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Investigating the Total phenolic contents, in-vitro Antioxidant activities and Elemental contents of *Annona muricata*, *Bryophyllum pinnatum* and *Syzygium aromaticum* in South-West Nigeria

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Abstract

Based on the existence of phytochemicals including alkaloids, flavonoids, saponins, tannins, carotenoids, phenols and steroids that are highly effective antioxidants helpful in medicinal plants, they are useful in disease control and management. Thus, medicinal plants are used as natural remedies for treating conditions like cancer, diabetes, hypertension, and other cardiovascular diseases. However, the actual ways of involvement of these herbs and plants remain unclear, but the presence of certain mineral elements has also been connected to the pharmacological efficacy of medicinal plants in addition to their phytochemicals. Exploring the precise pathways of involvement of these herbs in the management and control of various diseases may be a useful direction in developing pharmacologically active phytomedicines. This study was designed to investigate the phytochemical contents, total phenolic contents, antioxidant potentials and concentrations of mineral elements including selenium (Se), iron (Fe), zinc (Zn), copper (Cu), potassium (K), magnesium (Mg) and chemical elements, "lead (Pb) and cadmium (Cd)" in the ethanolic extracts of *Annona muricata* (Soursop fruits), *Bryophyllum pinnatum* (miracle leaf) and *Syzygium aromaticum* (cloves). The Total phenolic content was determined by the use of the Folin-Ciocalteu assay, whereas antioxidant analysis of the three extracts was conducted via the Ferric Reducing Antioxidant Potential (FRAP) and 2,2-diphenyl-1-picrylhydrazyl (DPPH) assay to determine their efficacy in preventing, controlling and managing health challenges. The elemental assessment was done by employing atomic absorption spectroscopy (AAS). Every test was conducted in triplicates and results were analyzed. Our

results showed that ethanolic extracts of *Annona muricata* (Soursop fruits), *Bryophyllum pinnatum* (miracle leaf) and *Syzygium aromaticum* (clove) exhibit outstanding antioxidant activities, showed the presence of dominant and helpful elements as well as phytochemicals in varying degrees. The extract of *Syzygium aromaticum* among the three tested extracts exhibits the highest phenolic content of 111.3 mg GAE/g, the highest DPPH scavenging efficiency of 81.3% and the highest Ferric ion reducing antioxidant potential (FRAP) of 77.9%. These results suggest that the pharmacological efficacies of the selected herbs may be a function of the presence of notable elements, phytochemicals and antioxidant activities observed in their ethanolic extracts.

***Keywords*:** antioxidants, phytochemicals, total phenols, *Annona, Bryophyllum, Syzygium.*

Introduction

In the healthcare system, the traditional medical approach relying on the use of herbal treatment is still significant because they are natural medicines with fewer adverse effects, less toxin synthesis and greater efficacy than alternatives that involve chemical combination (Batiha *et al*., 2020); thus, adoption of medicinal plants in health improvement has yielded more acceptability in past few years (Abushouk *et al*., 2017).The stems, leaves, flowers, roots, and fruits of plants contain functional compounds that have medicinal value. As a matter of significance, phytochemicals and active substances found in medicinal plants include tannins, flavones, triterpenoids, steroids, saponins, alkaloids, steroids, phenolics, flavonoids, glycosides, terpenes, and polyphenols (Batiha *et al*., 2020; Tungmunnithum *et al*., 2018; Abushouk *et al*., 2017). Therefore, Sun and Shahrajabian (2023) and Sofowora *et al*., (2013) indicated that the compounds found in beneficial herbs afford them their pharmacological activity and beneficial effects for relieving various health issues without causing adverse side effects. Moreover, these medicinal plants are also highlighted as beneficial herbs that are effective against inflammation, bacterial infection, hyperactivity, and pain (Beshbishy *et al*., 2019; Batiha *et al*., 2019).

Additionally, the administration of extracts from health-promoting plants is used as a safe therapy (antioxidants) against oxidation in tissues caused by reactive oxygen species (ROS) synthesis from macromolecules, including lipids, proteins, and DNA which is linked with the onset of most health impairments (Hassan *et al*., 2017). Thus, as the overproduction of oxidants in the body is connected to the occurrence and progression of chronic illnesses, the protective influence of phytochemicals on the health of living organisms may be linked to their antioxidant ability (Zhang

et al., 2015). The quantification of total phenolic compounds present in plants extract has also been widely considered as an indication of antioxidants potentials of these extracts (Sun *et al*., 2002). Hence, based on their ability to treat health challenges, medicinal plants are recognized as natural medicines in many nations (Sun and Shahrajabian, 2023; Batiha *et al*., 2020); therefore, this study adopted *Annona muricata*, *Bryophyllum pinnatum* and *Syzygium aromaticum* as the medicinal plants of interest whose phytochemical contents, antioxidant activities, total phenolic and mineral elements contents were determined for health management measures.

Annona muricata, sometimes referred to as graviola, soursop, guanabana and sirsak has the taxonomy Magnoliophyta division, Magnoliales order, Annonaceae family and genus Annona (Mutakin *et al*., 2022). The tropical plant named *Annona muricata* is well-identified for the presence of edible fruits with therapeutic benefits but also has some toxicological and cytotoxic effects (Coria-Téllez *et al*., 2018). Moreover, *Annona muricata* has been adopted as herbal medicine against illnesses including cancer, diabetes mellitus, hypertension, and bacterial infections; focusing on health improvement (Mutakin *et al*., 2022). Furthermore, according to Zubaidi *et al*., (2023), *Annona muricata* has pharmacological effects against cancer, microorganisms, oxidative stress, ulcers, diabetes, hypertension, and wounds. However, future investigations are required to fill the current knowledge gap concerning the potency of *Annona muricata* by carrying out in-depth analyses and obtaining a deeper understanding of the basis for the pharmacological potentials of *Annona muricata*.

Bryophyllum pinnatum is a herb existing as a member of the Crassulaceae family and it may reach a height of 120 cm having simple or trifoliolate leaves with petioles (Bakare *et al*., 2021). Furthermore, *Bryophyllum pinnatum* also referred to as the miracle leaf, the life plant, and the never-die plant (Ogidi *et al*., 2019) is endemic to Madagascar, and it is valued for its medicinal and ornamental qualities (Kumar *et al*., 2020). It is also known as Ewe Abamoda in Yoruba, "odaa opue" in Igbo, and Shuka halinka in the Hausa language (Adeola *et al*., 2023). Interestingly, the bark and leaves of *Bryophyllum pinnatum* are utilized for treating insect stings, diarrhoea, vomiting, and stomach ulcers. Also, *Bryophyllum pinnatum* leaf juice is used to treat asthma, colds, palpitations, chickenpox, ear infections and to get rid of kidney stones. Additionally, Elufioye *et al*., (2022), Fernandes *et al*., (2019) and Yadav *et al*., (2021) highlighted that the plant *Bryophyllum pinnatum* is commonly used for its immunosuppressive and

immunomodulatory properties, which help to fight inflammation, treat oedema, treat liver and vascular health, and act as a muscle relaxant. Hence, it is hypothesized that the benefits of *Bryophyllum pinnatum* plant may be associated with chemical compounds including alkaloids, saponins, flavonoids and triterpenoids; which may aid in the development of safe pharmaceuticals and this thus influenced the conduct of this current research.

Clove or *Syzygium aromaticum* is a typical spice that is high in hydrocarbons, phenolic compounds, monoterpenes, and sesquiterpenes (Batiha *et al*., 2020). In addition, the tree of the clove plant comprises flowering buds, stems, shoots and leaves (Batiha *et al*., 2020; Cortés-Rojas *et al*., 2014). The phytochemical components of *Syzygium aromaticum* have enabled the plant to be used pharmacologically to treat infections caused by pathogens such as bacteria, viruses (hepatitis C, *Herpes simplex*) and parasites (*Babesia spp., Plasmodium spp.*, *Theileria spp.* (Tiwari and Dubey, 2022). Similarly, sesquiterpenes extracted from cloves were revealed to exhibit anti-carcinogenic potential (Batiha *et al*., 2020), whereas eugenol has also been widely employed by dentists due to its permeability through the pulp of dental tissues, thereby interacting with blood and eventually enters systemic circulation (Martínez-Herrera *et al*., 2016). Significantly, cloves have been used for treating medical conditions such as nausea, vomiting, and flatulence, for inhibiting food-borne pathogens; as well as for managing liver, bowel, and stomach disorders.

Crucially, for this investigation, the discovery of potential pharmacologically active natural products that are easily affordable and likewise presenting fewer side effects than synthetic analogues to manage several diseases is of utmost importance. Synthetic drugs have been utilized in the management of diseases with relative success but the presence of significant biologically active chemicals in medicinal herbs can be harnessed for the development of practical therapeutic solutions with exceptional qualities against inflammation, cancer, diabetes, and hypertension. In addition to the bioactive substances found in beneficial herbal plants, it has also been observed that certain beneficial elements such as iron, zinc, copper and selenium are present in variable degrees (Swain and Rautray, 2021). Although medicinal plants have been used as therapeutic interventions for managing diseases, the report by Li and Weng (2017) has shown that the precise mechanism of their beneficial activity is still unclear. Hence, it is crucial to evaluate the influence of essential elements and/or minerals obtained from medicinal plants and ascertain their precise

involvement in the management of various diseases. Therefore, as bioactive substances (Ugboko *et al*., 2020) and mineral elements (Anand *et al*., 2022) are reported to grant immuno-protective effect against diseases in minute quantities, investigating their precise importance in metabolic activities may help to adequately understand disease incidence and evolution (Ausländer *et al*., 2017). Thus, this *in vitro* study is focused on observing the anti-oxidant ability, amount of total phenols and mineral elements contained in the ethanolic extracts of *Bryophyllum pinnatum*, *Annona muricata* and *Syzygium aromaticum* plants grown and cultivated in South-West Nigeria.

MATERIALS AND METHODS

Materials

Annona muricata (Soursop fruit), *Bryophyllum pinnatum* (miracle leaf) and *Syzygium aromaticum* (cloves) are the major materials used in this study

Chemicals and reagents

The chemicals and reagents utilized in this study were of pure and analytical grade. Gallic acid, DPPH, Ascorbic acid, and hydrogen peroxide were purchased from Sigma Aldrich (Gillingham, UK). Sodium phosphate, sodium hydroxide, Folin-Ciocalteu reagent, $FeCl_3$, and potassium ferricyanide were products of Randox Laboratories Ltd. (Crumlin, UK).

Plant collection, Authentication and Extraction

Annona muricata (Soursop fruits), *Bryophyllum pinnatum* (miracle leaf) and *Syzygium aromaticum* (clove) were sourced from three (3) different locations in Ogun state, Nigeria. These samples were identified at the Department of Plant Science, Faculty of Sciences, Olabisi Onabanjo University, Ago-Iwoye, Ogun State, Nigeria; after which the leaves, buds and fruits specimens were placed in the herbarium. *Annona muricata* fruits, the clove buds and *Bryophyllum pinnatum* leaves were carefully collected and were rinsed using water from the tap before rinsing with distilled water. Leaves of *Bryophyllum pinnatum* plant and the clove buds samples were chopped into small sizes and subsequently air-dried for two weeks while the *Annona muricata* fruits were dried to a constant weight using the food dehydrator (Bosch food dehydrator) at 70 degrees. Thereafter, there was pulverization of the dried samples into coarse powder from which 300 g

each of the plant samples was taken and mixed in 1000 mL of ethanol. The sample was soaked in 99.7% ethanol for five days to achieve adequate extraction. The herbs were decanted and a wool funnel was used to filter in order to ensure that the filtrate was used. Evaporation of the solvent from the herbs was done by RotoVap 110 and the slurry form was collected into a beaker, concentrated at 40°C using a rotary evaporator. There was lyophilization of the concentrated samples to obtain dry powders which were weighed and thereafter stored in different dry containers that were air-tight.

Methods

Phytochemistry

The qualitative assay for the phytochemical contents of ethanolic extracts of *Annona muricata* (Soursop fruits), *Bryophyllum pinnatum* (miracle leaf) and *Syzygium aromaticum* (clove) was carried out by following the method of Harborne, (1984) and Sofowora, (1993).

Determination of Total Phenolic Content (TPC)

Folin-Ciocalteu assay, as reported by Singleton *et al.,* (1999), was used to ascertain the phenolic contents in the ethanolic extract of each plant [*Annona muricata* (soursop fruits), *Bryophyllum pinnatum* (miracle leaf), *Syzygium aromaticum* (clove)]. 100 µL of the ethanolic extract was neutralized with 2.0 ml of 7.5% sodium carbonate followed by oxidization with 2.5 ml of 10% Folin-Ciocalteau's reagent (v/v). The reaction mixture was incubated for 40 minutes at 45°C, after which a spectrophotometer was used to measure the absorbance at 765 nm. By comparing the absorbance with a standard curve created using (10 mg/100 ml) of Gallic acid as an equivalent reference compound, the total phenol content was ascertained. Gallic acid equivalent (mg GAE /g extract) was used to express the extract's total phenol content.

In-vitro Antioxidant Activity Assays

The determination of *in vitro* free radical scavenging (anti-oxidant) ability was carried out with two different chemical methods including the Ferric Reducing Antioxidant Potential (FRAP) and 2,2-diphenyl-1-picrylhydrazyl (DPPH) assays.

Determination of DPPH antioxidant ability

The DPPH assay was determined using the method introduced by Mensor *et al.,* (2001) after brief modification. The sample stock, initially at a concentration of 0.004g /10 ml, was diluted to final concentrations of 200, 100, 50, 25, and 12.5 µg/ml using methanol. 1 ml of a 0.3 mM DPPH methanol solution was subsequently mixed with 1 ml of the sample solution at varying concentrations. Incubation of the resulting mixture to achieve effective reaction was done at 100°C for 30 minutes. Consequently, determination of the absorbance values at 517nm was done using a spectrophotometer. However, the DPPH free radical scavenging activity of the ethanolic plant extracts was relatively determined by using ascorbic acid as a standard, and the outcomes were presented as the percentage of antioxidant activity.

Determination of Ferric Reducing Antioxidant Potential (FRAP)

The ferric-reducing antioxidant potential (FRAP) was determined using the method of Oboh and Omoregie (2011). For this assay, 50 µl aliquot extract was added to 450 µl of 200 mM sodium phosphate buffer (pH 6.6), followed by the addition of 250 µl 1% potassium fericyanide. The resulting mixture was allowed to incubate at 50 ^{0}C for 20 minutes. Thereafter, there was addition of 250 µl of 10% trichloroacetic acid. Then, centrifugation of the mixture was done at 2000 rpm for 10 minutes. Afterwards, 10 µl of the supernatant was collected and mixed with 500 µl of 0.1% of iron (III) choride ($FeCl_3$). The absorbance value of the final mixture was measured spectrophotometerically at 700nm. All tests were conducted in triplicates. An increase in the absorbance value of the resulting (final) mixture reflects an elevated reducing power of the plant samples. However, to ascertain the relative antioxidant potential of the ethanolic plant extracts, ascorbic acid was also used as a standard.

Elemental Analysis

Atomic absorption spectroscopy, as outlined by Gaya and Ikechukwu (2016), was used to perform the elemental analysis and estimate the concentration of Fe, Zn, Se, Cu, Mg, Cd, K and Pb in the extracted sample. After wet digestion, the filtrate was subjected to analysis using an Atomic Absorption Spectrophotometer (AAS) from Hitachi Z 5000 flame. The usage of AAS was supported with acetylene standards and the results were analyzed as required.

Statistical Analysis

Data derived from respective assays were analyzed using the statistical package for social sciences (SPSS version 16.0 for Windows) and expressed as mean ± SD with each experiment independently conducted in triplicates. Statistical significance was determined with one-way analysis of variance (ANOVA) test and comparisons across groups were considered significant at $p < 0.05$.

RESULTS

This study investigated the phytochemical components, antioxidant potential, and levels of mineral element in the ethanolic extracts of *Annona muricata* (soursop fruit), *Bryophyllum pinnatum* (miracle leaf) and *Syzygium aromaticum* (clove). By using the methodologies as detailed under materials and methods, useful and valid insights about the tested medicinal plants were gathered.

1. PHYTOCHEMICAL ANALYSIS

In this study, qualitative phytochemical screening of *Annona muricata* fruits, *Bryophyllum pinnatum* leaves and *Syzygium aromaticum* buds were determined and the findings are shown in Table 1.

Table 1: Qualitative screening of phytochemicals from the ethanolic extracts of *Annona muricata* fruits, *Bryophyllum pinnatum* leaves and *Syzygium aromaticum* buds.

Phytochemical	*Syzygium aromaticum*	*Bryophyllum pinnatum*	*Annona muricata*
Alkaloids	-	+++	+
Flavonoids	+++	++	-
Tannins and phenolic compounds	+++	++	++
Saponins	+	+	+
Steroids	+++	+++	-
Terpenoids	+	-	-
Triterpenoids	+++	-	Trace

KEYS: + Present ++ High concentration +++ Very high concentration - Absent

2. TOTAL PHENOLIC CONTENT

The quantification of total phenolic compounds present in plants extract has been widely considered as an indication of antioxidants potentials of these extracts. The outcome of this assessment in the three tested plants is presented in Figure 1.

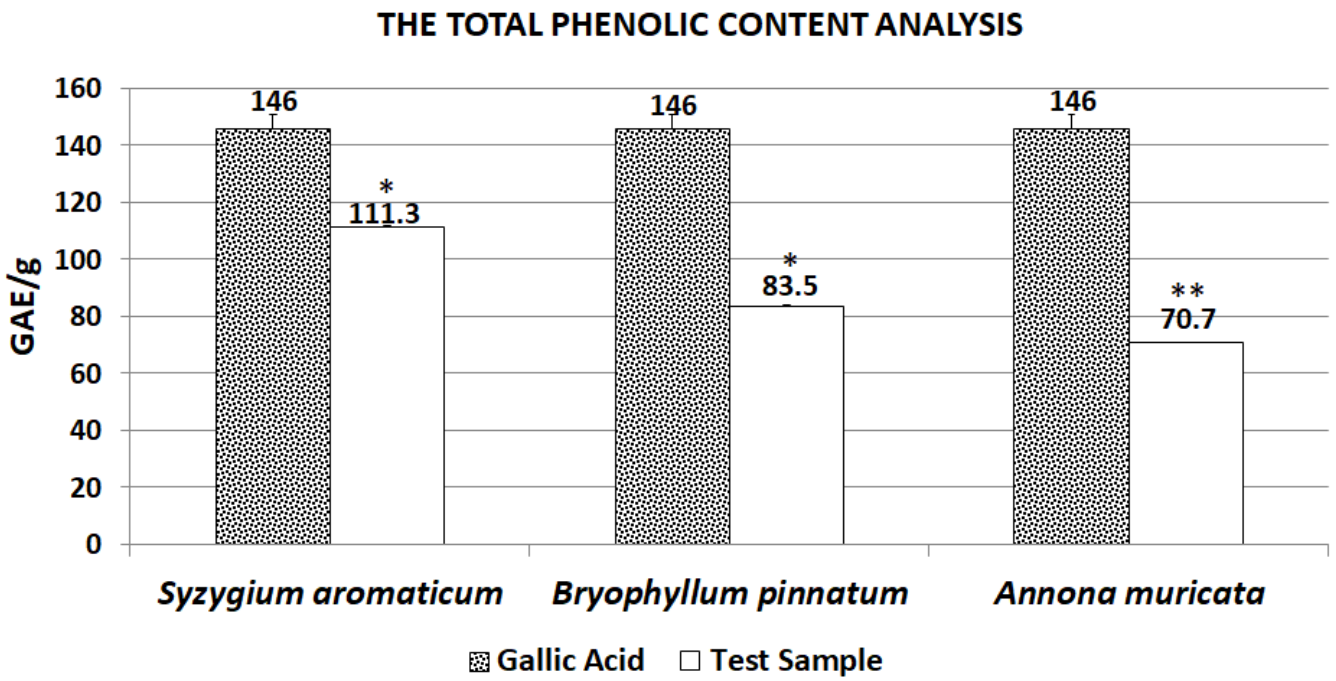

Figure 1: Bar chart illustration of the Total phenolic content in the ethanolic extracts of *Annona muricata* fruits, *Bryophyllum pinnatum* leaves and *Syzygium aromaticum* buds in comparison to Gallic acid. Results are expressed as mean ± SD Gallic acid equivalent (GAE)/g with each experiment independently conducted in triplicates. Two asterisks show significance at $p<0.01$, and one asterisk indicates significance at $p<0.05$.

3. ANTIOXIDANT ANALYSIS RESULTS

Antioxidant evaluation was conducted using two distinct chemical methodologies, namely DPPH and FRAP assays as delineated in the materials and methods section. The antioxidant/radical scavenging ability of ethanolic extracts of *Annona muricata* (fruits), *Bryophyllum pinnatum* (leaves) and *Syzygium aromaticum* (buds) were investigated and the findings are presented in Figures 2 and 3.

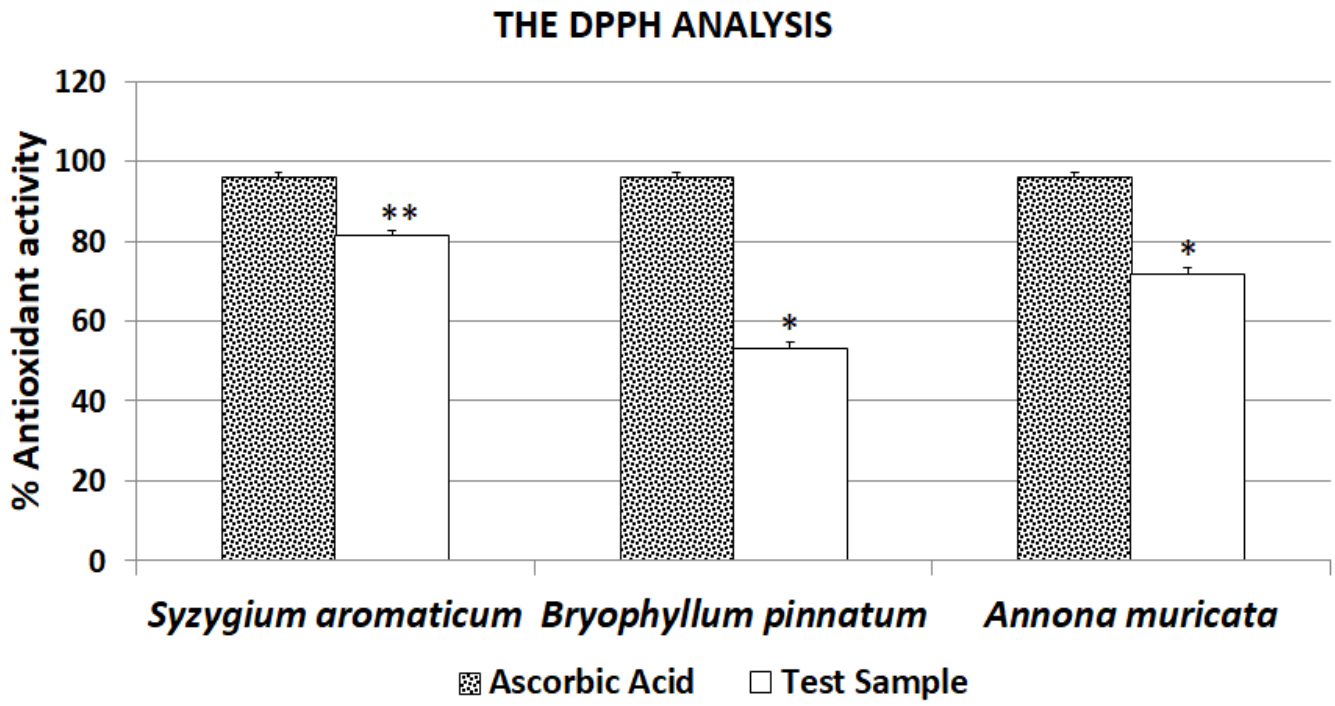

Figure 2: Bar chart illustration showing the comparison of percentage DPPH radical scavenging activity between the ethanolic extracts of *Annona muricata, Bryophyllum pinnatum* and *Syzygium aromaticum* with Ascorbic acid. Results are expressed as mean percentage antioxidant activity ± SD with each experiment independently conducted in triplicates. Two asterisks show significance at $p<0.01$, and one asterisk indicates significance at $p<0.05$.

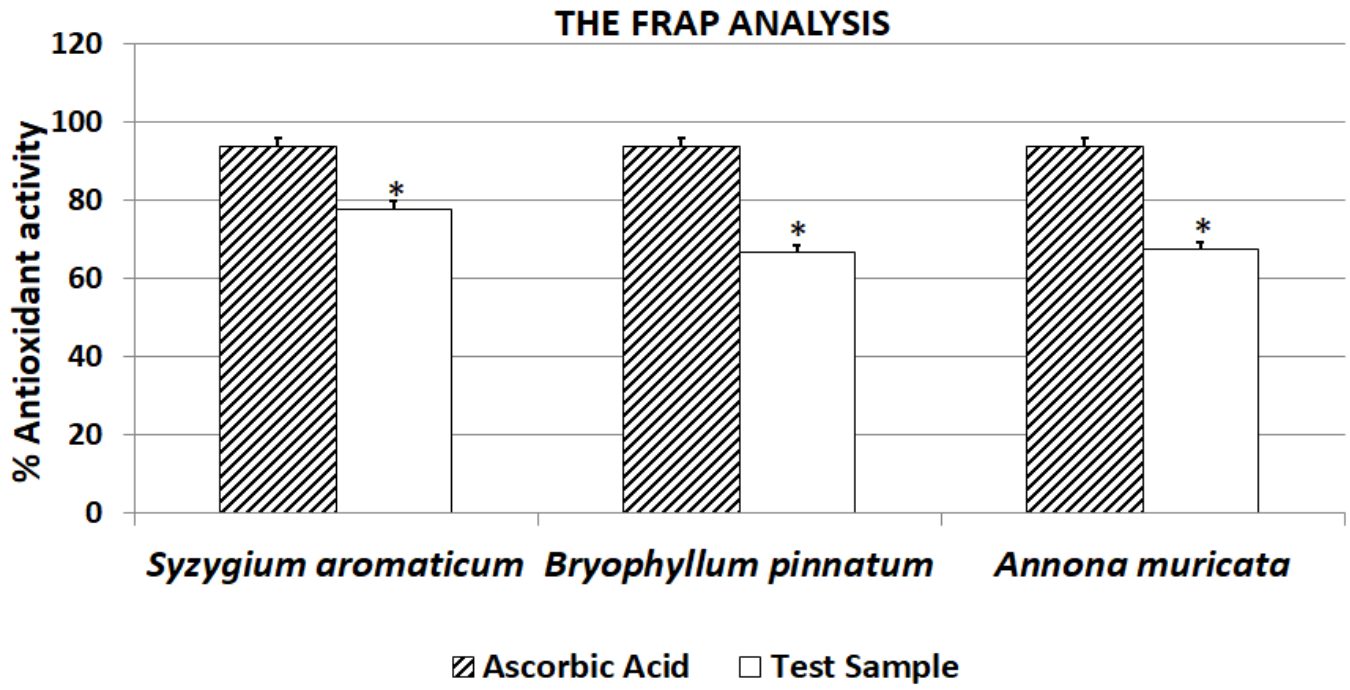

Figure 3: Bar chart representation showing the comparison of percentage FRAP between the ethanolic extracts of *Annona muricata, Bryophyllum pinnatum* and *Syzygium aromaticum* with Ascorbic acid. Results are expressed as mean percentage antioxidant activity ± SD with each experiment independently conducted in triplicates. One asterisk indicates significance at $p<0.05$.

4: **ELEMENTAL CONTENT DETERMINATION**

The outcome of evaluating the mineral elements "zinc, copper, iron, selenium, magnesium, sodium" and chemical elements "cadmium and lead" in the ethanolic extracts of *Annona muricata***,** *Bryophyllum pinnatum and Syzygium aromaticum* are presented in Figure 4.

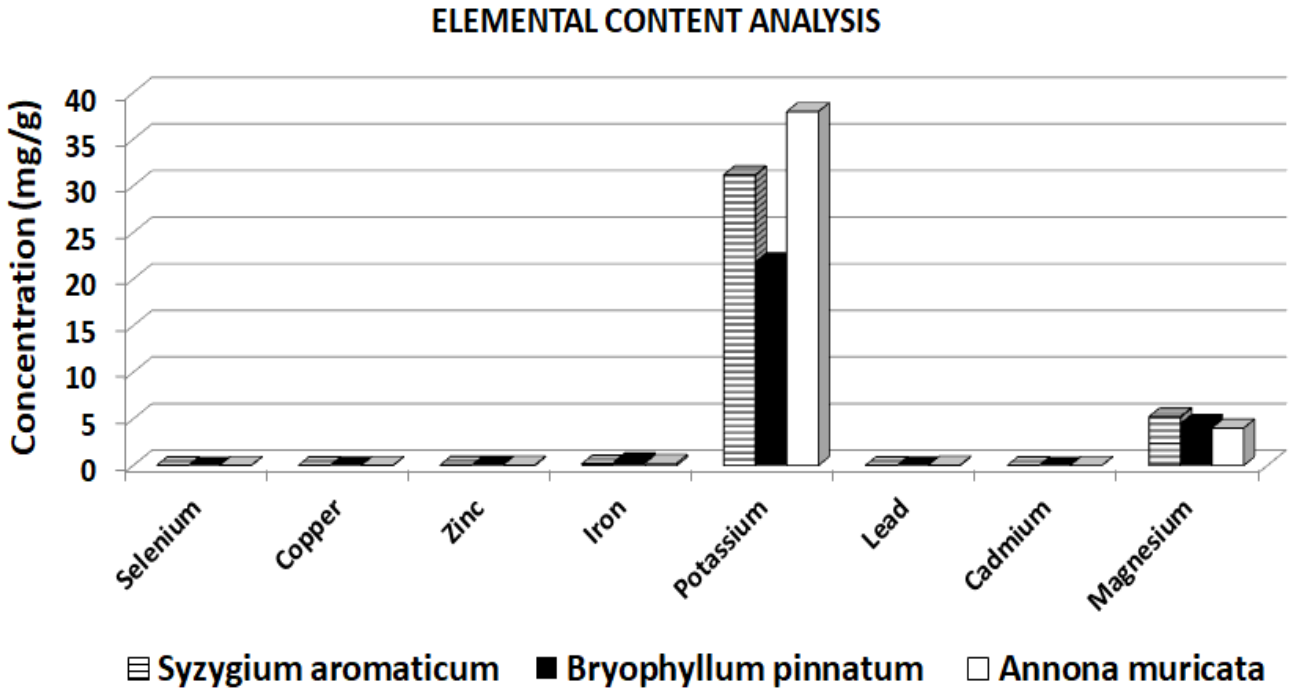

Figure 4: Bar chart illustration of the assessment of elements in the ethanolic extracts of *Annona muricata, Bryophyllum pinnatum* and *Syzygium aromaticum.* Results are presented as bar charts with each experiment independently conducted in triplicates and expressed in mg/g.

DISCUSSION

The qualitative screening of *Annona muricata, Bryophyllum pinnatum* and *Syzygium aromaticum* plants extracts (Table 1) indicates that phytochemicals including tannins, alkaloids, terpenoids, flavonoids, phenolic compounds, steroids, saponins and triterpenoids are present in these extracts. Laboratory studies were carried out and the result shows that flavonoids, tannins, phenolic compounds and steroids are highly present in *Syzygium aromaticum*. In *Bryophyllum pinnatum* phytochemicals showing very high concentrations are alkaloid and Steroid; while in *Annona muricata*, tannins and phenolic compounds appear to be in high concentration which corroborate the findings of Gavamukulya *et al*., (2023) and Coria-Téllez *et al*., (2018). Few of the phytochemicals in these plants are sometimes present in trace amounts. The presence of these beneficial phytochemicals may be an important tool in the treatment and management of several illnesses such as ophthalmia, menorrhagia, boils, epilepsy, hemorrhoids, piles, cancer, lung infections and rheumatoid arthritis (Elufioye *et al*., 2022). Plants of important phytochemical

contents have been employed in the treatment of allergic reactions, as an antihistamine, an antifungal, and to improve vascular integrity (Elufioye *et al*., 2022). The observed presence of phytochemicals in *Annona muricata, Bryophyllum pinnatum* and *Syzygium aromaticum* plants in this study and the suggested usefulness in disease management conform to the research by Adegbola *et al.,* (2017) and Singleton *et al*., (1999), wherein they both claimed that phenol and flavonoid-rich plants reduce the incidence of coronary heart disease, shield bio-systems from free radicals, prevent oxidative cell damage, and had highly effective anti-cancer potential.

As highlighted in the report by Aguilera *et al.,* (2016), phenolic compounds are regarded as strong and very significant antioxidants found in a various plant species. Hence, phenols and phenolic compounds have been reported to be associated to defense responses in plants such as acting against pathogens found in the soil and/or insects that feed on plant roots (Ndakidemi and Dakora, 2003) as well as being associated with anti-microbial activity against bacterial and fungi in humans (Anand *et al.,* 2019). Following this study output, the total amount of phenolic contents were significantly expressed as Gallic acid equivalent per gram (GAE /g) (Figure 1) with 111.3, 83.50 and 70.7 mg GAE/g respectively recorded for *Syzygium aromaticum, Bryophyllum pinnatum* and *Annona muricata* extracts with the highest value observed in *Syzygium aromaticum* extract followed by *Bryophyllum pinnatum* and thereafter *Annona muricata* extracts. Phenolic compounds have been reported to perform significant roles in prevention of chronic diseases (Aguilera *et al.,* 2016). Thus, our results on the total phenolic contents of these tested extracts revealed their antioxidative potentials and consequently suggest their therapeutic and pharmacological abilities.

In addition, according to this study results, the extracts of *Syzygium aromaticum, Bryophyllum pinnatum* and *Annona muricata* possess significant DPPH scavenging efficiencies (Figure 2) of 81.3%, 53.2%, and 71.9% respectively when compared with the standard (Ascorbic acid); whereas the Ferric Reducing Antioxidant Potential (FRAP) results revealed that *Syzygium aromaticum* also has the highest reducing antioxidant potential of 77.9% followed by *Annona muricata* (67.4%) and 66.6% value for *Bryophyllum pinnatum* (miracle leave*)*, having the lowest ferric ion reducing antioxidant potential (Figure 3).

The findings of this research show *Syzygium aromaticum* (clove) with the highest phenolic content of 111.3 mg GAE/g; highest DPPH scavenging efficiency of 81.3% and the highest

Ferric ion reducing antioxidant potential (FRAP) of 77.9%. These results corroborate the report of Mutlu-Ingok *et al*., (2020) and Parham *et al*., (2020) that *Syzygium aromaticum* (clove) has drawn global attention because of its strong antioxidant and antimicrobial properties. The observed results for antioxidant and total phenolic content in *Syzygium aromaticum* from this study also corroborate the works of Tiwari and Dubey (2022) who noted that *Syzygium aromaticum* oil (clove oil) contains three notable phytochemicals including β-caryophyllene, eugenol and eugenyl acetate. Moreover, the study by Batiha *et al*., (2020) conferred on eugenol an antimicrobial, antiseptic, analgesic, antioxidant, anticancer, antidepressant, antispasmodic and antiviral properties against various pathogenic bacteria. The observed significant total phenolic content of 83.5 mg GAE/g and DPPH scavenging efficiency of 53.2% with *Bryophyllum pinnatum* leaves extract in this study also corroborate the study of Elufioye *et al*., (2022), Fernandes *et al*., (2019), Okhale *et al*., (2021), Selvakumar (2022) and Yadav *et al*. (2021) who reported that the plant *Bryophyllum pinnatum* is commonly used for its immunosuppressive and immunomodulatory properties, which help to fight inflammation, treat oedema, treat liver and vascular health, involved in wound healing, treatment of smallpox, asthma, palpitations, cuts, burns, scrapes, diarrhoea, get rid of kidney stones and act as a muscle relaxant.

Furthermore, the total phenolic content of 70.7 mg GAE/g and DPPH scavenging efficiency of 71.9% recorded for *Annona muricata* extract corroborate the reports of Mutakin *et al*., (2022) who reported that *Annona muricata* is utilized as medicinal/herbal therapy for treating and improving illnesses including cancer, bacterial infections, diabetes mellitus (DM) and hypertension. Also, the research by Zubaidi *et al*., (2023) indicated that *Annona muricata* had pharmacological properties against oxidation, cancer, ulcers, microbial infection, diabetes, hypertension and wounds.

The mineral elemental analysis of this study (Figure 4) revealed that potassium (K) and magnesium (Mg) were the major elements with potassium being more abundant in all the three tested extracts while other elements investigated [copper (Cu), cadmium (Cd), iron (Fe), selenium (Se), zinc (Zn) and lead (Pb)] exist in trace amounts in the extracts. *Annona muricata* extract contains the highest value of potassium (37.9 mg/g) followed by *Syzygium aromaticum* extract (31.2 mg/g) and the least value of 21.9 mg/g in *Bryophyllum pinnatum* extract. *Syzygium aromaticum* extract contained the highest value of magnesium with a concentration of 5.0 mg/g followed by *Bryophyllum pinnatum* extract with a concentration of 4.6 mg/g and the least concentration of 3.0 mg/g in *Annona muricata* extract. The presence of potassium in the ethanolic

plant extracts studied in this research suggests their therapeutic property in controlling osmotic pressure, nerve impulse conduction, muscular contraction, and cell membrane integrity. High concentration of magnesium suggests that the extracts may be useful in maintaining brain and heart functions which in diet may have health benefits including lowering blood sugar level and consequently protect against heart attack, stroke and diabetes. Our results on mineral content analysis corroborate the report of Zubaidi *et al*., (2023) who reported that *Annona muricata* exhibits outstanding pharmacological properties including cancer prevention, abrogation of oxidative damage, control of diabetes, microbial prevention and control, wound healing, treatment of ulcer and management of hypertension. The presence of magnesium in the extracts is an indication of their anti-diabetic effect possibly by having influence on the pathways involved in inhibition of glucose absorption following the inhibition of α-amylase and α-glucosidase activities. Consequently, decreasing glucose absorption rate via enzyme inhibition will promote insulin release, enhance glucose re-uptake by peripheral tissues and improve glucose tolerance.

CONCLUSION

The findings from this research suggest that pharmacological efficacies attributed to *Annona muricata*, *Bryophyllum pinnatum* and *Syzygium aromaticum* may be a function of the presence of notable elements, phytochemical expression and antioxidant activities observed in their ethanolic extracts. These results have indications for the development of new drugs and therapies emerging from natural products which are believed to be less expensive and present fewer side effects than the synthetic analogues.

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