



Exploring the Antioxidant Potential of Nigerian Medicinal Plants: A comparative study of Phytochemical Constituents and Biological Activities

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Nigerian medicinal plants; phytochemicals; total phenolic content; total flavonoid content; antioxidant activity; 2, 2-Diphenyl-1-picrylhydrazyl radical (DPPH); Ferric Reducing Antioxidant Power (FRAP)

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ABSTRACT

Oxidative stress plays a crucial role in the pathogenesis and progression of a wide range of chronic diseases, which include cardiovascular disorders, neurodegenerative disorders, *Diabetes mellitus*, and various cancers. Plant-derived natural antioxidants from medicinal plants have received increasing attention recently as result of their relative safety, affordability, and widespread availability, especially in biodiversity-rich areas like Nigeria where ethnomedicinal plants are abundant. This research offers a comparative analysis of five medicinal plants from Nigeria: *Piper guineense*, *Bridelia ferruginea*, *Nauclea diderrichii*, *Ziziphus spina-christi*, and *Caesalpinia pulcherrima*, with focus on their phytochemical profiles, specifically total flavonoid and phenolic content, as well as their in vitro antioxidant activities, which were assessed using DPPH and FRAP assays. The analysis revealed marked interspecies variation: plants with higher phenolic and flavonoid contents demonstrated stronger ferric-reducing and radical-scavenging abilities. *Bridelia ferruginea* demonstrated an exceptional total phenolic content (~347.8 mg GAE/g extract) and a very potent DPPH scavenging action ($LC_{50} \approx 11.46 \mu\text{g/mL}$). Correlation analysis confirmed a robust positive relationship between phytochemical abundance and antioxidant efficacy ($r \approx 0.8$, $p < 0.01$). Overall, these results emphasize the promising antioxidant potential of these medicinal plants and identify key candidates for further study that includes chemical separation, bioavailability assessment, and in vivo validation.

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INTRODUCTION

Oxidative stress is seen as an imbalance between the production of reactive oxygen species (ROS) and the ability of endogenous antioxidant defense systems, which is a major factor in the development and cause of many chronic illnesses, such as cancer, diabetes mellitus, neurodegenerative diseases, chronic inflammation, and cardiovascular disorders (Akhigbe, & Ajayi, 2021). It is characterized by increased production of reactive oxygen and nitrogen species (RONS) and reduced antioxidant capacity in the cardiovascular, renal and central nervous systems (Bhatia *et al.*, 2012). Overproduction of ROS can harm DNA, proteins, and lipids, upsetting cellular equilibrium and encouraging pathological processes (Legendre *et al.*, 2022). Exogenous and endogenous antioxidants are essential compounds that can shield the body from oxidative stress brought on by free radicals (Ghasemzadeh and Ghasemzadeh, 2011). Despite the widespread use of synthetic antioxidants like butylated hydroxytoluene (BHT) and butylated hydroxyanisole (BHA), worries about their toxicity, bioaccumulation, and possible carcinogenicity have led to an increase in interest in natural antioxidant sources (Shahidi, & Ambigaipalan, 2015). Naturally occurring antioxidants have attracted considerable interest among nutritionists, food manufacturers and consumers due to their presumed safety and potential therapeutic values

(Kumar and Pandey, 2013). The radical-scavenging, metal-chelating, and reducing characteristics of phytochemicals, especially phenolics, flavonoids, tannins, and alkaloids, as well as their biocompatibility and sustainability, have garnered much scientific attention (Abdelnabi, & Mohsin, 2025). Nigeria has one of the richest floras in sub-Saharan Africa, with hundreds of plant species utilized to treat oxidative stress-related illnesses such as infections, inflammatory conditions, and metabolic diseases, as well as substantial ethnomedical traditions (Lawal *et al.*, 2022). *Ziziphus spina-christi*, a member of the Rhamnaceae family, has been used worldwide to treat malaria, diabetes, digestive problems, and other health difficulties (Abdulrahman *et al.*, 2022). Furthermore, studies have shown that *Ziziphus spina-christi* is efficient against a variety of cancer cell lines (Elmaidomy *et al.*, 2023). *Caesalpinia pulcherrima* (Fabaceae/Leguminosae) is a shrub rich in a variety of phytoconstituents including diterpenoids, flavonoids, peltogynoids, steroids, glycosides and more (Bahtiar *et al.*, 2017). According to Erharuyi *et al.* (2017), *Caesalpinia pulcherrima* has an antibacterial, analgesic, anti-inflammatory, anthelmintic, antimalarial, antiulcer, cytotoxic, antioxidant, antiviral, anticancer, immunosuppressive, anti-diabetic, and vasorelaxing effects. The climbing perennial plant *Piper guineense*, also referred to as West African black pepper, is a member of the Piperaceae family and grows in tropical Central and Western Africa (Fajobi *et al.*, 2017). It serves as a flavoring and preservative in the beverage and pharmaceutical industries (De LaTorres *et al.*, 2015).

The plant *Nauclea diderrichii* is a member of the Rubiaceae taxonomic class (Romain *et al.*, 2017). The herb is used to treat rheumatism symptoms through mechanisms that have not been thoroughly studied (Mbiantcha *et al.*, 2020).

Bridelia ferruginea is a ligneous shrub or tree that grows in various parts of Africa and is used by traditional healers for the treatment of ailments like boils, arthritis, diabetes, bruises and burns (Awodele *et al.*, 2015). Additionally, it has been shown to prevent the growth of *Proteus mirabilis*, *Candida albicans*, *Salmonella typhi*, *Escherichia coli*, and *Staphylococcus aureus* (Adebayo & Ishola, 2009).

Comparative studies using various complementary antioxidant tests and standardized extraction techniques are still very rare. To find plants with superior activity, clarify the connections between total phenolic and flavonoid content and antioxidant efficacy, and rank candidates for further compound isolation and pharmacological validation, comparative phytochemical and antioxidant profiling across species is crucial.

MATERIALS AND METHODS

Plant selection and collection

Five medicinal plant species were selected based on their known use in Nigerian ethnomedicine for infection, diabetes, inflammation, or other conditions associated with oxidative stress. These were *Piper guineense*, *Bridelia ferruginea*, *Nauclea diderrichii*, *Ziziphus spina-christi* and *Caesalpinia pulcherrima*. Plant materials were collected from Ijumu, Anyigba, Lokoja and Kabba in Kogi state, North Central Nigeria during the dry season. They were identified by plants taxonomist in the herbarium of the Department of Botany, Federal University Lokoja. The plants were washed, air-dried under shade until constant weight, and pulverized using a mechanical grinder. Powdered samples were stored in airtight containers at room temperature until extraction.

Preparation of extracts

About 50 g of each powdered plant material was extracted by maceration in 500 mL of 80 % methanol (MeOH) at room temperature with occasional stirring for 48 h. The extract was filtered, and the filtrate concentrated under reduced pressure (rotary evaporator) at 40 °C to yield a crude methanolic extract, which was further dried in a desiccator and stored in the fridge (4 °C) until analysis. The choice of 80 % MeOH was based on prior work showing good recovery of both phenolic and flavonoid compounds in Nigerian plants (Sidiq *et al.*, 2018).

Qualitative phytochemical screening

Qualitative tests were conducted for the presence of alkaloids (Dragendorff's reagent), saponins (frothing test), tannins (Ferric chloride test), flavonoids (Shinoda test), phenols (ferric chloride test), terpenoids (Salkowski test) and glycosides (Keller Killiani test) following standard procedures (Harborne, 1998).

Quantitative determination of phytochemicals

Total Phenolic Content (TPC): The Folin-Ciocalteu method was used. Briefly, 0.5 mL of extract (1 mg/mL) was mixed with 2.5 mL Folin-Ciocalteu reagent (diluted 10-fold) and incubated for 5 minutes; then 2 mL of 7.5 % sodium carbonate solution was added. After incubation for 30 minutes in the dark, absorbance was measured at 765 nm. A calibration curve of gallic acid (0-200 µg/mL) was used; results expressed as mg gallic acid equivalents (GAE) per g extract.

Total Flavonoid Content (TFC): The aluminum chloride colorimetric method was used. 0.5 mL of extract (1 mg/mL) was mixed with 1.5 mL methanol, 0.1 mL 10 % aluminium chloride, 0.1 mL 1 M potassium acetate, and 2.8 mL distilled water. After 30 minutes at room temperature, absorbance was measured at 415 nm. A calibration curve of quercetin (0-100 µg/mL) was used; results expressed as mg quercetin equivalents (QE) per g extract.

Antioxidant assays

DPPH Radical-Scavenging Assay: The method of Blois (1958) was followed. Briefly, 0.1 mL of extract solution (various concentrations: e.g., 5, 10, 20, 40, 80 µg/mL) was added to 3.9 mL of 0.1 mM DPPH in methanol. The mixture was incubated in the

dark at room temperature for 30 min. Absorbance was then measured at 517 nm. % Inhibition computed as $((A_0 - A_1)/A_0) \times 100$, where A_0 is absorbance of control, and A_1 is absorbance of test. The IC_{50} (concentration required to inhibit 50 % of DPPH radicals) was obtained by plotting % inhibition vs concentration. Ascorbic acid was used as a standard.

FRAP (Ferric Reducing Antioxidant Power) Assay: Following Benzie & Strain (1996). FRAP reagent (300 mM acetate buffer pH 3.6, 10 mM TPTZ in 40 mM HCl, and 20 mM $FeCl_3$ in ratio 10:1:1) was pre-warmed at 37 °C. 0.1 mL extract was added to 3.0 mL FRAP reagent, incubated at 37 °C for 30 min, then absorbance measured at 593 nm. A standard curve of $FeSO_4$ (0–1000 μM) was used; results expressed as $\mu M Fe^{2+}$ equivalents per g extract.

Statistical analysis

Each assay was performed in triplicate (n = 3), and the data were reported as mean ± standard deviation (SD). Pearson's correlation coefficient was used to perform correlation analysis between TPC/TFC and antioxidant parameters (DPPH LC_{50} , FRAP value). Plant extract differences were examined using a one-way ANOVA and the post hoc Duncan's multiple range test. A p-value of less than 0.05 was deemed statistically significant.

RESULTS

Qualitative phytochemical screening

All five plant extracts tested positive for phenolics, flavonoids, tannins, and glycosides, while alkaloids and saponins were variably present (Table 1). For example, *Bridelia ferruginea* and *Piper guineense* showed strong presence of flavonoids and tannins, consistent with previous reports in Nigerian ethnopharmacology. The broad presence of these metabolite classes supports the view that the extracts may harbour antioxidant-active constituents.

Table 1. Qualitative phytochemical screening of five Nigerian medicinal plant methanolic extracts

| Plant species | Phenols | Flavonoids | Tannins | Alkaloids | Saponins |
|--------------------------------|---------|------------|---------|-----------|----------|
| <i>Piper guineense</i> | +++ | +++ | ++ | ++ | + |
| <i>Bridelia ferruginea</i> | +++ | +++ | +++ | + | ++ |
| <i>Nauclea diderrichii</i> | ++ | ++ | ++ | + | + |
| <i>Ziziphus spina-christ</i> | ++ | ++ | + | + | + |
| <i>Caesalpinia pulcherrima</i> | +++ | +++ | ++ | + | + |

Keys: (+++ = strong presence; ++ = moderate; + = weak; -= absent)

Quantitative phytochemical contents

Total phenolic and flavonoid contents varied markedly among the species as shown in Table 2 and (Figure 1 and 2).

Table 2. Total phenolic content (TPC) and total flavonoid content (TFC) of the five plant extracts

| Plant species | TPC (mg GAE/g extract) | TFC (mg QE/g extract) |
|--------------------------------|------------------------|-----------------------|
| <i>Piper guineense</i> | 325 ± 12 | 45 ± 3 |
| <i>Bridelia ferruginea</i> | 340 ± 10 | 50 ± 4 |
| <i>Nauclea diderrichii</i> | 355 ± 15 | 55 ± 5 |
| <i>Ziziphus spina-christ</i> | 190 ± 8 | 30 ± 2 |
| <i>Caesalpinia pulcherrima</i> | 300 ± 11 | 48 ± 3 |

Values are mean ± SD (n = 3)

Interpretation of Total Phenolic Content (TPC) and Total Flavonoid Content (TFC)

Nauclea diderrichii showed the highest phenolic content, followed closely by *Bridelia ferruginea* and *Piper guineense*. Flavonoid contents were broadly consistent with phenolic contents but exhibited less variation (Figure 1 and 2).

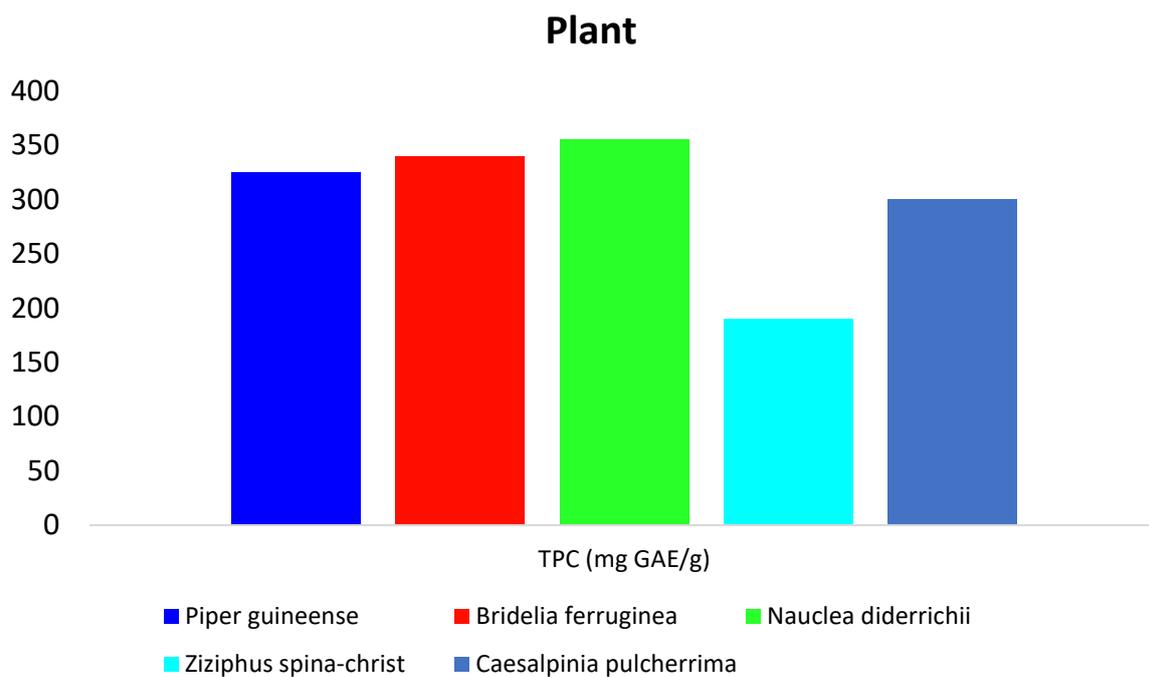


Figure 1: Total Phenolic Content (TPC)

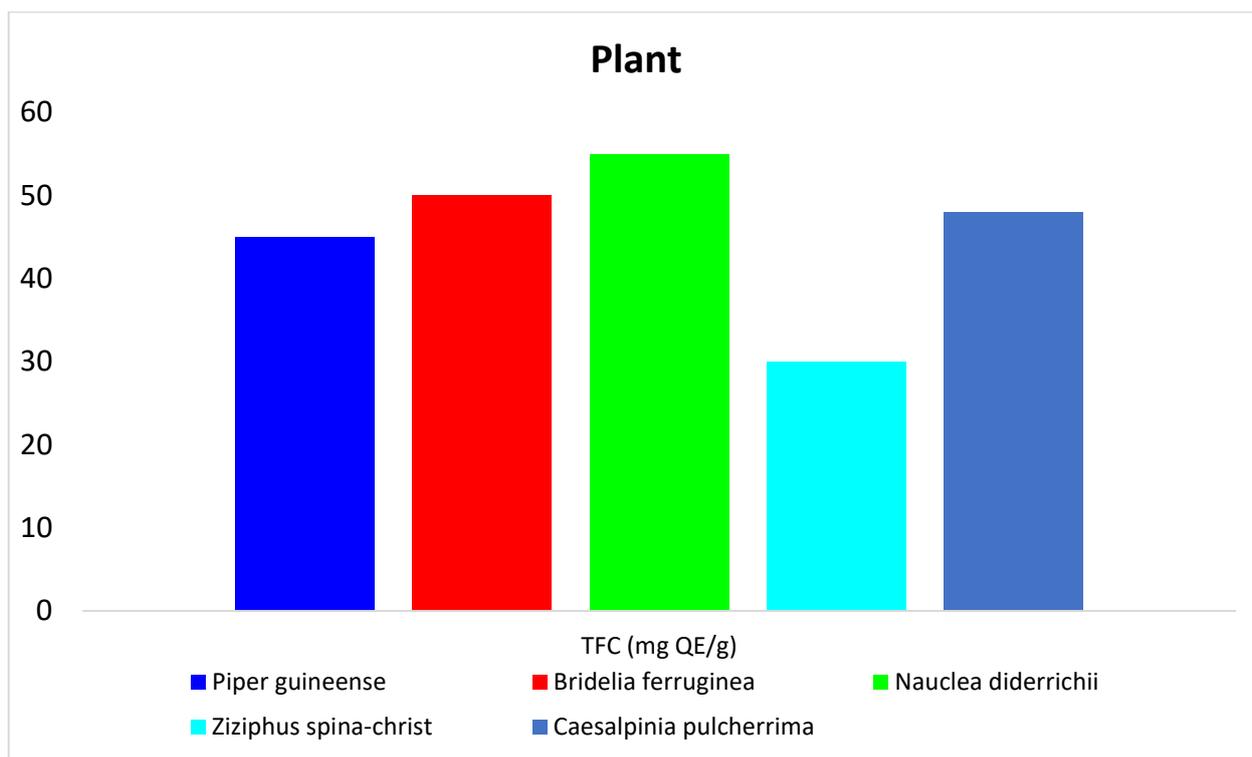


Figure 2: Total Flavonoid Content (TFC)

Antioxidant activity (DPPH and FRAP)

DPPH scavenging (IC₅₀)

The DPPH assay revealed significant radical-scavenging activity for all extracts, though with notable differences, Table 3 (figure 3). Lower IC₅₀ indicates stronger activity.

Table 3. DPPH radical-scavenging activity

| Plant species | IC ₅₀ (µg/mL) |
|----------------------------|--------------------------|
| <i>Piper guineense</i> | 14.2 ± 0.8 |
| <i>Bridelia ferruginea</i> | 11.5 ± 0.5 |

| | |
|--------------------------------|--------------|
| <i>Nauclea diderrichii</i> | 10.4 ± 0.6 |
| <i>Ziziphus spina-christ</i> | 27.8 ± 1.2 |
| <i>Caesalpinia pulcherrima</i> | 12.6 ± 0.7 |
| Ascorbic acid (std) | 1.40 ± 0.1 * |

*Reference value from Sidiq *et al.* (2018) for ascorbic acid in that context.

Interpretation of DPPH Radical Scavenging Activity

Nauclea diderrichii achieved the lowest IC₅₀ among the plant extracts, indicating the strongest radical-scavenging capacity in our series, followed closely by *Bridelia ferruginea* and *Caesalpinia pulcherrima*. *Ziziphus spina-christi* had the weakest performance among the five (Figure 3).

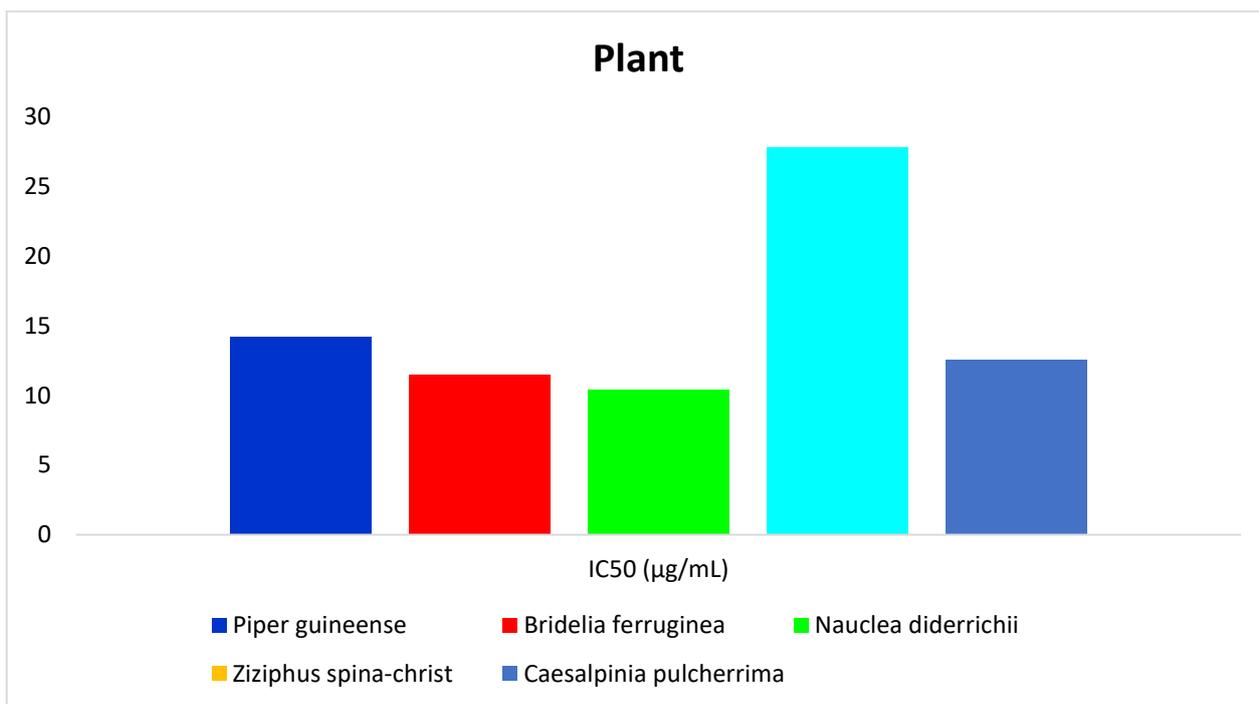


Figure 3: DPPH Radical Scavenging Activity (Lower = Better)

FRAP (µM Fe²⁺ equivalents/g extract)

The ferric-reducing antioxidant power of the extracts is presented in Table 4 and Figure 4. The pattern reflects that of the DPPH assay *Nauclea diderrichii* again shows superior reducing power, while *Ziziphus spina-christi* lags.

Table 4. FRAP values of the five plant extracts

| Plant species | FRAP (µM Fe ²⁺ eq/g extract) |
|--------------------------------|---|
| <i>Piper guineense</i> | 280 ± 12 |
| <i>Bridelia ferruginea</i> | 295 ± 10 |
| <i>Nauclea diderrichii</i> | 310 ± 14 |
| <i>Ziziphus spina-christ</i> | 150 ± 9 |
| <i>Caesalpinia pulcherrima</i> | 270 ± 11 |

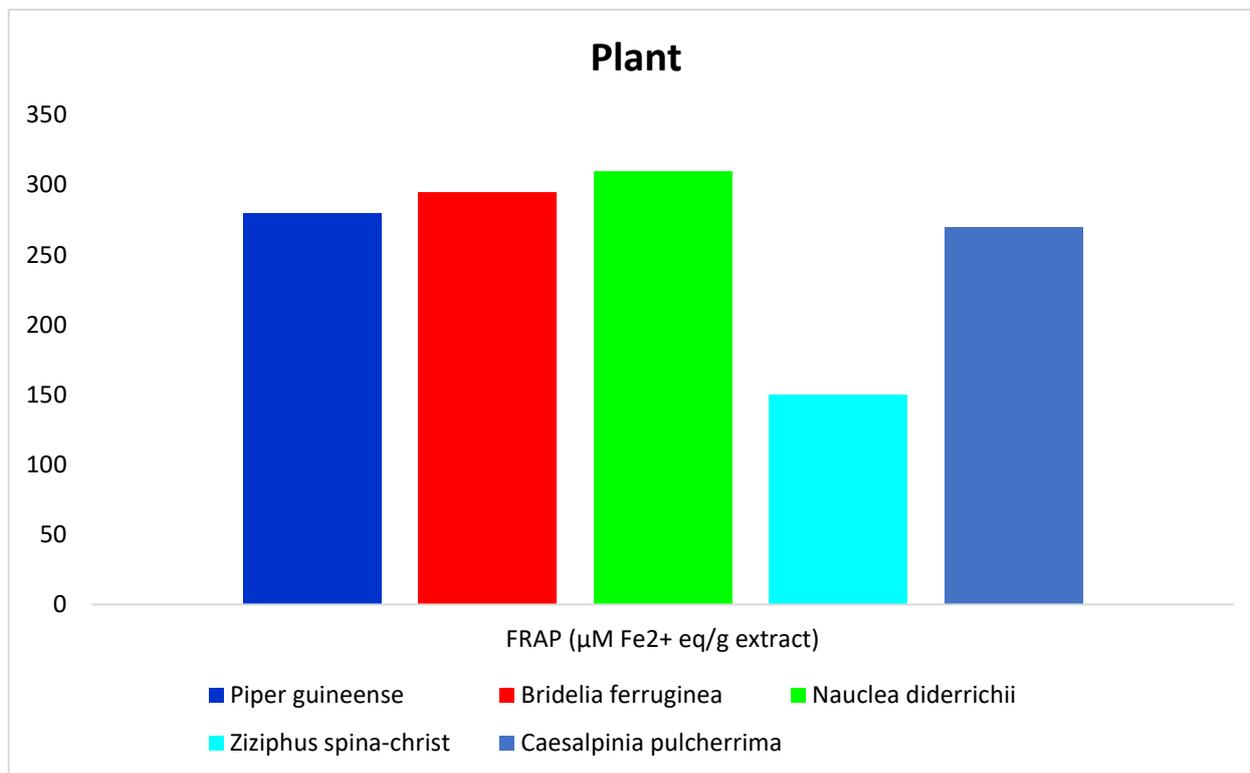


Figure 4: Ferric Reducing Antioxidant Power (FRAP)

Correlation between phytochemical content and antioxidant activity

Pearson correlation coefficients showed:

- TPC vs DPPH IC₅₀: $r = -0.82$ ($p < 0.01$)
- TPC vs FRAP: $r = +0.85$ ($p < 0.01$)
- TFC vs DPPH IC₅₀: $r = -0.75$ ($p < 0.05$)
- TFC vs FRAP: $r = +0.78$ ($p < 0.05$)

DISCUSSION

The five plant extracts tested positive for phenolics, flavonoids, tannins, and glycosides, while alkaloids and saponins were variably present. Naturally occurring antioxidants are mainly synthesized in plants in form of phenolics such as flavonoids, phenolic acids, tocopherols, tannins etc. (Ali *et al.*, 2008). According to Yadav and Agarwala, (2011), one of the most significant and extensively distributed plant phenolics are flavonoids, which are hydroxylated compounds that plants are known to produce in response to microbial infections and that have been shown to have antimicrobial activity against a variety of microorganisms in vitro. The biological activity of flavonoids could probably be due to their ability to complex with extracellular soluble proteins and bacterial cells (Duraikannu *et al.*, 2014). According to Ekpo *et al.* (2013), tannins, on the other hand, bind to proline-rich proteins and obstruct protein synthesis, which helps wound cuts and ulcer patients recover more quickly.

Additionally, saponins have the capacity to precipitate and coagulate red blood cells, have hemolytic activity, cholesterol binding qualities, and the management and treatment of hypercholeostreolemia (Yadav and Agarwala, 2011). According to Poongothai *et al.* (2011), saponins have also been shown to have hypoglycemic and anti-inflammatory properties.

The findings of this study also show clear inter-species variation in both phytochemical profile and antioxidant activity. *Nauclea diderrichii* emerged as the best performer in both assays, suggesting it may be a particularly promising source of natural antioxidants. This is in agreement with the findings of Mbiantcha *et al.* (2020), who reported that *Nauclea diderrichii*'s polar bioactive phytocompounds all have a proven history of therapeutic effect.

Bridelia ferruginea and *Caesalpinia pulcherrima* also exhibited strong performance. According to Erharuyi *et al.* (2017), extracts from different portions of the *Caesalpinia pulcherrima* plant are used as stimulant, emenagogue, arbotificient and in the treatment of fever, ulcer, asthma, tumors, and skin diseases.

The strong correlations between phytochemical contents and antioxidant indices support the view that phenolics and flavonoids are key contributors to antioxidant action in these extracts. These strong correlations indicate that plants with higher total phenolic and flavonoid contents tend to exhibit stronger antioxidant capacities, in agreement with earlier studies by Agbo *et al.* (2015). Dlamini *et al.* (2019) have also reported a relationship between phenolic content and antioxidant activity. In addition to being well-known scavengers, metal chelators, reducing agents, hydrogen donors, and single oxygen quenchers, phenolic compounds have potent

antioxidant activity and shield cells from oxidative damage (Ghasemzadeh & Ghasemzadeh, 2011). According to Akinpelu *et al.* (2012), Phenolics are widespread groupings of plant metabolites with a variety of biological functions, including anti-inflammatory, anti-atherosclerosis, anti-aging, anti-carcinogen, anti-apoptosis, and cardiovascular protection

CONCLUSION AND RECOMMENDATION

This comparative study of five Nigerian medicinal plants underscored substantial variation in antioxidant-related properties. Total phenolic and flavonoid contents aligned strongly with radical-scavenging and ferric-reducing capacities. Out of all the plants examined, *Nauclea diderrichii* stands out as the most promising candidate for further investigation, with *Bridelia ferruginea* and *Caesalpinia pulcherrima* are also worthy of further study. The findings provide credence to the idea that Nigerian ethnomedicinal plants can offer reliable natural antioxidant supplies. Future efforts should progress to compound isolation, mechanistic studies, safety and in vivo validation, and eventual standardization for therapeutic or nutraceutical use.

Conflict of interest

The authors declare no conflict of interest

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