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# Qualitative and Quantitative Phytochemical Screening and Antioxidant Capacity of *Zingiber officinale, Ocimum gratissimum* and their Mixture

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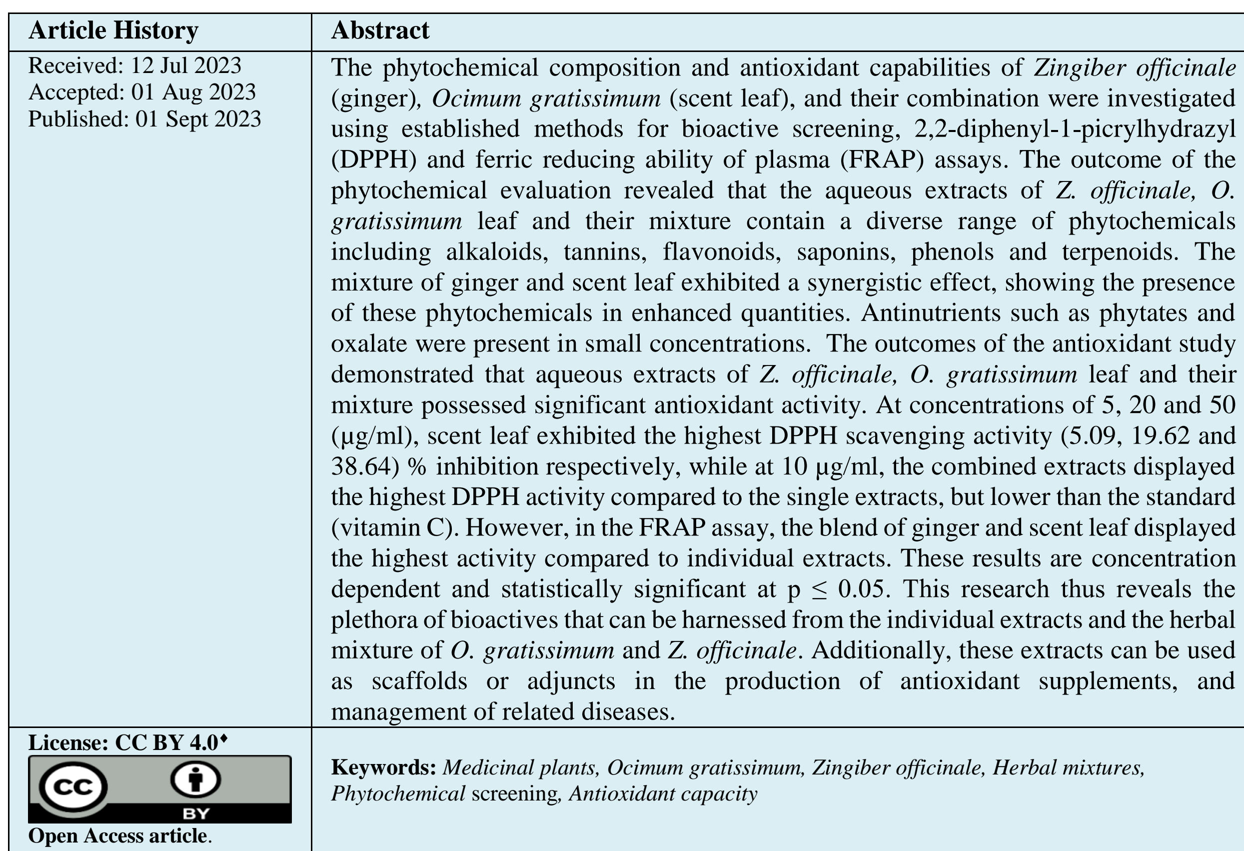

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**Abstract**
The phytochemical composition and antioxidant capabilities of *Zingiber officinale* (ginger), *Ocimum gratissimum* (scent leaf), and their combination were investigated using established methods for bioactive screening, 2,2-diphenyl-1-picrylhydrazyl (DPPH) and ferric reducing ability of plasma (FRAP) assays. The outcome of the phytochemical evaluation revealed that the aqueous extracts of *Z. officinale, O. gratissimum* leaf and their mixture contain a diverse range of phytochemicals including alkaloids, tannins, flavonoids, saponins, phenols and terpenoids. The mixture of ginger and scent leaf exhibited a synergistic effect, showing the presence of these phytochemicals in enhanced quantities. Antinutrients such as phytates and oxalate were present in small concentrations. The outcomes of the antioxidant study demonstrated that aqueous extracts of *Z. officinale, O. gratissimum* leaf and their mixture possessed significant antioxidant activity. At concentrations of 5, 20 and 50 (µg/ml), scent leaf exhibited the highest DPPH scavenging activity (5.09, 19.62 and 38.64) % inhibition respectively, while at 10 µg/ml, the combined extracts displayed the highest DPPH activity compared to the single extracts, but lower than the standard (vitamin C). However, in the FRAP assay, the blend of ginger and scent leaf displayed the highest activity compared to individual extracts. These results are concentration dependent and statistically significant at $p \leq 0.05$. This research thus reveals the plethora of bioactives that can be harnessed from the individual extracts and the herbal mixture of *O. gratissimum* and *Z. officinale*. Additionally, these extracts can be used as scaffolds or adjuncts in the production of antioxidant supplements, and management of related diseases.

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**Keywords:** *Medicinal plants, Ocimum gratissimum, Zingiber officinale, Herbal mixtures, Phytochemical* screening, *Antioxidant capacity*

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## Introduction

Oxidants are compounds that can receive electrons from other substances in a chemical reaction. They are commonly known as oxidizing agents, and also perform an important part in numerous chemical reactions and biological activities (Jamshidi-Kia *et al*., 2020). Chemical substances having unpaired electrons behave as oxidants in the body (Jamshidi-Kia *et al*., 2020), and are often exceedingly reactive, and capable of oxidatively deteriorating biological components such as proteins, lipids, and DNA, which ultimately results in mutation(Juan *et al*., 2021). Oxidants are typically characterized by their high electronegativity (Das *et al*., 2013), which means they have a strong tendency to attract electrons. This property allows them to oxidize other substances by removing electrons from them. Examples of common oxidants include oxygen, chlorine, hydrogen peroxide, and ozone.

In biological systems, oxidants are very crucial in cellular respiration (Buonocore *et al*., 2010), which is the process by which cells produce energy. They are also involved in the immune response (Hawkins, 2009), as white blood cells use oxidants to destroy invading pathogens. However, when the equilibrium between antioxidants and oxidants is disturbed, it can lead to oxidative stress. When there is an imbalance between the body's ability to undo the damage that reactive oxygen species (ROS) cause and the pace at which they are produced by the body, the result is a state known as oxidative stress (Preiser, 2012; Sies *et al*., 2017). ROS, often referred to as free radicals, are very reactive compounds that are connected to a variety of diseases, including cardiovascular

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diseases, cancer, and neurological problems. They may damage proteins, lipids, and DNA as well as induce cellular malfunction (Essick & Sam, 2010; Bhatti *et al*., 2017).

To combat free radicals and restore the body's equilibrium, antioxidants, which are chemicals which are able to neutralize reactive species (by giving an electron to stabilize them), can be utilized (Kunwar & Priyadarsini, 2011). This can prevent free radicals from damaging cells and causing oxidative stress (Rao *et al*., 2011). Antioxidants come in a variety of forms like flavonoids, selenium, beta-carotene, vitamins C and E, and others which are present in fruits, vegetables, nuts, and whole grains. Eating a diet rich in these foods can help boost antioxidant levels in the body.

### Medicinal plants as good sources of antioxidants

They include a wide range of active chemicals that can be utilized to cure or prevent illness (Singh, 2015). Since humans first began using medicinal plants thousands of years ago, they have played an important part in traditional medicine all across the world. They are a source of natural medicine and are often used as an alternative or complementary therapy to conventional medicine (Jamshidi-Kia *et al*., 2017), treating various diseases from minor conditions like headaches and stomach upset to more serious diseases like cancer and diabetes. They are also important for their cultural significance as many indigenous communities rely on them as their primary form of healthcare, and an integral part of their cultural identity (Kufer *et al*., 2005; Gold & Clapp, 2011). Researchers in their bid to discover substances utilized for medical reasons, have targeted numerous floras traditionally that are therapeutically beneficial.

The medicinal properties of plants depend on both nutrient and non-nutrient constituents. In West Africa, *Ocimum gratissimum* (OG) and *Zingiber officinale* (ZO) are well-known medicinal plants with various biological activities and therapeutic efficacy, as reported in ethnobotanical studies. These medicinal plants can be used as a" stand alone" medication or complexed with other medicinal plants as an herbal mixture. For example, OG has been investigated by some researchers as a single herb (Gupta *et al*., 2011; Olamilosoye *et al*., 2018) or as polyherbal mixtures (Ojewumi, 2021; Guleria, 2022; Oghenetekevwe & Orororo, 2022).

The herb (OG) is commonly used in conventional medical practice to address a range of conditions, including respiratory infections, skin ailments, headaches, conjunctivitis, diarrhea, pneumonia, fever, and cough. Its flowers and leaves contain valuable oils, which are utilized in making teas and infusions (Prabhu *et al*., 2009). Coastal areas of Nigeria utilize it to control high fever, epilepsy, and diarrhoea (Imosemi, 2020), whereas savannah zones use extracts from the leaves to address mental health issues (Imosemi, 2020). In Southeastern Nigeria, the Ibos use OG to manage the baby's cord and fungal infections (Prabhu *et al*., 2009). The herb has been used to relieve headaches, sunstroke, influenza, inflammation, and as an antipyretic and diaphoretic in India. The roots of OG are used as a sedative for children in Brazilian tropical forests (Agarwal & Varma, 2014), while the leaves are rubbed and sniffed to cure congested nostrils in coughs, sore eyes, abdominal pains, ear infections, and fever in Kenya and sub-Saharan Africa,. Additionally, the plant is employed as a tooth gargle, for regulating menstruation, and as a remedy for rectal prolapse (Naluwuge, 2013). The OG leaves' infusion is utilized as a pulmonary antiseptic, antitussive, and antispasmodic agent (Bhavani *et al*., 2019). OG also exhibits ovicidal activity (Pessoa *et al*., 2002) , leishmanicidal activity (Ueda-Nakamura *et al*., 2006), antidiarrhoeal effect (Owulade *et al*., 2004) , cytotoxic activity (Mahapatra *et al*., 2009) , cardiovascular effect (Lahlou *et al*., 2004), neuroprotective effect (Bora *et al*., 2011), antidiabetic effect (Okoduwa *et al*., 2017; Awwad *et al*., 2021) , nephroprotective (Ogundipe *et al*., 2017; Akara *et al*., 2021), hepatoprotective effect (Farombi, 2014; Chigozie *et al*., 2016; Huang *et al*., 2020). Treatment of hair loss, suspending activity, central nervous system activity, anticonvulsant activity, nematocidal activity, disintegrating activity and acne management (Prabhu *et al*., 2009).

*Zingiber officinale* (ZO), also known as ginger, is a tropical medicinal plant that is highly sought after for both its culinary and medical uses across the globe. According to (Ashraf *et al*., 2017 the plant is sterile by nature and only reproduces through rhizomes. The tropics of southern and southern-eastern Asia are home to many family members (Mintah *et al*., 2019). It is regarded to be the first vegetative farmed plant among them (Mans *et al*., 2019). It has a wide range of phytochemicals, which are physiologically active, non-nutritive substances. It has long been used in Chinese, Tibb-Unani herbal treatments, and Ayurvedic to treat rheumatism, catarrh, neurological illnesses, gingivitis, and discomfort in the teeth, stroke, asthma, diabetes, and constipation (Mintah *et al*., 2019). Ginger is now grown as a commercial crop in Africa, Latin America and South-east Asia (da Silveira Vasconcelos *et al*., 2019).

The nutritional contents and growth of a plant can be influenced by soil type, formulation, and susceptibility to harmful environmental issues (Khan *et al*., 2015). Sorrenti *et al*. (2016) verified, and Paetsch *et al*. (2018) supported, that environmental factors can alter soil physicochemical characteristics and impact plant nutrients and phytochemical components. Differences in the report of concentrations of chemical constituents of plants may be greatly dependent on the plant cultivar and region (An *et al*., 2016). The goals of this study wasto conduct phytochemical evaluations and assess the antioxidant activity of extracts from these healing plants grown in southern Nigeria.

## Materials and Methods

### Collection and processing of plant materials

*Zingiber officinale* roots and *Ocimum gratissimum* leaves were harvested in October 2022 from Okuzu Mbana village Oba Anambra state in the South-eastern part of Nigeria. The species were identified, categorized, and given a reference number at the University of Port Harcourt's Plant Science and Biotechnology Department: UPH/PSB/2023/014 and herbarium numbers: UPH/P/373, UPH/P/374 for later referring purposes. The plant parts were exposed to no direct sunlight for 15 days while being air dried and kept at ambient temperature (25± 2 °C).

### Extraction technique

The aqueous concentrates of the root and leaves were obtained following the methods of Onakurhefe *et al.* (2020); El-Borm *et al.,* 2018) and Badawy *et al.* (2019). Water was used as a solvent in the extraction. The plant samples were air dried until constant weights were obtained and coarsely ground.

They were put in a container with a lid and solvent and allowed to stand at room temperature for 72 hours with intermittent stirring to ensure that all of the soluble materials were dissolved. After straining the mixture, the Marc (solid, damp substance) was compressed. And then, Whatman's filter was used to clarify the mixed liquid. The filtrate from the crude plant aqueous extraction was mounted on the water bath at 100°C to evaporate the liquid part of the extract. This temperature was set based on the solvent boiling point to avoid denaturing the extracts. This filtrate obtained was then frozen at -40°C.

**Reagents Utilized**

Standard-grade chemicals from Merck and Sigma-Aldrich (Johannesburg, South Africa) were utilized in this work. The following reagents were used in the assays while taking chemical safety precautions: diethyl ether, octanol, sodium sulfite ($Na_2SO_3$), acetone, NaOH, sulfuric acid ($H_2SO_4$), boric acid, $KMnO_4$, HCl, $FeCl_3$ solution, 2,6-dichlorophenolindophenol, oxalic acid, - 0-dipyridyl reagent, ethanol and α-tocopherol.

**Phytochemical analysis**

Screening for Qualitative Phytochemicals

Aqueous extracts of ginger roots, scent leaf, and their mixture were used to assess the content of phytoconstituents such as flavonoids, terpenoids, alkaloids, tannins, saponins, steroids, and phenols. The analysis was conducted in accordance with the conventional qualitative methodologies given by Evans, (2002) and Adegoke *et al.* (2010).

**Flavonoids detection (Alkaline reagent test)**

To each 0.2 g extract, 6 drops of a 2% NaOH solution were added. When vivid yellow colouring first developed, flavonoids were present in the extracts; however, when diluted acid was added, the solution lost its intense yellow color. (Adegoke *et al.*, 2010).

**Alkaloids detection (Mayer's test)**

Each extract was diluted in 5ml of 1% diluted HCl solution before being filtered. Each filtrate was given a different treatment with Mayer's reagent (potassium mercuric iodide). The formation of a yellow precipitate revealed the presence of alkaloids in the extracts (Evans, 2002; Adegoke *et al.*, 2010).

**Terpene detection (Salkowski's test)**

A 0.5 ml of chloroform and 1 ml of concentrated sulphuric acid were added to 0.1 g of each extract first. The emergence of a reddish-brown precipitate in the extracts indicated the presence of terpenoids (Evans, 2002).

**Tannin identification (Ferric chloride test)**

Three drops of diluted ferric chloride were added to each test tube after 0.2 g of the single and combined extracts were mixed with an equal amount of distilled water. The presence of tannins in the extracts was indicated when a brownish blue or dark colour formed (Adegoke *et al.*, 2010).

**Steroid detection (Liebermann-Burchard test)**

0.5 g of each extract was dissolved in 2 ml of chloroform, and the test tubes were then filled with 2 ml of concentrated tetra oxo sulphate (vi) acid. Red coloration in the bottom chloroform layer signaled the presence of steroids in the extracts, which led to a successful analysis (Adegoke *et al.*, 2010).

**Saponin detection (Foam test)**

Distilled water (6ml) and 0.2 g of each extract were added, and the mixture was vigorously agitated in a graduated cylinder for 15 minutes. By creating bubbles or foam that lasted for 10 minutes, the extracts' saponin content was verified (Evans, 2002).

**Phenol detection (Ferric chloride test)**

A quantity (0.2g) of the extracts were combined with 2 ml of a 5% aqueous ferric chloride solution. The extracts' phenol concentration was high, as evidenced by the emergence of a bluish tint (Adegoke *et al.*, 2010)

**Determination of Resins (Color Test)**

Each sample was extracted with chloroform for 0.12 g, and the concentrated extracts were then dried. Resolving the residue required 3ml of concentrated HCl and acetone, which were then heated in a water bath for 30 minutes. Resins are present because resins cause the colour pink to change to magnet red.

**Quantitative Phytochemical Analysis**

**Tannin content determination**

The method outlined by Kajaria *et al.* (2011) was used to compute the total phenolic compounds. The findings were compared to the standard curve, which encompassed 0.0 to 600 g of tannic acid per milliliter after the composite sample had been diluted with distilled water to a known concentration. In a test tube, 250g of the diluted composite sample or tannic acid solution was added to 1 ml of distilled water. After that, 250 g of Folin-Ciocalteu reagent was added and thoroughly mixed into the tube. The combination was then allowed to sit at room temperature for 5mins so that the Folin-Ciocalteu reagent could thoroughly react with it. After that, 2.5 ml of a 7% sodium carbonate aqueous solution was added. Afterward, distilled water was added to make the whole volume which is 6.0 ml. The solution's absorbance at 760 nm was assessed using a spectrophotometer after 90 minutes of incubation. The composite sample's tannins were measured using the technique described by Makkar and Becker (1993). A tannin-containing extract and distilled water were added after the first weighting of 100 mg of polyvinyl poly-pyrrolidone (PVPP). The resultant mixture was agitated for 60 minutes before centrifuging it for 10 minutes and collecting the supernatant. Other simple phenolic compounds, besides tannins, were present in the supernatant since tannins would have precipitated with the PVPP.

The total phenol content was subtracted from the total tannin content to determine the overall tannin content. Tannic acid equivalent measurements were used to determine the amount of tannins, and a dry sample basis (y%) was used to indicate the fraction of non-tannin phenols.

**Flavonoid content determination**

The method developed by Kajaria *et al.* (2011) was used to determine the flavonoid content. A 10g composite sample was repeatedly extracted at room temperature to yield 100 ml of 80% aqueous methanol. The entire solution was then filtered using Whatman filter paper no. 42 (125 mm). The filtrate was then transferred to a crucible and dried in a water bath. The

dried residue was weighed, and the following formula was used to determine the sample's flavonoid content:
% Flavonoid = weight of saponin multiplied by 100/weight of sample

**Total saponin content determination**

To assess the total saponin concentration, a modified version of the vanillin-sulphuric acid colorimetric method reported by Adusei *et al*. (2019) was used. 100mL of diosgenin and precisely 5000mL of water were used. The combination was subsequently treated with 8 g of vanillin dissolved in 100 ml of 99.5% ethanol using a 500 L vanillin reagent solution. 5 mL of 72% sulphuric acid was also added and properly mixed. The final mixture was heated in a water bath for 10 minutes to 60$^{O}$C. The solution's absorbance at 544 nm was measured and recorded after it had been incubated for 10 minutes. Each of the three aqueous extracts that were the subject of the investigation underwent this procedure. Every measurement was made in three copies for every analysis.

**Total alkaloid content determination**

The total alkaloids content (TAC) of crude extracts was determined using the method outlined by Adegoke *et al*. (2010). A 100 ml beaker was filled with 1g each of crude extract made from OG, ZO and OG-ZO blend respectively. The beaker was then filled with 50 mL of a 10% solution of hydrochloric acid in ethanol and sealed for four hours. The mixture was then filtered, and the filtrate was concentrated to a quarter of its original volume using a rotary evaporator and 78°C. Then, 15 drops of concentrated ammonium hydroxide were added to the concentrate drop-wise until precipitation was complete. 20 ml solution of 0.1 M $NH_4OH$ was used to wash the precipitates after the mixture had been allowed to sediment for three hours. The resulting combination was then filtered, and the alkaloid content was indicated by the residue that was left behind. After being dried and weighed, the residue's alkaloid content was determined by dividing its weight by the weight of the sample it represented and multiplying the result by 100.

**Total phenols determination**

Total phenolic content was ascertained using a spectrophotometric technique. First, 100 ml of diethyl ether was used to defatten two grammes (g) of pulverised material in a Soxhlet device for two hours. After using the defatting method, the sample was heated for 15 minutes with 50 ml of diethyl ether to help in the extraction of phenolic chemicals. The resultant mixture was transferred along with 10 ml of distilled water after being separated into 5 ml sections in a 50 ml Erlenmeyer flask. Then, 5 ml of concentrated amyl alcohol and 2 ml of $NH_4OH$ solution were added to each test tube. Each test tube received 50 mL of the liquid, which was then let to stand for 30 minutes to develop colour. The resulting colour was then determined by spectrophotometry at a wavelength of 505 nm.

$$\text{Dilution Factor (DF)} = \frac{\text{Volume of flask (50ml)}}{\text{Volume of extract}}$$

$$\%\ \text{Phenol} = \frac{100 \text{ x AS x CS x DF x VF}}{\text{Wt. x AP x 1000 x VE}}$$

Where, AS = Absorbance of Sample Solution, AP = Absorbance of Standard Phenol Solution, CS = Standard Phenol Solution Concentration VF = Total extract volume, Wt = Sample weight, VE = Extract volume measured, DF = Dilution factor employed.

**Anti-Nutrient Quantification**

**Phytic acid determination**

Aina *et al*. (2012) approach was used to calculate the phytic acid content. In Erlenmeyer flask (250 mL) containing 2.0 g of crushed plant material and 100 mL of 2% HCl was macerated for 3 hours before the resulting liquid was filtered. With the use of a pipette, 25 mL of the filtrate was transferred into a conical flask that already had 5 mL of a 0.3% ammonium thiocyanate solution. A consistent brownish yellow tint appeared and lingered for a few minutes after being titrated against a typical $FeCl_3$ solution (0.001 95 g Fe/mL). The sample's concentration of phytic acid was calculated as follows: % Phytic acid = titre value × 0.00195 × 1.19 × 100

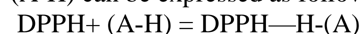
**Determination of oxalate content**

The method of Unuofin *et al*. (2017) was adopted for determining the oxalate content of materials. One gram (1g) of each sample was weighed twice and placed in an Erlenmeyer flask containing 75 mL of 3 M sulfuric acid. The mixture was agitated with a magnetic stirrer for one hour prior to filtration. After 5 mL of the filtrate was extracted and titrated against a 0.05 M KMnO4 solution, a reddish-brown colour could be seen. According to calculations, the sample had an oxalate concentration of 2.2 mg per 1 mL of a 0.05 M KMnO4 solution.

**Antioxidant capacity**

**DPPH assay for antioxidant activity**

DPPH (2,2-diphenyl-1-picrylhydrazyl) is a synthetic chemical that is commonly employed in laboratory studies to assess the antioxidant activity of an item, such as food or biological sample. When DPPH is exposed to a substance with antioxidant activity, such as vitamin C or a plant extract, it becomes reduced, resulting in a color change from purple to yellow. In this assay, a sample is mixed with a solution of DPPH and the rate of change in the solution's absorbance at a specific wavelength is gauged. The extent of the colour shift is related to the sample's antioxidant activity and capacity to emit hydrogen. The DPPH scavenging reaction with an antioxidant (A-H) can be expressed as follows.


DPPH+ (A-H) = DPPH—H-(A)

The ability of the extract to neutralize DPPH radicals was evaluated with a few slight modifications (Kedare & Singh, 2011). One milliliter (1.0 ml) of 0.3 mM DPPH methanol solution was added to 2.5 ml of the extract or standard solution (250 ug/ml), and the mixture was then allowed to react for 30 minutes at room temperature. Using a spectrophotometer, the mixture's absorbance at 518 nm was measured to calculate the percentage antioxidant activity (AA%) of the final mixture. Methanol (1.0 ml plus 2.5ml of the extract solution) was used as a control. 1.0ml of 0.3mM DPPH in methanol (2.5ml) was used as a negative control. A positive control was an ascorbic acid solution.

**Determination of reducing potential (FRAP)**

The reducing potential was estimated using Afolabi and Oloyede (2014) approach, with minor modifications. Acetate buffer (pH 3.6), (2,4,6-Tripyridyl-S-triazine (TPTZ) solution, and ferric chloride (FeCl3) solution were combined to make

the FRAP reagent. TPTZ was dissolved in 40 mM HCl to make the TPTZ solution, while FeCl3 was dissolved in water to make the FeCl3 solution. The plant extract was then diluted with the sample buffer to an appropriate concentration where the absorbance of the reaction mixture falls within the linear range of the spectrophotometer used for measurement. In a 1:9 ratio, the FRAP reagent was combined with the sample, and the reaction mixture was incubated at 37°C for 10 minutes. A spectrophotometer was used to measure the absorbance of the reaction mixture at 593 nm. The level of antioxidant lowering power in the sample is correlated with the intensity of the blue colour produced. Greater reaction mixture absorbance denotes a higher reductive potential. The amount of a standard antioxidant called Trolox that was known to be present in the sample was used to create a calibration curve that measured its antioxidant capacity. The results were represented as µl/g or µl/ml of Trolox equivalents of the sample.

**Statistical Analysis**

The outcomes of each test were performed in triplicate and shown as mean standard deviation. XLSTAT 2016 was used to statistically analyze the gathered data. Using a one-way ANOVA, mean values between extracts were compared. p-values less than 0.05 ($p \leq 0.05$) were regarded as statistically significant using Tukey's Multiple Comparison Test.

## Results and Discussion

The results of determined phytochemicals in the roots of *Zingiber officinale* GN, *leaves of Ocimum gratissimum* SL, and their mixture GNSL are presented in Table 1 and Figures 1 and 2. The result indicates that flavonoids, tannins, saponins, alkaloids, phenols, and steroids are present in *Zingiber officinale leaves of Ocimum gratissimum*, and their mixture GNSL aqueous extracts.According to Damodar *et al*. (2011), phytochemicals are non-nutritive plant compounds with protective and disease-prevention properties. Medicinal plants are highly valued for their economic benefits, particularly in enhancing health (Tungmunnithum *et al*., 2018), and they are frequently employed in the manufacture of drugs. Metabolite profiling studies have shown that specific plants contain highly therapeutic phytochemicals, which are impacted by the nutritional and essential oil composition of the plants (Efferth & Koch, 2011). Long-used plants like *Zingiber officinale* and *Ocimum gratissimum* have been proven to have anti-cancer, anti-inflammatory, antioxidant, and antibacterial effects. Primarily due to their bioactive components such as polyphenols and alkaloids (Ugbogu *et al*., 2021). This study, focused on investigating and comparing their phytochemical content and their antioxidant capacities.

**Table 1**: Qualitative Phytochemical Screening of GN, SL, and GNSL

| S/N | Phytochemicals | Sample ID | | |
|---|---|---|---|---|
| | | GN | SL | GNSL |
| 1 | Flavonoids | ++ | +++ | ++ |
| 2 | Saponins | ++ | ++ | ++ |
| 3 | Tannins | ++ | ++ | ++ |
| 4 | Phenols | + | + | ++ |
| 5 | Alkaloids | + | + | + |
| 6 | Steroids | + | + | ++ |
| 7 | Terpenoids | + | _ | + |
| 8 | Cyanogenic glycosides | _ | _ | _ |
| 9 | Resins | + | + | + |

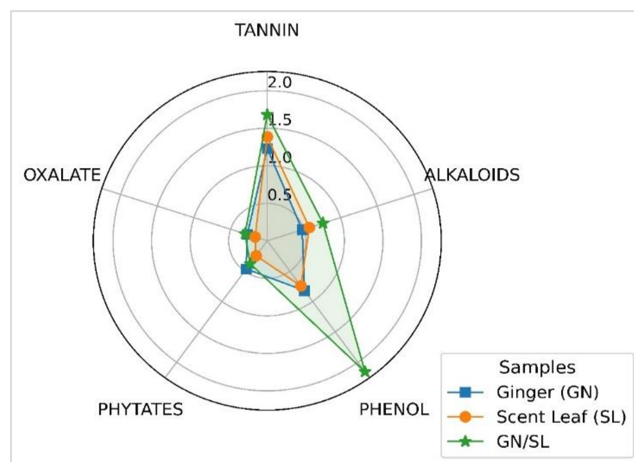



**Figure 1:** Radar plot showing the phytochemicals composition of the plant extract

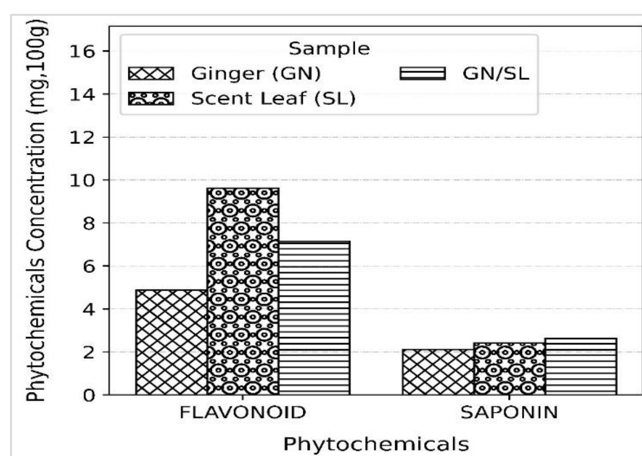



**Figure 2:** Bar plot showing the phytochemicals composition of the plant extract

Phytochemicals are phytonutrients found in vegetables and fruits, as well as whole grains, spices, herbs, and seeds, and many more with potent health benefits (Liu, 2004; Leitzmann, 2016). They are non-crucial nutrients that the human body does not require to sustain life, but studies have shown that they can protect humans against diseases (Biesalski *et al*., 2009; Granado-Lorencio & Hernández-Alvarez, 2016). Biologically active chemicals found in plants include flavonoids, saponins, terpenoids, anthraquinone, tannins,, sugars, glycosides, steroids, and alkaloids. These bioactive compounds have anti-inflammatory, anti-diabetic, antibacterial, anti-atherosclerotic, and anti-carcinogenic characteristics, which contribute to their therapeutic significance. Tannins, steroids, terpenoids, alkaloids, phenols, saponins, and cardiac glycosides have been found in preparations of ZO root and OG leaf. Certain chemicals are known to boost certain medicinal plants' antioxidant activity. Extensive research has been undertaken to demonstrate these chemicals' significant pharmacological and therapeutic benefits (Ashraf *et al*., 2017; Pandey, 2017; Ansari *et al*., 2021).

According to Lobo *et al*. (2010), antioxidants function as reducing agents by eliminating free radical intermediates and inhibiting further oxidation. Antioxidant potentials have been studied using the DPPH scavenging activity test (Igbinosa *et*

*al*., 2013). All extracts showed concentration-dependent percentage inhibitions of DPPH scavenging activity (Fig. 3). The herbal combination (GNSL) and ginger (GN) extracts were the next most effective at scavenging DPPH, followed by the scent leaf (SL) extract. At a concentration of 10µg/ml, the GNSL extract had the best DPPH scavenging activity, however at higher concentrations, the SL extract had the highest DPPH scavenging potential, followed by the GNSL and GN extracts. The three extracts exhibit potent DPPH scavenging effects at the highest concentration (50.0 µg/mL), as demonstrated by the fact that all of the aqueous extracts' concentration dependent curves exhibited the same pattern as the reference drug Vitamin C. The results of this study are in line with those of (Igbinosa *et al*., 2013), who attributed the high flavonoid content of SL to its strong antioxidant activity.

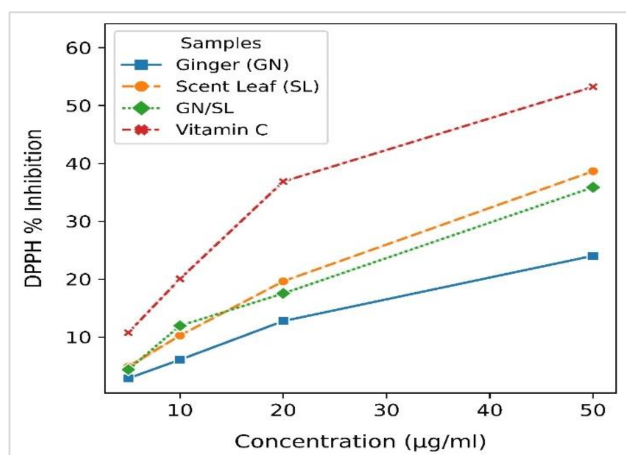



**Figure 3:** DPPH % Inhibition for different concentrations.

The plant extract's ability to convert $Fe^{3+}$ to $Fe^{2+}$ was utilized to gauge how effective an antioxidant was. Fig. 4 displays the ferrous-reducing antioxidant potencies of various aqueous extracts of ginger roots, scent leaf, and their mixtures. This assessment was concentration dependent, with rising inhibitory power proportionate to increasing concentration activity. The GNSL sample inhibited the most, followed by the scent leaf and ginger, which inhibited the least. These findings also suggest that the extracts have the capacity to lessen oxidative damage to various important tissues in the body. These plants' reducing capacity was shown to have a direct and linear correlation to their percentage antioxidant activity.

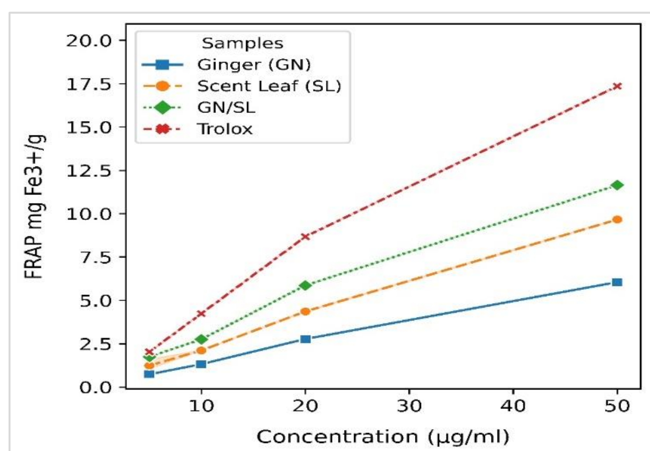



**Figure 4**: FRAP mg $Fe^{3+}$/g for different concentrations**.**

## Conclusion

The chemical makeup of a plant influences its physiological qualities and therapeutic efficacy. The study looked at the possible medical benefits of GN, SL, and the herbal combo GNSL, which may be ascribed to their unique chemical makeup and nutritional content. The current study concentrated on the phytochemical evaluation and antioxidant capacity of scent leaf, ginger and their mixtures established that these medicinal plants cultivated in the southern part of Nigeria are powerhouses of nutrients. The above results indicate that these extracts can form part of a complementary diet for total wellbeing. Additionally, owing to therapeutic potentials revealed by the considerable amounts of secondary metabolite present, the exploration of the mixtures of these extracts in drug development cannot be overemphasized.

**Authors' contributions**

CGI conceived and carried out the experiments, evaluated the findings, and wrote the report. JOA determined the antioxidant characteristics of the extracts, wrote the report, and helped plan the study. KPI took charge of the statistical analysis and text revision. The work was overseen by ENO. All authors evaluated and approved the final draught.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**Data and resource availability.**

The corresponding author may provide the data that supports the study's findings upon reasonable request.

**Competing interest**

There are no stated competing interests**.**

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