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Original Article

Phytochemical Analysis of *Ficus saussureana DC., Clerodendrum rotundifolium* Oliv., and *Microglossa pyrifolia* (Lam.) O. Ktze as Important Antidiabetic and Antihypertensive Plants

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ABSTRACT

Phytochemicals or secondary metabolites are key constituents of plants that play a major role in treating and alleviating human ailments. They possess numerous therapeutic and physiological properties, including disease prevention and treatment, symptom management, and the promotion of physical and mental well-being. Although 80% of the global population relies on plants for medicinal purposes, and analysing the phytochemical components of traditionally used plants is crucial for understanding their potential to treat specific diseases, many of these plants remain unresearched. This study assessed the phytochemical composition of *Ficus saussureana* DC., *Clerodendrum rotundifolium* Oliv., and *Microglossa pyrifolia* (Lam.) O. Ktze qualitatively and quantitatively via standard methods. The gas chromatography-mass spectrometry (GC-MS) method was used to detect general compounds in the methanol extracts. The results indicated the presence of most of the analysed compounds, such as alkaloids, steroids, phenols, tannins, flavonoids, coumarins, and terpenoids. The highest amounts of polyphenols (217±25.05 mg/g) and tannins (179.75±3.44 mg/g) were detected in the *Ficus saussureana* methanolic extract, whereas the highest amounts of flavonoids (28.75±0.98 mg/g), saponins (225.07±4.11 mg/g), and alkaloids (116.15±3.73 mg/g) were detected in the *Clerodendrum rotundifolium* methanolic extract. GC-MS profiles revealed 14 compounds in *Ficus saussureana* and 30 compounds in *Clerodendrum rotundifolium* and *Microglossa pyrifolia*. Several of the identified compounds demonstrate pharmacological activities relevant to the treatment of diabetes, hypertension, and various other human ailments. This research validates the assertions of traditional herbalists concerning the three selected plants, uncovering compounds with potential antidiabetic and antihypertensive properties. These findings not only support their traditional use but also highlight their potential for future drug discovery.

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INTRODUCTION

The use of plants to treat human ailments is deeply intertwined with the history of human existence. Since ancient times, when no alternative curative options have been available, plants have played a pivotal role in healthcare. This reliance on plants did not diminish with the advent of industrially manufactured drugs; rather, it remains significant in the modern era. Today, traditional medicine accounts for the healthcare needs of approximately 80% of the world's population, reflecting a substantial rise in use and demand. This enduring reliance is not solely due to poverty or the inability to afford expensive modern drugs. Traditional medicine is often more culturally acceptable and fulfils psychological needs in ways that modern medicine cannot (Agidew, 2022). Interestingly, even modern pharmaceuticals owe much to plant-derived compounds, with at least 25% of developed drugs originating from plants. Consequently, plant secondary metabolites are indispensable for human health and serve as the foundation for many pharmaceutical drugs (Yeshi et al., 2022). The enduring significance of plant-based medicine, both in traditional healing practices and modern pharmaceutical development, underscores the invaluable role of plant secondary metabolites in advancing human health.

Phytochemicals are essential bioactive compounds that contribute to plant survival by facilitating adaptation to environmental challenges and stressors. These active biomolecules are synthesised by specialised plant tissues and organs in response to altered environmental conditions to perform specific roles (Ahad et al., 2021). Even though they are not directly involved in growth (Kumar et al., 2024), these small organic compounds are crucial for various physiological and biochemical functions that contribute to plant fitness and survival, especially their interactions with the environment and adaptation to biotic and abiotic stressors (Yeshi et al., 2022). These roles include defending plants against herbivores and microbial infections, attracting pollinators and seed-dispersing animals, acting as allelopathic agents, providing UV protection, and serving as signalling molecules in the formation of nitrogen-fixing root nodules in legumes (Crozier et al., 2006). In addition to their ecological roles, these specialised biomolecules play crucial roles in plant defence, reproduction, and symbiotic interactions, highlighting their

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importance in both plant biology and broader ecological systems.

In addition to their essential roles in plants, secondary metabolites offer significant therapeutic benefits that contribute to human health and disease management. Scientific research has revealed that secondary metabolites are a common feature of all therapeutic agents. They possess numerous therapeutic and physiological properties, including disease prevention and treatment, symptom management, and the promotion of physical and mental well-being (Ahad et al., 2021). Notably, these compounds have been shown to reduce the risk of chronic or noncommunicable diseases (NCDs) (Liu, 2004; Heneman & Zidenberg-Cherr, 2008; Siyuan et al., 2018). Owing to their ability to prevent chronic diseases, manage their complications, and promote overall well-being, secondary metabolites are indispensable in both traditional medicine and modern strategies for managing noncommunicable diseases.

Despite these benefits and considering that analysing the phytochemical components of traditionally used plants is crucial for understanding their potential to treat specific diseases, many plants used in traditional medicine have not been analysed for their chemical compositions. Additionally, the potential of known medicinal plants is being diminished because of various natural and human-induced factors. This highlights the urgent need to accelerate the exploration and analysis of secondary metabolites from diverse plant sources. Documenting these compounds in lesser-known, unexplored, or newly discovered plant species is therefore essential (Kumar et al., 2024). The analysis of secondary metabolites could lead to the unearthing of novel therapeutic compounds for treating several diseases.

Ficus saussureana DC., *Clerodendrum rotundifolium* Oliv., and *Microglossa pyrifolia* (Lam.) O. Ktze are medicinal plants from the families Moraceae, Verbenaceae, and Asteraceae, respectively. These plants are important in Ugandan traditional medicine and are known for their use in treating diabetes and hypertension (Adia et al., 2016; Gang et al., 2023; Nnko et al., 2024). In addition to their antidiabetic and antihypertensive properties, they offer a range of other therapeutic benefits. *Ficus saussureana* is used to treat fallopian tube blockage, HIV/AIDS, male infertility, syphilis, typhoid fever, and ulcers (Gang et al., 2023). *Clerodendrum rotundifolium* is employed for malaria, intestinal parasites, stomach aches, deworming, and labour induction during childbirth. *Microglossa pyrifolia* is used to manage malaria, abdominal disorders, convulsions, skin allergies, syphilis, cough, and chest pain (Adia et al., 2016). Despite their recognised medicinal importance in Uganda and beyond, there is limited knowledge about their phytochemical compositions. Given their pharmacological importance and the need to understand their novel mechanisms of action (Priyadarshini et al., 2017), scientific investigations of these plants are crucial. Therefore, the objective of this study was to investigate the phytochemical compounds present in these three medicinal plants, with a focus on their roles as antidiabetic and antihypertensive agents. This research not only offers promising advantages but also serves as a key source of inspiration and models for synthesising new drugs with improved therapeutic, chemical, or physical properties (Priyadarshini et al., 2017). By exploring the phytochemical compositions of these medicinal plants, this study contributes valuable insights into their potential therapeutic applications. These findings will not only enhance the scientific understanding of their bioactive compounds but also pave the way for the development of novel, more effective treatments for diabetes and hypertension.

MATERIALS AND METHODS

Study Area

This study was conducted in Buwama Sub-County, which is located in Mpigi District, Central Uganda. Buwama sub-county comprises 10 parishes and 62 villages. Geographically, it is situated at a latitude

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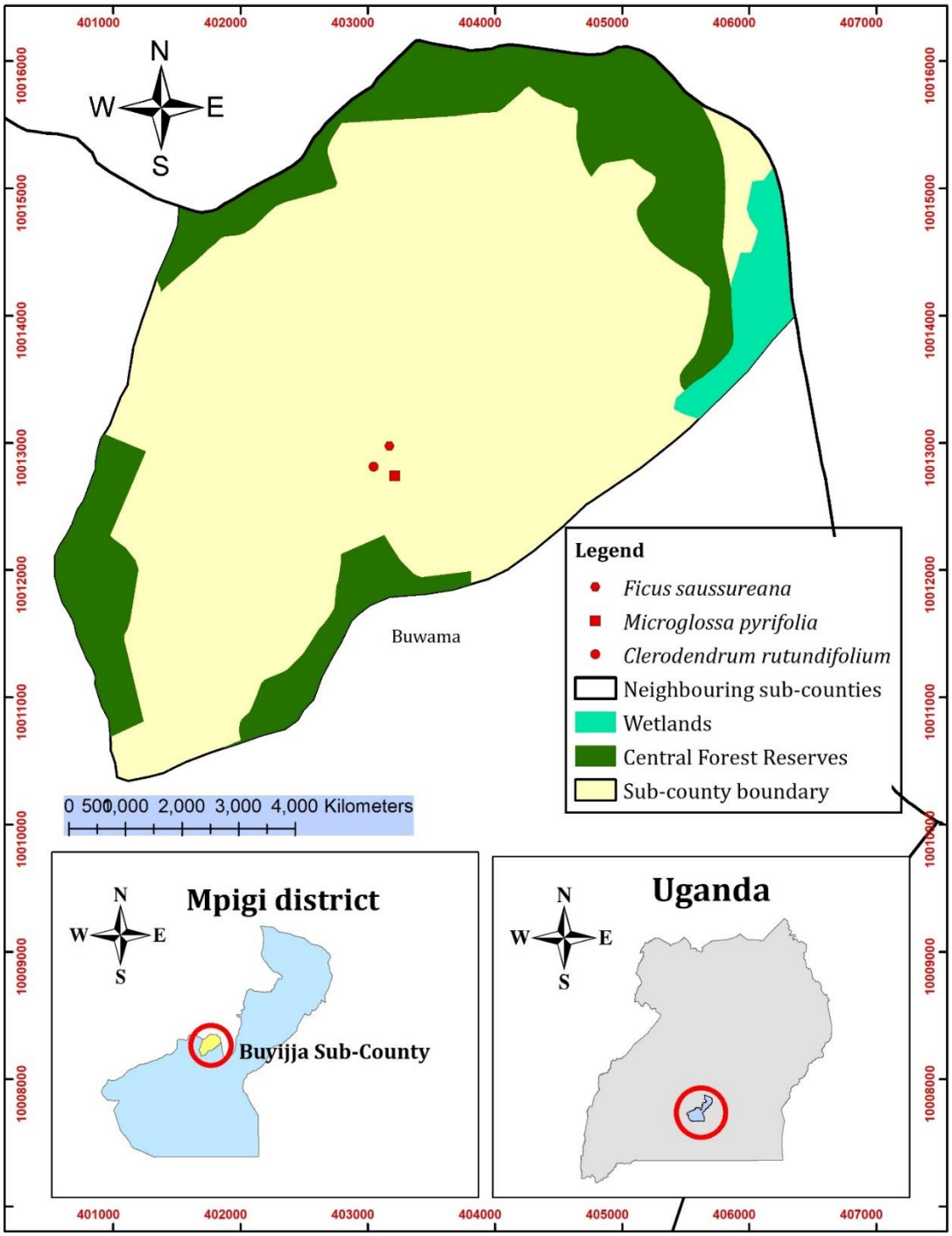
of 0°03′39.0″ N and a longitude of 32°05′47.0″ E. The subcounty is approximately 32 kilometres southwest of the district headquarters and approximately 71 kilometres southwest of Kampala, the capital city of Uganda. Predominantly rural, Buwama experiences an average temperature ranging from 22.5°C to 27°C, with an annual average rainfall of 1,320 mm.

Figure 1: Buwama Subcounty, Mpigi District, where the Collections were Performed



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Plant Sample Collection, Cleaning, and Drying

The plants selected for phytochemical analysis were collected from their natural habitats in Buyijja-Buwama. After collection, the plant samples were washed immediately with water to avoid the deterioration of phytochemicals (Banu & Cathrine, 2015). The samples were subsequently dried to remove water content from the plants, which was performed immediately after the samples were collected to avoid spoilage. They were dried under the shade for three to four weeks. The samples were then diced, crushed, and powdered via an electrical grinder, and the finished dried powdered samples were kept in polyethylene bottles for subsequent procedures (Hussain et al., 2019). The plant specimens were collected and subsequently authenticated by a taxonomist from the Makerere University Herbarium. The authenticated specimens were deposited in the herbarium under accession numbers 51272, 51273, and 51274, corresponding to *Ficus saussureana* DC, *Clerodendrum rotundifolium* Oliv., and *Microglossa pyrifolia* (Lam.) O. Ktze, respectively. All phytochemicals were analysed at the Department of Government Analytical Laboratory (DGAL), and at the Natural Chemotherapeutics Research Institute (NCRI), both of which are under the Ministry of Health, Uganda.

Sample Extraction

Maceration and decoction methods were used to obtain crude extracts from the plants. Two solvents (methanol and aqueous) were used for extraction. These two solvents were chosen for their exceptional effectiveness, as they not only yield the highest amounts but also maximise the concentration of phytochemical constituents (Truong et al., 2019). Additionally, methanol at different concentrations has proven to be an effective solvent for reliably extracting active compounds, as many of these components are readily soluble in methanol (Tejavathi & Sujatha, 2019). The maceration method is mainly used for fluid extracts. The obtained powdered plants were soaked in methanol (80%) for 48 hours in a stoppered jar, and the mixture was then incubated in the dark at room temperature with occasional agitation. After 48 hours of maceration and occasional shaking, the extracts were filtered separately with filter paper and a funnel. The obtained extract was then subjected to the following procedures. The decoction method was mainly used to obtain aqueous extracts. The powdered plant extracts were boiled in distilled water for 20 minutes. The obtained mixture was then cooled and finally filtered through filter paper and a funnel. The obtained liquid extract was then subjected to the following experimental procedures.

Preliminary Qualitative Phytochemical Screening

Preliminary phytochemical screening was performed via standard methods. The presence and abundance of the compound were determined by the change in colour and intensity of the colour after the addition of reagents to the sample extracts (Biapa et al., 2007). The procedures, reagents, and various tests used for each type of phytochemical are explained below.

Detection of Alkaloids

One millilitre of extract was mixed with 1 ml of 1% hydrochloric acid. The mixture was then warmed and filtered. Three drops of Mayer's reagent were subsequently added. A positive alkaloid test was shown by the development of a yellowish-white precipitate (Morsy, 2014).

Detection of Saponins

For the foam test, 5 ml of plant extract was combined with an equal amount of distilled water, shaken thoroughly, and then allowed to stand for 10 minutes. Positive saponins were detected by the formation of stable froth (Gracelin et al., 2013; Morsy, 2014; Longbap et al., 2018; Alqethami & Aldhebiani, 2021).

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Detection of Steroids

Five millilitres of plant extract was blended with 2 ml of chloroform and then with 2 ml of concentrated sulphuric acid. A positive steroid test was indicated by the appearance of a red colour (Gracelin et al., 2013; Morsy, 2014).

Detection of Phenols

For the ferric chloride test, 10 ml of plant extract was mixed with 3− 5 drops of ferric chloride solution. A positive phenol test revealed a bluish-black colour (Tyagi & Agarwal, 2017; Longbap et al., 2018; Nortjie et al., 2022).

Detection of Tannins

Braymer's test: Five millilitres of extracts were mixed with a few drops of 10% ferric chloride ($FeCl_3$). A positive tannin test was indicated by a dark blue/greenish-grey colour (Gracelin et al., 2013; Morsy, 2014; Tyagi & Agarwal, 2017; Longbap et al., 2018; Alqethami & Aldhebiani, 2021).

Detection of Flavonoids

For the Shinoda test, 5 ml of extract was mixed with a piece of magnesium ribbon, followed by 2 ml of concentrated hydrochloric acid (HCl). A positive flavonoid test was indicated by a red or pink–red colour (Morsy, 2014; Alqethami & Aldhebiani, 2021).

Detection of Resins

For the precipitation test, 10 ml of plant extract was blended with 15 ml of distilled water. A positive resin test was shown by the formation of a precipitate (Alqethami & Aldhebiani, 2021).

Detection of Glycosides

For Salkowski's test, 5 ml of extract was treated with 2 ml of concentrated sulphuric acid (H_2SO_4), carefully shaken, and left to stand for two minutes. A positive glycoside test was indicated by a red precipitate (Gracelin et al., 2013)**.**

Detection of Coumarins

Five millilitres of each extract was mixed with 1 N sodium hydroxide (NaOH) solution and placed in boiling water with the mouth of the test tube covered. After boiling, the test tube cover was removed and inspected under UV light for the occurrence of yellow fluorescence, which indicated a positive coumarin test.

Detection of Terpenoids

For the Salkowski test, 5 millilitres of plant extract were mixed with 2 millilitres of chloroform, and then 3 millilitres of concentrated sulphuric acid were added carefully to create a distinct layer. A positive terpenoid test revealed a layer of reddish-brown colour (Morsy, 2014; Tyagi & Agarwal, 2017; Longbap et al., 2018; Nortjie et al., 2022).

Quantitative Phytochemical Analysis

After the preliminary screening, selected groups of detected phytochemicals were quantitatively analysed via standard procedures. In this phase, alkaloids, flavonoids, saponins, tannins, and polyphenols were chosen on the basis of their potential efficacy in managing diabetes and hypertension, considering the constraints of research funding.

Quantitative Estimation of Total Alkaloid Contents

Harborne method: Five grams of each sample were measured and placed in a 250 ml beaker. Subsequently, 200 ml of 10% acetic acid in ethanol was added to the beaker, and the mixture was covered and left undisturbed for 4 hours. The extract was subsequently filtered and concentrated in a water bath until it reached one-quarter of the original volume. Concentrated ammonium hydroxide was then gradually added dropwise to the extract until the precipitation was complete. The entire solution was allowed to settle, and the precipitate was collected, washed with dilute ammonium hydroxide, and subsequently sieved. The resulting residue, identified as the alkaloid, was

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dried and weighed once complete dryness was attained. The determination of total alkaloids was carried out in triplicate (Khan et al., 2011; Morsy, 2014; Longbap et al., 2018; Alqethami & Aldhebiani, 2021; Nortjie et al., 2022).

Quantitative Estimation of Total Flavonoids

The method described by Ordoñez *et al*. (2006) was employed for the estimation of total flavonoids. One gram of the sample was extracted in 10 ml of 80% methanol. A 0.1 ml aliquot of the sample was then mixed with 0.5 ml of a 2% aluminium chloride ($AlCl_3$) ethanol solution. After one hour at room temperature, the absorbance was measured at 420 nm via a UV-visible spectrophotometer (U-2602, LaboMed-Inc. USA). The presence of flavonoids is indicated by a yellow colour. Reference standard solutions of rutin (0, 10, 20, 40, and 80 µg/mL) were prepared. The total flavonoid content, expressed as quercetin (mg/g), was calculated via the equation derived from the calibration curve. The estimation of total flavonoid content was conducted in triplicate, and the results for the extract samples were expressed as quercetin equivalent (QE) milligrams per gram of extract.

Quantitative Estimation of Saponin Content

The saponin content of the samples was assessed via the double extraction gravimetric method outlined by Hashmi *et al*. (2021) with some modifications. Five grams of each powdered sample was combined with 50 mL of a 20% aqueous ethanol solution in a flask. The mixture was heated with intermittent agitation in a water bath for 90 minutes at 55ºC, followed by filtration through Whatman filter paper (No. 42). The residue underwent a second extraction with 50 ml of 20% ethanol, and both extracts were combined. The resulting extract was reduced to approximately 40 ml at 90°C and transferred to a separating funnel, where 40 ml of diethyl ether was added and vigorously shaken. Re-extraction through partitioning was repeated until the aqueous layer became clear. Saponins were then extracted using 60 mL of normal butanol. The combined extracts were washed with a 5% aqueous sodium chloride (NaCl) solution, evaporated to dryness in a pre-weighed evaporation dish, and dried at 60°C in an oven. The dish was reweighed after cooling in a desiccator, and this process was repeated twice more to obtain an average. The saponin content was determined by the difference and expressed as a percentage of the original sample.

Quantitative Estimation of Total Tannin Content

The total tannins were determined via the Folin–Ciocalteu method with slight modifications. A 0.1g sample was extracted in 10 ml of distilled water. For the analysis, 0.1 mL of the sample extract was added to a 10 mL volumetric flask containing 7.5 mL of distilled water, 0.5 mL of Folin-Ciocalteu phenol reagent, and 1 mL of 35% sodium carbonate solution. The mixture was then diluted to 10 mL with distilled water, thoroughly shaken, and left at room temperature for 30 minutes. Reference standard solutions of tannic acid (0, 10, 20, 40, and 50 µg/mL) were prepared. The absorbances of both the test and standard solutions were measured via a UV/Visible spectrophotometer (U-2602, Labomed Inc, USA) at 725 nm against a blank (distilled water). The determination of total tannin content (TTC) was conducted in triplicate, and the tannin content was expressed in terms of mg/g gallic acid in the sample.

Quantitative Estimation of Total Polyphenols

The spectrophotometric determination of total phenolic contents in the extracts was conducted via the Folin–Ciocalteu method as outlined by Singleton *et al.* (1999). In this process, 0.1 g of the sample was extracted in 10 ml of distilled water. To the extract solution (0.1 ml), 0.5 ml of Folin–Ciocalteu reagent was added, and the total volume was adjusted to 8.5 ml with distilled water. After the tubes were maintained at room temperature for 10 minutes, 1.5 ml of sodium carbonate (20%) was added. The tubes were then incubated in a water bath at 40°C for 20 minutes, and the intensity of the

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developed blue colour was measured by recording the absorbance at 755 nm via a UV–visible spectrophotometer (U-2602, LaboMed, Inc., USA). A reagent blank was prepared using distilled water. To quantify the total phenolic content in the extract, a standard calibration curve was established using gallic acid. Reference standard solutions of gallic acid (0, 10, 20, 40, and 80 µg/mL) were prepared. The absorbances of both the test and standard solutions were measured with a UV/Visible spectrophotometer (U-2602, Labomed Inc., USA) against a blank (distilled water) at 755 nm. The total polyphenol content was determined in triplicate, and the results for the extract samples were expressed as gallic acid equivalent (GAE) milligrams per gram of extract.

Gas Chromatography–Mass Spectrometry (GC–MS) Analysis

For the general identification of the phytochemical compounds within the plants, the methanolic extracts of individual plants were analysed via GC-MS of Agilent Technologies (USA) model (Intuvo 9000 GC connected to 19091S-433UI-INT MS) with an HP-5MS UI column 30 m in length, 250 µm in dimension, and 0.25 µm in film thickness. Helium served as the carrier gas, and the flow rate was set at 3 mL/min. Sample injection was performed in splitless mode. The sample volume used was 5 µL. The temperature at the injector was set at 280°C. The oven temperature was programmed as follows: 70°C for 2 minutes, increased at a rate of 25°C/min to 150°C, held for 2 minutes, increased at a rate of 3°C/min to 200°C, held for 2 minutes, and finally increased at 8°C/min to 280°C, held for 10 minutes. The ionisation voltage of MS analysis was controlled by the EI procedure with an ion source heat of 280°C. The total GC–MS running time was 45.867 min. The relative proportional percentage of each component was calculated by comparing its average peak area to the total area. The analysis of the mass spectrum of the GC–MS instrument utilised the National Institute of Standards and Technology (NIST) database, which contains more than 62,000 patterns. The spectra of the unidentified components were matched against the spectra of known components stored in the NIST library. This process determines the names, molecular weights, and molecular formulas of the components present in the extracts. To ensure that the results were accurate, the instrument was tuned with perfluorotributylamine (PFTBA) to ensure that the calibration had not shifted. This was performed before injecting the sample. Additionally, the blank methanol solvent used for extraction was also injected into the machine.

Data Analysis

The preliminary phytochemical screening and GC–MS results were compiled in Excel and presented in tabular format. The quantitative phytochemical data were analysed via GraphPad Prism version 8 (GraphPad Software, San Diego, CA, USA). The results are expressed as the mean ± standard error of the mean (SEM).

RESULTS

Qualitative Phytochemical Analysis

The outcomes of the qualitative analysis are presented in Table 1. This study revealed the presence of most of the analysed phytochemicals, including alkaloids, steroids, phenols, tannins, flavonoids, coumarins, and terpenoids. Resins were absent in all of the extracts except the *Microglossa pyrifolia* methanolic extract (MPM), and glycosides were detected in all of the extracts except the *Clerodendrum rotundifolium* methanolic extract (CRM). Saponins were present in all four extracts except the *Ficus saussureana* methanolic extract (FSM) and MPM.

For the qualitative phytochemical analysis of the six plant extracts, quantitative analysis was performed on major phytochemicals, such as saponins, alkaloids, tannins, flavonoids, and polyphenols. The results are presented in Figure 2. The results of the quantitative analysis revealed that the highest amount of polyphenols (217 mg/g) was in FSM,

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followed by CRM (174 mg/g), MPM (152.05 mg/g), FSP (44.73 mg/g), MPP (40 mg/g), and CRP (29.04 mg/g). Flavonoids amount was measured in mg of RE/g of extract; the greatest amount was depicted in CRM (28.75 mg/g), followed by CRP (12.33 mg/g), MPM (11.88 mg/g), FSM (7.62 mg/g), MPP (3.72 mg/g), and FSP (0.78 mg/g). Tannins were measured in mg of GAE/g of extract, and the highest amount was recorded for FSM (179.77 mg/g), followed by CRM (67.47 mg/g), and the least in MPP (3.8 mg/g). The greatest amount of saponins was detected in CRM (225.07 mg/g), followed by FSM (92.36 mg/g), and the lowest amount was detected in CRP (29 mg/g). For alkaloids, the greatest amount was in the order of saponins, except that the lowest amount was associated with FSP (29.55 mg/g).

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Table 1: Qualitative Phytochemical Analysis of Ficus saussureana, Microglossa pyrifolia, and Clerodendrum rotundifolium

GROUPS	TEST	DETECTION	EXTRACTS					
			FSM	FSA	MPM	MPA	CRM	CRA
Alkaloids	Hager's test	Yellow precipitate	+	+	+	+	+	+
Saponins	Foam test	Stable froth	-	+	-	+	+	+
Steroids	Extract + chloroform +H_2SO_4	Red colour	+	+	+	+	+	+
Phenols	Ferric chloride test	Blue-black colour	+	+	+	+	+	+
Tannins	Braymer's test	Dark blue colour	+	+	+	+	+	+
Flavonoids	Shinoda test	Red fluorescence	+	+	+	+	+	+
Resins	Precipitate test	Green precipitate	-	-	+	-	-	-
Glycosides	Salkowski's test	Red ppt	+	+	+	+	-	+
Coumarins	1 N NaOH	Greenish-yellow fluorescence	+	+	+	+	+	+
Terpenoids	Salkowski's test	Reddish-brown layer	+	+	+	+	+	+

FSM= *Ficus saussureana* Methanol**, FSA=** *Ficus saussureana* Aqueous, **MPM=***Microglossa pyrifolia* Methanol, **MPA=** *Microglossa pyrifolia* Aqueous**, CRM=***Clerodendrum rotundifolium* Methanol and **CRA=***Clerodendrum rotundifolium* Aqueous**.** + Detected, - Not detected.

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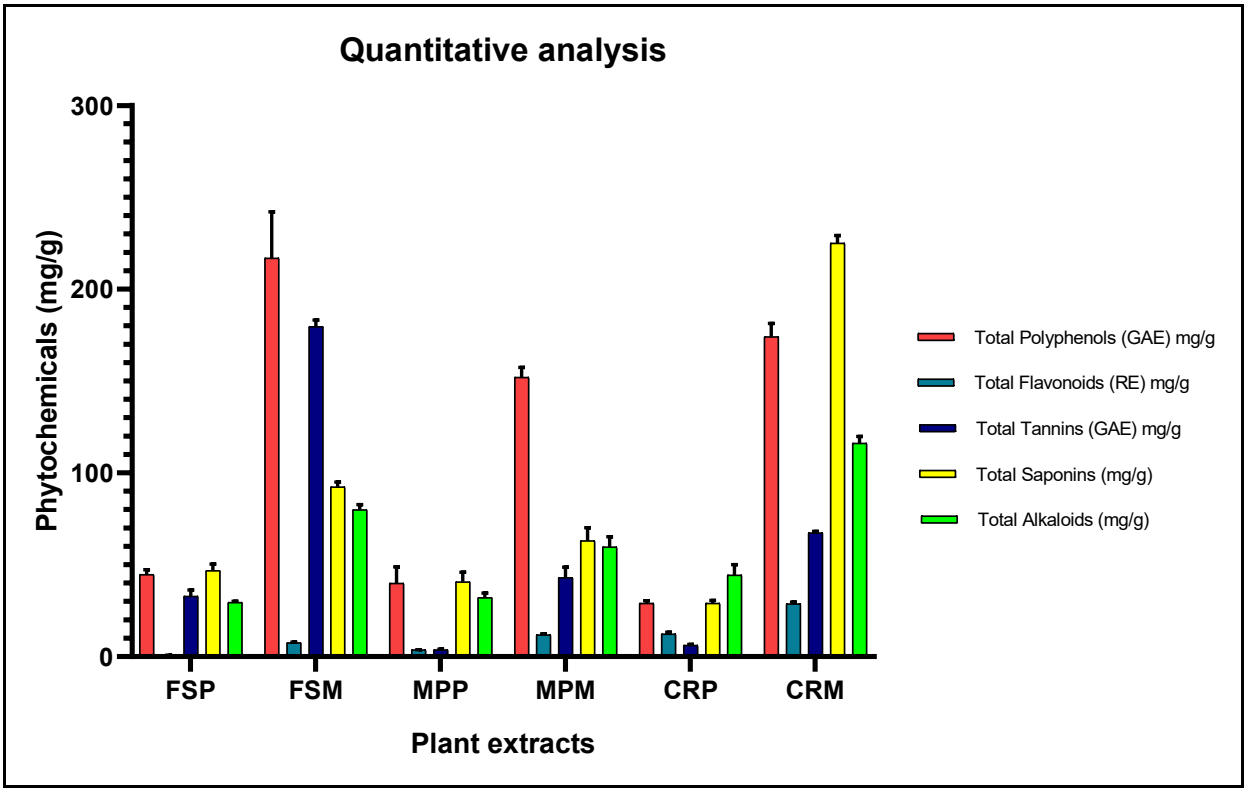
Figure 2. Quantitative Phytochemical Analysis of Ficus saussureana, Microglossa pyrifolia, and Clerodendrum rotundifolium. FSP = Ficus saussureana powder, FSM = Ficus saussureana methanol, MPP = Microglossa pyrifolia powder, MPM = Microglossa pyrifolia methanol, CRP = Clerodendrum rotundifolium powder, and CRM = Clerodendrum rotundifolium methanol.



Gas Chromatography–Mass Spectrometry Profiles

The findings from the GC–MS analysis of *Ficus saussureana*, *Clerodendrum rotundifolium*, and *Microglossa pyrifolia* are presented in Tables 2, 3, and 4, respectively. The compounds identified are systematically arranged in accordance with their RT. Additionally, a thorough literature search was conducted to elucidate the biological (pharmacological) activities associated with the identified biomolecules for each plant. The outcomes are succinctly documented in the tables.

GC-MS Profile of *Ficus saussureana*

The GC–MS results of methanolic extract of *Ficus saussureana* revealed the presence of fourteen compounds which included tridecanoic acid, 12-methyl-,methyl ester (7.7%), tetradecanoic acid (29.9%), hexadecanoic acid, 1-(hydroxymethyl)-1,2-ethanediyl ester (0.9%), milbemycin b, 13-chloro-5-demethoxy-28-deoxy-6,28-epoxy-5-(hydroxyimino)-25-(1-methylethyl)-,(6R,13R,25R)- (1.2%), 9,12-Octadecadienoic acid (Z,Z)- (5.5%), oleic acid (8.0%), Lup-20(29)-en-3-ol, acetate, (3.beta.)- (1.3%), squalene (5.3%), vitamin E (1.0%), 17.beta.-Acetoxy-1',1'-dicarboethoxy-1.beta., 2.beta.-dihydrocycloprop[1,2]-5.alpha.-androst-1-en-3-one (4.4%), 7aH-cyclopenta[a]cyclopropa[f]cycloundecene-2,4,7,7a,10,11-hexol, 1,1a,2,3,4,4a,5,6,7,10,11,11a-dodecahydro-1,1,3,6,9-pentamethyl-,2,4,7,10,11-pentaacetate (1.2%), .alpha.-Amyrin (0.4%), Betulinaldehyde (0.8%), and Lupeol (7.5%) (Table 2).

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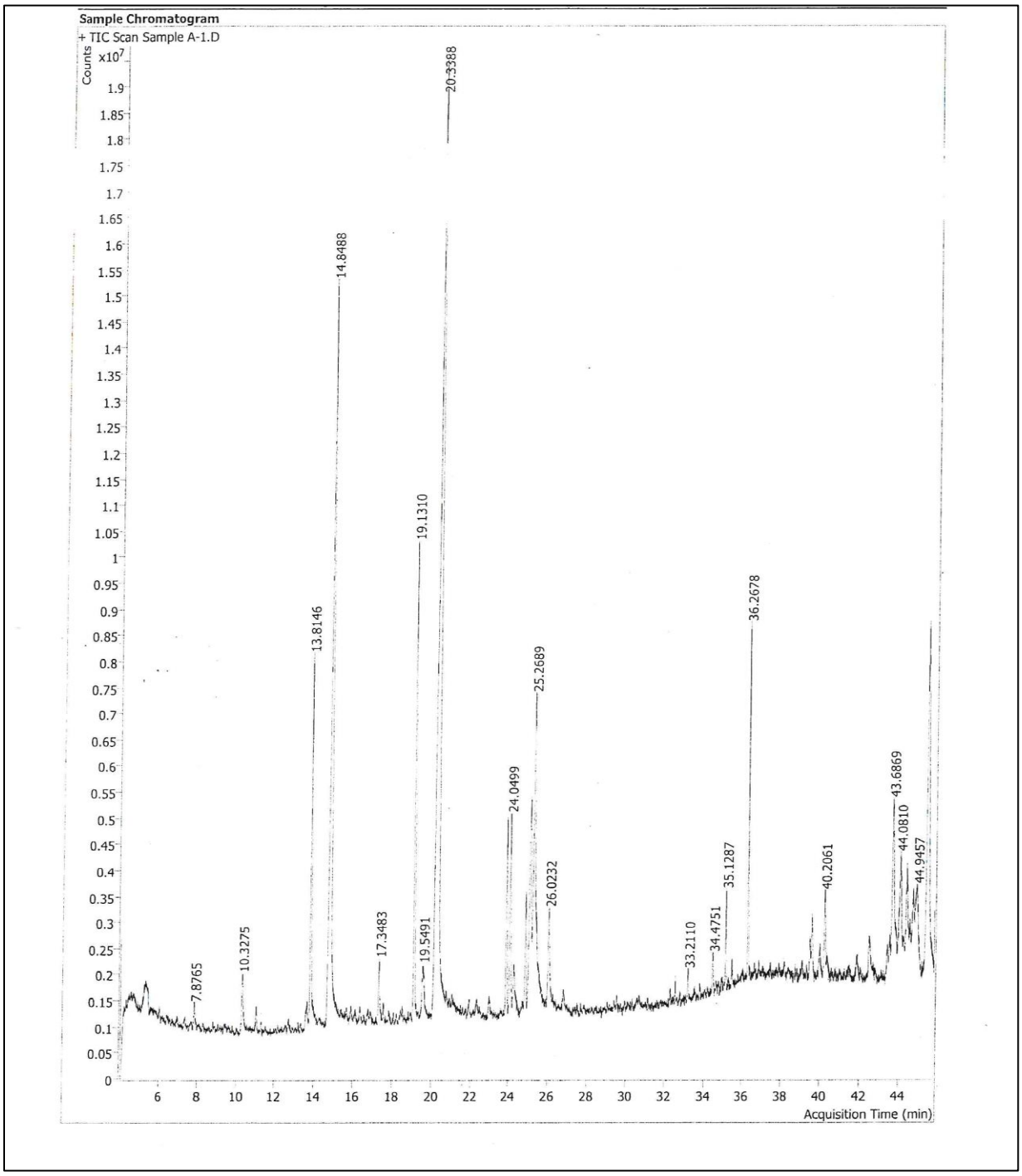
Figure 3. Gas Chromatography–Mass Spectrometry Chromatograms of Ficus saussureana Showing the Compounds Detected



Article DOI: https://doi.org/10.37284/eajhs.9.1.4294

Table 2: GC –MS Profile of Ficus saussureana Showing the Chemical Compounds Identified

Retention Time (RT)	Compound Name	Molecular Formula	% Area	Biological activities	Reference
13.8146	Tridecanoic acid, 12-methyl-,methyl ester	$C_{15}H_{30}O_2$	7.67586	NR	NR
14.8488	Tetradecanoic acid	$C_{14}H_{28}O_2$	29.8613	**Antioxidant**** Larvicidal and repellent activity, Lubricant, Hypercholesterolemic, Cancer-preventive, Cosmetic	(Ponnamma & Manjunath, 2012; Arora & Meena, 2017; Vijayalingam & Rajesh, 2020)
19.5491	Hexadecanoic acid, 1-(hydroxymethyl)-1, 2-ethanediyl ester	$C_{35}H_{68}O_5$	0.86858	Lubricants, emollients. Acidifiers, acidulants, and arachidonic acid inhibitors, increase aromatic amino acid decarboxylase activity and inhibit the production of uric acid	(Sivakumaran et al., 2019; Vijayalingam & Rajesh, 2020)
24.2421	Milbemycin b, 13-chloro-5-demethoxy-28-deoxy-6,28-epoxy-5-(hydroxyimino)-25-(1-methylethyl)-,(6R,13R,25R)-	$C_{33}H_{46}ClNO_7$	1.17701	NR	NR
25.0654	9, 12-Octadecadienoic acid (Z,Z)-	$C_{18}H_{32}O_2$	5.52681	**Antioxidant**** Antiarthritic, Antieczemic, Hepatoprotective, Anti-inflammatory, Anticane, Nematicide, anticancer, Insectifuge, Antihistaminic	(Ponnamma & Manjunath, 2012; Arora & Meena, 2017)
25.2689	Oleic Acid	$C_{18}H_{34}O_2$	7.9841	**Antidiabetic**. Antibacterial	(Vassiliou et al., 2009)

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Retention Time (RT)	Compound Name	Molecular Formula	% Area	Biological activities	Reference
35.1287	Lup-20(29)-en-3-ol, acetate, (3.beta.)-	$C_{32}H_{52}O_2$	1.25243	**Antidiabetic**.** Anticancer, anti-inflammatory, antituberculosis, antimalarial, antimicrobial, antinociceptive	(Javaid et al., 2021; Amankwaah et al., 2023)
36.2678	Squalene	$C_{30}H_{50}$	5.29692	**Hypoglycemic, Antioxidant**.** Antibacterial, cancer-preventive, immunostimulant, Antitumor, Anti-inflammatory, Antinociceptive, Potential antiplatelet components, Hypolipidemic effects, Sedative action, Antihistaminic, Hepatoprotective activities, lipoxygenase-inhibitor, perfumery, pesticide, sunscreen	(Ingole, 2016; Arora & Meena, 2017)
40.2061	Vitamin E	$C_{29}H_{50}O_2$	0.97196	**Antidiabetic and Antioxidant**.** Hypocholesterolemic, hepatoprotective, Anti-inflammatory, anticancer, anti-coronary, antiulcerogenic, antidermatitic, antiaging, analgesic, antidermatitic, antileukemia, antitumor, vasodilator, antispasmodic, anti-bronchitic.	(Ponnamma & Manjunath, 2012; Arora & Meena, 2017)
43.6869	17.beta.-Acetoxy-1',1'-dicarboethoxy-1.beta., 2.beta.-dihydrocycloprop[1,2]-5.alpha.-androst-1-en-3-one	$C_{28}H_{40}O_7$	4.41163	NR	NR
44.081	7aH-Cyclopenta[a]cyclopropa[f]cycloundecene-2,4,7,7a,10,11-hexol, 1,1a,2,3,4,4a,5,6,7,10,11,11a-dodecahydro-1,1,3,6,9-pentamethyl-,2,4,7,10,11-pentaacetate	$C_{30}H_{44}O_{11}$	1.21305	Immune system enhancement and antimicrobial property	(Verma et al., 2021)

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Retention Time (RT)	Compound Name	Molecular Formula	% Area	Biological activities	Reference
44.3854	.alpha.-Amyrin	$C_{30}H_{50}O$	0.38601	Antimicrobial, anti-inflammatory	(Priyadarshini et al., 2017)
44.729	Betulinaldehyde	$C_{30}H_{48}O_2$	0.84042	NR	NR
45.4779	Lupeol	$C_{30}H_{50}O$	7.54766	**Antihyperglycemic, Antioxidant**.** Anticancer, anti-arthritic, Anti-inflammatory, anti-mutagenic, Antiviral, anti-HIV, Antitumor, Antihyperlipidemic, Anti-flu, Prostaglandin-synthesis and Topoisomerase II-inhibitor, Antimalarial, Pesticide, Cytotoxic	(Arora & Meena, 2017; Perumal et al., 2021)

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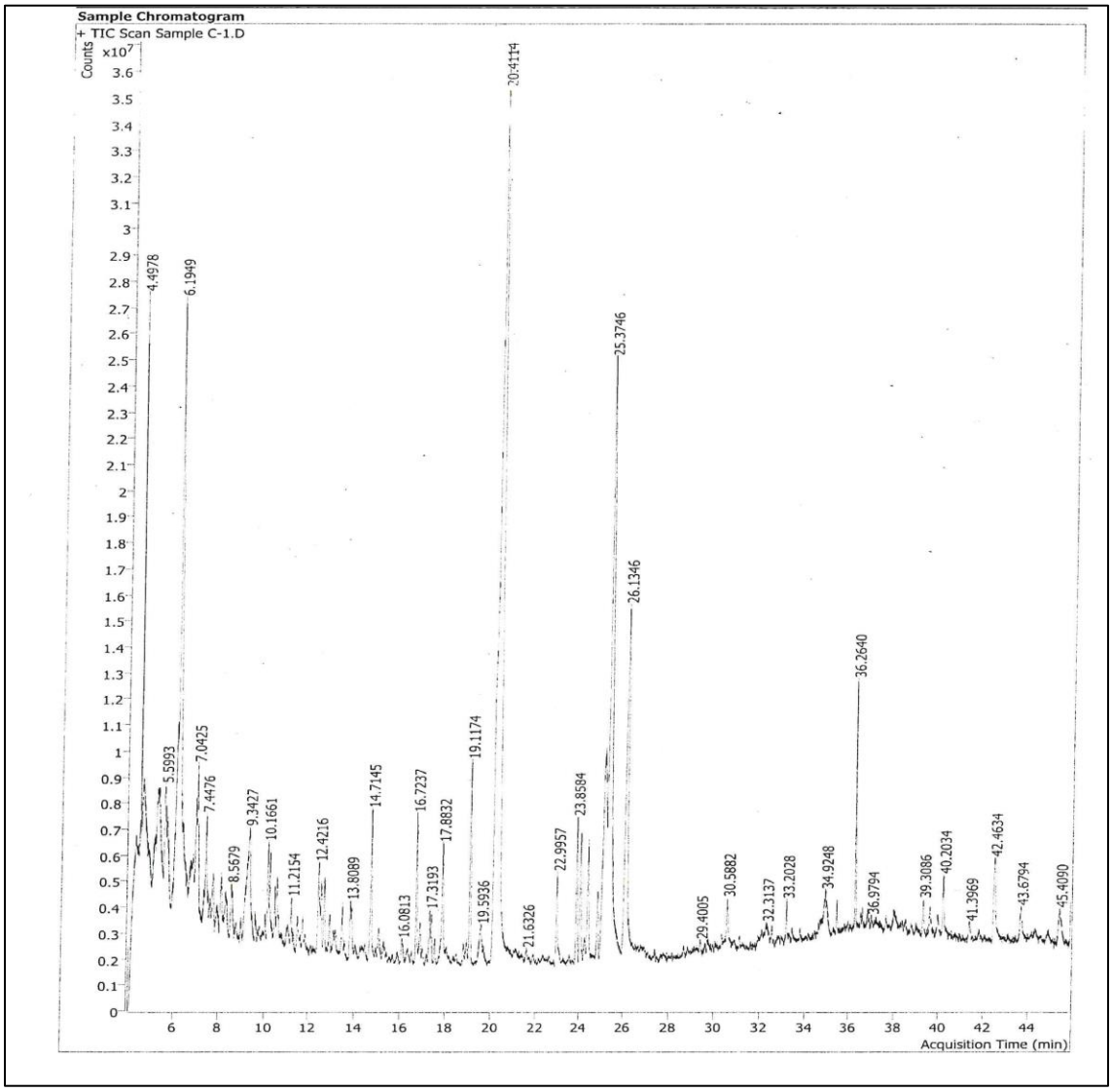
GC-MS Profile of Clerodendrum rotundifolium

Clerodendrum rotundifolium methanolic extract included thirty chemical compounds, including 9,12,15-Octadecatrienoic acid, (Z,Z,Z)- (49.37%), 5-Hydroxymethylfurfural (5.04%), octadecanoic acid (4.29%), tetradecanoic acid (1.36%), benzene, 1,2-dichloro- (1.31%), squalene (1.23%), phytol (1.06%), 2-hexadecen-1-ol,3,7,11,15-tetramethyl-,acetate, [R-[R*,R*-E)]]- (1.03%), stigmasterol (0.94%), heptadecanoic acid (0.72%), 9,12-octadecadienoic acid (Z,Z)- (0.58%), betulinaldehyde (0.49%), vitamin E (0.42%), .psi.,.psi.-Carotene, 1,1',2,2'-tetrahydro-1,1'-dimethoxy (0.29%), and 2-Methoxy-4-vinyl phenol (0.23%). Details of all the other compounds detected in *Clerodendrum rotundifolium* are presented in Table 3 below.

Figure 4: Gas Chromatography– Mass Spectrometry Chromatograms of Clerodendrum rotundifolium Showing the Compounds Detected



Article DOI: https://doi.org/10.37284/eajhs.9.1.4294

Table 3: GC –MS Profile of Clerodendrum rotundifolium Showing the Chemical Compounds Identified

Retention Time (RT)	Compound Name	Molecular Formula	% Area	Biological activities	Reference
4.4978	Benzene, 1,2-dichloro-	$C_6H_4CI_2$	1.31	Fumigants, Insecticides	(Suganthy & Gajendra, 2020)
6.1949	5-Hydroxymethylfurfural	$C_6H_6O_3$	5.04	Antioxidant. Anti-proliferative activity. It is reported to stop neuron apoptosis (Hai)	(Gu et al., 2013; Nandhini et al., 2021)
6.979	2-Methoxy-4-vinyl phenol	$C_9H_{10}O_2$	0.23	Antioxidant. Antimicrobial & anticancer	(Kim et al., 2019; Nandhini et al., 2021)
10.0578	1b,4a-Epoxy-2H- cyclopenta [3,4]cycloundec[1,2-b]oxiren-5(1aH)-one,2,7,10-tetrakis(acetyloxy)decahydro-3,6,8,8,10a-pentamethyl-	$C_{28}H_{38}O_{11}$	0.09	NR	NR
10.1661	Homovanillyl alcohol	$C_9H_{12}O_3$	0.66	NR	NR
10.2669	Dodecanoic acid	$C_{12}H_{24}O_2$	0.35	Antimicrobial	(Anbuselvi et al., 2012)
12.4216	Androsta-1,4-dien-3-one,17-hydroxy-17-methyl-, (17. alpha.)-	$C_{20}H_{28}O_2$	0.97	NR	NR
12.8959	Naphthalene, 1,6-dimethyl-4-(1-methylethyl)-	$C_{15}H_{18}$	0.23	NR	NR
13.4598	2-Cyclohexen-1-one, 4-(3-hydroxybutyl)-3,5,5-trimethyl-	$C_{13}H_{22}O_2$	0.39	NR	NR
13.8089	Tridecanoic acid, 12-methyl-,methyl ester	$C_{15}H_{30}O_2$	0.34	NR	NR

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Retention Time (RT)	Compound Name	Molecular Formula	% Area	Biological activities	Reference
14.7145	Tetradecanoic acid	$C_{14}H_{28}O_2$	1.36	Antioxidant. Nematicidal and repellent activity, Lubricant, Hypercholesterolemic, Cancer-preventive, Cosmetic	(Arora & Meena, 2017; Vijayalingam & Rajesh, 2020)
13.8631	2-Cyclohexen-1-one,3-(3-hydroxybutyl)-2,4,4-trimethyl-	$C_{13}H_{22}O_2$	0.31	NR	NR
16.7237	2-Hexadecen-1-ol,3,7,11,15-tetramethyl-,acetate, [R-[R*,R*-E)]]-	$C_{22}H_{42}O_2$	1.03	NR	NR
16.8861	9-Hexadecenoic acid, 9-octadecenyl ester, (Z,Z)-	$C_{34}H_{64}O_2$	0.02	NR	NR
17.3193	Pentadecanoic acid	$C_{15}H_{30}O_2$	0.25	Lubricants, Adhesive agents	(Arora & Meena, 2017)
22.9957	Heptadecanoic acid	$C_{17}H_{34}O_2$	0.72	Antioxidant. Anti-fungal, surfactant	(Ponnamma & Manjunath, 2012; Arora & Meena, 2017)
24.2468	9-Octadecenoic acid, 1,2,3-propanetriyl ester, (E,E,E)-	$C_{57}H_{104}O_6$	0.14	Anti-spasmodic and immune modulators	(Al-marzoqi et al., 2016)
24.3943	Phytol	$C_{20}H_{40}O$	1.06	Antioxidant. Anticancer, Antimicrobial, Decreases the autoimmune response and ameliorates both acute and chronic phases of arthritis, Anti-inflammatory, Diuretic	(Arora & Meena, 2017; Vijayalingam & Rajesh, 2020)
25.0983	9,12-Octadecadienoic acid (Z,Z)-	$C_{18}H_{32}O_2$	0.58	Antioxidant. Hepatoprotective, Hypocholesterolemic, antiarthritic, anti-inflammatory, anti-arteriosclerotic, anti-anaphylactic, antieczemic, Cancer preventive, anti-prostatic, Metastatic, Nematicide	(Ponnamma & Manjunath, 2012)

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Retention Time (RT)	Compound Name	Molecular Formula	% Area	Biological activities	Reference
25.3746	9,12,15-Octadecatrienoic acid, (Z,Z,Z)-	$C_{18}H_{30}O_2$	49.37	Anti-inflammatory, Hypocholesterolemic, Cancer preventive, Hepatoprotective, Nematicide Insectifuge, Antihistaminic Antieczemic, Antiacne, 5-Alpha reductase inhibitor, Antiandrogenic, Antiarthritic, Anticoronary, insectifuge	(Rao et al., 2019; Vijayalingam & Rajesh, 2019)
26.1346	Octadecanoic acid	$C_{18}H_{36}O_2$	4.29	Hypocholesterolemic, Antimicrobial, Antifungal, Antitumor, Antibacterial, Cosmetic, Flavour, Lubricant, Perfumery, Propecic, Suppository	(Ponnamma & Manjunath, 2012; Arora & Meena, 2017)
30.5882	.psi.,.psi.-Carotene, 1,1',2,2'-tetrahydro-1,1'-dimethoxy-	$C_{42}H_{64}O_2$	0.29	Antioxidant. Nutrient, Cytotoxic activity	(Kavitha R, 2021)
32.3137	4H-Cyclopropa[5',6']benz[1'2':7,8]azuleno[5,6-b]oxiren-4-one, 8,8a-bis(acetyloxy)-2a-(acetyloxy)methyl]-1,1a,1b,1c,2a,3,3a,6a,6b,7,8,8a-dodecahydro- 6b- hydroxy-3a-methoxy-1,1,5,7-tetramethyl-[1aR(1a.alpha.,1b.beta.,1c.alpha.,2a.alpha.,3a.alpha.,6a.alp	$C_{27}H_{36}O_{10}$	0.02	NR	NR
33.8191	2,4,6,8,10-Tetradecapentaenoic acid, 9a-(acetyloxy)-1a,1b,4,4a,5,7a,7b,8,9,9a-	$C_{36}H_{46}O_8$	0.05	NR	NR

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Retention Time (RT)	Compound Name	Molecular Formula	% Area	Biological activities	Reference
	decahydro-4a,7b-dihydroxy-3-(hydroxymethyl)-1,1,6,8-tetramethyl-5-oxo-1H-cyclopropa[3,4]benz[1,2-e]azulen-9-yl ester, [1aR-(1a.alpha.,1b.beta.,7a.alpha.,7b.alpha.,8.alp				
36.264	Squalene	$C_{30}H_{50}$	1.23	Antioxidant. Antibacterial, antitumor, chemo-preventive, immunostimulant, lipoxygenase-inhibitor, perfumery, pesticide, sunscreen	(Ingole, 2016; Zayed & Samling, 2016)
36.5778	Hexadecanoic acid, 1a,2,5,5a,6,9,10,10a-octahydro-5a-hydroxy-4-(hydroxymethyl)-1,1,7,9-tetramethyl-6,11-dioxo-1H-2,8a-methanocyclopenta[a]cyclopropa[e]cyclodecen-5-yl ester, [1aR-[1a.alpha.,2.alpha.,5.beta.,5a.beta.,8a.alpha.,9.alpha., 10a.alpha.)]-	$C_{36}H_{56}0_6$	0.06	NR	NR
39.9549	5H-Cyclopropa[3,4]benz[1,2-e]azulen-5-one,3,9,9a-tris(acetyloxy)-3-[(acetyloxy)methyl]-2-chloro-1,1a,1b,2,3,4,4a,7a,7b,8,9,9a-dodecahydro-4a,7b-dihydroxy-1,1,6,8-tetramethyl-,[1aR-(1a.alpha.,1b.beta.,2,alpha.,3.beta.,4a.beta.,7a.alpha.,7b.alpha.,8.alpha	$C_{28}H_{37}ClO_{11}$	0.14	NR	NR

Article DOI: https://doi.org/10.37284/eajhs.9.1.4294

Retention Time (RT)	Compound Name	Molecular Formula	% Area	Biological activities	Reference
40.2034	Vitamin E	$C_{29}H_{50}O_2$	0.42	Antidiabetic, Antioxidant. Hypocholesterolemic, anti-inflammatory, antiaging, anti-coronary, antiulcer, hepatoprotective, antidermatitic, antileukemia, antiaging, anti-alzheimeran, antitumor, anticancer, immunostimulant, analgesic, vasodilator, antispasmodic, anti-bronchitic.	(Ponnamma & Manjunath, 2012; Arora & Meena, 2017)
42.4634	Stigmasterol	$C_{29}H_{48}O$	0.94	Hypoglycemic effects, antioxidant. Antihypercholesterolemic, thyroid inhibitory, antiperoxidative, Precursor of progesterone, acts as intermediate in the biosynthesis of androgens and oestrogens, anti-osteoarthritic, cytotoxic, antitumor, antimutagenic, anti-inflammatory, analgesic	(Sivakumaran et al., 2019; Perumal, et al., 2021)
45.409	Betulinaldehyde	$C_{30}H_{48}O_2$	0.49	Anti-tumor	(Huang et al., 2023)

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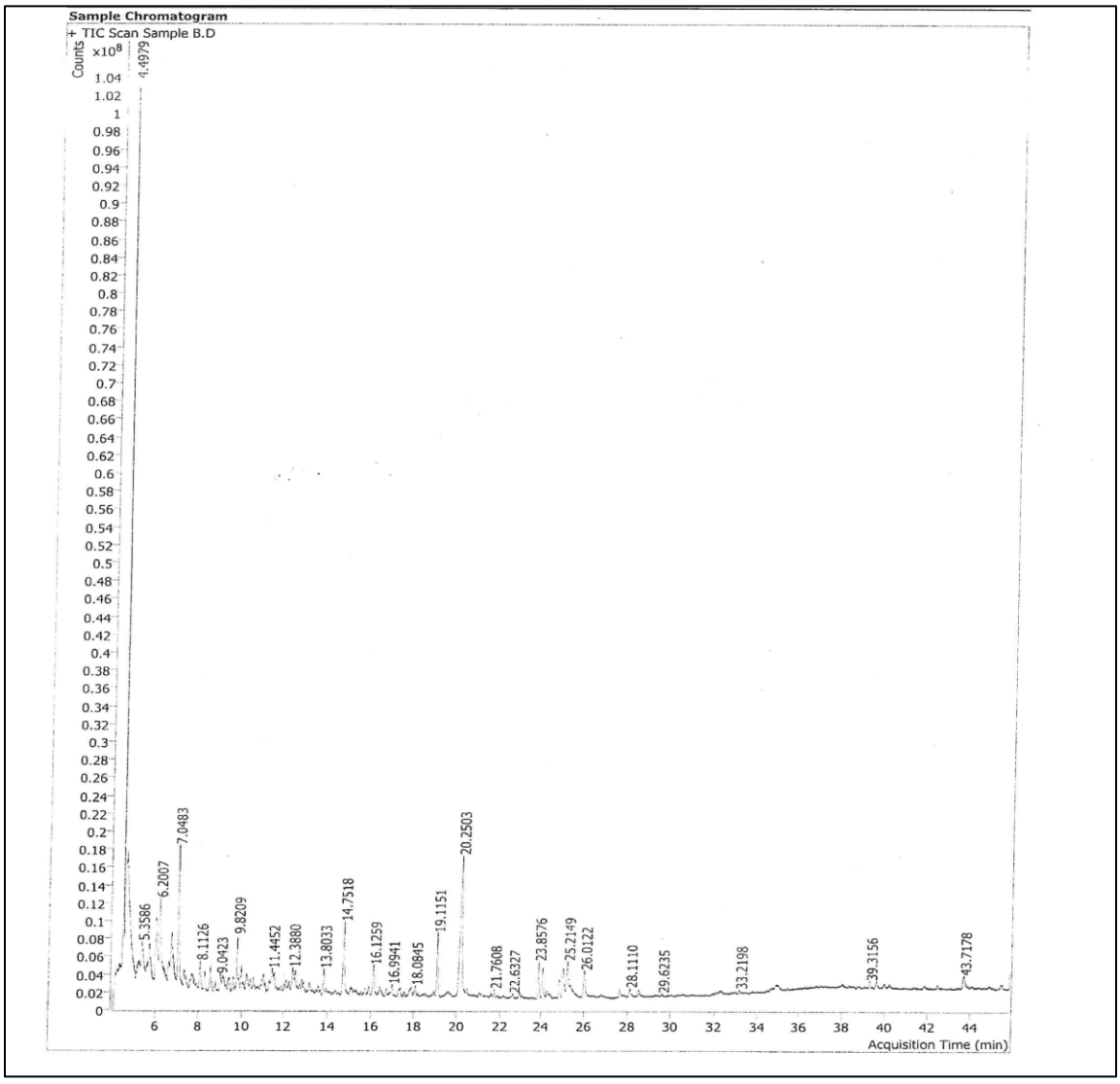
GC-MS Profile of Microglossa pyrifolia

GCMS analysis of the methanolic extracts of the *Microglossa pyrifolia* revealed thirty compounds, including benzene,1,2-dichloro- (15.70%), n-hexadecanoic acid (14.50%), 1,2-benzenedicarboxylic acid (8.50%), 5-hydroxymethylfurfural (6.10%), catechol (5.40%), tetradecanoic acid (4.70%), hexadecanoic acid, methyl ester (3.50%), and oleic acid (2.00%) 1,2-benzenediol,3-methyl- (1.80%), bicyclo[5.2.0]nonane,2-methylene-4,8,8-trimethyl-4-vinyl- (1%), 1-heptatriacotanol (0.70%), and .psi.,.psi.-Carotene, 1,1',2,2'-tetrahydro-1,1'-dimethoxy- (0.40%). Details of all the other compounds in *Microglossa pyrifolia* are presented in Table 4.

Figure 5: Gas chromatography–mass spectrometry chromatograms of Microglossa pyrifolia, Showing the Compounds Detected



Article DOI: https://doi.org/10.37284/eajhs.9.1.4294

Table 4: GC – MS Profile of Microglossa pyrifolia Showing the Chemical Compounds Identified

Retention Time (RT)	Compound Name	Molecular Formula	% Area	Biological activities	Reference
4.4978	Benzene, 1,2-dichloro-	$C_6H_4CI_2$	1.31	Fumigants, Insecticides	(Suganthy & Gajendra, 2020)
6.1949	5-Hydroxymethylfurfural	$C_6H_6O_3$	5.04	Antioxidant. Anti-proliferative activity. It is reported to stop neuron apoptosis (Hai)	(Gu et al., 2013; Nandhini et al., 2021)
6.979	2-Methoxy-4-vinyl phenol	$C_9H_{10}O_2$	0.23	Antioxidant. Antimicrobial & anticancer	(Kim et al., 2019; Nandhini et al., 2021)
10.0578	1b,4a-Epoxy-2H- cyclopenta [3,4]cycloundec[1,2-b]oxiren-5(1aH)-one,2,7,10-tetrakis(acetyloxy)decahydro-3,6,8,8,10a-pentamethyl-	$C_{28}H_{38}O_{11}$	0.09	NR	NR
10.1661	Homovanillyl alcohol	$C_9H_{12}O_3$	0.66	NR	NR
10.2669	Dodecanoic acid	$C_{12}H_{24}O_2$	0.35	Antimicrobial	(Anbuselvi et al., 2012)
12.4216	Androsta-1,4-dien-3-one,17-hydroxy-17-methyl-, (17. alpha.)-	$C_{20}H_{28}O_2$	0.97	NR	NR
12.8959	Naphthalene, 1,6-dimethyl-4-(1-methylethyl)-	$C_{15}H_{18}$	0.23	NR	NR
13.4598	2-Cyclohexen-1-one, 4-(3-hydroxybutyl)-3,5,5-trimethyl-	$C_{13}H_{22}O_2$	0.39	NR	NR
13.8089	Tridecanoic acid, 12-methyl-,methyl ester	$C_{15}H_{30}O_2$	0.34	NR	NR
14.7145	Tetradecanoic acid	$C_{14}H_{28}O_2$	1.36	Antioxidant. Nematicidal and repellent activity, Lubricant, Hypercholesterolemic, Cancer-preventive, Cosmetic	(Arora & Meena, 2017; Vijayalingam & Rajesh, 2020)
13.8631	2-Cyclohexen-1-one,3-(3-hydroxybutyl)-2,4,4-trimethyl-	$C_{13}H_{22}O_2$	0.31	NR	NR
16.7237	2-Hexadecen-1-ol,3,7,11,15-tetramethyl-,acetate, [R-[R*,R*-E)]]-	$C_{22}H_{42}O_2$	1.03	NR	NR
16.8861	9-Hexadecenoic acid, 9-octadecenyl ester, (Z,Z)-	$C_{34}H_{64}O_2$	0.02	NR	NR

Article DOI: https://doi.org/10.37284/eajhs.9.1.4294

Retention Time (RT)	Compound Name	Molecular Formula	% Area	Biological activities	Reference
17.3193	Pentadecanoic acid	$C_{15}H_{30}O_2$	0.25	Lubricants, Adhesive agents	(Arora & Meena, 2017)
22.9957	Heptadecanoic acid	$C_{17}H_{34}O_2$	0.72	Antioxidant. Anti-fungal, surfactant	(Ponnamma & Manjunath, 2012; Arora & Meena, 2017)
24.2468	9-Octadecenoic acid, 1,2,3-propanetriyl ester, (E,E,E)-	$C_{57}H_{104}O_6$	0.14	Anti-spasmodic and immune modulators	(Al-marzoqi et al., 2016)
24.3943	Phytol	$C_{20}H_{40}O$	1.06	Antioxidant. Anticancer, Antimicrobial, Decreases the autoimmune response and ameliorates both acute and chronic phases of arthritis, Anti-inflammatory, Diuretic	(Arora & Meena, 2017; Vijayalingam & Rajesh, 2020)
25.0983	9,12-Octadecadienoic acid (Z,Z)-	$C_{18}H_{32}O_2$	0.58	Antioxidant. Hepatoprotective, Hypocholesterolemic, antiarthritic, anti-inflammatory, anti-arteriosclerotic, anti-anaphylactic, antieczemic, Cancer preventive, anti-prostatic, Metastatic, Nematicide	(Ponnamma & Manjunath, 2012)
25.3746	9,12,15-Octadecatrienoic acid, (Z,Z,Z)-	$C_{18}H_{30}O_2$	49.37	Anti-inflammatory, Hypocholesterolemic, Cancer preventive, Hepatoprotective, Nematicide Insectifuge, Antihistaminic Antieczemic, Antiacne, 5-Alpha reductase inhibitor, Antiandrogenic,	(Rao et al., 2019; Vijayalingam & Rajesh, 2019)

Article DOI: https://doi.org/10.37284/eajhs.9.1.4294

Retention Time (RT)	Compound Name	Molecular Formula	% Area	Biological activities	Reference
				Antiarthritic, Anticoronary, insectifuge	
26.1346	Octadecanoic acid	$C_{18}H_{36}O_2$	4.29	Hypocholesterolemic, Antimicrobial, Antifungal, Antitumor, Antibacterial, Cosmetic, Flavour, Lubricant, Perfumery, Propecic, Suppository	(Ponnamma & Manjunath, 2012; Arora & Meena, 2017)
30.5882	.psi.,.psi.-Carotene, 1,1',2,2'-tetrahydro-1,1'-dimethoxy-	$C_{42}H_{64}O_2$	0.29	Antioxidant. Nutrient, Cytotoxic activity	(Kavitha R, 2021)
32.3137	4H- Cyclopropa[5',6']benz[1'2':7,8]azuleno[5,6-b]oxiren-4-one, 8,8a-bis(acetyloxy)-2a-(acetyloxy)methyl]-1,1a,1b,1c,2a,3,3a,6a,6b,7,8,8a-dodecahydro- 6b- hydroxy-3a-methoxy-1,1,5,7-tetramethyl-[1aR(1a.alpha.,1b.beta.,1c.alpha.,2a.alpha.,3a.alpha.,6a.alp	$C_{27}H_{36}O_{10}$	0.02	NR	NR
33.8191	2,4,6,8,10-Tetradecapentaenoic acid, 9a-(acetyloxy)-1a,1b,4,4a,5,7a,7b,8,9,9a-decahydro-4a,7b-dihydroxy-3-(hydroxymethyl)-1,1,6,8-tetramethyl-5-oxo-1H-cyclopropa[3,4]benz[1,2-e]azulen-9-yl ester, [1aR-(1a.alpha.,1b.beta.,7a.alpha.,7b.alpha.,8.alp	$C_{36}H_{46}O_8$	0.05	NR	NR
36.264	Squalene	$C_{30}H_{50}$	1.23	Antioxidant. Antibacterial, antitumor, chemo-preventive, immunostimulant, lipoxygenase-inhibitor, perfumery, pesticide, sunscreen	(Ingole, 2016; Zayed & Samling, 2016)
36.5778	Hexadecanoic acid, 1a,2,5,5a,6,9, 10,10a-octahydro-5a-hydroxy-4-(hydroxymethyl)-1,1,7,9-tetramethyl-6,11-dioxo-1H-2,8a-methanocyclopenta[a]cyclopropa[e]cyclodecen-5-yl ester, [1aR-[1a.alpha.,2.alpha.,5.beta.,5a.beta.,8a.alpha.,9.alpha., 10a.alpha.)]-	$C_{36}H_{56}0_6$	0.06	NR	NR

Article DOI: https://doi.org/10.37284/eajhs.9.1.4294

Retention Time (RT)	Compound Name	Molecular Formula	% Area	Biological activities	Reference
39.9549	5H-Cyclopropa[3,4]benz[1,2-e]azulen-5-one,3,9,9a-tris(acetyloxy)-3-[(acetyloxy)methyl]-2-chloro-1,1a,1b,2,3,4,4a,7a,7b,8,9,9a-dodecahydro-4a,7b-dihydroxy-1,1,6,8-tetramethyl-,[1aR-(1a.alpha.,1b.beta.,2,alpha.,3.beta.,4a.beta.,7a.alpha.,7b.alpha.,8.alpha	$C_{28}H_{37}ClO_{11}$	0.14	NR	NR
40.2034	Vitamin E	$C_{29}H_{50}O_2$	0.42	Antidiabetic, Antioxidant. Hypocholesterolemic, anti-inflammatory, antiaging, anti-coronary, antiulcer, hepatoprotective, antidermatitic, antileukemia, antiaging, anti-alzheimeran, antitumor, anticancer, immunostimulant, analgesic, vasodilator, antispasmodic, anti-bronchitic.	(Ponnamma & Manjunath, 2012; Arora & Meena, 2017)
42.4634	Stigmasterol	$C_{29}H_{48}O$	0.94	Hypoglycemic effects, antioxidant. Antihypercholesterolemic, thyroid inhibitory, antiperoxidative, Precursor of progesterone, acts as intermediate in the biosynthesis of androgens and oestrogens, anti-osteoarthritic, cytotoxic, antitumor, antimutagenic, anti-inflammatory, analgesic	(Sivakumaran et al., 2019; Perumal, et al., 2021)
45.409	Betulinaldehyde	$C_{30}H_{48}O_2$	0.49	Anti-tumor	(Huang et al., 2023)

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DISCUSSION

Qualitative and Quantitative Phytochemical Analysis

This study revealed the presence of alkaloids, steroids, phenols, tannins, flavonoids, coumarins, terpenoids, resins, glycosides, and saponins in plant extracts. In contrast to the study by Adia *et al*. (2016), which reported the absence of alkaloids and flavonoids in MP extracts and alkaloids in CR extracts, this study detected their presence in both methanolic and aqueous extracts. In the process of quantification, noteworthy concentrations of total phenols and tannins were detected in FSM. Conversely, elevated levels of total flavonoids, saponins, and alkaloids were observed in the CRM. Compared with studies conducted in Kenya by Odhiambo *et al.* (2019), this study detected the highest amounts of flavonoids and phenols in CRM.

Many of these phytoconstituents are effective at managing diabetes and hypertension. Alkaloids are a class of organic compounds that contain nitrogen and are produced by a variety of organisms, including plants, animals, fungi, and bacteria. They have a wide range of biological activities and pharmacological effects and are essential bioactive compounds with significant potential in diabetes management. Their mechanisms of action include promoting the translocation of glucose transporter type 4 (GLUT4), enhancing insulin secretion by regenerating pancreatic β-cells, and increasing glucose-stimulated insulin secretion (GSIS) rather than basal insulin secretion (Kumar et al., 2019; Shehadeh et al., 2021). Additionally, alkaloids function as inhibitors of α-amylase and α-glucosidase—key enzymes involved in carbohydrate digestion—making them a critical component in strategies to lower blood glucose levels (Shehadeh et al., 2021). Phenolic compounds exhibit substantial pharmacological potential in the management of diabetes and its associated complications. They possess antioxidant properties, protect pancreatic β-cells, reduce β-cell apoptosis, promote β-cell proliferation, stimulate the pancreas to increase insulin secretion, inhibit glucose absorption, and suppress the activity of digestive enzymes (Sun et al., 2020). The diverse biological activities of alkaloids and phenolic compounds, from increasing insulin secretion to inhibiting carbohydrate digestion, highlight their crucial role in the development of natural and effective strategies for diabetes management.

Bioactive compounds such as flavonoids and saponins play a significant role in diabetes management by targeting multiple physiological pathways to regulate glucose metabolism and enhance insulin function. Flavonoids, prominent secondary metabolites identified in plant extracts, have well-documented potential in preventing diabetes and its complications. Their mechanisms of action include regulating glucose metabolism, modulating hepatic enzyme activity, and improving lipid profiles. Flavonoids support carbohydrate digestion regulation, insulin signalling, insulin secretion, glucose uptake, adipose deposition, β-cell proliferation, apoptosis prevention, and antioxidant activity (Al-Ishaq et al., 2019). Saponins are a group of organic compounds that are found in many plants and have a variety of uses. In diabetes management, saponins play crucial roles in restoring insulin responsiveness, enhancing insulin signalling, and increasing plasma insulin levels. They stimulate insulin release from the pancreas, activate glycogen synthesis, upregulate GLUT4 expression, and inhibit gluconeogenesis and α-glucosidase activity (Elekofehinti, 2015). The diverse mechanisms of flavonoids and saponins highlight their potential as natural therapeutic agents in the prevention and management of diabetes and its associated complications.

Naturally occurring plant compounds such as tannins and coumarins offer promising antidiabetic properties by targeting key metabolic pathways involved in glucose regulation and insulin function. Tannins contribute to diabetes management by enhancing insulin signalling pathways, facilitating GLUT4 translocation to increase glucose uptake, and reducing intestinal glucose absorption. They also promote β-cell regeneration, improve insulin activity in adipose

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tissues, and inhibit the enzymatic activities of α-amylase and α-glucosidase (Shehadeh et al., 2021). Coumarins are a class of organic compounds that are found in plants, fungi, and microorganisms. Coumarins exhibit significant antidiabetic effects by regulating hepatic enzymes, protecting pancreatic β-cells from damage, improving abnormal insulin signalling, and providing antioxidant benefits (Li et al., 2017). The ability of tannins and coumarins to increase insulin signalling, protect pancreatic β-cells, and regulate glucose metabolism underscores their potential as valuable natural agents in diabetes management.

Terpenoids are small secondary plant metabolites and are arguably the most widespread group of natural products. Terpenoids increase insulin levels and glucose uptake in tissues while inhibiting several signalling pathways involved in carbohydrate metabolism. They promote glucose uptake through the upregulation of GLUT4 translocation, strengthen insulin signalling pathways, stimulate insulin secretion, protect pancreatic cells, and increase glycogen storage (Shehadeh et al., 2021). These bioactive compounds collectively demonstrate diverse and complementary mechanisms in addressing diabetes and its complications.

By targeting multiple physiological pathways to regulate blood pressure and improve cardiovascular health, plant-derived phytochemicals play crucial roles in hypertension management. For example, flavonoids have been shown to lower blood pressure and provide protection against coronary heart disease and stroke. They may reduce the formation of atherosclerotic plaques and decrease arterial stiffness, making arteries more responsive to endogenous vasodilation stimuli. Polyphenols exert antihypertensive effects by increasing endothelial nitric oxide bioavailability, which is facilitated by their antioxidant properties and ability to activate vascular endothelial nitric oxide synthase (Kooshki & Hoseini, 2014). Studies have demonstrated that polyphenols and flavonoids, such as those analysed in red wine, can lower blood pressure through in vivo vasorelaxation mechanisms and increase nitric oxide synthesis (Sajjad et al., 2020). Additionally, bioactive compounds such as polyphenols, flavonoids, alkaloids, and saponins have shown in vitro potency in inhibiting angiotensin-converting enzyme (ACE) activity, a key target in hypertension treatment (Sajjad et al., 2020). Generally, various mechanisms have been established for the actions of secondary metabolites and their antihypertensive properties. The potential mechanisms through which plant secondary metabolites contribute to hypertension management include ACE inhibition, diuretic effects, increased nitric oxide production, radical scavenging activity, calcium channel blockade, reduced proliferation of vascular smooth muscle cells, enhanced vasodilation, direct blood pressure-lowering effects, renin inhibition, anti-inflammatory properties, and increased hydrogen sulfide (H_2S) production leading to vasorelaxation. These metabolites may also inhibit the activity of β-adrenoceptors and autonomic ganglion receptors, all of which are critical for therapeutic blood pressure control (Obode et al., 2020; Verma et al., 2021). The diverse mechanisms of action of flavonoids, polyphenols, alkaloids, and other bioactive compounds highlight their potential as natural therapeutic agents for controlling hypertension and reducing the risk of cardiovascular complications.

GCMS Profiles: Antidiabetic and Antihypertensive Potential

GC–MS analysis of *Ficus saussureana* (FS) revealed the presence of fourteen compounds. The methanolic extract of *Clerodendrum rotundifolium* (CR) contained thirty chemical compounds, whereas the extract of *Microglossa pyrifolia* (MP) contained thirty compounds. Most of the compounds identified in these three extracts have also been detected in other medicinal plants, such as *Justicia wynaadensis* (Ponnamma & Manjunath, 2012), *Ceropegia bulbosa* (Arora & Meena, 2017), *Cymodocea serrulata*, *Syringodium isoetifolium,* and *Enhalus acoroides* (Vijayalingam & Rajesh, 2019).

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Some of the compounds identified in this study, such as stigmasterol, squalene, vitamin E, lupeol, and oleic acid, have been investigated for their pharmacological potential. Stigmasterol has also been detected in the leaves of *Pseuderanthemum palatiferum* (Nualkaew et al., 2015) and *Bridelia duvigneaudii* (Credo et al., 2018). This compound has hypoglycemic potential, such as the ability to lower blood glucose levels (Nualkaew et al., 2015; Credo et al., 2018), augment glucose uptake, alleviate insulin resistance, and improve oral glucose tolerance (Wang et al., 2017). Additionally, stigmasterol facilitates the translocation of GLUT4, reduces fasting blood glucose levels, and supports the regeneration of pancreatic β-cells (Bakrim et al., 2022). This compound has a range of other remarkable properties, including antihypercholesterolemic, thyroid-inhibitory, and antiperoxidative properties. It serves as a precursor to progesterone and functions as an intermediate in the biosynthesis of androgens and oestrogens. Additionally, it has anti-osteoarthritic, antitumor, antimutagenic, anti-inflammatory, and analgesic effects (Sivakumaran et al., 2019; Perumal et al., 2021).

Squalene has also been detected in other medicinal plants, such as *Syzygium polyanthum* (Widyawati et al., 2023). This compound has noteworthy potential in the treatment of diabetes and hypertension, such as its ability to reduce fasting blood glucose levels and its antioxidant ability (Widyawati et al., 2023). In addition to its role in regulating blood glucose levels and providing antioxidant benefits, the diverse pharmacological properties of squalene, including its antibacterial, antitumor, chemopreventive, and immunostimulatory activities (Ingole, 2016; Zayed & Samling, 2016), underscore its broad therapeutic potential.

A study by Shreenithi *et al*. (2019) explored the therapeutic potential of lupeol in the treatment of diabetes. This study revealed that lupeol has the capacity to regulate insulin receptor and GLUT4 protein expression. In addition to its role in glucose metabolism, lupeol has broad-spectrum therapeutic effects, including anticancer, anti-inflammatory, anti-arthritic, anti-inflammatory, anti-mutagenic, anti-HIV, antitumor, antihyperlipidemic, anti-flu, antimalarial, and antiviral activities, highlighting its potential as a valuable natural compound for various medical applications (Arora & Meena, 2017; Perumal et al., 2021).

Studies have revealed the therapeutic benefits of vitamin E in diabetes management. It is apparent that supplementation with vitamin E plays a crucial role in delaying the onset of diabetic complications and slowing their progression, lowering fasting blood sugar, postprandial blood sugar, and total cholesterol levels (Jain & Jain, 2012). Vitamin E has been detected in other medicinal plants, such as *Justicia wynaadensis* (Ponnamma & Manjunath, 2012) and *Ceropegia bulbosa* (Arora & Meena, 2017). In addition to its role in glucose regulation, the extensive pharmacological properties of vitamin E, including its hypocholesterolemic, antiinflammatory, antiaging, anticoronary, antiulcer, hepatoprotective, antidermatitic, antileukemia, antiAlzheimer's, antitumor, anticancer, immunostimulatory, analgesic, vasodilatory, antispasmodic, and antibronchitic properties, highlight its broad therapeutic potential in various health conditions (Arora & Meena, 2017; Ponnamma & Manjunath, 2012).

This study revealed the presence of oleic acid, which has been reported to have antidiabetic potential, such as increasing insulin production, reversing the repressive effect of insulin production, and decreasing glucose levels (Vassiliou et al., 2009). Oleic acid also possesses antibacterial activities.

Octadecanoic acid, commonly known as stearic acid, has demonstrated significant potential in the context of diabetes management. Research indicates that it lowers LDL cholesterol levels and improves the total-to-HDL cholesterol ratio (Hunter et al., 2010). Additionally, it has been shown to significantly reduce visceral fat, blood glucose, and leptin concentrations (Shen et al., 2014). A reduction in excess visceral fat is particularly advantageous for individuals with

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type 2 diabetes (Mgbeje & Abu, 2020). In addition to its role in lowering blood glucose and visceral fat, the diverse bioactive properties of stearic acid, including antimicrobial, antibacterial, antifungal, hypocholesterolemic, and antitumor effects, underscore its therapeutic significance in promoting overall health (Ponnamma & Manjunath, 2012; Arora & Meena, 2017).

GC–-MS Profiles: Antioxidant Potential

Oxidative stress, resulting from an imbalance between free radicals and antioxidants, contributes to tissue damage and the development of various health disorders, including diabetes and hypertension. The excessive generation of free radicals exacerbates oxidative stress, leading to complications such as reduced pancreatic beta cell function, endothelial damage, inflammation, and impaired glucose uptake (Vijay & Vimukta, 2014). Furthermore, oxidative stress exacerbates insulin resistance and disrupts insulin secretion, thereby aggravating the progression of diabetes (Bajaj & Khan, 2012). Moreover, evidence suggests a decrease in antioxidant defences in diabetes, attributable to various factors such as diminished levels of specific antioxidants such as vitamin E, a reduction in the overall antioxidant capacity or free radical scavenging activity in plasma/serum, and an increase in plasma oxidizability as well as a decline in the activities of antioxidant enzymes (Bajaj & Khan, 2012). Antioxidants are key chemical and biological compounds that protect cells and tissues against possible injuries caused by reactive radicals. They play a vital role in neutralising reactive species, thereby shielding cells from their harmful effects.

Studies have assessed the value of plant-based antioxidants, which offer potential benefits for disorders linked to oxidative stress, such as diabetes and hypertension (Rahimi et al., 2005). This study detected key phytochemicals with antioxidant activities. Examples of these notable compounds include vitamin E, which prevents lipid peroxidation and protects cells against oxidative damage, contributing to diabetes prevention (Khan et al., 2015; Rajendiran et al., 2018). Squalene, a hydrophilic natural antioxidant, effectively reduces lipid peroxidation in heart tissue (Amarowicz, 2009). Lupeol can reduce protein levels in the pancreas and increase protein levels under diabetic conditions, indicating its ability to decrease oxidative stress and prevent oxidative protein damage associated with diabetes. Lupeol also has the ability to improve pancreatic antioxidants and reduce lipid peroxidation (Gupta et al., 2012). 5-Hydroxymethylfurfural, which was detected in this study, has novel antioxidant activities (Zhao et al., 2013) because of its ability to scavenge ABTS and DPPH radicals; to inhibit AAPH-induced hemolysis; to reduce ROS and MDA; and to increase SOD, CAT, and GPx enzyme activities, which show its ability to prevent peroxidation and protect erythrocytes (Zhao et al., 2013). Phytol has demonstrated significant in vitro antioxidant activity by efficiently scavenging hydroxyl radicals and nitric oxide, while also inhibiting the formation of thiobarbituric acid reactive substances (TBARS) (Santos et al., 2013). Stigmasterol effectively reduces the production of reactive oxygen species (ROS) and enhances the activity of key antioxidant enzymes, such as catalase (CAT), superoxide dismutase (SOD), and nitric oxide synthase enzymes (iNOS and nNOS) (Bakrim et al., 2022). Additionally, n-hexadecanoic acid exhibited DPPH radical scavenging activity (Purushothaman et al., 2024).

The bioactive compounds vitamin E, squalene, lupeol, 5-hydroxymethylfurfural, phytol, stigmasterol, and n-hexadecanoic acid have promising antioxidant potential and can be harnessed in the management of diabetes and hypertension. These compounds exhibit various antioxidant mechanisms, such as scavenging free radicals, reducing reactive oxygen species (ROS), and enhancing the activity of key antioxidant enzymes such as catalase (CAT) and superoxide dismutase (SOD). The ability of these compounds to mitigate oxidative stress—a major contributing factor in the pathogenesis of both diabetes and hypertension—indicates that these natural compounds hold significant therapeutic value. The incorporation of these compounds into

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treatment strategies may help reduce oxidative damage, improve vascular health, and potentially alleviate complications associated with these chronic conditions.

Other compounds have also been reported to possess antioxidant activities, such as 2-methoxy-4-vinyl phenol (Nandhini et al., 2021), tetradecanoic acid (Vijayalingam & Rajesh, 2019), heptadecanoic acid (Ponnamma & Manjunath, 2012), .psi.,.psi.-Carotene, 1,1',2,2'-tetrahydro-1,1'-dimethoxy- (Kavitha 2021), 1,2-benzenediol,3-methyl- (Naksing et al., 2021), bicyclo[5.2.0]nonane,2-methylene-4,8,8-trimethyl-4-vinyl- (Prakasia & Nair, 2015), 1-heptatriacotanol (Kotteswari et al., 2020), hexadecanoic acid, and methyl ester (Ali et al., 2017).

GC–-MS Profiles: Other Potentials

The methanolic extracts of the three plants under investigation revealed the presence of numerous additional bioactive compounds. In addition to their potential utility in managing diabetes and hypertension, these compounds exhibit a wide range of pharmacological activities, contributing significantly to the treatment of various human diseases. Detailed pharmacological profiles of each compound, supported by relevant references, are provided in Tables 3, 4, and 5.

In total, more than 40 other pharmacological activities were identified among the detected chemical compounds. These activities include antimicrobial, antifungal, antitumor, antibacterial, anticancer/chemopreventive, hypocholesterolemic, anti-inflammatory, antigenic, anticoronary, antiarthritic, anthistaminic, hemolytic, antimalarial, antiviral, antihyperlipidemic, thyroid-inhibitory, antimutagenic, antaging, antiulcer, antidermatitic, antileukemic, anti-Alzheimer's, immunostimulant, analgesic, vasodilatory, antispasmodic, antibronchitic, hepatoprotective, antiacne, antandrogenic, antiarteriosclerotic, antianaphylactic, antiprostatic, antimetastatic, diuretic, antiproliferative, anti-HIV, antiulcerogenic, antinociceptive, and antituberculosis activities. These findings underscore the therapeutic versatility and pharmacological potential of the identified compounds in addressing a broad spectrum of human health challenges.

CONCLUSION

The efficacy of medicinal plants in treating human health issues is attributed to the presence of secondary metabolites or phytochemicals. The findings of this study substantiate the assertions of traditional herbalists regarding these three selected plants, revealing compounds with potential antidiabetic and antihypertensive properties, along with their antioxidant capabilities. It could be concluded that *F. saussureana*, *C. rotundifolium*, and *M. pyrifolia* are effective at managing diabetes and hypertension because of their pharmacologically active compounds. Additionally, this study validates the claims of these plants in treating other significant human ailments, as reported in previous studies. However, further research is essential to explore the bioactivity of these compounds, thoroughly understand their mechanisms of action, and evaluate their efficacy in model organisms and in clinical trials. This will enable the development of targeted interventions aimed at enhancing patient outcomes.

List of Abbreviations

AAPH-: 2,2′-azobis(2- amidinopropane) dihydrochloride, ABTS: 2,2′-azinobis-3-ethylbenzothiazolin-6-sulfonic acid, ACE: Angiotensin-converting enzyme, CAT: Catalase, DPPH: 1,1-diphenyl-2-picryhydrazyl, GC–MS: gas chromatography– mass spectrometry, GLUT4: glucose transporter type 4, GPx: glutathione peroxidase, GSIS: glucose-stimulated insulin secretion, HDL: high-density lipoprotein, HIV: human immunodeficiency virus, LDL: low-density lipoprotein, MDA: malondialdehyde, PFTBA: perfluorotributylamine, ROS: reactive oxygen species, RT: retention time, SOD: superoxide dismutase, TBARS: thiobarbituric acid reactive substances.

Ethics Approval and Consent to Participate

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In compliance with ethical standards, research approval and clearance were obtained from the Uganda Christian University Research Ethics Committee (REC) prior to the start of the study (Registration Number: **UCUREC-2023-504**).

Availability of Data and Materials

The datasets used and/or analysed during the current study are available from the corresponding author upon request.

Competing Interests

The authors declare that they have no competing interests to disclose.

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Authors' Contributions

SSN conceptualised the research idea, formulated the methodology, conducted the data collection and analysis, and prepared the manuscript. JK contributed to the review of the research idea, participated in data collection, and critically reviewed the manuscript. MK contributed to shaping the research idea and provided a critical review of the manuscript. All the authors read and approved the final manuscript.

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