE/g of sample) differ significantly ($p<0.05$), as defined in Fig 3a. This may further suggest that *G. kola* may be a better source of polyphenolic compounds than *C. nitida*.

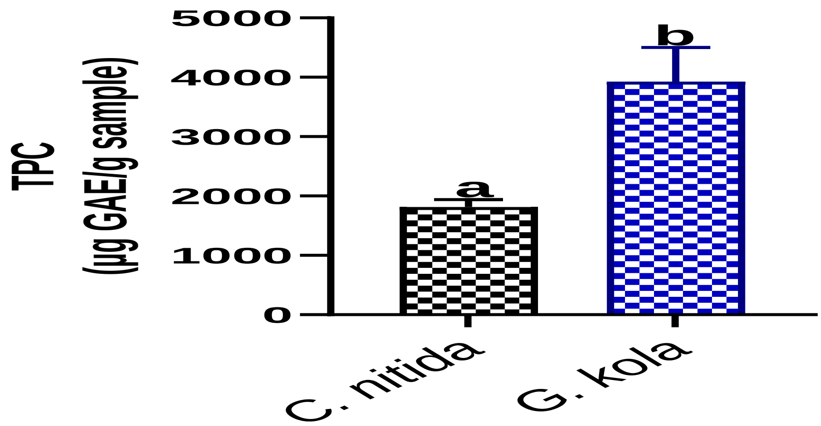


Figure 3a: Compositions of phenolic contents of two species of kola. Bars designated by different letters differ significantly at p<0.05, as a function of triplicate (n=3) determinations. *R-squared value accounted for 96.1 % of the experimental data

To extrapolate and quantify TFCs of *C. nitida* and *G. kola*, the quercetin experimental data were used to generate a standard calibration curve based on the earlier mentioned assay. For the purposes of measuring the aforementioned outcome, the generated regression model was deemed to be sufficient and reliable such that the regression coefficient ($R^2 = 0.887$) corroborated these findings and it explained 87.7% of the experimental data, establishing the model's credibility for the evaluative purpose. Considering the practicability of the aforementioned standard calibration curve developed for quercetin and the outcomes of the aforementioned comparative investigations of TFC in the two kola varieties, an adequate statistical correlation between the samples was established, as shown in Fig. 3b). Consequently, there was a marked variation between the TFC of *C. nitida* (385.80 ± 37.24 µg QE/g sample) and *G. kola* (1007 ± 8.02 µg QE/g sample) at $p<0.05$ level (Fig. 3b). Given the need for bioactive chemicals with drug-like potentials, such as those found in *G. kola* and *C. nitida,* the latter may be highly advised due to the former's abundance of flavonoid-related biological features.

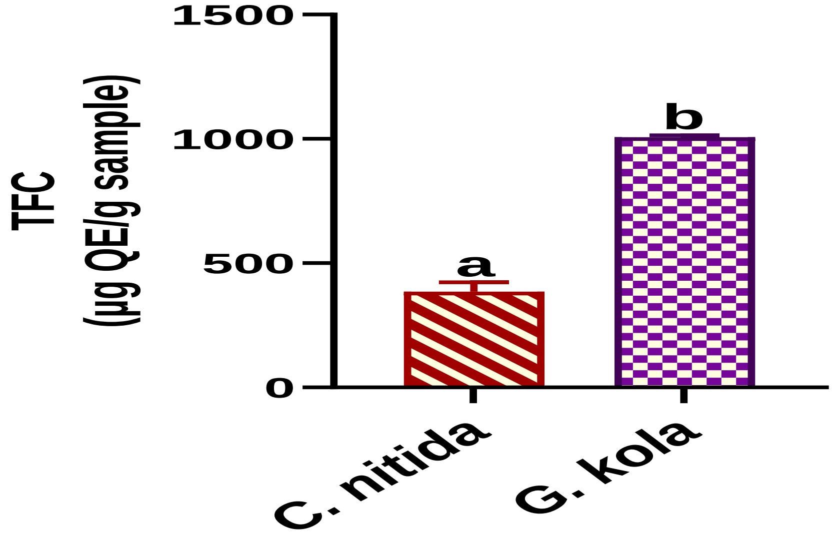


Figure 3b: Compositions of flavonoid contents of two species of kola. Bars designated with different letters differ significantly at p<0.05, as a function of triplicate (n=3) determinations. *R-squared value accounted for 87.7 % of the experimental data

From the data obtained for tannic acid, a standard calibration curve developed was employed for the extrapolation and quantifications of TTCs of *C. nitida* and *G. kola*. The regression model developed was considered adequate for the appraisal of the earlier mentioned endpoint: This was further substantiated with a regression coefficient ($R^2 = 0.9718$), which accounted for 97.18% of data generated during the experimentation, and this made the model reliable for the evaluation process. The comparative studies of TTC in the two kola varieties (*C. nitida* & *G. kola*) can be explained in Fig. 3c, and considering the feasibility of the standard calibration curve developed for tannic acid, the statistical correlation of the two kola varieties in terms of TTC was adequately established. Thus, the TTC of *C. nitida*

(2612.50 ± 31.25 µg TAE/g sample) and *G. kola* (6300 ± 31.25 µg TAE/g sample) differ significantly at p<0.05 (Figure 5). In view of drug-like requirement for bioactive compounds rich in tannic acid related biological properties, *G. kola* may be highly suggested in relation to *C. nitida*.

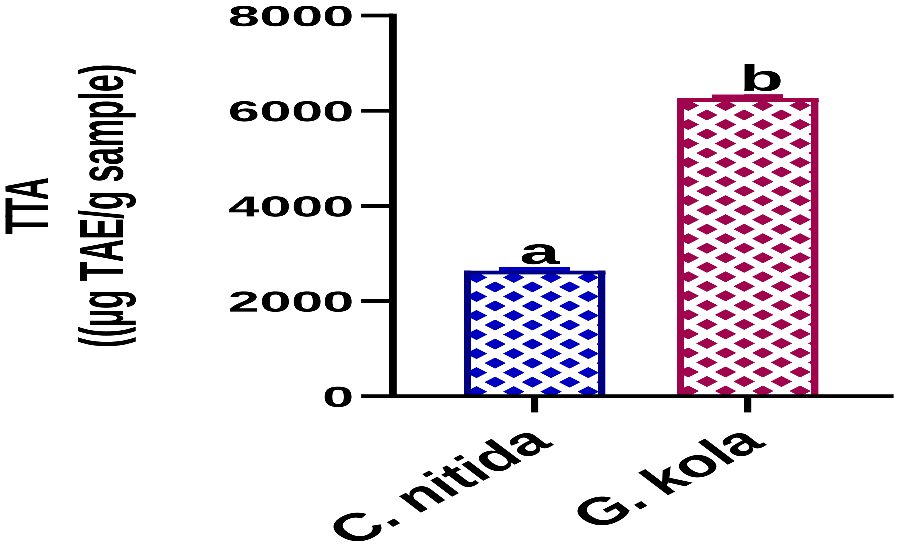


Figure 3c: Compositions of tannin contents of two species of kola. Bars designated by different letters differ significantly at p<0.05, as a function of triplicate (n=3) determinations. *R-squared value accounted for 99.92 % of the experimental data

**Disparity of the free radical-scavenging potentials**

Increasing quantities of samples and standard ascorbic acid clearly explain the free radical-scavenging capacity of the two species of kola investigated in the study built on the proportion of DPPH radical inhibition (Fig. 4a). Founded on the data presented in Figure 4a, the concentration of 500 g/mL of the test samples resulted in maximum inhibition of 68.77, 87.25, and 82.38% for *G. kola*, *C. nitida*, and ascorbic acid, respectively; these values were then converted into $IC_{50}$ values of 133.700.11 and 226.700.21 g/mL for *G. kola* Heckel and *C. nitida*. Since the $IC_{50}$ values decreased with increasing antioxidant activity, this study suggested that *G. kola* Heckel is more potent than *C. nitida* in this regard.

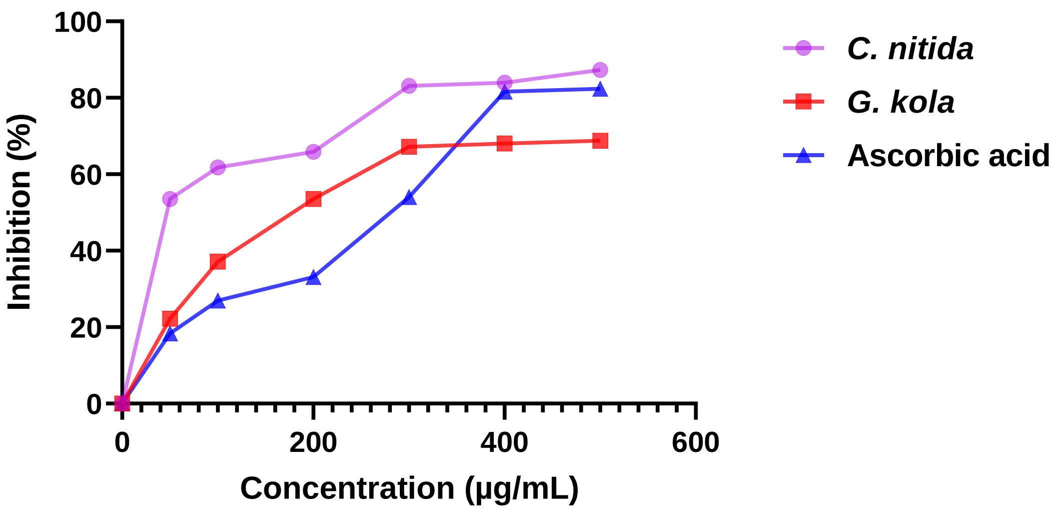


Figure 4a: Inhibitory actions of methanol extracts of *C. nitida* and *G. kola* on DPPH radical with respect to standard antioxidant (ascorbic acid) and the extrapolated mean $IC_{50}$ values, which differed markedly at p<0.05.

The increasing amounts of samples and standard ascorbic acid provided a clear explanation for the free radical scavenging ability of *C. nitida* and *G. kola* Heckel as measured by the proportion inhibition of the potassium ferricyanide radical (Fig. 4b). In Fig. 4b, the maximum inhibitions of 65.80, 78.16, and 94.25% was achieved at 500 g/mL of the test samples for *C. nitida*, ascorbic acid, and *G. kola*, respectively. These values were then converted to $IC_{50}$ values of 300.10 ± 0.52 and 255.70 ± 1.02 µg/mL for *C. nitida* and *G. kola* Heckel, respectively, and these differ significantly at p<0.05. It was possible to deduce from this research that *G. kola* Heckel has more antioxidant activity than *C. nitida* due to its low $IC_{50}$ value.

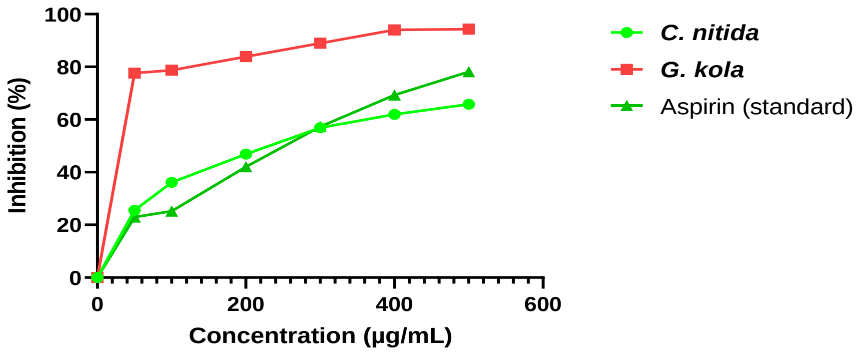


Figure 4b: Inhibitory effects of potassium ferricyanide radical on methanol extracts of two Nigerian kola nuts with respect standard antioxidant (ascorbic acid) and the extrapolated mean $IC_{50}$ values, which differed markedly at $p<0.05$.

**Disparity in anti-inflammatory potentials**

For albumin denaturation, Fig. 5a clearly presents the inhibitory patterns of aspirin (standard ant-inflammatory drug) and methanol extracts of *C. nitida* and *G. kola*. The results showed that increasing concentrations (0-500 µg/mL) of the extracts and aspirin caused a progressive inhibition against albumin denaturation. Consequently, the highest inhibitory activity was found with the administration of *G. kola* ($IC_{50}$ = 155.10 µg/mL) followed by *C. nitida* ($IC_{50}$ = 210.20 µg/mL) and least with aspirin ($IC_{50}$ = 347.20 µg/mL). This may further suggest that *C. nitida* and *G. kola* were better anti-inflammatory sources, though with much preference for the latter when compared with the standard drug.

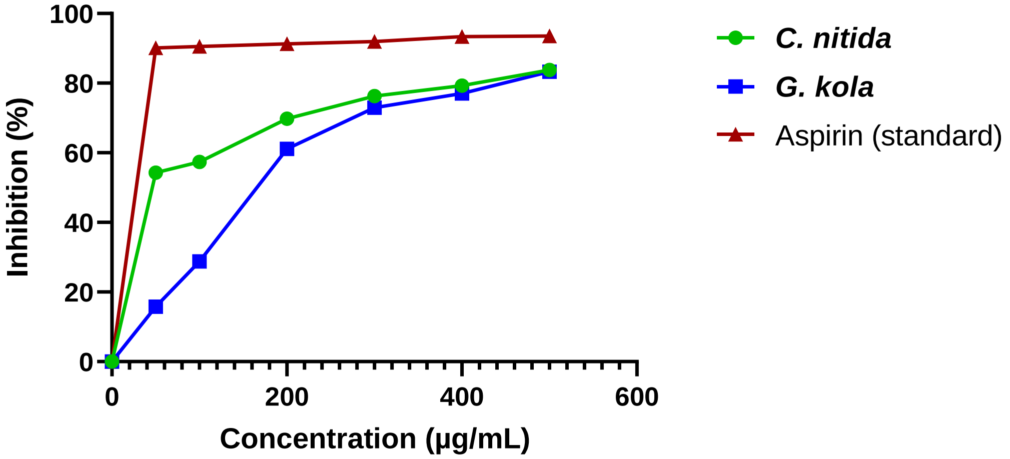


Figure 5a: The inhibitory actions of two kola varieties against albumin denaturation with respect to a standard anti-inflammatory drug (aspirin) and the extrapolation of mean $IC_{50}$ value, which differed significantly at $p<0.05$

Since leukocyte proteinases are essential in the development of tissue damage during inflammatory responses, proteinases are engaged in these processes. Therefore, proteinase inhibitors can afford a high degree of safety. Antiproteinase activity was used as an indicator of anti-inflammatory efficacy in this investigation of *G. kola* and *C. nitida* methanol extracts. Methanolic extracts of both *G. kola* and *C. nitida* showed antiproteinase action (Fig. 5b), with the former having an $IC_{50}$ of 162.9 g/mL and the latter of 223.6 g/mL. While both extracts showed some inhibitory action against proteinase, it was significantly less than that of aspirin (which inhibited 81.5.77% of proteinases at a dosage of 500 g/mL).

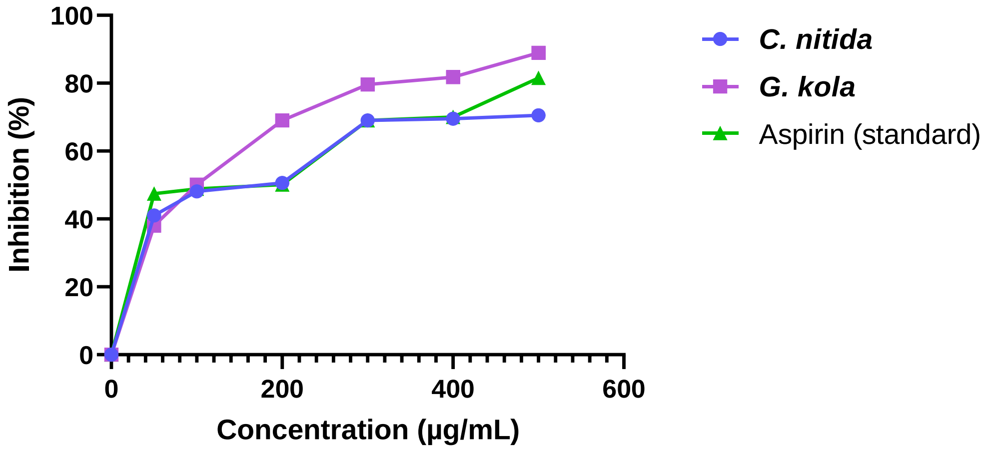


Figure 5b: The antiproteinase actions of two kola varieties with respect to a standard anti-inflammatory drug (aspirin) and the extrapolation of mean $IC_{50}$ value, which differed significantly at $p<0.05$.

The study reveals the effects of methanol extracts of *C. nitida* and *G. kola* on membrane stabilization (Fig. 5c), which is a protective indication against heat-induced haemolysis of RBCs. Increase in the concentrations of the methanol extracts of *C. nitida* and *G. kola* from 0 to 500 μg/mL caused an increased protection by inhibiting heat-induced haemolysis of RBCs in relation to aspirin. Thus, an outstanding inhibitory activity was observed using methanol extract of *G. kola* ($IC_{50}$ = 183.5 μg/mL) followed by *C. nitida* ($IC_{50}$ = 271.70 μg/mL) and aspirin ($IC_50$ = 467.2 μg/mL) respectively

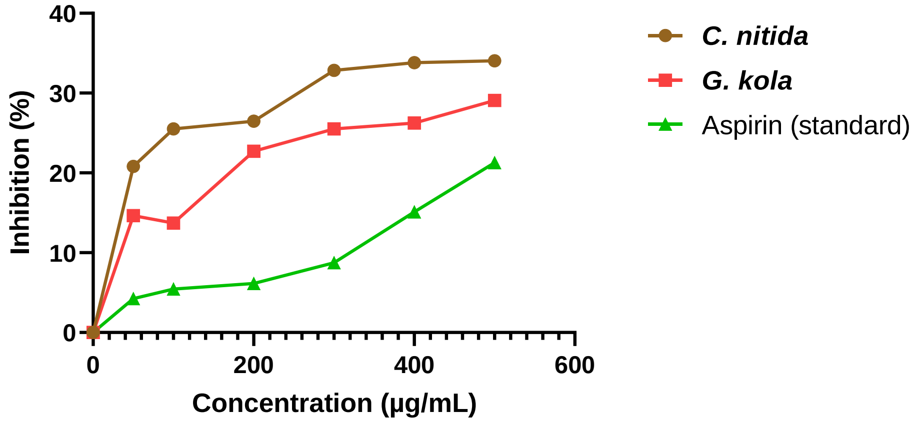


Figure 5c: The protective actions of two kola varieties against heat-induced haemolysis of RBCs with respect to a standard anti-inflammatory drug (aspirin) and the extrapolation of mean $IC_{50}$ value, which differed significantly at $p<0.05$.

**DISCUSSION**

Using herbs and other plants as medicine is common among rural residents of developing and poor countries. Traditional plant treatments have been used to treat and prevent illnesses in humans for thousands of years, suggesting that they are the ancestors of today's pharmaceuticals (Khan *et al.*, 2023; Attah *et al.*, 2022; Apiamu *et al.*, 2018). Even in regions where modern treatment is the norm, including the poor and industrialized nations, a deal of people still use traditional medicine (Attah *et al.*, 2022; Apiamu *et al.*, 2018; Himal *et al.*, 2008). To this end, the study evaluated the phytochemical, antioxidant, free radical-scavenging capability, and anti-inflammatory properties of *C. nitida* and *G. kola* Heckel nuts to establish which is more useful in traditional medicine. The saccharides, or carbs, are the most common organic compounds in living organisms. Only carbon, hydrogen, and oxygen make up these compounds, making them more chemically basic than nucleotides and amino acids. They are employed for energy production and tissue building, both of which are crucial for the survival of multicellular biosystems (Voet *et al.*, 2008). They control the nervous system and offer quick bursts of energy for work outs (Whitney and Rolfes, 2005). The results of this investigation demonstrated that, when compared to *C. nitida*, *G. kola* Heckel is the superior energy source. Proteins, which are also known as "nitrogen-containing natural products," are crucial to the efficient functioning of all organisms (Voet *et al.*, 2008), and the results of this study clearly demonstrated the superiority of *G. kola* Heckel over *C. nitida* as a protein source for the health of complex organisms. *C. nitida* was found to have more water activity than *G. kola* Heckel. According to Apiamu *et al.* (2017), it was highlighted that a high concentration of water in food samples may foster the development of harmful bacteria and moulds. Inferring from this, *G. kola* Heckel is more robust against spoilage caused by microbes while being stored than *C. nitida*. According to Okeke *et al.* (2015), the ash level of a meal may be utilized as a stand-in for the food's mineral content. This data also indicated that *C. nitida*, as opposed to *G. kola* Heckel, may play a more important role in the provision of minerals required for normal physiological and metabolic functions.

It has been established that phytochemicals, have an important role in human health by preventing disease and promoting recovery (Oguntibeju, 2018; Evuen *et al.*, 2016). Some researchers noted that previous epidemiological studies have accounted for the fact that nuts are rich in minerals and vitamins, as well as the presence of phytochemicals with proven antioxidant potentials (Evuen *et al.*, 2022; Ogunlade *et al.*, 2014). Quantification of phenolics, flavonoids, and tannins in the current study confirmed their inherent predominance in *C. nitida* and *G. kola* Heckel, both of which have demonstrated antioxidant capacity (Evuen *et al.*, 2022; Apiamu *et al.*, 2017; Saxena *et al.*, 2013), suggesting that they contribute to human health, though the latter was more preferred. These bioactive chemicals may promote health by, for example, directly interacting with and quenching free radicals (FRs), chelating transition metals, reducing peroxides, and/or increasing the antioxidative defence enzyme system (Evuen *et al.*, 2016; Ogunlade *et al.*, 2014). Thus, the results of the comparative investigation confirmed that *G. kola* Heckel is more effective than *C. nitida* as an antioxidant because of its higher levels of TPC, TFC and TTC in sequestering and inhibiting lipid peroxidation. In addition to their many other biological roles, such as stimulating bile synthesis, lowering blood cholesterol levels, and inhibiting *S. aureus* growth (Apiamu *et al.*, 2015; Saxena *et al.*, 2013; Silva *et al.*, 2007), phenolic acids were also known for a number of properties that make them useful for treating a wide range of medical conditions, including ulcers, inflammation, cytotoxicity, cancer, spas, etc. Although studies directly comparing *G. kola* Heckel and *C. nitida* are lacking, the aforementioned functional role of polyphenolic chemicals suggested that *G. kola* Heckel may be more effective in lowering blood cholesterol, acting as a superior antibacterial agent, and stimulating bile production in humans (Arsul *et al.*, 2022; Evuen *et al.*, 2016). There has been a resurgence of interest in flavonoid-rich compounds among scientists and medical professionals in light of recent publications on the wide range of pathological conditions that may be treated with these compounds due to their well-documented pharmacological actions (Apiamu *et al.*, 2015). Therefore, the best-described action of nearly all types of flavonoids was shielding the body from damaging effects of ROS: Flavonoids have been demonstrated to have antimicrobial, cytotoxic, anti-inflammatory, and anticancer properties (Saxena *et al.*, 2013). Therefore, the results of this study strongly recommended using *G. kola* Heckel in conjunction with *C. nitida*. In addition to its superiority over *C. nitida* in terms of clarifying wines and fruit juices, *G. kola* Heckel is recommended because of the tannin-rich compounds it contained, which were used to treat gastrointestinal problems like diarrhoea, stomach pain, and duodenal tumours.

The methanolic extract of *G. kola* had the lowest $IC_{50}$ value for DPPH-scavenging activity while the methanolic extract of *C. nitida* had the highest value. This observation validated the fact that the former extract had the highest concentrations of the bioactive compounds, namely phenolic, flavonoid, and tannin than the latter (Asagba and Apiamu 2019; Saeed *et al.*, 2015), and this may account for its effect. Also, several reactive oxygen species (ROS) in the body were affirmed to be neutralized by these chemicals, and thus prevented oxidative damage (Chao *et al.*, 2014). The DPPH-scavenging activity of the methanolic extract of *G. kola* was almost double that of the methanolic extract of *C. nitida*. These findings suggested that the *G. kola* methanolic extract may be more potent than the *C. nitida* counterpart in protecting cells from oxidative damage. Reducing $Fe^{3+}$ to $Fe^{2+}$ was the basis of the FRAP test, and the fundamental antioxidant mechanism of phenolics (Onocha *et al.*, 2011). Antioxidants are able to diminish free radicals because they break free radical chains via protonation, as was previously established by Arsul *et al.* (2022) and Meir *et al.* (1995). When more methanolic extracts of *C. nitida* and *G. kola* were used, the percentage of inhibition rose. *G. kola* and *C. nitida* were shown to have the highest reducing power of all the species tested. The $IC_{50}$ values determined for the extracts were compared to the reference compound, ascorbic acid, demonstrating that *G. kola* was more effective in blocking the protonation of lipid peroxidation chains, and this was ascribed to its higher reducing power potential (Odewade and Odewade, 2023; Apiamu *et al.*, 2018; Meir *et al.*, 1995).

Inflammation is a complex reaction of tissues to harmful substances, irritants, or injuries. Oyeleke *et al.* (2018) listed several conditions in which inflammation plays a role, including asthma, diabetes, cancer, arthritis, neurological disorders, and autoimmune diseases. There were significant risks connected with the use of NSAIDs or otherwise known as “nonsteroidal anti-inflammatory drugs” to treat inflammation. Novel anti-inflammatory drugs derived from natural sources have been the subject of increased study in recent years (Moreno-Quiros *et al.*, 2017) due to the prevalent perception that these compounds were safer and more tolerated than their synthetic equivalents. Extracts of *C. nitida* and *G. kola* Heckel were tested for their anti-inflammatory effects *in vitro* by assessing how well they prevented albumin denaturation, which was considered as one of the fundamental causes of inflammation. Aspirin, as well as methanolic extracts of *C. nitida* and *G. kola* Heckel, was shown to be effective in preventing albumin denaturation. All of these extracts, but especially *G. kola* Heckel, was shown to have protein-protecting properties that were significantly higher than those of aspirin, indicating their potential as novel anti-inflammatory medications. Since proteinases play a role in inflammation, the study also tested the extracts of *C. nitida* and *G. kola* Heckel for their anti-inflammatory potential by quantifying their antiproteinase activity. Proteinase activity was inhibited in methanol extracts of both *G. kola* Heckel and *C. nitida*, suggesting the latter might be effective as an anti-inflammatory medication. Methanolic extracts of *C. nitida* and *G. kola* Heckel were tested for their anti-inflammatory effects on the integrity of the red blood cell membrane due to their structural similarity to the lysosomal membrane. Lysosomes are broken down during inflammation, producing compounds that further inflame the body and contribute to a variety of disorders (Troung *et al.*, 2019). Lysosomal membrane stabilization against heat-induced haemolysis may contribute to inflammation attenuation. This prevents the release of fluids and serum proteins into the tissues that would otherwise be caused by inflammatory mediators (Oyeleke *et al.*, 2018). Based on the findings, it appeared that the methanolic extract of *C. nitida* and *G. kola* Heckel may effectively stabilize lysosomal membranes and protect red blood cell membranes from heat-induced haemolysis. The extract's bioactive components may inhibit phospholipase activation, protecting lysosomal membranes from injury. Anti-inflammatory effects via membrane stabilization were found to be most prominent in the methanolic extract of *G. kola* Heckel, followed by *C. nitida* and finally aspirin.

## CONCLUSION

The *in vitro* analyses of the two Nigerian kola nuts used in this study showed that both had a high volume of nutrients and inherent bioactive compounds with powerful energetic, antioxidant, and anti-inflammatory potentials; nevertheless, *G. kola* heckel was the clear winner. Therefore, the results of this study clearly suggested that the nutraceutical and pharmaceutical sectors should explore *G. kola* Heckel as an alternative to *C. nitida*, as a dietary supplement, functional food, and medication.

## FUTURE CONSIDERATION

Despite the promising results of this study, additional research into bioactive components, regional comparisons, pharmacological and clinical investigations, the development of functional foods and nutraceutical products, and conservation and sustainable utilization are all possible avenues to pursue. Thus, researching *C. nitida* and *G. kola* along these lines can help shed light on the inequalities that existed in relation to their purported health benefits and practical uses.

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## CONFLICT OF INTEREST

The authors confirmed that no competing interests, financial or otherwise, have influenced the design, execution, analysis or reporting of this study.

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