

Phytochemical Screening and Nutritional Constituents of *Cleome Viscosa* Root

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Abstract: *Cleome viscosa* known as asian spider flower or yellow spider flower in English, it's known as "wild mustard" the plant is called *Namijin yar anguwa* in hausa and 'eya'zo' in nupe. The plant is an annual sticky herb with, it belongs to capparaceae family it's grow up to 30 - 60 cm height the flower is yellow, they occur in the road side, grassland and sandy soil. The plant has many ethnomedicinal uses. This study investigated the phytochemical screening, proximate analysis and mineral analysis of *C. viscosa* root with the view of the nutritional values. The result of the phytochemical screening shows that alkaloids, flavonoids, phenols, terpenoids, steroids, glycosides and tannins are present but saponins are absent using ethanol as solvent extract. The proximate analysis result shows that the ash content was obtained as (5.0%), moisture content (6.5%), percentage crude protein (2.77%), percentage carbohydrate (15.73%), crude lipid (35.0%), and crude fibre (35.0%). The crude lipid and fibre with higher value shows that the *C. viscosa* is rich source of fibre and lipid. The estimated energy value was calculated as 355.76 kca/g show that *C. viscosa* root can be considered as calorie – dense food source. The result of mineral analysis of the has shown that *C. viscosa* root is rich in mineral, seven micro and macro mineral were detected which their concentrations were detected as Ca (0.74µg/ml), K (1.32µg/ml), Na (1.95µg/ml), Cu (0.044µg/ml), Mn (0.1905µg/ml), Fe (2.592µg/ml) and Zn (0.0028 µg/ml). Therefore, this study recommended that *C. viscosa* root can be included into day to day diet to improve nutrient intake and promote overall health and growth.

Keywords: *Cleome Viscosa*, Root, Ethnomedicinal, Phytochemicals, Proximate, Nutritional Value, Mineral, Analysis

1. Introduction

For many centuries plants are used as source of medicines in traditional and modern system. Many plants serve as herbs that have been investigated to contain phytochemicals are used to cure several diseases and also serve as a source of medicine by human kind since the ancient times. The indigenous knowledge of many of these plants has been documented and eventually become organized systems of medicine such as Ayurveda, Siddha, Unani, and other many more [1]. In Nigeria, in all her six geopolitical regions more especially among the rural people used some plants parts as source of medicine for treatment of many illness and diseases such as fever, headache, diarrhea, for wound healing and many more [2]. Plants also serve as sources of nutrients for proper growth and development of the body systems. Several medicinal plants have been documented to have essential macro and micro nutrients which can also augment the

pharmacological functionality of the phytochemicals found in the plants.

Cleome viscosa Linn commonly known as Asian spider flower or yellow spider flower in English, it is also known as "wild or dog mustard". The plant is called '*namijin yar anguwa*' in Hausa and 'èyà'zo' in Nupe. It belongs to the Capparaceae family that comprises of about 150 - 200 species of which 50 are indigenous to Africa. It is an annual sticky herb that grows to about 30 - 60cm in height, the stem is grooved, densely clothed with glandular and simple hairs [3]. Leaves of the plant are 3 - 5 foliolate. Lower leaves petioles are 2.5 - 5 cm long gradually becoming shorter upwards. The bracts are subsessile. Leaflets are elliptical-oblong or obovate, acute or obtuse. Petioles are short and hairy. Flowers are yellow in colour, axillary, growing out into a lax raceme. Pedicels are slender, terete and hairy. Sepals are 4.5 cm long oblong-lanceolate, glandular-pubescent outside. Petals are oblong-obovate, about 12 mm

long, veined. Stamens are more than 20 in number. Capsules 5 - 6.3 by 0.4 cm. erect, hairy, obliquely striate, compressed, tapering towards both ends, terminated by a style 3 mm. The leaves are diaphoretic, rubefacient and vesicant, the plant is capable of withstanding high daytime temperatures, intense sunlight, and drought [4].

C. viscosa plant has been considered as vegetable which contain both macro and micro nutrients that include fatty acids; palmitic, stearic, oleic, linoleic, etc, vitamin C and A, minerals; calcium, iron, etc and protein. The seeds are used to make feeds for livestock [5]. The plant also contains phytochemicals such as free gallic acid, gallotannins, iridoid, alkaloids, flavonoids, saponins, terpenoids and polyphenolic compounds [6-8]. Extracts especially from the leaves and the seeds of the plant have been used traditionally for treatment of various disorders such as diarrhea, fever, inflammation, liver diseases, bronchitis, skin diseases, and malarial fever [9]. These extracts have been investigated to be anthelmintic, antiseptic, carminative, antiscorbutic, sudorific, febrifuge, [1] cardiac stimulant, immunomodulatory, antipyretic, psychopharmacological, anti-diarrheal, anti-inflammatory, anticonvulsant [10] hepatoprotective, and anti-diarrheal activity [11, 12].

2. Materials and Methods

2.1. Collection of Plant Material

The fresh plant *Cleome viscosa* were collected from Sokoto state. Nigeria. The collected specimen was authenticated by Department of plant science and Biotechnology, Federal University Gusau, Zamfara state with voucher no.: FUG/BIO/HEB/2022/0079 and it was identified by Aliyu Sharhabilu Aminu a senior technologist.

2.2. Preparation of Plant Extracts

The *Cleome viscosa* was grounded in a clean and dry mortar and pestle into fine powder, the grounded sample was weighed as 100g and then stored in polyethene bag for further analysis.

2.3. Methods

The methods used in this work were described and recommended by Association of Official Analytical Chemists (AOAC).

2.3.1. Soaking of Plant Material

20gm of the powered sample of roots the *C. viscosa* was added into 250ml conical, 150ml ethanol was added to the plant material and then cover with aluminium foil paper for 24hrs, after 24hrs the extract was filtered using whatman No. 42 filter paper and filtrate for further use.

2.3.2. Phytochemical Screenings

Phytochemical screening was carried out on the filtrate (the crude extract) of the root of *C. viscosa* to check the presence of the following phytochemicals; saponins, flavonoids, tannins, glycoside, steroids and alkaloids.

(i). Test for Alkaloids (Wagner's and Hager's Test)

2ml of the ethanol extract (filtrate) was measured and transferred into a test tube and 5 drop of Wagner's reagent was added. A reddish brown precipitate indicates the presence of alkaloids [13]. And similarly same method was performed by drop 5 drop of Hager's reagent. A yellow precipitate indicates the presence of alkaloids [14].

(ii). Test for Flavonoids (Alkaline Test)

2ml of the ethanol extract was measured and then transferred into a test tube, 2ml of 2% NaOH was added, and the mixture turn to yellow and 2 drop of dilute acid was added the colour then disappeared. This indicates the present of flavonoids [15].

(iii). Test for Phenols (Ferric Chloride Test)

2ml of the ethanol extract was measured and then transferred into a test tube, 2ml of distilled water was added, followed by 10% FeCl₃ solution. A bluish black colour indicates the presence of phenol [16].

(iv). Test for Terpenoids (Sulphuric Acid Test)

2ml of the ethanol extract was measured and then transferred into a test tube, 2ml of chloroform was added into it, 2ml of H₂SO₄ was also added then it formed a layer in the test tube. The presence of a reddish brown colouration at the interface indicates presence of terpenoids [17].

(v). Test for Steroids

2ml of the ethanol extract was measured and then transferred into a test tube, 2ml of chloroform was added into it, and 2ml of conc H₂SO₄ was also added. In the lower chloroform layer, a red colour indicates the presences of steroids [18].

(vi). Test for Glycoside (Salkowski's and Keller-Kilani Test)

Salkowski's Test

2ml of the ethanol extract was measured and then transferred into a test tube, Then 2ml of chloroform was added and the few drops of conc H₂SO₄ was added and the mixture was shake. Formation of reddish brown colour indicates the presence of glycosides [16].

Keller-kilani Test

2ml of the ethanol extract was measured and then transferred into a test tube, 2ml of acetic acid was added and few drops of 2% FeCl₃ solution. The mixture was poured into a test tube containing 2ml conc H₂SO₄. A brown ring at the interface/junction indicates the presence of glycosides [17].

(vii). Test for Tannins (Ferric Chloride Test)

2ml of the ethanol extract was measured and then transferred into a test tube, few drops of 2% FeCl₃ solution was added. Formation of blue colour indicates the presence of hydrolysable tannins [15].

(viii). Test for Saponins (Emulsion Test)

2ml of the ethanol extract was measured and then transferred into a test tube then 2ml of distilled water was also added and shake vigorously for a stable persistent froth. Formation of emulsion indicates the presence of saponins

when few drops of olive oil was added [17].

2.3.3. Proximate Analysis

Proximate analysis refers to the determination of the major food constituents of plant extract. The analysis partitions nutrients into six components: moisture, ash, and crude protein, crude lipid, crude fiber and carbohydrate.

(i). Determination of the Percentage of Moisture Contents

The weight of crucible was measured using the digital weighing balance and recorded as W_1 (g), 2g of the powdered *C. viscosa* sample was added in the crucible, then weighed and recorded as W_2 . The crucible containing the sample was placed into the oven and heated at 105°C for 2 hours. The sample was then removed from the oven, it was cooled in a desiccator for at least an hour and then weighed as W_3 . The equation (1) was used to determine the percentage of moisture content was calculated using [19].

$$\% \text{ moisture} = \frac{W_2 - W_3}{W_2 - W_1} \times 100 \quad (1)$$

W_1 = Weight of crucible

W_2 = Weight of the crucible and sample before heating

W_3 = Weight of crucible and sample after heating

(ii). Determination of the Percentage of Ash Contents

The weight of crucible was measured using the digital weighing balance and recorded as W_1 (g), 2g of the powdered *C. viscosa* sample was added in the crucible, then weighed and recorded as W_2 . The crucible containing the sample was placed into the muffle furnace and burnt at 550°C for 2 hours. The sample was then removed from the muffle furnace, it was cooled in a desiccator for at least an hour and then re-weighed as W_3 . The equation (2) was used to determine the percentage of ash content was calculated using [19].

$$\% \text{ Ash} = \frac{W_3 - W_1}{W_2 - W_1} \times 100 \quad (2)$$

W_1 = Weight of crucible

W_2 = Weight of the crucible and the sample

W_3 = Weight of crucible after ashing

(iii). Determination of Percentage Crude Lipid

2g of the powdered *C. viscosa* sample was weighed using weighing balance and then transferred into a 200ml beaker, 100ml of petroleum ether was measured and added to the beaker the sample, the beaker is then cover with aluminium foil paper and then kept for 24hrs in a room temperature 25°C. After soaking for 24hrs the sample was then transferred into the crucible and was weighed as W_1 . It was then placed into the oven and heated at the 105°C for 1hr and the sample was then removed in the oven, it was then cooled in a desiccator for at least an hour and was then re-weighed and recorded as W_2 . the crude lipid was then calculated using [19].

$$\% \text{ Crude Lipid} = \frac{W_1 - W_2}{W_1} \times 100 \quad (3)$$

W_0 = Weight of dried sample taken for the test

W_1 = Weight of crucible and 2g of sample after soaking

W_2 = Weight of crucible and 2g of sample after soaking and heating

(iv). Determination of Percentage Crude Fiber

2g of the powdered *C. viscosa* sample was weighted using weighing balance and then transferred into 200ml beaker, 100ml of H_2SO_4 was added into the beaker containing the sample and boiled, after boiling it then filtered using sieve and then the hot distilled water was poured into the sieve with the sample, the procedure was repeated using 10% NaOH, then petroleum ether was poured into the sieve, ethanol also poured and then filtered after it then transferred into crucible and then take into oven and heated at 105°C for 2hrs, after taken out from the oven it then take into desiccator to cooled for about 5 minutes and then it then weighted and recorded in the weighing balance as W_1 , after recorded it then take into muffle furnace and burnt at 550°C for 2hrs, and then take out and then put in desiccator to cooled for about 5 minutes, after cooling it was then weighted and recorded using weighing balance as W_2 . The crude fiber was then calculated using [20].

$$\% \text{ Crude fibre} = \frac{W_1 - W_2}{W_0} \times 100 \quad (4)$$

W_0 = Weight of dried sample taken for the test

W_1 = Weight of crucible and sample before ashing

W_2 = Weight of crucible and sample after ashing

(v). Determination of the Percentage of Crude Protein

The crude protein of the sample was determined using the micro-Kjeldahl method described by AOAC [21].

1) Digestion of Sample

2g of powered *C. viscosa* sample was weighed into a Kjeldahl flask and 1 drop of Kjeldahl catalyst was added followed by 20ml of sulfuric acid. This was heated on a hot plate for 2 hours until the solution became clear and then cooled and dried in a desiccator for about 1hour. After cooling, the digested sample was filtered, washed several times, transferred to a 100ml volumetric flask, and distilled water was added to the mark.

2) Distillation and Titration

A 50 ml Erlenmeyer flask containing 10 ml of 4% boric acid and 2 drops of mixed indicator was placed under the condenser so that the top of the condenser was below the liquid. 10 ml of digestion product was weighed through a small funnel opening into the body of the device, washed with distilled water, then 10 ml of NaOH solution was added and steamed for approximately 5-7 minutes to collect sufficient ammonium sulfate. The receiving flask was removed and the tip of the condenser was poured into the flask. A 25ml portion of the solution in the receiving flask was titrated against 0.1N HCl, the amount of acid used was recorded, and finally a blank was analyzed using the same procedure. The nitrogen content of a sample is determined by the formula:

$$\% \text{ Nitrogen} = \frac{T_v - V_b \times N_{\text{acid}} \times 0.01401}{W} \times 100\%$$

Where

T_v = volume (ml) of acid required to titrate sample (Titre value)

Vb = volume (ml) of acid required to titrate the blank

N acid = normality of acid (0.1N)

W = weight of sample used in grams

% crude protein = $N \times (\text{conversion factor})$

Conversion factor = 6.25

(vi). Crude Carbohydrate Determination by Difference

This can be determined when all other analysis has been carried out on the sample. The available carbohydrate of the sample (% dry matter) was determined by adding % ash + % moist + % crude protein + crude lipid + % fibre content and the total subtracted from 100%

$$\% \text{ carbohydrate} = 100\% - \% \text{ Ash} + \% \text{ Moist} + \% \text{ crude protein} + \% \text{ crude lipid} + \% \text{ crude fibre}$$

(vii). Estimation of Energy Value

The sample calorific value was estimated (in kcal/g) by multiplying the percentages of crude protein, crude lipid and carbohydrate with the recommended factors (2.44, 8.37 and 3.57 respectively) as proposed by [22].

2.3.4. Determination of Mineral Content

Mineral element both micro minerals and macro mineral were determined by using of atomic absorption spectrophotometric and flame photometer by using different method at different proportion.

(i). Sample Digestion

2g of powered *C. viscosa* sample was weighed using weighing balance and then transferred into 250ml beaker 5ml of Conc HNO_3 and 15ml of Conc H_2SO_4 was added to the sample in the beaker the concentration of the acid is 1:1 ratio and then heated in the hot plate for about 2 hours until the acids liberated and turn into a clear blackish liquid, the beaker was removed from the hot plate to cooled and 50ml of distilled water was added after cooling, then filtered using Whatman No. 42 filter paper. The digested sample was stored in container and was taken further for Atomic Absorption spectrophotometry and Flame spectrophotometry Analysis.

(ii). Mineral Analysis

1ml was injected in the AAS machine to determine Cu, Fe, Mn, and Zn (micro minerals) and also 1ml was injected in the flame spectrophotometer to determine Ca, K, Na and (macro minerals).

3. Result

The result obtained in the laboratory analysis in Tables 1 to 3 for the powdered sample of *C. viscosa* root. All the analyses were conducted accordingly, and values were determined.

Table 1. Preliminary phytochemicals screening of *Cleome viscosa* root ethanolic extract.

S/N	Phytochemical	Present	Absent
1	Alkaloids	+	
	i. Wagner's Test	+	
	ii. Hager's Test		
2	Flavonoids	+	
	Alkaline Test		
3	Phenols	+	
	Ferric Chloride Test (FeCl_3)		
5	Steroids (Phytosterol)	+	
	Glycosides		
6	i. Sakowski's Test	+	-
	ii. Keller - kilani Test		
7	Tannins		-
	Ferric Chloride Test (FeCl_3)		
8	Saponins	+	

Table 2. Proximate constituent (% dry sample) of the root of *C. viscosa*.

Parameters	Composition (%)
Ash	5.00
Carbohydrate	15.73
Fibre	35.0
Lipid	35.0
Moisture	6.50
Protein	2.77

Estimated Energy Value: 355.76 kcal/g

Table 3. Mineral compositions (% dry sample) of the root of *C. viscosa*.

Minerals (macro and micro)	Concentration ($\mu\text{g/ml}$) of root	RDA (mg)
Calcium (Ca)	0.74	1200
Potassium (K)	1.32	2000
Sodium (Na)	1.95	500
Copper (Cu)	0.044	3-1.5
Iron (Fe)	2.592	15-1.10
Manganese (Mn)	0.190	5-2
Zinc (Zn)	0.0513	15-12

Recommended Dietary Allowance, (NRC, 1989).

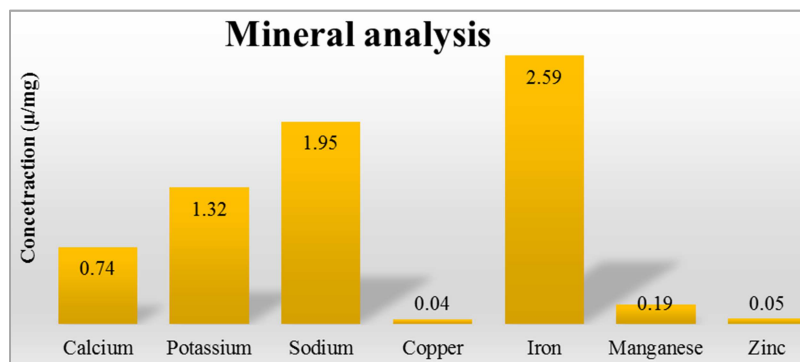


Figure 1. Chart representation of atomic absorption spectroscopy results.

4. Discussions

Phytochemical screening

The results of preliminary phytochemical examination are shown in Table 1. Phytochemicals constituents detected in *C. viscosa* root powdered sample using ethanol as solvent showed the presence of steroids, saponins, phenols, alkaloids, terpenoid, glycoside, and flavonoids but Tannin was absent. Similarly compared to the research conducted by [23] which reported the presence of phytochemicals in the *C. viscosa* of the whole plant using the methanol as a solvent extract which shows the presence of flavonoids, phenols, steroids, alkaloids, lignin, tannins, glycosides, terpenoids, saponins, quinines, and coumarins. Therefore, this shows that the plant contains phytochemicals with pharmacological potentials.

Proximate Analysis

The results of proximate analysis examined are shown in Table 2. *C. Viscosa* root. The ash content was obtained as (5.0%) compared to the research conducted by [24] which there is slightly different in the ash content value of (4.62±0.04%) in *C. viscosa* root. However, the ash content of leaf was high with the value of (9.25±0.10%). Ash level generally signifies the total quantity of inorganic minerals present in the plant. The percentage carbohydrate was calculated as (15.73%) compared to the research conducted by [24] the carbohydrate concentration was detected to be (64.43±0.40%) in stem which was higher when compared with this work. The percentage fibre was obtained as (35.00%) which was lower to the report conducted by [24] in which their crude fibre value was detected as (40.41±0.65%). However, the percentage fibre of this work showed higher percentage when compared with the values of stem (21.67±0.34%) and leaf (6.43±0.01%) in [24] work. The percentage protein was obtained as (2.77%) and it's higher than the percentage protein value of the stem (1.98±0.01%) of [24] research. The crude protein content indicated the total amount of protein in plant. Moisture content was obtained as (6.50%) was lower to the report conducted by [24] in which their Moisture content value was detected as (6.98±0.04%). This indicated that the root of *C. viscosa* is relatively low in water content and low moisture content may indicate shelf life and potential for drying and preservation of the root. The percentage lipid was obtained as (35.00%) which was higher when compared with the value of the leaf (20.45±0.01%) and leaf (6.43±0.01%) in [24] work. The estimated energy value was calculated as (355.76 kcal/g), the estimated value represents the potential caloric content of the plant. Therefore *C. viscosa* can be considered as a calorie - dense food source.

Mineral Analysis

Table 3 and figure 1 revealed the present of mineral analysis with the concentration of both the macro minerals and the trace element varies. Calcium concentration was detected as (0.74µg/ml) which was higher when compared with the research conducted by [25] in the *C. viscosa* seed which the concentration was (0.383 µg/ml). Calcium is essentials for nerve impulses conduction and activates some

enzymes, which generate neurotransmitters [25]. Potassium concentration was detected as (1.32µg/ml) which was higher when compared to the report conducted with the concentration of potassium which was detected as (0.072µg/ml) reported by [25] in the *C. viscosa* seed. Potassium is associated with the movement of water, nutrients and carbohydrates in plant tissues, it play important role in treatment of diabetes as it has effect on secretion of insulin. Sodium with the concentration of (1.95µg/ml) close to the one with higher concentration among the macro minerals and trace element detected from the root, the concentration of the sodium was higher when compared to the report conducted by [25] in the seed of *C. viscosa* which the concentration was detected (0.0182µg/ml). Sodium maintains the acid alkali (pH) balance in the body. It is necessary to maintain electrical potentials of the nervous system. Copper with the lower concentration of (0.044µg/ml) among all the macro minerals and trace element detected, the copper concentration is higher when compared to the research conducted by [25] in the seed of *C. viscosa* with wide range difference in concentration which was detected as (0.00064 µg/ml). Copper is required for many enzymatic activities in plants for chlorophyll and seed production. Iron with the higher concentration of (2.592µg/ml) among the macro minerals and trace element detected, the iron concentration is higher when compared to the research conducted by [25] with far difference in the concentration which was detected as (0.0406 µg/ml). Iron is responsible in the synthesis of chlorophyll, and it's essential for the maintenance of chloroplast structure and function. Manganese concentration was detected as (0.1905µg/ml) which is higher when compared to the research conducted in the seed by [25] which the concentration was detected as (0.0064 µg/ml). Manganese play important role in plant as major contributor to various biological systems including photosynthesis, respiration and nitrogen assimilation. Manganese deficiency cause skeletal abnormalities, retarded bone growth, change in hair colour to growth, abnormalities in pancreas, and disturbances in lipid and carbohydrate metabolism [26]. Zinc concentration was detected as (0.051µg/ml) which is higher when compared to the research conducted in the seed of *C. viscosa* by [25] which the concentration was detected as (0.0028 µg/ml). Zinc activates the enzymes that are responsible for certain protein. It's used in the formation of chlorophyll and carbohydrate and is used in the conversion of starch to sugar.

5. Conclusions

The present study of phytochemicals screenings in *Cleome viscosa* root showed that the plant is a very good source of secondary metabolites such as alkaloids, steroids, terpenoids, tannins, glycosides, flavonoids, phenols and saponins. The presence of various proximate parameters (ash content, moisture content, carbohydrate, crude protein, crude fibre and crude lipid) in *C. viscosa* root. The plant is a rich source

of mineral nutrients (micro and macro minerals), and nutritional agents could be utilized in the formation of therapeutic drugs/ medicines to treat various diseases that are mainly caused due to the deficiency of these minerals.

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