

Phytochemical Analysis, Antioxidant properties, Anti-nutrient Contents and Antimicrobial study of Aqueous Extract of Three Plants of Medicinal benefits

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Abstract

The emergence of antibiotic-resistant microorganisms poses a significant challenge in clinical practice by complicating treatment options for infections. Exploring natural products as alternative drugs presents a promising approach to addressing these issues. Medicinal plants, acknowledged by the World Health Organization (WHO) as a preferred and safest source of various medicines, have gained considerable attention. Therefore, this study was conducted to investigate the phytochemical constituents, antioxidant properties, anti-nutrient content, and antimicrobial activities of *Morinda lucida*, *Solanum erianthum*, and *Dryopteris expansa*. The phytochemical analysis indicates that while flavonoids, saponins, and reducing sugars are common across the studied plants, *Morinda lucida* and *Dryopteris expansa* exhibit higher levels of saponins and the presence of alkaloids, suggesting potential unique medicinal properties. The antimicrobial study revealed that the aqueous leaf extract of *Morinda lucida* made *Streptococcus pyogenes* and *Staphylococcus aureus* susceptible to its effects, while *Dryopteris expansa* made only *Streptococcus pyogenes* susceptible and the aqueous leaf extract of *Solanum erianthum* made the fungal species *Penicillium chrysogenum* susceptible to its action. All the studied plants demonstrated very strong antioxidant activity. The anti-nutrient analysis showed that *Morinda lucida* had the lowest cyanide content at 5.50 mg/100 g, while *Solanum erianthum* had the highest cyanide content at 6.89 mg/100 g. *Dryopteris expansa* exhibited the lowest levels of oxalates and phytates, with values of 0.28 mg and 0.20%, respectively. The results of this study demonstrate that each of these plants possesses significant potential as antimicrobial agents and antioxidants, with low toxicity levels. Consequently, they hold promise for medicinal applications and drug development.

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Keywords: Antimicrobial Resistance, Antioxidant Properties, Medicinal Plants, Natural Products, Anti-Nutrient Content

Introductions

In recent years, there has been a growing interest in exploring the bioactive properties of plant extracts for their potential health benefits. Among the diverse array of plants studied, *Morinda lucida*, *Solanum*

erianthum, and *Dryopteris expansa* have garnered attention due to their rich phytochemical profiles [1,2,3]. These plants are recognized for containing phytochemicals such as flavonoids, saponins, and alkaloids, which possess antioxidant properties capable of neutralizing harmful free radicals and protecting against oxidative stress-related diseases [4]. Beyond their therapeutic applications, these compounds also serve as valuable precursors for synthesizing pharmaceutical drugs [5]. Throughout history, traditional medicines have relied heavily on natural products, and today, plants are increasingly recognized as a source for discovering new drugs [6]. Compared to synthetic drugs, plant-based phytochemicals such as phenols, alkaloids, coumarins, and terpenes offer potent agents capable of combating multidrug-resistant microbial strains. These phytochemicals exhibit diverse medicinal properties and can target mechanisms of drug resistance, including bacterial cell communication, membrane proteins, efflux pumps, and biofilm formation [7].

Morinda lucida, abundantly found in the tropical regions of Central and West Africa, is a prominent medicinal plant widely utilized in African traditional medicine [8,9,10]. Belonging to the family Rubiaceae, it is an evergreen shrub that can reach heights of up to 18 meters [11]. The leaves are typically 7-15 cm long and 3.5-7.5 cm wide, forming a glossy foliage. Various parts of *M. lucida*, including the leaves, seeds, twigs, and stem bark, have been traditionally used across Africa to treat a wide range of ailments such as inflammation, typhoid fever, diabetes, abdominal pains, dysmenorrhea, splenomegaly, helminthiasis, trypanosomiasis, and sickle cell disease [12]. These ethnomedicinal properties have led several researchers to investigate the phytochemical, antioxidant, antimicrobial, anti-inflammatory and anti-diabetes characteristics of various parts of the plant though with inconsistent results with respect to phytochemical contents, antimicrobial activity and acute toxicity [13,14,15].

Solanum erianthum, also known as the potato tree, is a significant source of natural antioxidants, which play a crucial role in mitigating various oxidative stresses [16]. Belonging to the family Solanaceae, it is a fast-growing evergreen shrub or small tree, reaching heights of 4 to 10 meters with stems up to 20 cm in diameter. The plant is unarmed and densely covered with woolly, soft stellate hairs [17]. In West Africa, a decoction of *Solanum erianthum* leaves is traditionally used *Solanum erianthum* as a diuretic and purgative to treat malaria, leprosy, venereal diseases, and to stimulate liver functions [18]. Essential oils extracted from the fruits and leaves of *S. erianthum* have been studied for their cytotoxic properties and traditional

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medicinal uses, particularly for skin diseases and stomach-related ailments [19]. Research has demonstrated that contains phytochemicals with potent antioxidant activities, effectively preventing or reducing oxidative stress [20].

Dryopteris expansa, belonging to the family Dryopteridaceae and commonly known as Spreading Wood Fern, is a deciduous fern native to cool temperate and subarctic regions of the Northern Hemisphere, including Spain and Greece. This fern species has been traditionally harvested from the wild for local consumption as food and medicine, and as a source of materials. Pteridophytes, including ferns like *Dryopteris expansa*, have long played a significant role in traditional medicines across various systems, including Unani, Ayurvedic, and homeopathic practices [21]. These plants have evolved diverse morphological characteristics and secondary metabolites to thrive in harsh terrestrial environments. *Dryopteris expansa* is known to be enriched with a variety of phytochemicals that have potential medicinal applications. These phytochemicals could serve as supplements alongside traditional antibiotics, showing promising activity against multidrug-resistant bacterial strains [22]. While extensive studies specifically on *Dryopteris expansa* are limited, research on related fern species within the Dryopteridaceae family, such as *Dryopteris filix-mas*, has demonstrated medicinal uses, including treatment for worm infections and diarrhea [23]. Hence, exploring the phytochemical constituents, antioxidant properties, anti-nutrient contents, and antimicrobial activities of these plants will provide insights into their therapeutic applications.

Methods and materials

Collection and authentication of the plant's samples: Fresh samples of the *Morinda lucida* (Brimstone tree), *Solanum erianthum* (Potato tree) and *Dryopteris expansa* (Alabama streak sorus Fern) plants were obtained from Anchor University, Ayobo, Lagos State, Nigeria. The leaves were sourced from various sites within the location between July and September 2020 by detaching large quantities of leaves from the plants. The plants were authenticated at the Plant Science Department of Ekiti State University, Ado-Ekiti, Nigeria by the Chief Technologist with voucher numbers UHAE 2020066 *Morinda lucida* Benth (Rubiaceae), UHAE 2020084 *Solanum erianthum* D. Don and UHAE 2020065 *Dryopteris expansa* (C. Presl) Fraser-Jenkins & Jermy. The leaves of the plants were cleansed and air-dried in the open laboratory, crushed, and subsequently ground to powder separately with a Marlex Excella laboratory blender.

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Phytochemical and in vitro antioxidant analyses of *Morinda Lucinda*, *Solanum Erianthun*, and *Dryopteris Expansa*,

Preparation of plant extract

20 g of each plant leaf was weighed and blended in 100 mL of distilled water and filtered to obtain the aqueous solution which was used for the determination of the various parameters.

Phytochemical screening

The qualitative phytochemical screening of [flavonoids, saponins, alkaloids, and reducing sugar] was conducted using the methods described by Sofowora [24] and Trease & Evans [25] to identify the active constituents.

Determination of 2,2 Diphenyl-1-picrylhydrazyl (DPPH) free radical scavenging ability

The 1,1-diphenyl-2-picrylhydrazyl (DPPH) free radical scavenging ability of the extract was determined using the modified method of Gyamfi [26]. Briefly, 1.0 mL of different concentrations (20, 40, and 80 mg/mL) of the extracts were placed in respective test tubes. A 1.0 mL of 0.1 mM methanolic DPPH solution was added to the samples. These samples were vortexed and incubated in the dark at room temperature for 30 minutes. The respective solutions were thoroughly mixed and incubated in the dark for 30 minutes before measuring absorbance at 516 nm. Decreased absorbance of the sample indicated DPPH free radical scavenging capability. Distilled water was replaced for the extract in the control. Percentage radical scavenging ability was calculated using the following expression:

$$\% \text{ DPPH radical scavenging ability} = 1 - \text{Abs}_{\text{Sample}} / \text{Abs}_{\text{Control}} \times 100$$

Determination of Nitric Oxide (NO) radical scavenging ability

The modified methods of Baliga [27] were used to determine the nitric oxide radical scavenging ability. Sodium nitroprusside in aqueous solution at physiological pH 7.0 spontaneously generated NO, which interacted with oxygen to produce nitrite ions that were estimated by the use of Greiss reagent [1.0 mL sulfanilic acid reagent (0.33%) prepared in 20% glacial acetic acid at room temperature for 5 minutes with 1 mL of naphthyl ethylenediamine dichloride (0.1% w/v)]. The reaction mixture contained 2.0 mL of 5 mM sodium nitroprusside in phosphate-saline solution, 0.2 ml of the extract, and 0.5 mL of the Greiss reagent. The absorbance of the blank, test, and control solutions were measured at 546 nm with a spectrophotometer and radical scavenging ability was determined.

$$\text{Nitric oxide radical scavenging ability} = (\text{Abs}_{\text{Control}} - \text{Abs}_{\text{Sample}} / \text{Abs}_{\text{Control}}) \times 100.$$

Determination of ferric-reducing antioxidant power (FRAP)

The reducing property of the three aqueous extracts of the leaves was determined by the modified method of Pulido [28]. This method is based on the reduction of (Fe³⁺) ferricyanide in stoichiometric excess relative to the antioxidants. Different concentrations of the aqueous extract of the sample and its various fractions (10-50 µg/mL) were added to 1.0 mL of 200mM of sodium phosphate buffer pH 6.6 and 1.0 mL of 1% potassium ferricyanide. [K₃Fe (CN)₆]. The mixture was incubated at 50°C for 20 min, thereafter 1.0 mL of freshly prepared 10% TCA was quickly added and centrifuged at 2000 rpm for 10 min, 1.0 mL of the supernatant was mixed with 1.0 mL of distilled water and 0.25 mL of 0.1% of

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FeCl₃ solution was added. Distilled water was used for blank without the test sample while the control solution contained all other reagents except the 0.1% potassium ferricyanide. Absorbances of these mixtures were measured at 700 nm using a spectrophotometer. Decreased absorbance indicated ferric reducing power capability of the sample. The percentage ferric reducing antioxidant power (%) was subsequently calculated = $(Abs_{control} - Abs_{sample}) / Abs_{control} \times 100$.

Determination of total flavonoid

The total flavonoid content of the three aqueous extracts was determined using a colorimeter assay method developed by Bao [29]. A 0.2 mL of the extract was added to 0.3 mL of 5% NaNO₃ at zero time. After 5 min, 0.6 mL of 10% AlCl₃ was added and after 6 mins, 2 mL of 1M NaOH solution was added to the mixture followed by the addition of 2.1 mL of distilled water. Absorbance was read at 510 nm against the reagent blank and flavonoid content was expressed as mg gallic acid equivalent: $Y = 0.005x + 0.464$ (R² = 0.961) mg of GAE/mg of dry extract

The anti-nutrient analyses of the plants

The anti-nutrient analyses of phytate were done according to the Wheeler and Ferrel method [30], oxalate by the procedure of Day and Underwood [31] while the method of AOAC [32] was used to determine the total cyanide content.

Antimicrobial analysis of *Morinda Lucinda*, *Solanum Erianthun*, and *Dryopteris Expansa* Leaf Extracts

The antimicrobial ability of the three aqueous crude extracts was determined using selected bacteria and fungi that were grown in nutrient agar (28 g of nutrient agar in 1000 mL of distilled water and autoclave at temperature 121 °C for 15 minutes) and potato dextrose agar (39 g of potato dextrose agar in 1000 mL of distilled water and autoclave at temperature 121° C for 15 minutes) respectively. The respective agar was brought out of the autoclave to cool a bit before pouring about 15 mL of the agar into the petri dish that was allowed to gel.

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Absorbing the extract into the disc absorbent

2 mL of each extract was placed into respective sterile evaporating dishes and a few adsorbent discs were poured into the evaporating dishes for the extract to be adsorbed.

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Introducing the organism into the media in the petri dish

About 0.1 mL from the slant of the pure organism was introduced with an inoculating loop by streaking into the petri dish. Afterward, the extract-adsorbed discs were introduced into the respective agar plates and incubated at 37 °C for 18 to 24 hours for bacteria in nutrient agar and fungi in potato dextrose agar at 26 °C to 28 °C for 18 to 24 hours.

Standard antibiotics for both gram-positive and gram-negative bacteria (PEF = Pefloxacin, GN = Gentamycin, APX = Ampliclox, Z = Zinnacef, AM = Amoxicillin, R = Rocephin, CP/CPX = Ciprofloxacin, STN = Streptomycin, SXT = Septrin, E = Erythromycin and PEF = Pefloxian, CN = Gentamicin, CH = Chloramphenicol, AU = Augmentin were also used. The plates were observed for antimicrobial activity as

presented in **Tables**. The results obtained from the various analyses were expressed as Mean \pm standard deviation of three determinations.

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Results

Table 1: Phytochemical Analysis of the Samples Aqueous extracts

Samples	Reducing sugar	Saponins	Flavonoids	Alkaloid
<i>Morinda lucida</i>	+	+++	+	+
<i>Solanum erianthum</i>	+	+	+	-
<i>Dryopteris expansa</i>	+	++	+	++

- = Not present, + = Present, ++ =High, +++ =Higher

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Table 2: Total Phenolic content of aqueous leaf extracts(mg garlic acid equivalents/g)

Samples	20mg(GAE/g)	40mg(GAE/g)	80mg(GAE/g)
<i>Morinda lucida</i>	42.00 \pm 0.20	52.40 \pm 0.00	61.40 \pm 0.10
<i>Solanum erianthum</i>	48.00 \pm 0.10	52.30 \pm 0.30	63.40 \pm 0.40
<i>Dryopteris expansa</i>	52.80 \pm 0.20	73.10 \pm 0.10	80.40 \pm 0.10

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Table 3: Total Flavonoids content of aqueous leaf extracts (mg quercetin equivalents/g)

Samples	20mg(QE/g)	40mg(QE/g)	80mg(QE/g)
<i>Morinda lucida</i>	24.00 \pm 0.10	51.80 \pm 0.30	56.00 \pm 0.10
<i>Solanum erianthum</i>	34.50 \pm 0.40	35.50 \pm 0.20	36.00 \pm 0.20
<i>Dryopteris expansa</i>	36.30 \pm 0.10	74.40 \pm 0.20	87.20 \pm 0.10

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Table 4: Concentration of 2,2 Diphenyl-1-picrylhydrazyl (DPPH) radical scavenging ability of aqueous leaf extracts (%)

Samples	20mg	40mg	80mg
<i>Morinda lucida</i>	60.20 \pm 0.00	89.30 \pm 0.10	94.50 \pm 0.10
<i>Solanum erianthum</i>	15.70 \pm 0.00	35.30 \pm 0.30	50.60 \pm 0.00
<i>Dryopteris expansa</i>	38.00 \pm 0.40	85.28 \pm 0.00	98.00 \pm 0.20

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Table 5: Ferric Reducing Antioxidant Power (FRAP) activity of aqueous leaf extracts (%)

Samples	20mg	40mg	80mg
<i>Morinda lucida</i>	70.10 \pm 0.30	82.30 \pm 0.10	97.50 \pm 0.00

<i>Solanum erianthum</i>	75.40±0.00	88.10±0.10	98.30±0.20
<i>Dryopteris expansa</i>	33.71±0.10	34.42±0.20	39.28±0.00

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Table 6: Anti-nutrient Analysis of the Samples Aqueous extracts

Samples	Cyanide content (mg/100g)	Oxalate content (mg/100g)	Phytate content (mg/100g)
<i>Morinda lucida</i>	5.50±0.01	0.33±0.01	0.22 ± 0.02
<i>Solanum erianthum</i>	6.89 ±0.01	0.99 ±0.01	0.22 ± 0.02
<i>Dryopteris expansa</i>	5.75 ±0.01	0.28 ±0.01	0.20 ± 0.01

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Table 7: Antibacterial effects of aqueous leaf extracts compared to standard antibiotic discs against Gram-positive bacteria.

Samples	Organisms	Plants	Antibiotics									
			PEF	CN	APX	Z	AM	R	CP	S	SXT	E
<i>Morinda lucida</i>	Staphylococcus aureus	S	S	S	S	R	R	S	S	S	S	S
	Streptococcus pyrogenes	S	R	S	R	R	R	R	R	R	R	R
<i>Solanum erianthum</i>	Staphylococcus aureus	R	S	S	S	R	R	S	S	S	S	S
	Streptococcus pyrogenes	R	R	S	R	R	R	R	R	R	R	R
<i>Dryopteris expansa</i>	Staphylococcus aureus	S	S	S	S	R	R	S	S	S	S	S
	Streptococcus pyrogenes	R	R	S	R	R	R	R	R	R	R	R

Antimicrobial activity of the extracts on the organisms: S= Susceptible/Sensitive; R= Resistant
PEF: Pefloxacin, CN: Gentamycin, APX: Ampliclox, Z: Zinnacef, AM: Amoxacillin,
R: Rocephin, CP: Cirpofloxan, S: Sterptomycin, SXT: Seprin, E: Erythromycin

Table 8: Antibacterial effects of aqueous leaf extracts compared to standard antibiotic discs against Gram-negative bacteria.

Samples	Organism	Plant	Antibiotics
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		s	PEF	CN	CH	AU	AM	OFX	CPX	SP	SXT	E
<i>Morinda lucida</i>	Escherichia coli	R	R	R	R	R	R	R	S	R	R	R
<i>Solanum erianthum</i>	Escherichia coli	R	R	R	R	R	R	R	S	R	R	R
<i>Dryopteris expansa</i>	Escherichia coli	R	R	R	R	R	R	R	S	R	R	R

Antimicrobial activity of the extracts on the organisms: S= Susceptible/Sensitive; R= Resistant
 PEF: Pefloxian, CN: Gentamycin, CH: Chloranphenicol, AU: Augmentin, AM: Amoxicillin
 OFX: Tarivid, CPX: Ciprofloxacin, SP: Sparfloxacin, SXT: Septrin, E: Erythromycin

Table 9: Anti-fungal effect of aqueous leaf extracts

Samples	Organisms	
	Aspegilluscandidius	Penicillium chrysogenum
<i>Morinda lucida</i>	R	R
<i>Solanum erianthum</i>	R	S
<i>Dryopteris expansa</i>	R	R

Antimicrobial activity of the extracts on the organisms: S= Susceptible/Sensitive; R=Resistant

Discussion

From Table 1.0, the phytochemical analysis revealed that while flavonoids, saponins, and reducing sugars are common across the studied plants, *Morinda lucida* and *Dryopteris expansa* exhibit higher levels of saponins and the ~~present~~ of alkaloids, suggesting potential unique medicinal properties. The medicinal effects of various plant extracts have been linked to their inherent phytochemicals. For example, alkaloids have always been known to exhibit diverse physiological effects in man and have contributed to the development of certain analgesics [33]. Similarly, Adelani-Akande linked their observed antimicrobial activity of watermelon seeds to the presence of saponins [34] while flavonoids have been found to exhibit an interesting array of biological activities such as antimicrobial, antioxidant, analgesic, anti-allergic, anti-inflammatory amongst others [35,36,37]. These tested phytochemicals are known to exhibit medicinal and physiological activities by strengthening the immune system, reducing inflammation, preventing DNA damage, and inhibiting cancer cell growth [38].

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Table 2.0 demonstrates that as the samples concentration increases, the total phenolic content also increases. This trend indicates that higher sample concentrations are associated with greater phenolic content and antioxidant power, whereas lower concentrations result in diminished phenolic content and antioxidant capacity. Since the higher the phenolic content of any plant, the more powerful antioxidant property it ~~contained~~, *Dryopteris expansa* exhibits the greatest antioxidant power, consistently showing the highest phenolic content across all concentrations. This is followed by *Solanum erianthum* and lastly

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Morinda lucida. Additionally, the phenolic content measured in this study for these three plants surpasses the levels reported by Song [39] for the antioxidant capacities of selected Chinese medicinal plants.

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From Table 3.0, the result is also concentration-dependent, indicating that as the samples concentration increases, the total flavonoids content also increases. This indicates that higher sample concentrations

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correspond to higher total flavonoid content and antioxidant power, while lower sample amounts result in lower total flavonoid content and antioxidant power. Since higher flavonoids content indicates a stronger

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antioxidant property, *Dryopteris expansa* exhibits the greatest antioxidant power, consistently showing the highest phenolic content across all concentrations. This is followed by *Morinda lucida* and lastly *Solanum*

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erianthum. The flavonoids contents in these plants were observed higher than what Di Carlo [40] reported in the antioxidant activity of some selected traditional Indian medicinal plants for *Cyperus*

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rotundus and *Vitex negundo*.

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Table 4.0 illustrates that as the concentration of samples increases, their antioxidant scavenging power also rises. This means that higher concentrations lead to greater antioxidant scavenging abilities, while

lower concentrations result in reduced scavenging power. Among the samples, *Morinda lucida* demonstrates the highest antioxidant scavenging ability, consistently achieving peak values of

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60.20±0.00 at 20 mg and 89.30±0.10 at 40 mg. This is followed by *Dryopteris expansa*, which shows the highest value of 98.00±0.20 at 60 mg, and then *Solanum erianthum*. Similar findings were reported by

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Miliauskas [41], who observed comparable radical scavenging activities in their study on medicinal plant extracts.

Table 5.0 demonstrates that as samples concentrations increases, so does the ferric-reducing antioxidant power. Higher values indicate greater ferric-reducing antioxidant power, while lower values signify lower

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reducing antioxidant power. Among the samples, *Solanum erianthum* consistently shows the highest ferric-reducing antioxidant power across all concentrations, followed by *Morinda lucida*, and then

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Dryopteris expansa. These findings are consistent with earlier studies, such as those by Koksai [42], which examined the antioxidant activity of *Melissa officinalis* leaves, and Song [39], which investigated

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the antioxidant capacities of selected Chinese medicinal plants. Overall, the antioxidant analysis results demonstrate that the aqueous extracts of these plants exhibit strong antioxidant activity. This suggests

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their effectiveness in neutralizing or scavenging free radicals, such as reactive nitrogen species (RNS)

and reactive oxygen species (ROS). These free radicals play a critical role in neurodegenerative diseases and oxidative stress by causing damage to biomolecules, including lipids, proteins, and DNA. Such damage can impact cell survival, inflammation, and stress responses [43,44].

Table 6.0 displays the cyanide content in *Morinda lucida*, *Dryopteris expansa*, and *Solanum erianthomas* 0.055 ± 0.01 mg/g, 0.057 ± 0.01 mg/g, and 0.068 ± 0.01 mg/g, respectively. All these values exceed the World Health Organization's acceptable limit for cyanide in medicinal plants [45], which is 0.01 mg/g. However, various plant processing methods, such as boiling, abrasion, and dehulling, have been shown to reduce cyanide levels significantly [46,47,48]. Consequently, the observed cyanide concentrations are unlikely to pose any harmful effects. In terms of oxalate content, *Dryopteris expansa*, *Morinda lucida*, and *Solanum erianthum* contain 0.28 ± 0.01 mg/100g, 0.33 ± 0.01 mg/100g, and 0.99 ± 0.01 mg/100g, respectively. These levels are considerably lower than the threshold of 50 mg/100g that defines high-oxalate plants, as reported by Judprasong [49]. Therefore, these plants are unlikely to form insoluble calcium salts that could lead to kidney stones, even if consumed in excess. The phytate levels in *Dryopteris expansa*, *Solanum erianthum*, and *Morinda lucida* are 0.20± 0.01 mg/100g, 0.20± 0.01 mg/100g, and 0.22± 0.01 mg/100g, respectively. These amounts are relatively low and should not significantly hinder the absorption of essential minerals such as iron, zinc, and calcium. Overall, the concentrations of cyanide, oxalate, and phytate in these plants are not high enough to affect nutrient bioavailability or inhibit growth.

Table 7.0 reveals that the aqueous extracts of *Morinda lucida* and *Dryopteris expansa* exhibit susceptibility to the Gram-positive bacterium *Staphylococcus aureus*. This bacterium is resistant to only two standard antibiotics, Ampiclox and Zinnacef, but is susceptible to the remaining eight antibiotics tested. Additionally, the aqueous extract of *Morinda lucida* demonstrates notable antibacterial activity against the Gram-negative bacterium *Streptococcus pyogenes*. This bacterium shows resistance to nine standard antibiotics (Pefloxacin, Ampiclox, Zinnacef, Amoxicillin, Rocephin, Ciprofloxacin, Streptomycin, Septrin, Erythromycin) and is susceptible to only one. The aqueous extract of *Solanum erianthum* did not show effectiveness against any of the Gram-positive bacteria tested. However, it may be effective against other Gram-positive bacteria. Its ethanol, methanol, and other solvent extracts could potentially exhibit enhanced antimicrobial activity. Alawode's research [50] supports this, indicating that hexane extracts of

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Solanum erianthum leaves and stems have significant activity against *S. aureus*, with MIC values as low as 1.25 mg/ml, demonstrating a broad spectrum of antibacterial and antifungal properties. Overall, *Morinda lucida* proved effective ~~ness~~ against two Gram-positive bacteria, while *Dryopteris expansa* was effective against one. These findings underscore their considerable antibacterial potential. This is consistent with Okwute's report [15] that the methanol extract of *Morinda lucida* exhibits high activity against *S. aureus*, *Bacillus subtilis*, *E. coli*, *Salmonella typhi*, *Klebsiella pneumoniae*, and *Pseudomonas aeruginosa*. Additionally, Femi-Adepoju reported [51] antimicrobial activity in ethanolic, methanolic, and acetone extracts of *Dryopteris filix-mas*, which belongs to the same family ~~with~~ *Dryopteris expansa*.

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Table 8.0 reveals that the aqueous extracts of *Morinda lucida*, *Dryopteris expansa*, and *Solanum erianthum* are resistant to the Gram-negative bacterium *Escherichia coli*. Additionally, *E. coli* shows resistance to all standard antibiotics tested ~~, with the exception of~~ Ciprofloxacin, which is effective against this bacterium. This resistance may be attributed to the structural components of the Gram-negative bacteria membrane [52]. Although the aqueous extracts show resistance to *E. coli*, alternative solvent extracts such as ethanol and methanol may offer improved antimicrobial activity. Okwute [15] reports that the methanol extract of *Morinda lucida* exhibits substantial activity against *E. coli*, *Salmonella typhi*, *Klebsiella pneumoniae*, and *Pseudomonas aeruginosa*. Moreover, further chemical modifications and synthesis could potentially enhance the therapeutic effectiveness of these plant extracts, thereby improving their efficacy against *E. coli* and other Gram-negative bacteria.

Comment [h65]: replace "with" with "as"

Table 9.0 presents the results of the antifungal activity of the aqueous extracts of *Morinda lucida*, *Dryopteris expansa*, and *Solanum erianthum* against *Aspergillus candidus* and *Penicillium chrysogenum*. The findings show that the fungi were resistant to the aqueous extracts of all the plants, except for *Solanum erianthum*, which was susceptible to *Penicillium chrysogenum*. This indicates that *Solanum erianthum* possesses notable antifungal activity. The results for *Morinda lucida* are consistent with those reported by Okwute [15], which found that *Morinda lucida* extracts had no activity against *Candida albicans* and *Aspergillus niger*, suggesting that *Morinda lucida* may lack antifungal properties. ~~I believe~~ further chemical modifications and synthesis of these plant extracts could potentially enhance their therapeutic effectiveness and improve their antifungal activity.

Comment [h66]: delete and replace with except for

Comment [h67]: rephrase this sentence

Conclusion

Conclusively, this study underscores the significant medicinal potential of *Morinda lucida*, *Dryopteris expansa*, and *Solanum erianthum*, based on their phytochemical profiles and biological activities. The analysis reveals high levels of flavonoids, saponins, and alkaloids, especially in *Morinda lucida* and *Dryopteris expansa*. While cyanide levels in all the studied plants exceed WHO limits, which however, they are not expected to pose a substantial risk when appropriate processing methods are applied. Overall, these findings highlight the therapeutic potential of these plants and suggest the need for further research to optimize their efficacy and safety.

Comment [h68]: insert

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during the writing or editing of this manuscripts.

Comment [h69]: you should be specific whether the article is written by an author or authors. Hence, rephrase the statement.

Comment [h70]: add

Comment [h71]: insert

Comment [h72]: insert

Comment [h73]: delete

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