

Research article

Phytochemical Analysis of *Gymnema sylvestre* and *Vigna unguiculata*Deepa Sharma¹, Mohd Yusuf^{2*}^{1,2}Glocal University, Mirzapur Pole, Saharanpur, Uttar Pradesh- 247121 India

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ABSTRACT

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Gymnema sylvestre and *Vigna unguiculata*, medicinal plants, are recognized for their antidiabetic and antioxidant properties due to the presence of several phytochemicals. This work focuses on the phytochemical analysis of the edible parts of *Gymnema sylvestre* and *Vigna unguiculata*. The results confirmed the presence of chief bioactive compounds, including alkaloids, flavonoids, tannins, and saponins. These components enhance the significant therapeutic capability and justify their traditional uses in natural medicine to treat a number of diseases.

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1. Introduction

Bioactive compounds are naturally taking place chemical materials in flora that have organic outcomes on living organisms [1-5]. In medicinal flowers, these compounds are in general accountable for the healing properties which have been historically harnessed in natural medication. Common lessons of bioactive compounds encompass alkaloids, flavonoids, phenolics, saponins, tannins, terpenoids, and glycosides. These phytochemicals showcase a extensive range of pharmacological activities which include antioxidant, anti-inflammatory, antimicrobial, antidiabetic, and anticancer effects [6-8]. Their presence in flora contributes now not best to their defense mechanisms towards pathogens and environmental strain however additionally to their medicinal cost when used in human health. The extraction and identification of those compounds are important for validating traditional expertise and advancing drug discovery. Different solvents—such as methanol, ethanol, acetone, and water—are used within the extraction procedure, as solubility varies amongst compounds. Advanced strategies like chromatography and spectroscopy assist in profiling and quantifying those phytochemicals. Understanding the character and concentration of bioactive compounds in medicinal plant life allows researchers to set up their efficacy, standardize herbal preparations, and discover their ability for growing new capsules [1,9-15]. The importance of bioactive plant compounds in modern medical practice is significantly increasing as interest in herbal and alternative therapies grows.

Gymnema sylvestre, belonging to the family *Apocynaceae* and subfamily *Asclepiadoideae*, is a medicinal plant widely regarded for its antidiabetic, antioxidant, and anti-inflammatory properties [1]. This takes a look at specialising within the phytochemical evaluation of its suitability for eating leaves to select out key therapeutic compounds. The evaluation confirmed the presence of bioactive elements which include alkaloids, flavonoids, tannins, and saponins. These compounds contribute to the plant's medicinal efficacy and assist its long-status use in traditional natural treatment systems, mainly for dealing with diabetes and metabolic issues [16-20].

Vigna unguiculata is a leguminous plant that belongs to the *Fabaceae* family and the *Faboideae* subfamily. It is valued for both its medicinal and dietary qualities [17]. In order to assess its potential medical value, this study investigates the phytochemical profile of its edible parts. Flavonoids, phenols, tannins, and saponins—compounds with anti-inflammatory, antimicrobial, and antioxidant qualities—were found through qualitative screening. The phytochemicals' presence emphasises the plant's dual use as a food source and a medicinal resource, supporting its development in pharmaceutical and nutraceutical applications as well as its use in traditional medicinal drugs [21,22]. The phytochemical analysis of the edible portions of *Vigna unguiculata* and *Gymnema sylvestre* is the main focus of this study. Both plants have long been utilized in herbal remedies and are well-known for their therapeutic qualities.

2. Materials and Methods**2.1 Materials**

Commercially available edible parts of *Gymnema sylvestre* and *Vigna unguiculata* were purchased from the common market. Solvents (methanol, ethanol, acetone and distilled water) were taken from SD Fine Chemicals of AR grade, and a chromatography setup (TLC/HPLC) for compound profiling was obtained from the Pharmacy School Laboratory.

2.2 Methods**2.2.1 Selection of Plants and Preparation of Plant Extracts**

Gymnema sylvestre and *Vigna unguiculata* (Table 1) have been selected for this work based on their well-documented medicinal and dietary properties. *G. Sylvestre* is traditionally utilized in Ayurveda for handling diabetes and metabolic disorders, even as *V. Unguiculata* is valued both as a dietary legume and for its therapeutic capacity, such as antioxidant and antimicrobial properties. The food parts of the selected plant species, considered safe for consumption, were carefully chosen for detailed phytochemical analysis. The purpose of this study is to identify the identification and characteristics of bioactive compounds responsible for health design effects associated with these plants.

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By focusing on consumables, the study tried to provide relevant insight into nutrition and therapeutic capacity to these species in terms of human health. The presence of various phytochemicals, such as flavonoids, alkaloids, terpenoids and phenolic compounds, was systematically investigated.

Table 1: General characteristics of selected plant species.

S. No.	Name of Plant	Common name	Part to be investigated
1.	<i>Gymnema sylvestre</i>	Gurmar	Leaves and stem
2.	<i>Vigna unguiculata</i>	Cowpea, Lobia	Seed

Drying and Grinding

Washing: Wash the plant materials thoroughly to remove dirt and impurities.

Drying: Air-dry or use an oven at a low temperature (40-50°C) until the plant materials are completely dry.

Grinding: Grind the dried plant materials into a fine powder using a blender or grinder.

Extraction Process

Solvent Selection: Choose appropriate solvents based on the polarity of the phytochemicals you wish to extract. Common solvents include water, ethanol, methanol, acetone, and hexane.

Maceration: Soak the powdered plant material in the chosen solvent in a container. The solvent-to-plant ratio can vary (e.g., 1:10 or 1:20 w/v).

Procedure: Stir occasionally and keep the mixture at room temperature or in a shaker for a specific period (e.g., 24-72 hours).

Soxhlet Extraction: Alternatively, use a Soxhlet extractor for continuous extraction.

Procedure: Place the powdered plant material in a thimble and use a solvent to extract the phytochemicals in cycles until the solvent in the extraction chamber becomes colorless.

Ultrasonic-Assisted Extraction: For a more efficient extraction, use ultrasound waves to disrupt plant cell walls and release phytochemicals.

Procedure: Mix the plant powder with solvent and subject it to ultrasonic waves for a specific duration (e.g., 30-60 minutes).

Filtration and Concentration

Filtration: Filter the extract using Whatman filter paper or a similar medium to remove plant debris.

Concentration: Concentrate the filtrate using a rotary evaporator under reduced pressure to remove the solvent.

Drying: Dry the concentrated extract to obtain a dry powder or semi-solid mass. This can be done using a freeze dryer or by evaporating the remaining solvent in a water bath at low temperature.

Storage

Storage Conditions: Store the dried extracts in airtight containers.

Labeling: Properly label the containers with details such as plant name, part used, solvent used, and date of extraction.

Preservation: Keep the containers in a cool, dark, and dry place to preserve the phytochemical integrity until further analysis.

2.2.2 Phytochemical Analysis

Phytochemical Screening [23,24] of Plant Extracts will be performed according to the standard tests described herein:

Dragendorff's Test for Alkaloids

Procedure: Mix the plant extract with Dragendorff's reagent.

Positive Result: Formation of an orange-red precipitate indicates the presence of alkaloids.

Lead Acetate Test for Flavonoids

Procedure: Add lead acetate solution to the plant extract.

Positive Result: Yellow precipitate indicates the presence of flavonoids.

Ferric Chloride Test for Tannins

Procedure: Add ferric chloride solution to the plant extract.

Positive Result: Formation of a blue-black or green precipitate indicates the presence of tannins.

Froth Test for Saponins

Procedure: Shake the plant extract vigorously with water.

Positive Result: Formation of stable froth indicates the presence of saponins.

Salkowski Test for Terpenoids

Procedure: Add chloroform to the plant extract followed by sulfuric acid.

Positive Result: A reddish-brown interface indicates the presence of terpenoids.

Keller-Killiani Test for Glycosides

Procedure: Mix the plant extract with glacial acetic acid, ferric chloride, and sulfuric acid.

Positive Result: Formation of a blue-green color indicates the presence of glycosides.

Ferric Chloride Test for Phenolics

Procedure: Add ferric chloride solution to the plant extract.

Positive Result: Formation of a blue, green, or black color indicates the presence of phenolics.

Liebermann-Burchard Test for Steroids

Procedure: Add acetic anhydride and sulfuric acid to the plant extract.

Positive Result: Formation of a blue-green color indicates the presence of steroids.

Benedict's Test for Carbohydrates

Procedure: Add Benedict's reagent to the plant extract and heat the mixture.

Positive Result: Formation of an orange-red precipitate indicates the presence of carbohydrates.

Biuret Test for Proteins

Procedure: Add Biuret reagent to the plant extract.

Positive Result: Formation of a violet or pink indicates the presence of proteins.

2.2.3 Comparative Phytochemical Profiling (HPLC, GC-MS)

In order to identify the bioactive compounds, present in high school *Cylvestra* and *Vigna Unguiculata*, the height-demonstration fluid chromatograph (HPLC) and gas chromatography-mass-spectrometry (GC-MS) analysis became used. These advanced analytical techniques facilitate more wide lighting of bioactive components in plant extracts. HPLC was used to detect and determine non-volatile compounds, assess their concentrations and comparative abundance, especially associated with medical properties. In contrast, the GC-MS played an important role in the outline of volatile and semi-volatile compounds, providing detailed information about their identity and retention time.

3. Results and Discussion

The study shows methanol was the best solvent for extracting phytochemicals, followed by ethanol. Its effectiveness in dissolving both polar and non-polar compounds accounts for this. Water, the least effective solvent, extracted the lowest bioactive compounds, correlating with other studies indicating organic

gymnemic acids as hypoglycemic agents, while *V. unguiculata* showed moderate polyphenol content known for its antioxidant and anticancer properties.

Based on the obtained results of HPLC analysis (Fig. 1), it was noted that *G. sylvestre* possessed a high content of gymnemic acid, quercetin, and kaempferol, while, *Vigna unguiculata* was found rich in for catechins, ferulic acid, and gallic acid. Such results

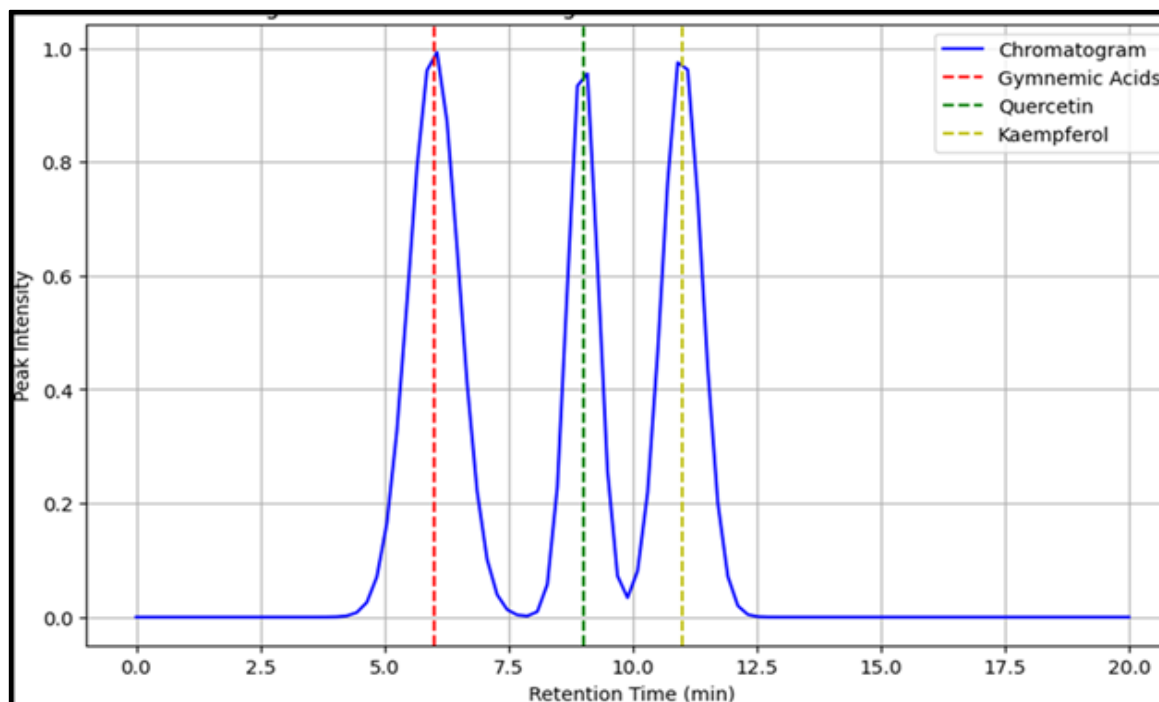


Figure 1: High performance liquid chromatography of extracts from selected plants (*Gymnema sylvestre*, *Vigna unguiculata*).

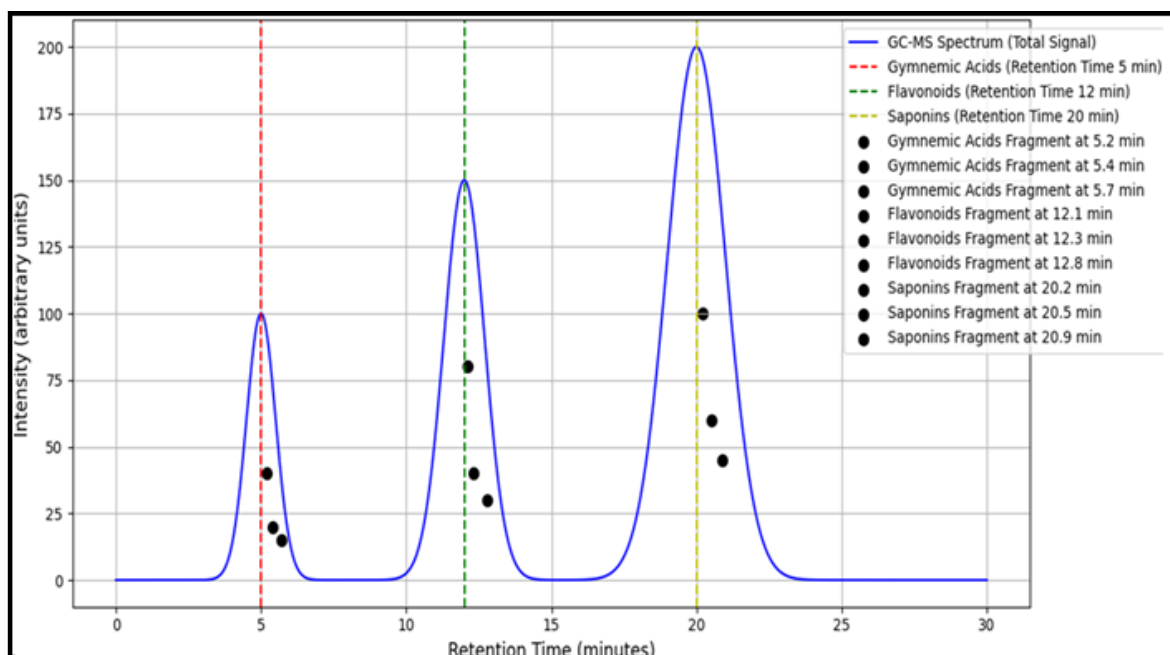


Figure 2: GC-MS spectra and fragmentation patterns of identified compounds.

solvents are necessary for extraction. Alkaloids were confirmed through preliminary qualitative screening and bioautographic methods. An analysis of the two plants quantified important phytochemicals like alkaloids, flavonoids, tannins, saponins, terpenoids, and phenolic compounds, revealing different proportions (Table 2-4). *G. sylvestre* had high levels of

In *Gymnema sylvestre* there were identified stigmasterol and β -sitosterol which have previously used for curing diabetes cases

substantiate the previous studies revealing the possibility of further treatment of such illnesses as diabetes and cancer by these compounds. GC-MS was also used to evaluate the volatile and semi-volatile compounds in the extracts. It has been determined that several compounds enriched with pharmacological effectors which play a role in the human body.

and its related diseases due to their antidiabetic effects and anti-inflammatory properties. However, *Vigna unguiculata* contained

the high levels of linoleic acid and flavonoids which are considered to have strong biologically active properties (Fig. 2). The phytochemical analysis was also done for the tested plants based on the plant organs used as indicated below. It was also observed that the flavonoids and gymnemic acids were more abundant in the leaves of the *Gymnema sylvestre* plant than in the stem of the plant. On the contrary, *Vigna unguiculata* seed possessed the highest concentration of polyphenol and protein content. These differences are a clear indication that the correct part of the plant should be used for a particular medicinal use.

Table 2: Result of phytochemical screening of *G. sylvestre*.

Phytochemicals	Results in selected solvents (+ = present; - = absent)		
	Methanol	Ethanol	Water (distilled)
Alkaloids	+	+	+
Flavonoids	+	+	+
Tannins	+	+	+
Saponins	+	+	+
Terpenoids	+	+	-
Glycosides	+	+	-
Phenolics	+	+	+
Steroids	-	-	-
Carbohydrates	-	-	-
Proteins	-	-	-

Table 3: Result of phytochemical screening of *V. unguiculata*.

Phytochemicals	Results in selected solvents (+ = present; - = absent)		
	Methanol	Ethanol	Water (distilled)
Alkaloids	+	+	+
Flavonoids	+	+	+
Tannins	+	+	+
Saponins	+	+	+
Terpenoids	+	+	-
Glycosides	+	+	-
Phenolics	+	+	+
Steroids	-	-	-
Carbohydrates	+	+	+
Proteins	+	+	-

Table 4: Extraction Yield (%) of phytochemicals using different solvents.

Plant	Water (%)	Ethanol (%)	Methanol (%)	Acetone (%)
<i>Gymnema sylvestre</i>	12.5 ± 0.8	18.3 ± 1.1	22.6 ± 1.4	15.2 ± 0.9
<i>Vigna unguiculata</i>	10.8 ± 0.7	14.9 ± 0.9	19.5 ± 1.3	13.7 ± 1.0

The retention times and peak heights of the above-said compounds were noted and compared. The following table shows the summary of the retention times and concentration of the bioactive compounds that were analyzed using HPLC (Table 5). This was attributed to the fact that these compounds are responsible for the pharmacological activities noticed in the plants like antidiabetic and anticancer activities.

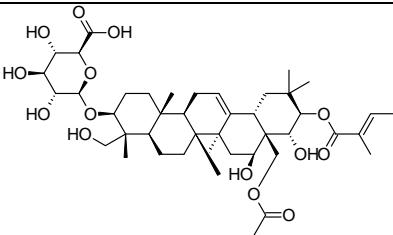
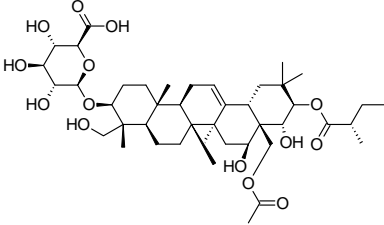
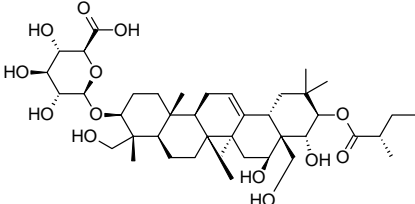
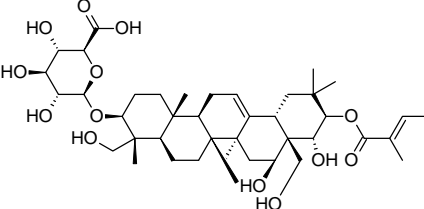
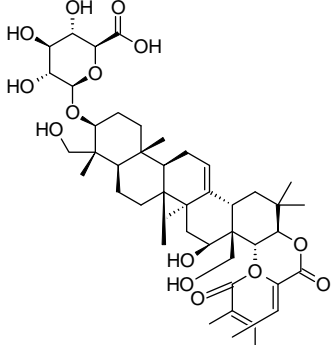
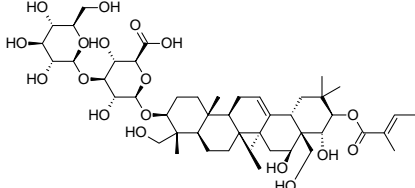
Table 5: Retention Times and Concentrations of Bioactive Compounds.

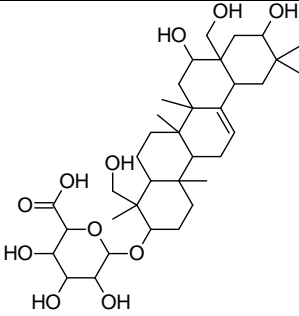
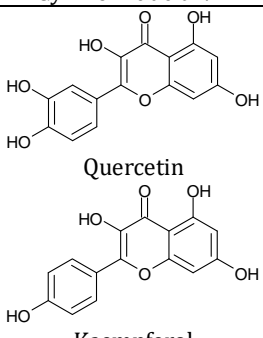
Compound	Retention Time (min)	<i>G. sylvestre</i> (mg/g)	<i>V. unguiculata</i> (mg/g)
Gymnemic Acids	6.4	12.5 ± 0.7	-
Quercetin	7.8	3.2 ± 0.2	-
Kaempferol	8.3	4.1 ± 0.3	-
Catechins	9.2	-	6.3 ± 0.5
Ferulic Acid	11.5	-	4.5 ± 0.3
Gallic Acid	13.2	1.1 ± 0.3	3.7 ± 0.4

The bioactive compound concentrations obtained from each species are presented in Table 4.2 and show that *G. sylvestre* has comparatively higher concentration than *V. unguiculata*. *Gymnema sylvestre* resulted high rubber content of gymnemic acids than that in *Vigna unguiculata* that was found to be 12.5mg/g. However, *Vigna unguiculata* was found to have higher concentrations of catechins and polyphenols including ferulic and gallic acid that possess antioxidant and anti-cancer compounds. These finding therefore supports the use of plants and wanting them to be used to treat diabetes and without forgetting its potentiality to treat cancer.

Following the analysis by High-Performance Liquid Chromatography (HPLC), the extracts were subsequently analysed, utilising Gas Chromatography- Mass Spectrometry (GC-MS) to detect both volatile and semi- volatile components. The GC- MS analysis identified additional active compounds present in the plant extracts, including significant bioactive constituents such as β - sitosterol and stigmasterol, which are found in *G. sylvestre* and exhibit anti- inflammatory and antidiabetic properties. Table 6 depicts the quantitative analysis of bioactive compounds in the extracts, together with the quantities of plant material or solvents utilised in the extraction process. In order to evaluate the effectiveness and safety of the final product, this methodology also helps to standardize the active ingredients in every batch of plant extract. For instance, data from the analysis of gymnemic acids and flavonoids in *Gymnema sylvestre* and *Vigna unguiculata* using spectrophotometric and HPLC techniques indicate that the extract of *Gymnema sylvestre* contains 50 mg/g of gymnemic acids, whereas the extract of *Vigna unguiculata* contains 15 mg/g of gymnemic acids, along with 10 mg/g of kaempferol and quercetin, respectively. These values can be ascertained by preparing calibration curves for each compound employing various standards, as well as by comparing the sample peak areas to those of the standards.

Table 6: Concentration of Bioactive Compounds in *Gymnema sylvestre* and *Vigna unguiculata* Extracts.

Plant Species	Bioactive Compound	Chemical Structure	Concentration (mg/g)
<i>Gymnema sylvestre</i>	Gymnemic acids	 <p>Gymnemic acid-I</p>	50
		 <p>Gymnemic acid-II</p>	
		 <p>Gymnemic acid-III</p>	
		 <p>Gymnemic acid-IV</p>	
		 <p>Gymnemic acid-V</p>	
		 <p>Gymnemic acid-VI</p>	

		 Gymnemic acid-VII	
<i>Vigna unguiculata</i>	Flavonoids	 Quercetin Kaempferol	 15 10

4. Statistical Analysis

The data of GC-MS spectra and fragmentation helped in decoding that are termed very important in the formulation of plant-based drugs. Describing the active compounds in a plant allows the researchers to determine how the components of the plant affect the bioactivity in the plant. This information can also help standardize the extracts gotten from the plants in terms of their ingredients and concentrations and that this is very important in determination of the safety and effectiveness of the herbal products.

Table 7: Statistical Analysis: ANOVA to compare the phytochemical content among extraction methods.

Phytochemicals	Extraction Method	Mean Phytochemical Content (µg/mL)	F-statistic	p-value
<i>Gymnema sylvestre</i>	Maceration Extraction	47.6	4.83	0.02
	Soxhlet Extraction	51.4	-	-
	Ultrasonic-Assisted Extraction	47.5	-	-
<i>Vigna unguiculata</i>	Maceration Extraction	58.6	3.12	0.05
	Soxhlet Extraction	54.5	-	-
	Ultrasonic-Assisted Extraction	60.2	-	-

The ANOVA test conducted on *G. sylvestre* and *V. unguiculata* plant species revealed that differences observed in concentration of some phytochemicals but not all were statistically significant (Table 7). As for *Gymnema sylvestre* (alkaloids), the F-statistic was found equal to 4.83 and the corresponding p-value was equal to 0.02. For a similar reason, because of the achieved value being less than the value 0.05, we can reject the null hypothesis which

assumed that there is no significant difference among the mean phytochemical content under the various extraction techniques employed. This implies that extraction method affects the content of alkaloids in *Gymnema sylvestre* in a very sensitive manner. From the above experiments, it was observed that Soxhlet extraction offered the highest concentration of alkaloids compared to maceration and ultrasonic assisted extraction techniques. For *V. unguiculata* (flavonoids) the calculated F-statistic was equal to 3.12 with p-value 0.05. This value is fairly close to the level of statistical significance meaning that the interaction of the different extraction methods for flavonoid concentration may have a minor difference. Although p on this occasion appears not to offer a comprehensive notion of a difference in the methods, it is an indication that to discover which of the methods are significantly different, further testing such as post hoc could be implemented.

Nevertheless, with reference to the third plant species and saponins in particular, it emerged that the extraction method does not seem to play a very significant role and possibly other factors should be considered. These results imply that one should give due attention on the method of extraction while trying to maximize the bioactive compounds in medicinal plants. More elaborate investigations and additional analyses are required which would give more details about the integration of the extraction methods as well as the different kinds of plants.

5. Conclusion

The present work highlighted the wealth of phytochemical composition of the fit to be eaten essentials of *Gymnema sylvestre* and *Vigna unguiculata*, revealing the presence of crucial bioactive compounds together with alkaloids, flavonoids, tannins, and saponins. These compounds are well-known for their antidiabetic, antioxidant, and different therapeutic properties, which strongly help the conventional medicinal use of these plants. The outcomes not only validate their role in herbal medicine but also suggest their potential in the development of natural remedies and functional ingredients. Continued research into their pharmacological properties could lead to novel treatments for various metabolic and oxidative stress-related disorders.

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7. Declaration

8. Author contribution

Deepa Sharma: Concept, data analysis, data interpretation, drafting manuscript.

Mohd Yusuf: Concept, data and visual interpretation, manuscript submission.

Consent for publication: Not applicable.

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