

eISSN: 2581-3250 CODEN (USA): GBPSC2 Cross Ref DOI: 10.30574/gscbps Journal homepage: https://gsconlinepress.com/journals/gscbps/

(RESEARCH ARTICLE)

GSC Biological and Pharmaceutical Sciences GSC Conline Press RUDIA

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Phytochemical screening of extracts cotton plants (*Gossypium Barbadense*), pumpkin plants (*Cucurbita Pepo*) and valavet bean (*Mucuna Pruriens*) in Fufore Local government of Adamawa State, Nigeria

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GSC Biological and Pharmaceutical Sciences, 2022, 18(03), 100-107

Publication history: Received on 25 January 2022; revised on 03 March 2022; accepted on 05 March 2022

Article DOI: https://doi.org/10.30574/gscbps.2022.18.3.0092

Abstract

Alkaloids, saponins, anthraquinones, glycosides, phenolics, terpenoids and flavonoids distribution in ten medicinal plants belonging to different families were assessed and compared. The medicinal plants investigated were cotton (Gossypium hursitum), Cucurbita pepo (Pumpkin) and velvet bean (Mucuna prupriens). The results of the phytochemical analysis showed the presence of major classes of secondary metabolites such as alkaloids, tannins, saponins, flavonoids, glycosides, phytosterols, carbohydrates, proteins, terpenoids and anthraquinones in ethanolic and aqueous extract. The results of cotton (Gossypium hursitum) leaves and roots in ethanolic extract revealed the presence of alkaloids, tannins, saponins, flavonoids, glycosides, phytosterols, carbohydrates, proteins, and anthraquinones. Terpenoids were absent in the leaves of cotton (Gossypium hursitum), meanwhile Glycosides were absent the roots of cotton (Gossypium hursitum) in ethanolic extracts. The result of aqueous extracts of cotton (Gossypium hursitum) leaves and roots showed the presence of alkaloids, tannins, saponins, flavonoids, glycosides, phytosterols, carbohydrates, proteins, terpenoids and anthraquinones. While the roots revealed the absence of protein and terpenoids. Similarly, the Phytochemical screening result of Cucurbita pepo (Pumpkin) leaves and roots of ethanolic and aqueous extracts revealed the presence of some bioactive components in the extracts such as alkaloids, tannins, phytosterols, carbohydrates, proteins and anthraquinones. Whereas saponins, flavonoids, glycosides, terpenoids were absent in the ethanolic extracts of Cucurbita pepo (Pumpkin) leaves. And also, saponins, glycosides and terpenoids were absent in the roots of Cucurbita pepo (Pumpkin). While the aqueous extract of the leaves revealed all the phytochemical being carried out except saponins, flavonoids and Terpenoids are absent. Also, the aqueous extracts of Cucurbita pepo (Pumpkin) roots revealed the presence of some secondary metabolites as shown in table IV such as alkaloids, tannins, flavonoids, phytosterols, carbohydrates, proteins, terpenoids and anthraquinones. Saponins and glycosides were absent. The results of the velvet bean (*Mucuna prupriens*) leaves and roots of ethanolic extract showed that alkaloids, saponins, phytosterols, carbohydrates, proteins, terpenoids and anthraquinones were present both in leaves and roots with tannins present only in the roots. However, flavonoids, glycosides, were absent in both leaves and roots of velvet bean (Mucuna prupriens). The result of aqueous extract of velvet bean (Mucuna prupriens) leaves and roots revealed the presence of alkaloids, saponins, flavonoids, phytosterols, carbohydrates, proteins, terpenoids and anthraquinones while tannins, glycosides were found absent. Similarly, the aqueous roots extract revealed all the phytochemical tested except flavonoids, terpenoids were absent in the roots of velvet bean (Mucuna prupriens). Results of this study may provide a foundation for designing new drugs for several diseases.

Keywords: Phytochemical screening; Ethanoic extract; Aqueous extract; Medicinal plants

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1. Introduction

Medicinal plants are of great importance to the health of individuals and communities. The medicinal value of these plants lies in some chemical substances that produce a definite physiological action on the human body). The most important of these bioactive constituents of plants are alkaloids, tannins, flavonoids, and phenolics compounds. Many of these indigenous medicinal plants are used as spices and food plants. They are also sometimes added to foods meant for pregnant and nursing mothers for medicinal purposes [6].

Over the years, plant extracts and plant-derived medicines have made immense contributions to the overall health and well-being of human beings. The antimicrobial ability of plant extracts and oils has established a platform for the processing and transformation of these plant products into pharmaceuticals, preservatives and natural medicine. From time immemorial, the use of plants for the treatment of diseases has been widely accepted due to their healing properties. According to World Health Organization, many microorganisms are developing resistance to several drugs which is posing severe threat to the general public health hence requires actions across all sectors to curb this menace [16].

Gossypium hirsutum known as upland cotton belongs to the family *Malvaceae*. It is a perennial shrub that grows to approximately 1.5-2 m and has the potential to develop leaves, stem, flowers, fruits and seeds all at the same time. The leaves have long petiole, cordate, weakly 3-5 lobed, the lobed are broadly triangular to ovate, acute to acuminate. The flowers are large and showy with cream to pale yellow petals, sometimes with red spot at the base of the petals. The capsules have 3-5 valves with a smooth surface and many black gland dots. This yield white or brown lint with seeds embedded. The seed vary from black and smooth to green with tightly adhering fuzz [6]. *Gossypium hirsatum* is propagated by seed. The seed will germinate in 14-21 days at 20-23°C [5].

Pumpkin is one such plant that is frequently being used as food as well as traditional medicine for long days (Muchirah *et al.*, 2018). Cucurbita pepo is an herbaceous plant, belonging to a gourd family, Cucurbitaceae. The plant is good source of nutrients such as vitamin A and C. In many parts of the world, *C. pepo* has been used to treat tapeworm infection, hypertrophy of the prostate, urinary problems, and burns Plants are known to contain innumerable biologically active compounds, which possess antibacterial, antidiabetic, anticancer, antioxidant activities. The seeds are used as a vermifuge, treat problem of the urinary system, hypertensions, prevents the formation of kidney stones, alleviate prostate diseases, and enhanced the erysipelas skin infection [19]. One hundred and nineteen secondary plant metabolites derived from plants are used as drugs globally. Therefore, it is imperative and of utmost significance to carry out a screening of these plants in order to validate their use in folk medicine and reveal the active principle by isolation and characterization of their constituents. However, bioactive compounds present in this plant are yet to be identified. Hence, the present study was designed to investigate for the presence of various phytochemicals in the leaf of C. pepo which evokes various therapeutic effects [2].

The genus *Mucuna*, belonging to the Fabaceae family, sub family Papilionaceae, includes approximately 150 species of annual and perennial legumes. Among the various under-utilized wild legumes, the velvet bean *Mucuna pruriens* is widespread in tropical and sub-tropical regions of the world. It is considered a viable source of dietary proteins due to its high protein concentration (23–35%) in addition its digestibility, which is comparable to that of other pulses such as soybean, rice bean, and lima bean. It is therefore regarded a good source of food [12].

Velvet beans (*Mucuna pruriens var.* utilis (Wall. ex Wight) L.H.Bailey) are an important constituent in the Ayurvedic system of medicine, and forward to be having aphrodisiac effect on against, parkinsonism, lymphoedema and it is a good source of L-DOPA. With reference to the nutritional value, this wild legume can supply significant amount of energy, vitamins and minerals in addition to protein (24-31%). More importantly other parts of this plant are also used for medicinal purposes, e.g. trichomes of pods are used for deworming, decoction of root to contain delirium, root powder as a diuretic and anti-inflammatory agent. Similarly, the paste of fresh root is used in the treatment of lymphoedema (Kalidass and Mahapatra 2014). The toxicity of unprocessed velvet bean may explain why the plant exhibits low susceptibility to insect pests. Velvet bean is well known for its nematicidic effects; it also reportedly possesses notable allelopathic activity, which may function to suppress competing plants [12].

2. Material and methods

2.1. Description of Study Area

This study was conducted in Fufore local Government Area of Adamawa State; Fufore is located in Adamawa State, it has a land area of 4972Km2 and a population of 209,460 and a density of 42.1 inch. It is 26km away from Yola and lies between latitude 9° 13'N and longitude 12° 39'E of Green which meridian. The area experiences distinct dry and wet season with temperature and humidity varying with season, average annual rainfall of 750-100mm between April to October is experienced and dry season period between November to march characterized by dry, dusty and hazy north east trade wind. Temperatures are relatively high through the year about 30 -40°c. The site of this study include: Parda, Dasin Bata and Dasin Hausa Wuro biriji, Beli chutti and Giere all in Fufore Local Government Area of Adamawa State.

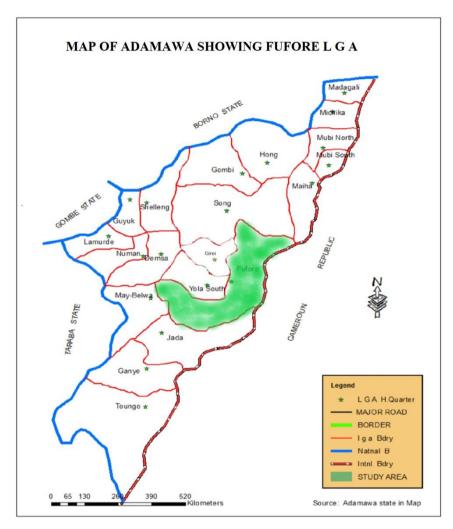


Figure 1 Map of Adamawa State showing the study Area

2.2. Chemicals and Reagents

Unless otherwise stated all chemicals and reagents used were of analytical grade and purchased from Scientific Laboratory, Jimeta-Yola, Adamawa State-Nigeria.

2.2.1. Sample collection

Cotton (*Gossypium hursitum*), *Cucurbita pepo* (Pumpkin) and velvet bean (*Mucuna prupriens*) leaves and roots were collected from the bush area of Fufore Local Government Area of Adamawa State, Nigeria.

2.2.2. Preparation of sample

The leaves and the roots of cotton (*Gossypium hursitum*), *Cucurbita pepo* (Pumpkin) and velvet bean (*Mucuna prupriens*) plant were washed and air dried in Chemistry laboratory, School of Science and Technology, Adamawa State Polytechnic, Yola under shade at room temperature and was weighed and ground to get a coarse powder form using sterile mortar and pestle. The powder was stored in an air tight container and was used for successive analysis [10]

2.2.3. Plant preparation and extraction

100 g each of the leaves and roots of cotton (*Gossypium hursitum*), *Cucurbita pepo* (Pumpkin) and velvet bean (*Mucuna prupriens*) leaves powder were weighed and extracted using ethanol and aqueous solvent in air tight container for 24 hours. The resultant mixture was filtered with filter paper (Whatman No. 1) under gravity. The filtrate was dried at 60°C on a water bath to yield cotton (*Gossypium hursitum*), *Cucurbita pepo* (Pumpkin) and velvet bean (*Mucuna prupriens*) leaf aqueous extract residue [1].

2.3. Phytochemical Screening

Phytochemical screening was performed using standard procedures. By using different specific reagents, the presences of main groups of natural products were detected in ethanolic and aqueous extracts of cotton (*Gossypium hursitum*), *Cucurbita pepo* (Pumpkin) and velvet bean (*Mucuna prupriens*) leaves and roots.

2.3.1. Test for alkaloids

Mayer's test: To a few ml of plant sample extract, two drops of Mayer's reagent are added along the sides of test tube. Appearance of white creamy precipitate indicates the presence of alkaloids [21].

2.3.2. Test for phenolic compounds and tannins

Ferric Chloride test: The extract (50 mg) is dissolved in 5 ml of distilled water. To this, few drops of neutral 5% ferric chloride solution are added. A dark green colour indicates the presence of phenolic compound alkaloids [22].

2.3.3. Detection of saponins

Foam test: small amount of extract was shaken with little quantity of water. If foam produced persists for ten minutes it indicates the presence of saponins [9].

2.3.4. Test for flavonoids

Dilute ammonia (5 ml) was added to a portion of an aqueous filtrate of the extract. Concentrated sulphuric acid (1 ml) was added. A yellow coloration that disappears on standing indicates the presence of flavonoids [4].

2.3.5. Detection of cardiac glycosides

Killer-killani test: To 0.5 gm of extract diluted to 5ml with distilled water and add 2 ml of glacial acetic acid and containing one drop of ferric chloride solution. This was underplayed with 1 ml of conc. sulphuric acid. Brown ring at the interface indicates the presence of a deoxy sugar characteristic of cardenolides [9].

2.3.6. Detection of phytosterols

Salkowski's Test: Extracts were treated with chloroform and filtered. The filtrates were treated with few drops of Conc. Sulphuric acid, shaken and allowed to stand. The appearance of golden yellow colour indicates the presence of triterpenes [9].

2.3.7. Test for reducing sugars (fehling's test)

The aqueous ethanol extract (0.5 g in 5 ml of water) was added to boiling Fehling's solution (A and B) in a test tube. The solution was observed for a colour reaction [4].

2.3.8. Detection of proteins and aminoacids

Xanthoproteic Test: The extracts were treated with few drops of concentrated Nitric acid solution.

Formation of yellow colour indicates the presence of proteins [9].

2.3.9. Test for terpenoids (salkowski test)

To 0.5 g each of the extract was added 2ml of chloroform. Concentrated H2S04 (3 ml) was carefully added to form a layer. A reddish brown colouration of the interface indicates the presence of terpenoids [4].

2.3.10. Test for anthraquinones

0.5 g of the extract was boiled with 10 ml of sulphuric acid (H₂SO4) and filtered while hot. The filtrate was shaken with 5 ml of chloroform. The chloroform layer was pipetted to another test tube and 1 ml of dilute [4].

2.3.11. Solvent extraction

A quantity of 200g plant powder was extracted in 200ml of ethanol (95% w/v) for 24hours, strained and the extract was concentrated to dryness at 60° c in a water bath. The same procedure was used for aqueous extraction. The extracts were then refrigerated at 4° c prior to use [20].

3. Results

Table 1 Phytochemical Screening Result of cotton (*Gossypium hursitum*), *Cucurbita pepo* (Pumpkin) and velvet bean(*Mucuna prupriens*) leaves Ethanolic Extract

S/N	Phytoconstituent	Cotton	pumpkin	Velvet bean
1	Alkaloids	+	+	+
2	Tannins	+	+	-
3	Saponins	+	-	+
4	Flavonoids	+	-	_
5	Glycosides	+	-	-
6	Phytosterols	+	+	+
7	Carbohydrates	+	+	+
8	Proteins	+	+	+
9	Terpenoids	-	_	+
10	Anthraquinones	+	+	+

Key: +: Present; -: Absent

Table 2 Phytochemical Screening Result of cotton (*Gossypium hursitum*), *Cucurbita pepo* (Pumpkin) and velvet bean(*Mucuna prupriens*) roots Ethanolic Extract

S/N	Phytoconstituent	Cotton	pumpkin	Velvet bean
1	Alkaloids	+	+	+
2	Tannins	+	+	+
3	Saponins	+	_	+
4	Flavonoids	+	+	_
5	Glycosides	_	_	_
6	Phytosterols	+	+	+
7	Carbohydrates	+	+	+
8	Proteins	+	+	+
9	Terpenoids	+	_	+
10	Anthraquinones	+	+	+

Table 3 Phytochemical Screening Result of cotton (G	ossypium hursitum), Cucurbita pepo (Pumpkin) and velvet bean
(Mucuna prupriens) leaves Aqueous Extract	

Phytoconstituent	Cotton	pumpkin	Velvet bean
Alkaloids	+	+	+
Tannins	+	+	-
Saponins	+	_	+
Flavonoids	+	_	+
Glycosides	+	+	-
Phytosterols	+	+	+
Carbohydrates	+	+	+
Proteins	+	+	+
Terpenoids	+	-	+
Anthraquinones	+	+	+
	Alkaloids Tannins Saponins Flavonoids Glycosides Phytosterols Carbohydrates Proteins Terpenoids	Alkaloids+Tannins+Saponins+Flavonoids+Glycosides+Phytosterols+Carbohydrates+Proteins+Terpenoids+	Alkaloids++Tannins++Saponins+-Flavonoids+-Glycosides++Phytosterols++Carbohydrates++Proteins++Terpenoids+-

Key: +: Present; -: Absent

Table 4 Phytochemical Screening Result of Cotton (Gossypium hursitum), Cucurbita pepo (Pumpkin) and velvet bean(Mucuna prupriens) roots Ethanolic Extract

S/N	Phytoconstituent	Cotton	pumpkin	Velvet bean
1	Alkaloids	+	+	+
2	Tannins	+	+	+
3	Saponins	+	_	+
4	Flavonoids	+	+	-
5	Glycosides	+	-	+
6	Phytosterols	+	+	+
7	Carbohydrates	+	+	+
8	Proteins	_	+	+
9	Terpenoids	_	+	-
10	Anthraquinones	+	+	+
Key: +: Present; -: Absent				

4. Discussion

4.1. Preliminary phytochemical screening

The extracts of cotton (*Gossypium hursitum*), Pumpkin (*Cucurbita pepo*) and velvet bean (*Mucuna prupriens*) leaves and roots were screened for the qualitative determination of secondary metabolites. Results of the phytochemical analysis showed the presence of major classes of secondary metabolites such as alkaloids, tannins, saponins, flavonoids, glycosides, phytosterols, carbohydrates, proteins, terpenoids and anthraquinones in ethanolic and aqueous extract.

The results of cotton (*Gossypium hursitum*) leaves and roots in ethanolic extract revealed the presence of alkaloids, tannins, saponins, flavonoids, glycosides, phytosterols, carbohydrates, proteins, and anthraquinones. Terpenoids were absent in the leaves of cotton (*Gossypium hursitum*). The roots of cotton (*Gossypium hursitum*) in ethanolic extracts revealed all the phytochemicals above except Glycosides were absent. The preliminary phytochemical screening of aqueous extracts of cotton (*Gossypium hursitum*) leaves and roots as in table III and IV showed the presence of alkaloids, tannins, saponins, flavonoids, glycosides, phytosterols, carbohydrates, proteins, terpenoids and anthraquinones. While

the roots revealed the absent of protein and terpenoids. This work is in line with the reported by Chinweuba *et al.*, (2013). Preliminary Study on the Phytochemical Constituents of *Gossypium hirsatum* Leaves. Also agreed with the work reported by Ezeifeka *et al.*, (2017) In-vitro Antiviral Activity of *Gossypium hirsatum* on Newcastle Disease Virus.

Similarly, the Phytochemical screening result of *Cucurbita pepo* (Pumpkin) leaves ethanolic extract extracts revealed the presence of some bioactive components in ethanolic extracts such as alkaloids, tannins, phytosterols, carbohydrates, proteins and anthraquinones. Whereas saponins, flavonoids, glycosides, terpenoids were absent in the ethanolic extracts of Cucurbita pepo (Pumpkin) leaves. The phytochemical result of *Cucurbita pepo* (Pumpkin) roots ethanolic extracts revealed the presence of alkaloids, tannins, flavonoids, phytosterols, carbohydrates, proteins and anthraquinones. Were saponins, glycosides and terpenoids are absent in the roots of *Cucurbita pepo* (Pumpkin). The preliminary phytochemical screening of aqueous extracts of *Cucurbita pepo* (Pumpkin) leaves showed the presence of following phytochemicals alkaloids, tannins, glycosides, phytosterols, carbohydrates, proteins, and anthraquinones. Were saponins, flavonoids are absent.

The result of aqueous extracts of *Cucurbita pepo* (Pumpkin) roots revealed the presence of some secondary metabolites as shown in table IV such as alkaloids, tannins, flavonoids, phytosterols, carbohydrates, proteins, terpenoids and anthraquinones. Saponins and glycosides were absent. This research agreed with work reported by Arul and Saravanan (2017) on the Phytochemical and GC-MS Studies on Traditional Herbaceous Plant Pumpkin (*Cucurbita Pepo*) Leaf.

Phytochemical screening of the velvet bean (*Mucuna prupriens*) leaves and roots ethanolic extract showed that alkaloids, saponins, phytosterols, carbohydrates, proteins, terpenoids and anthraquinones were present both in leaves and roots with tannins present only in the roots. However, flavonoids, glycosides, were absent in both leaves and roots of velvet bean (*Mucuna prupriens*). Tannins were not present in the leaves of ethanolic extract. The results from the phytochemical analysis of aqueous extract velvet bean (*Mucuna prupriens*) leaves and roots revealed the presence of various bioactive secondary metabolites that were confirmed from their chemical colour reaction tests as shown in Table III and IV.

Velvet bean (*Mucuna prupriens*) leaves extract showed the presence of alkaloids, saponins, flavonoids, phytosterols, carbohydrates, proteins, terpenoids and anthraquinones while tannins, glycosides were found absent. Similarly, the aqueous roots extract revealed the presence of alkaloids, tannins, saponins, glycosides, phytosterols, carbohydrates, proteins, and anthraquinones while flavonoids, terpenoids were absent in the roots of velvet bean (*Mucuna prupriens*). Similarly, the results obtained in the present study are seen to be accordance with the previous studies on Phytochemical Analysis and Antimicrobial Activity of methanol Extract of the Leaves of *Mucuna utilis* (Velvet Beans) by Mercy and Sunday (2015). And also, in accordance with the work on *Hibiscus Esculentus* reporting change in phytochemical constituents due to variation in geographical location by [2].

5. Conclusion

The plants studied here can be seen as a potential source of useful drugs. Further studies are recommended on these plants in order to isolate, identify, characterize and elucidate the therefore the plants in this study are validated as medicinal. This present work seen to be accordance with the previous work on "Phytochemical Screening and Antioxidant Activities of Seed and Callus Extracts of *Mucuna Pruriens* (L) by Ramachandra Kumar.

Compliance with ethical standards

Acknowledgments

We are grateful to the, Tertiary Education Trust Fund (TETFUND) management, Adamawa State Polytechnic, Management of Fufore Local Government Area for the permission to undertake this work. Immense gratitude is also extended to the Department of Science Laboratory Technology, Adamawa State Polytechnic, Yola for liberal provision of laboratory facilities.

Disclosure of conflict of interest

We declare that there are no conflicts of interest in connection with this paper.

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