A LEGACY OF LEADERSHIP: A SPECIAL ISSUE HONOURING THE TENURE OF OUR VICE CHANCELLOR, PROFESSOR ARMAYA'U HAMISU BICHI, OON, FASN, FFS, FNSAP



FUDMA Journal of Sciences (FJS) ISSN online: 2616-1370 ISSN print: 2645 - 2944 Vol. 9 April Special Issue, 2025, pp 336 - 342 DOI: https://doi.org/10.33003/fis-2025-09(AHBSI)-3755



PHYTOCHEMICAL PROFILING, ANTIOXIDANT POTENTIAL, AND LC-MS ANALYSIS OF BIOACTIVE COMPOUNDS IN *Ipomoea batatas (L.)* LAM *LEAVES*

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ABSTRACT

Leaves of *Ipomoea batatas* (sweet potato) are abundant sources of phytochemicals with possible therapeutic benefits. This study investigated the phytochemical profile, total phenolic content (TPC) and Total flavonoid content (TFC), antioxidant activity, and bioactive compounds in ethanol extracts of *I. batatas* leaves using qualitative and quantitative methods. The phytochemical screening revealed the presence of alkaloids, flavonoids, terpenoids, tannins, and glycosides in separate solvent fractions, namely methanol, hexane, dichloromethane, ethyl acetate, and aqueous. Quantitative analysis revealed the highest TPC (172.58 \pm 2.37 mg GAE/g) in aqueous fraction and the highest TFC (59.89 \pm 4.70 mg QE/g) in ethyl acetate fraction. DPPH assay showed a strong antioxidant effect using the ethyl acetate fraction, which was observed to have the lowest IC₅₀ (18.42 \pm 1.29 µg/mL), comparable to quercetin. LCMS analysis has identified up to 15 bioactive compounds such as luteolin, quercetin, kaempferol, and chlorogenic acid among others as likely contributors to the observed bioactivities. This foliage has the potential to act as a source of natural antioxidants and further support the traditional ethnomedicine use of *I. batatas* leaves. The extraction solvent also shows a differential effect on phytochemical yield and bioactivity, highlighting polar solvents as ideal for the extraction of phenolic and flavonoid phytochemicals.

Keywords: Ipomoea batatas, Phytochemicals, Antioxidant activity, LC-MS, Total phenolic content, Total flavonoid content

INTRODUCTION

Due to increased global demand, there is now a much broader scope of research into the myriad of medicinal plants that have been tested as sources of bioactive compounds and which have antioxidant and anti-inflammatory as well as antimicrobial properties (Azwanida 2015). A sample of such versatile crops among many is Ipomoea batatas (L.) Lam., commonly known as sweet potato. The leaves of this crop have been used in African and Asian ethnomedicine for treating health-related impediments like metabolic disorders, wounds, and infections (Truong et al., 2019). This notwithstanding, much documentation on the nutritional values of the tubers of sweet potatoes is in the records; however, very little to no studies are available on the awesome pharmacological potential of the leaves, which have to their credit an abundance of polyphenols, flavonoids, and alkaloids (Singh et al., 2017). Recent studies have put forward that the leaves of I. batatas have notable antioxidant activities attributed to their high phenolic contents, but systematic evaluations of solvent-extracted fractions and their bioactive constituents are somewhat rare (Jayaprakasha et al., 2001).

Free radicals, and oxidative stress blamed by these, form the basis of chronic diseases like diabetes, cancer, and neurodegeneration (Amarowicz et al., 2004). Because of the risks posed by toxicity, synthetic antioxidants like BHT and BHA were unearthed as safer alternatives, especially from plant sources (Keshari et al., 2016). The diverse phytochemical profile harbors *I. batatas* leaves to such an extent that such chemicals will be a potential remedy, but their efficacies depend on the extraction solvents of choice since polarity also plays a part in the yield of specific antioxidants (Umar et al., 2010). For instance, polar solvents (methanol,

water, etc.) are more selective in extracting phenolics while semi-polar solvents (ethyl acetate, etc.) are better in recovering flavonoids (Liu et al., 2008). A holistic analysis capturing these discrepancies is essential to perfect extraction protocols for nutraceutical applications. Recently, more advancements in liquid chromatography-mass spectrometry (LC-MS) have enabled the identification of bioactive plant molecules with great accuracy linking particular compounds in certain bioactivities performed (Sarian et al., 2017). Some of these compounds possibly identified in *I. batatas* include chlorogenic acid, quercetin, and kaempferol. (Eleazu et al., 2012).

Moreover, comparative studies on the antioxidant efficacy of different leaf extracts using standardized assays, such as DPPH, and FRAP, are scanty and at times give different opinions as regards the most productive solvent systems (Klepacka et al., 2011). This study comprehensively analyzes Ipomoea batatas (sweet potato) leaf extracts to unlock their phytochemical potential. Using ethanol extraction and sequential solvent partitioning (hexane, DCM, ethyl acetate, aqueous), the research profiles bioactive compounds quantifies total phenolic (TPC) and flavonoid (TFC) content and evaluates antioxidant activity via DPPH scavenging (with IC50 values for each fraction). Advanced LC-MS analysis identifies key antioxidant contributors, bridging traditional phytochemical methods with modern techniques. By highlighting the leaves' rich bioactive properties, this work lays the foundation for their future use in functional foods and herbal medicines. The findings also open sustainable avenues for agriculture and pharmaceuticals, promoting underutilized plant parts as viable sources of health-promoting compounds.

MATERIALS AND METHODS Extraction of the plant sample

The *Ipomoea batatas* leaves were obtained from the Dutsin-Ma local government area of Katsina State, Nigeria, at the irrigation area in January, 2024. It was identified at the Department of Biological Sciences at the Federal University of Dutsin-Ma Katsina. Thereafter, the leaves were ground with a pestle and mortar. The powdered samples were maintained at ambient temperature in clean, airtight containers until they were needed. Using ethanol, 1 Kg of the powdered leaf was extracted for 7 days. The solvent was collected by a rotary evaporator at the end of the period. After that, the extracts were placed in a desiccator and allowed to dry and other subsequent experiments.

Phytochemical screening

The phytoconstituents in the plant extracts were detected using conventional qualitative techniques (Bhandari et al., 2021; Gul et al., 2017; Iqbal et al., 2015). These are as described below:

Detection of alkaloids

After adding a small amount (2 mL) of the extract to a few milliliters of diluted HCl, the mixture was filtered. To find out if there were any alkaloids in the filtrate, the steps listed below were carefully followed.

Mayer's test

A brown or orange-colored precipitate was seen when two drops of potassium mercuric iodide (Mayer's reagent) solution were added to a small quantity (2 mL) of the extract solution from the test tube's side.

Wagner's test

After adding Wagner's reagent (iodine potassium iodide solution) to 2 mL of ethanolic extract, the presence of a reddish-brown precipitate was a positive test.

Dragendorff's test

A reddish-brown or orange-red precipitate formed when 2 mL of extract solution is added to a few drops of potassium bismuth iodide solution (Dragendorff's reagent).

Detection of flavonoids

Lead acetate test

The presence of flavonoids was indicated by the yellow precipitation that appeared when 2 ml of lead acetate solution was added to the ethanolic extract solution.

Ferric chloride test

A few drops of FeCl₃ solution were added to the extract's alcoholic solution. The presence of flavonoids was indicated by the formation of a greenish solution.

Alkaline test

By adding roughly 2 milliliters of 10% NH4OH to the ethanolic extract solution and observing for the appearance of a yellow tint, the presence of flavonoids was verified.

Detection of tannins Ferric chloride test

Two milliliters of the extract and ten percent alcoholic FeCl₃ solution were mixed equally; the presence of tannins was indicated by the mixture's dark blue or greenish-grey color. *Gelatin test*

A white precipitate formed after the extract was treated with 10% NaCl and 1% gelatin solution, was a positive test for the presence of tannins.

Detection of steroids Salkowski test

After shaking each extract separately with chloroform and adding concentrated H_2SO_4 , a reddish-brown hue developed, suggesting the presence of steroids.

Liebermann-Burchard test

A test tube containing 3 milliliters of extracts, 2 milliliters of chloroform, and a few drops of acetic anhydride was heated in a water bath and quickly cooled in cold water. There was a brown ring where the two liquids met when conc. H_2SO_4 was mixed with the test tube wall. The top layer turned green to indicate the presence of steroids, and a dark red tint formed to show the presence of terpenoids.

Test for Phenol Ferric chloride test

The extracts (2 ml) were combined with 10% FeCl₃ solution and purified water. The presence of phenolic compounds was indicated by the emergence of a dark green coloring.

Fehling test

A small quantity of crude extracts was added after Fehling Solution 1 and Fehling Solution 2 were combined in equal parts (2 mL each). After a little more heating, the test tube's bottom turned brick-red, indicating that carbs had successfully entered the mixture.

Benedict's test

A very tiny amount (1 mL) of the extract solution was combined with the Benedict reagent, and the mixture was heated in the water bath for two minutes. Brick-red precipitate appeared as sugar was reduced.

Keller-Killiani test

Glacial acetic acid (1 ml) and FeCl₃ (1 ml) were mixed with the extract- containing aqueous solution (2 ml). When a few drops of H_2SO_4 were poured from the test tube's side, a brown ring formed at the intersection of the two liquids, signifying the presence of cardiac glycoside.

Bromine water test

When test solutions were dissolved in bromine water, a yellow precipitate formed, this indicates a positive glycoside test.

Determination of Total Phenolic Contents (TPC)

The method of (Singleton et al., 1999) was used to determine the extract's total phenol content. 2.5 ml of 10% Folin-Ciocalteau's reagent and 2 ml of 7.5% sodium carbonate were combined with 0.2 ml of the extract. Garlic acid was used as the standard phenol after the reaction mixture was incubated at 45 $^{\rm O}$ C for 40 mins, and the absorbance was measured at 700 nm in the spectrophotometer.

Preparation of reagents

Folin-Ciocaltau reagent (10 ml) (1:10 v/v) was produced after 5.29 g of sodium carbonate was dissolved in 50 mL of distilled water to achieve 1 M.

Preparation of standard gallic acid solution

Ethanol (10 ml) and 5 mg of gallic acid were combined to make the standard stock solution (500 μ g/mL). To make solutions at different concentrations (10, 20, 30, 40, 50, 60, 70, and 80 μ g/mL), the mixture was then diluted with distilled water.

The stock solutions of each extract (50 mg/mL) were diluted ten times in 50% solvent to make test solutions of plant extracts (500 μ g/mL).

Procedure

With a small modification, using 96-well plate methods, the extracts' TPC was evaluated using the Folin Ciocalteu reagent (FCR) (Ainsworth and Gillespie, 2007; Liu et al., 2008). To summarize, a 96-well plate was filled with three copies of each extract (20 $\mu L;~500~\mu g/mL)$ and standard gallic acid solutions (10, 20, 30, 40, 50, 60, 70, and 80 µg/mL). Subsequently, 100 µL of 1:10 v/v FCR prepared in distilled water was added to the spectrophotometer to acquire the first findings. Subsequently, 80 microliters of 1 M sodium bicarbonate were added one by one to every well containing the standard gallic acid and plant extract solution. The final absorbance was then measured at 700 nm when this mixture had been left at room temperature in the dark for fifteen minutes, using a microplate reader (Synergy LX, BioTek, Instruments, Inc., USA). Using a standard calibration curve, the concentration of all polyphenolic compounds in the extracts was determined. The results were expressed as milligrams of gallic acid equivalent per gram of dry mass (mgGAE/g) of the extract. Gallic acid absorbance was utilized to create a standard calibration curve (10-80 µg/mL).

Determination of Total Flavonoid Contents (TFC)

The extract's total flavonoid content was ascertained by the use of a colorimeter assay created by Bao (2005). At zero time, 0.2 ml of the extract was mixed with 0.3 ml of 5% NaNO₃. 0.6 ml of 10% AlCl₃ were added after 5 mins, and after 6 mins, the mixture received 2 ml of 1M NaOH and 2.1 milliliters of distilled water. Flavonoid concentration was represented as milligrams of rutin equivalent after absorbance was measured at 510 nm in comparison to the reagent blank.

Preparation of reagents

One gram of AlCl3 was dissolved in ten milliliters of distilled water to yield 10% aluminum trichloride, and 0.98 grams of potassium acetate were dissolved in the same amount of water to yield 1 M potassium acetate.

Preparation of standard guercetin solution

One milligram of quercetin was dissolved in ten milliliters of methanol to create a stock solution (0.1 mg/mL). Subsequently, the standard solution was prepared at various concentrations of 10, 20, 30, 40, 50, 60, 70, and 80 µg/mL by diluting the 0.1 mg/mL stock solution.

Preparation of plant extracts

Each plant extract solution was diluted to 500 µg/mL in a 50% DMSO solution starting from an initial solution of 50 mg/mL.

Procedure

Using a 96-well plate reader and the AlCl₃ colorimetric approach (Chang et al., 2002), the TFC of each extract was measured. The stock solution (100 µg/mL) was diluted with distilled water to yield 130 µL of various concentrations of standard quercetin solutions (10, 20, 30, 40, 50, 60, 70, and 80 µg/mL), which were then loaded in triplicate onto 96-well plates. Subsequently, a 96-well plate was filled in triplicate with 20µL (500µg/mL) of each plant sample. Adding 110 µL of distilled water, the total volume of each well containing a plant sample was 130 μ L. Then, 60 μ L of ethanol, 5 μ L of AlCl₃, and 5 µL of potassium acetate were added separately to each well that held the plant sample and standard quercetin. After 30 minutes in the dark, this combination was then tested for absorbance at 415 nm using a 96-well plate reader. United States, BioTek Instruments Inc., Epoch 2. The quercetin normal calibration curve (10-80 μ g/mL) is used to measure the total amount of flavonoids, which is expressed as milligrams of quercetin equivalent per gram of dry extract mass (mgGAE/g).

Determination of total carotenoid content (TCC)

Weigh 2.5 g of finely blended sample into a conical flask, 30 ml of hexane, 20 ml of ethanol, and 2 ml of 2% NaCl were added. Mix very well and transfer content into a separating funnel, allowing it to stand for about 10 mins to allow for the extraction of carotenoid run the lower content off and collect the upper layer (extractant phase). Measure the absorbance (a)436 nm using a UV-visible spectrophotometer.

Statistical analysis

The first analysis and data collection were conducted using Microsoft Excel. The standard error of the mean, shown as \pm mean, was used to depict the experimental data. In addition, the molecular formulae and structures of compounds were found and assigned using additional databases such as ChemDraw, PubChem, Dictionary of Natural Products, and ChemSpider.

RESULTS AND DISCUSSION Phytochemical Analysis

The results of the phytochemical study verified that the plant extracts contained a wide range of chemicals, including phenols, carbohydrates, steroids, alkaloids, flavonoids, terpenoids, saponins, tannins, and essential oils. Phytochemicals such as phenolics, flavonoids, carbohydrates, terpenoids, tannins, and glycosides were present in all plant extracts. The n-hexane fraction of the plants was the only one containing steroids. anthraquinones, tannins, and saponins were not present in all plant DCM fractions.

Table 1: Phytochemical Screening for the ethanol extract of the leaves of <i>I. batatas</i>

Dhada ah and a la	Leaves of Ipomoea batatas						
Phytochemicals	Performed test	MeOH	n- Hexane	DCM	EA		
Alkaloids	Dragendorff's	+	+		+		
Flavonoids	Alkaline reagent	+	+	+	+		
Phenols	Ferric chloride	+	+	+	+		
Steroids	Salkowski's	_	+	_	_		
Terpenoids	Liebermann Burchard's	+	+	+	+		
Tannins	Gelatin test	+	+	_	+		
Glycosides	Keller-Kiliani's	+	+	+	+		
Saponins	Foam	+	+	_	+		
Carbohydrates	Fehlings test	+	+	+	+		
Anthraquinones	Anthraquinones	+	+		+		

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The basic phytochemicals flavonoids and alkaloids were found in almost all plants. They are the strongest scavengers of free radicals and antioxidants, protecting cells from oxidative damage. Furthermore, their properties include antibacterial, anti-inflammatory, anti-tumor, and antidiabetic effects, which are therapeutically significant (Eleazu *et al.*, 2012; Roy, 2017). Antiviral, antimicrobial, antiinflammatory, anti-cholesterol synthesis, anti-malarial, and anti-inflammatory qualities are just a few of the noteworthy pharmacological effects that terpenoids exhibit (Agidew, 2022).

Quantitative phytochemical analysis Determination of Total Phenolic Contents

A Gallic acid calibration curve (10 μ g/mL to 100 μ g/mL) was generated, and the findings were computed using the regression equation (y = 0.0271x, R² = 0.9655).

Table 2: Total Phenolic Content values for various plant extracts

Madiainal Dlant		Total Phenolic Content (mg QE/g)					
Medicinal Flant	MeOH	Hexane	DCM	EA	Aqueous		
Ipomoea Batatas	155.83±1.51	128.24±1.22	87.86±2.45	168.24±1.17	172.58±2.37		
The total phenolic	content values wer	e represented as	milligrams of gallic	acid equivalent per	gram mass of dry extract		
(mg GAE/gm).							

For plant extracts extracted in methanolic and other solvents, the TPC (mean \pm standard error) varies from 172.58 \pm 2.37 to 41.45 ± 2.6 mg GAE/gm dry weight. Out of all the extracts, the Ipomoea batatas aqueous fraction had the highest TPC value (172.58 \pm 2.37 mg GAE/gm), while the DCM fraction had the lowest (87.86 \pm 2.45 mg GAE/gm). The results showed that the solvents significantly affected the polyphenol content. Polar solvent fractions contained more phenolics than non-polar solvent fractions did. According to (Jayaprakasha et al., 2001), methanol and ethanol were the most effective solvents tested and produced the maximum amount of total phenolics discovered from Ipomoea batatas leaves. In a different study, the TPC values for the methanolic extracts of I. batatas (128.3 \pm 0.47) were discovered. Prior research by Singh et al. (2017) revealed that the TPC of I. batatas was $375.9 \pm 17.5 \text{ mg GAE/g}$ from the methanolic fraction and 249.7 \pm 1.3 mg GAE/g from the EA fraction. Contrary to this, our study showed 163.54 ± 1.31 mg GAE/g of TPC from methanol and 168.24 ± 1.17 mg GAE/g of TPC from EA fraction. Variations in the amount of bioactive phytochemicals can also be caused by genetic factors, storage conditions, harvesting time, type (wild or farmed), solvent type, extraction process, and climate (Klepacka *et al.*, 2011; Zhao *et al.*, 2015). Klepacka and associates, 2011; Liu and associates, 2008. The presence of polar phenolic compounds was suggested by the increased TPC values in the extracts prepared with aqueous solvent and EA. Because of their combined action, methanolic extracts contained several phenolic compounds with high total phenolic content values.

Determination of Total Flavonoid Content (TFC)

Table 3 displays the TFC values for the different plant extract fractions. Using a calibration curve of standard quercetin (y = 0.024x + 0.004, R² = 0.971), total flavonoid content (TFC) was determined and expressed as quercetin equivalent (mg QE/gm dry mass of the extract). There is a range of 6.52 ± 0.53 mg to 7.84 ± 3.62 mg QE/gm dry mass in the TFC (mean \pm standard error). *Ipomoea batatas*'s EA extract had the greatest QE/gm TFC value at 59.89 ± 4.70 mg, while the DCM extract had the lowest flavonoid concentrations were those with DCM fractions.

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Madiainal Dlant		Tota	al flavonoid Content	(mg QE/g)	
Medicinal Plant	MeOH	Hexane	DCM	EA	Aqueous
Ipomoea Batatas	47.26 ± 1.21	26.25±1.63	11.28±0.10	28.49±0.67	19.60±3.10

Our findings showed that the crude methanolic extract had a high TFC concentration, while the EA fractions had the maximum flavonoid content. According to a study, semipolar solvents like acetone and EA, as well as more polar solvents like methanol, are effective in extracting most flavonoids and phenolic compounds during the extraction and fractionation processes (Umar et al., 2010). Other variables that may affect the TFC in plant extracts include the concentration and type of solvent and flavonoids, pH, extraction temperature and duration, pressure, material sizes, and plant harvesting time (Juntachote et al., 2006). Due to the combined effects of the different elements in the crude methanolic extract, the crude methanolic fraction of Ipomoea batatas exhibited the highest TFC value. In contrast to our investigation's 47.26 ± 1.21 mg QE/g from methanol and 28.49 ± 0.67 mg QE/g from EA fractions, a previous study on Ipomoea batatas found TFC of $89.9 \pm 0.1 \text{ mg QE/g}$ from methanol and 208.4 \pm 0.6 mg QE/g from EA (Singh et al., 2017).

DPPH assay for antioxidant activity

Using the DPPH test, the antioxidant capacities of methanolic extracts and different solvent fractions were evaluated. The

percentage of scavenging activity for each fraction was computed before calculating the IC₅₀, or the lowest concentration of the antioxidant required to scavenge 50% of the DPPH. A standard quercetin with an IC $_{50}$ value of 2.86 \pm 0.51 µg/mL was used to compare the results. EA, crude, and aqueous fractions demonstrated high radical scavenging efficacy as compared to normal quercetin. Additionally, the findings showed that Ipomoea batatas had moderate to strong IC50 values, ranging from $11.98 \pm 0.36 \ \mu\text{g/mL}$ in EA to $141.53 \pm 10.73 \ \mu g/mL$ in the hexane fraction (Table 4). The extract's phenolic and flavonoid molecules significantly impact the extract's capacity to scavenge free radicals, which is dependent on the quantity and positioning of hydroxyl groups that supply hydrogen atoms or electrons to the free radicals (Amarowicz et al., 2004). Both flavonoids and phenolics significantly contribute to antioxidant activity because of their redox properties, which include acting as reducing agents or donors of hydrogen atoms (Keshari et al., 2016). With somewhat little activity, the DCM fractions were ascertained. Table 4 presents the tabulated IC50 values of crude and different solvent extracts, quercetin was employed as positive standard.

Table 4: Antioxidant activity of various fractions of *I. batatas*

Madiainal Dlant	DPPH radical scavenging (IC ₅₀ values in µg/mL)					
Medicinal Plant	MeOH	Hexane	DCM	EA	Aqueous	
Ipomoea batatas	52.60±3.63	60.24±2.19	108.20±2.73	18.42±1.29	16.99 ± 2.56	

We found that Ipomoea batatas exhibited antioxidant activities of 52.60 \pm 3.63 µg/mL (methanol), 60.24 \pm 2.19 $\mu g/mL$ (hexane), and 108.20 \pm 2.27 $\mu g/mL$ (DCM), 18.42 \pm 1.29 µg/mL (EA), based on IC50 values from our study (Tunna et al., 2015). The antioxidant activity of Ipomoea batatas methanolic extract was reported to be 26.5 ± 1.1 , 23.7 ± 0.4 , and $13.2 \pm 0.1 \ \mu g/mL$ in our earlier work (Sapkota et al., 2021). It has been discovered that the quantity, location, and arrangement of hydroxyl groups within the structures of flavonoids determine their antioxidant action. The majority of the flavonoids in our investigation had the catechol moiety, which has several -OH groups in its structure and is essential for strong antioxidant activity, in the EA fraction of the plant extracts. Antioxidant activity was found to be increased by hydroxylations in rings A and B, especially in the 4' and 5' locations, and decreased by methylation and acetylation of phenolic groups at positions C-5, C-7, and C-8 (Sarian et al., 2017). The EA fraction $(18.42 \pm 1.29 \ \mu g/mL)$ in our study on

LC-MS Analysis

Ipomoea batatas shown the highest DPPH scavenging activity, with the crude (22.36 \pm 1.95 $\mu g/mL)$ and aqueous $(22.34 \pm 2.71 \ \mu g/mL)$ fractions exhibiting nearly identical activity. After conducting our investigation, we discovered that the IC50 values for EA and aqueous extracts were 12.6 μ g/mL and 142.6 \pm 5.3 μ g/mL, respectively (Liu et al., 2008). Ipomoea batatas has antioxidant potential since it contains phenolic components. (Kumar and others, 2006). These extracts have larger amounts of phytochemicals that are responsible for the DPPH scavenging activity, as observed by the lower IC50 values and increased phenolic content. The presence of polar antioxidant molecules in the EA fractions of the plant extract was likely the cause of the higher antioxidant activity in these fractions. Because of the compounds' synergistic effects, a wide spectrum of chemicals are extracted by the methanolic solvent and exhibit increased scavenging action in the crude (methanolic) extract.



Figure 1: spectrum for Ipomoea Batatas

The detected compounds are listed in Table 5 below, which also includes details about their predicted mass, molecular formula, and fragments. The measured mass spectrum was used to identify the compounds, and the outcomes were assessed by contrasting them with information found in published works.

Table 5: Secondary metabolites detected in Ipomoea Batatas using LC-MS

S/N	Identified Compounds	Molecular formula	Calculated mass (g/mol)	Fragmentation
1.	Luteolin	$C_{15}H_{10}O_{6}$	286.24	
2.	Zeaxanthin	$C_{40}H_{56}O_2$	568.9	127.03 [M+H-CO ₂] +
3.	Coumestrol	$C_{15}H_8O_5$	268.22	313.07 [M+Na] +, and 139.03
4.	Quercetin	$C_{15}H_{10}O_7$	302.23	313.07 [M+Na] +, and 139.03
5.	Kaempferol	$C_{15}H_{10}O_{6}$	286.24	273.12, 257.13
6.	2-(3,4-dihydroxyphenyl) chroman - 3, 5, 7-triol	$C_{15}H_{14}O_{6}$	290.27	259.13, 165.09, 153.12
7.	Naringenin	$C_{15}H_{12}O_5$	272.25	303.21, 274.20,153.12
8.	Cyanidin		287.24	427.08, 169.07,
9.	Ellagic acid	$C_{14}H_6O_8$	302.19	181.05; 153.01
10	Chlorogenic acid	$C_{16}H_{18}O_9$	354.31	170.97; 127.03
11	Scopoletin	$C_{10}H_8O_4$	192.17	193.02

12	Apigenin	$C_{15}H_{10}O_5$	270.24	298, 288, 192, 191
13	Folic acid	$C_{19}H_{19}N_7O_6$	441.4	285; 163; 267; 159; 239
14	Umbelliferone	C9H6O3	162.14	
15	Ferulic acid	$C_{10}H_{10}O_4$	194.18	

Discussion

The qualitative screening indicated a rich presence of different kinds of phytochemicals such as alkaloids, flavonoids, terpenoids, tannins, and glycosides, in the leaf extracts of I. batatas. The absence of steroids in methanol and DCM fractions but their presence in hexane suggests that they are, by character, nonpolar, this is similar to the findings of Agidew (2022), who reported a similar trend in solubility in plant extractions. Flavonoids and phenolics are abundant in polar solvents (methanol, ethyl acetate, and aqueous), this also agrees with report of Jayaprakasha et al., (2001) who reported the efficiency of methanol and ethyl acetate as solvents for the extraction of hydroxyl-rich antioxidants. The highest TPC value was recorded in the aqueous fraction $(172.58 \pm 2.37 \text{ mg GAE/g})$, while the EA fraction showed the highest flavonoid content (59.89 \pm 4.70 mg QE/g), which is comparable to Singh et al. (2017) who reported 375.9 ± 17.5 mg GAE/g TPC in methanolic extracts of I. batatas. Discrepancies in TPC and TFC in differing studies can be attributed to the difference in regions of plant sources, extraction methods, and the environments lepacka et al. (2001). The highest antioxidant activity by EA fraction, as proven by the DPPH radical scavenging assay (IC50 = 18.42) \pm 1.29 µg/mL), was better than that of the crude methanolic extract (52.60 \pm 3.63 µg/mL). This was consistent with Sarian et al. (2017), reporting that hydroxylated flavonoids having catechol moieties such as quercetin and luteolin exhibit strong radical-scavenging activities due to the capacity to donate hydrogens. Such an observation is further substantiated by the lower IC50 values of polar fractions, suggesting an enhancement of antioxidant potential owing to the hydroxylation pattern in ring B of flavonoids as previously studied by Amarowicz et al. (2004). The strong correlation is indicative of TPC/TFC being a contributor to free radical neutralization by phenolic compounds (Keshari et al., 2016). The moderate activity observed in hexane and DCM fractions indicates that nonpolar antioxidants (Carotenoids, terpenoids) may play a minor role in the antioxidant activity.

The 15 compounds detected by LC-MS analysis include luteolin, quercetin, kaempferol, and chlorogenic acid, and all are associated with various pharmacological importance. Quercetin and kaempferol are very abundant compounds and are well documented for their anti-inflammatory and anticancer effects (Roy, 2017). The presence of chlorogenic acid, a strong antioxidant, further supports the claim for the therapeutic value of the plant, which has been linked to antidiabetic and neuroprotective activities (Truong et al., 2019). The presence of ellagic acid and coumestrol suggests other pharmacological activities namely, the protection of DNA and estrogenic activity, respectively (Eleazu et al., 2012). These observations correspond with the report of Liu et al. (2008) indicating similar compounds found in I. batatas leaves and suggesting a synergistic interaction for their health benefits. The TPC showed lower values compared with those of Singh et al. (2017) and the later's TFC results were similar, indicating various methodological and ecological influences. Higher antioxidant activities shown in EA fractions according to our study stand in contrast with research done by Umar et al. (2010), which showed methanol extracts to be more potent, probably due to variations in solubility of the phytocompounds. The aforementioned results indicate that I.

batatas leaves offer a viable source of nutraceuticals and this in support of its traditional application for wound healing and management of metabolic disorders (Azwanida, 2015).

CONCLUSION

This research has shown that the leaves of *I. batatas* are endowed with significant levels of phenolics, flavonoids, and other bioactive compounds with a considerable amount of antioxidant potential. The ethyl acetate and the aqueous fractions depict the highest bioactivity, further proving the role of solvent polarity in the extraction of phytochemicals. The profiling by LC-MS of various compounds responsible for these effects provided further support for the plant's ethnomedicinal potential. Thus, this work gives a good reason for incorporating *I. batatas* leaves into the list of functional foods and herbal formulations.

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