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Studies on physicochemical and nutritional properties of aerial parts of *Cassia occidentalis* L.

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ABSTRACT

In the present, work chemical composition and nutritional value of aerial parts of *Cassia occidentalis* L. was studied. The aerial parts of *C. occidentalis* possess favorable physico-chemical properties with good nutritional value, such as high energy value, crude fibers, and vitamin levels. The X-ray fluorescence spectrophotometry data revealed that the sample is rich in minerals, especially in Fe, Ca, K, and Mn. Further, minerals such as Mg, Zn, Cu, Na, P, and S are present in good amount and depicted the nutritional value of the selected material. The plant sample is rich in phytochemicals such as flavonoids, alkaloids, lignin, tannins, and phenols. The presence of phytochemical constituents was confirmed by gas chromatography–mass spectrometry profile and high-performance thin layer chromatography fingerprinting techniques. The findings stimulate the on-farm cultivation of *C. occidentalis* on a large scale to relieve the iron deficiency in local community, and it can be used as a dietary supplement to treat anemia.

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1. Introduction

Herbs are staging a comeback, and herbal “renaissance” is happening all over the world. In India, drugs of herbal origin have been used since ancient times in traditional systems of medicine such as Siddha and Ayurveda [1]. Indian traditional medicinal systems like Ayurveda, Unani, Siddha and modern medicine uses about 700, 600, 600 and 30 plant species, respectively [2]. Even the allopathic system of medicine has adopted a number of plant-derived drugs, which form an

important segment of the modern pharmacopoeia. Some important chemical intermediates (e.g., diosgenin, solasodine, etc.) that are needed for manufacturing the modern drugs are also obtained from plants [3]. Population growth, inadequate supply of drugs, prohibitive cost of treatments, side effects of several allopathic drugs, and development of resistance to currently used drugs for infectious diseases have led to an increased emphasis on the use of plant materials as a source of medicine for a wide variety of human ailments. As part of the strategy to reduce the financial burden on the

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human population in developing countries, increased use of plant drugs will be recommended.

Cassia occidentalis L. (Leguminosae) is regarded as an “edible weed of agriculture” or “famine food” [4]. *C. occidentalis*, also known as “kasamarda,” has been mentioned in various nighandus, viz. Rajnighantu, Dhanwantari, Bhavaprakasa, Rajballaba, etc. [5]. This plant is widely consumed by local people as a coffee substitute. The seeds are brewed into a coffee-like beverage for asthma and a flower infusion is used to treat bronchitis [6]. The roots are considered as tonic, febrifuge, and diuretic; they are also used for menstrual problems, tuberculosis, anemia, and liver complaints, and as a tonic for general weakness and illness [7]. The leaves are used in gonorrhoea, fevers, urinary tract disorders, and edema [8]. *C. occidentalis* extract is used to cure eye inflammations in Ayurveda. It is also used in Jamaican folk medicines for curing diarrhea, dysentery, constipation, fever, cancer, eczema, and venereal diseases. It is a main ingredient of Liv. 52 [9]. Herbolax, a polyherbal formulation that is commonly used in treating constipation, contains *C. occidentalis* as one of the ingredients [10]. A new indigenous metabolic corrective for newborns and infants, called “Bonnisan,” is also made up of *C. occidentalis* (0.5 mg/5 mL), which helps bring immediate relief from the discomfort caused by gastric wind [11].

C. occidentalis has been found to possess significant antibacterial, antifungal [12,13], antimalarial [14], anti-inflammatory [15], antimutagenic/anticarcinogenic [16], immunostimulant [8], laxative, analgesic, chloretic, and diuretic properties [17]. The main phytochemicals in *C. occidentalis* include achrosin, aloe-emodin, emodin, islandicine, kaempferol, obtusifolin, obtusin, physcion [18], anthraquinones, apigenin, aurantiobtusin, campesterol, cassiollin, chrysoobtusin, chrysophanic acid, chrysarobin, chrysophanol [19], chrysoeriol, funiculosin [20], quercetin, rhamnosides, rhein, rubrofusarin, sitosterols, tannins, and xanthorine [21]. A study of phytochemicals of *C. occidentalis* reveals that the nature and amount of phytochemicals vary according to the climate and soil conditions of the growing location. For example, stems, leaves, and root bark of the plant from Ivory Coast, Africa, contain no alkaloids, while a large amount of alkaloids was found in the samples collected from Ethiopia [22].

Even though *C. occidentalis* L. finds a large number of uses in traditional medicine as well as in ethnic food, its physicochemical properties and nutritional value remains unexplored. Hence, in the present paper, attempts were made to analyze the physicochemical properties and nutritional profile of *C. occidentalis* L. employing sophisticated techniques.

2. Methods

2.1. Collection of plant material

Aerial parts of *C. occidentalis* L. were collected from the herbal garden of STET Women's College, Mannargudi, Tamil Nadu, India in May 2014. Identification and authentication were performed at the Department of Centre for Advanced Research in Indian System of Medicine (CARISM), SASTRA University, Thanjavur, India. The collected materials were

cleaned, shade dried, and powdered. These pulverized materials were used for further analysis.

2.2. Physicochemical analysis

The sensory nature of dry powder of selected plant material was observed by keeping a small quantity in a Petri dish placed on a white background, and the organoleptic characters except audition were observed and tabulated. The physicochemical characters such as foreign matter; loss on drying; contents of total ash, acid-insoluble ash, water-soluble ash, and sulfated ash; and extractive values were determined according to the Ayurvedic Pharmacopoeia [23]. For the determination of foreign matter, 100 g of the sample was weighed and separated out in a thin layer. The foreign matter was detected by inspection with the unaided eye. For the determination of foreign matter, 100 g of the sample was weighed and separated out in a thin layer. The loss on drying was analyzed after drying the powdered sample in an electrical oven at 110°C until it reached a constant weight. Total ash was determined by taking ~2 g of accurately weighed sample; it was incinerated in a silica dish at a temperature not exceeding 450°C until free from carbon and weighed. The percentage of ash with reference to the air-dried drug was calculated. To determine the acid-insoluble ash, the residue of the total ash was boiled for 5 minutes with 25 mL of dilute HCl. The insoluble matter in the crucible was collected on an ash-less filter paper, washed with hot water, and ignited until a constant weight was obtained. The percentage of acid-insoluble ash was calculated with reference to the air-dried material. Water-soluble ash was determined by boiling the ash obtained from total ash for 5 minutes with 25 mL of water, and the insoluble matter was collected in an ash-less filter paper, washed with hot water, and ignited for 5 minutes at a temperature not exceeding 450°C. The difference in weight represents the water-soluble ash. For the estimation of sulfated ash, a silica crucible was heated to redness for 10 minutes, allowed to cool in a desiccators, and weighed. One gram of the substance was placed into the crucible, ignited gently at first until the substrate is thoroughly charred, and cooled; then the residue was moistened with 1 mL of sulfuric acid, heated gently until white fumes no longer evolved, and ignited at 800°C until all black particles disappeared. The crucible was allowed to cool and a few drops of sulfuric acid were added and weighed repeatedly until a constant weight was obtained in two successive measurements.

2.3. Phytochemical profile

Nearly 5 g of shade-dried, coarsely powdered material was subsequently extracted with sufficient volume of various organic solvents in the order of increasing polarity, using Soxhlet extraction apparatus for 6 hours. The extract was then concentrated and the solvent was removed completely under reduced pressure, and then the yield of the extract was calculated. Different qualitative chemical tests were performed in the aqueous extract of *C. occidentalis* using standard procedures to identify the major constituents, as described by Trease and Evans [24], Harborne [25], and Edeoga et al [26]. Quantification of phytochemicals such as alkaloids [27],

flavonoids [28], total phenols [26], tannins [29], and lignin [30] was performed by adopting standard protocols.

2.4. Chemical composition

The carbohydrate [31], free amino acid [32], protein [33], fat [34], fiber [35], cholesterol [36], energy value [33], thiamine [37], riboflavin [38], niacin [39], vitamin E [40], and vitamin C [41] contents were estimated according to the standard procedures. Activities of enzymes such as catalase [42], lipase [43], amylase [44], acid phosphatase [45], and alkaline phosphatase [46] were also estimated. X-ray fluorescence spectrophotometry and atomic absorption spectroscopy techniques were employed to analyze the mineral composition, while phytochemical profile of selected sample was determined using gas chromatography–mass spectrometry (GC–MS) and high-performance thin layer chromatography (HPTLC) fingerprinting techniques.

3. Results and discussion

Green plants synthesize and preserve a variety of phytochemical constituents, many of which are extractable and used as raw material for various scientific investigations. Many secondary metabolites of plants are commercially important and find use in a number of pharmaceutical applications. However, a sustained supply of the source material often becomes difficult due to factors such as environmental changes, cultural practices, diverse geographical distribution, labor cost, selection of the superior plant stock, and over-exploitation by pharmaceutical industries [47]. Hence standards must be evaluated for this source material such as ash value, extractive values, chemical composition, mineral composition, GC–MS profile, and HPTLC fingerprinting. In this connection, physicochemical properties of an Indian traditional medicinal plant *C. occidentalis* were investigated and its nutritional value was also explored in the present study.

The data obtained in the present work revealed interesting chemical features, which were tabulated. The selected part of *C. occidentalis* is bitter in taste and brown in color, and has a pleasant odor (Table 1). Tests for identity, purity, and strength were also conducted for *C. occidentalis*. The moisture content (loss on drying) is 10.17%, which implies that the shelf life of

this plant material appears to be longer. Ash content (14.27%) reveals that the plant is rich in mineral contents. Solubility in water (22.69%) is greater than that in alcohol (20.56%; Table 1). This extractive value suggests that the sample satisfies purity standards and is also rich in highly polar compounds. Among all the extracts, water extract of *C. occidentalis* was found to have the maximum yield (17.53%), followed by ethanol, ethyl acetate, hexane, and chloroform solvent extracts (Table 2).

Preliminary phytochemical analysis on an aqueous extract of *C. occidentalis* exhibits the presence of alkaloids, carbohydrates, flavonoids, phenolic compounds, tannins, and lignins (Table 3). Flavonoids recorded a higher percentage of yield (2.45 mg/g sample) when compared with alkaloids (1.56 mg/g sample), lignin (0.34 mg/g sample), tannins (0.21 mg/g sample), and phenols (0.16 mg/g sample) in the aerial part of *C. occidentalis* (Table 4). Secondary metabolites play both a defensive role against herbivore, pathogen attack, and inter-plant competition, and an attractant role toward beneficial organisms such as pollinators or symbionts [48]. Plant secondary products also have protective actions in relation to abiotic stresses such as those associated with changes in temperature, water status, light levels, UV exposure, and mineral nutrients. Furthermore, previous work has indicated potential role of secondary products at the cellular level as plant growth regulators and modulators of gene expression, and in signal transduction [49].

Flavonoids present in the plant might be responsible for its anti-inflammatory properties [50]. Alkaloids are a diverse group of secondary metabolites found to exhibit antimicrobial activity. Alkaloids are also known for decreasing blood pressure, balancing the nervous system in case of mental illness, and possessing antimalarial properties [51]. Tannins help in wound healing, act as an antiparasitic agent, and can reduce the risk of coronary heart diseases. Phenolic compounds are one of the largest and most ubiquitous groups of plant metabolites [52]. Natural antioxidants mainly come from plants in the form of phenolic compounds such as flavonoids, phenolic acids, etc. [53]. A number of studies have focused on the biological activities of phenolic compounds, which are potential antioxidants and free radical scavengers. Modern clinical studies have supported the role of steroids as anti-inflammatory and analgesic agents [54].

Nutritional value of the plant is clearly depicted in Tables 5 and 6. Energy value of the selected plant material was 34.44 kcal, and its crude fiber content was 5.69 mg/g. Intake of dietary fibers present in the selected plant can lower the serum cholesterol level, risk of coronary heart disease, hypertension, constipation, diabetes, and colon and breast cancer [55]. The recommended dietary allowances of fibers essential for children, adults, and pregnant and lactating mothers are 19–25 g/

Table 1 – Physicochemical properties of *Cassia occidentalis*.

S. No.	Parameters	Physicochemical properties
1	Taste	Bitter
2	Color	Brown
3	Odor	Pleasant
4	Foreign matter (%)	0.37
5	Loss on drying (%)	10.17
6	Total ash (%)	14.27
7	Acid-insoluble ash (%)	3.12
8	Water solubility (%)	0.36
9	Sulfated ash (%)	1.56
10	Solubility in alcohol (%)	20.56
11	Solubility in water (%)	22.69

Table 2 – Extractive values of *Cassia occidentalis*.

S. No.	Solvent	Extractive values (%)
1	Hexane	6.89
2	Chloroform	6.79
3	Ethyl acetate	7.74
4	Ethanol	9.08
5	Water	17.53

Table 3 – Preliminary phytochemical screening of various extracts of *Cassia occidentalis*.

Test	Reagents used	Hexane	Chloroform	Ethyl acetate	Ethanol	Water
Alkaloids	Dragendorff	–	–	–	–	+
	Mayer	–	–	–	–	+
	Wagner	–	–	–	–	+
	Hager	–	–	–	–	+
Reducing sugar	Fehling	–	–	–	+	–
Carbohydrates	Molisch	–	–	–	+	+
Saponins	Foam	–	–	–	–	–
Glycosides	Anthrone	–	–	–	–	–
Steroids	Liebermann–Burchard	+	+	+	+	–
Flavonoids	Shinado's	–	–	–	+	+
Phenolic compound	Ferric chloride	+	+	–	–	+
Tannin	Lead acetate	–	+	+	–	+
Quinone	Sulfuric acid	–	–	–	–	–
Anthraquinone	Aqueous ammonia	–	+	–	–	–
Lignin	Phloroglucinol	–	+	+	+	+
Proteins	Millon	–	–	–	+	–
Amino acids	Ninhydrin	–	–	–	+	–

Table 4 – Estimation of major phytoconstituents of *Cassia occidentalis*.

S. No.	Phytoconstituents	Content (mg/g sample)
1	Flavonoid	2.45
2	Alkaloid	1.56
3	Lignin	0.34
4	Tannin	0.21
5	Phenol	0.16

d, 21–38 g/d, and 28–29 g/d, respectively. Thus, *C. occidentalis* can act as a valuable source of dietary fibers in human nutrition. Other nutritional constituents found are free amino acids and carbohydrates. Total fat and cholesterol contents are as little as 0.03 mg/g. In addition to this, the plant is rich in vitamins, such as thiamine, niacin, and riboflavin, and in enzymes, especially catalase, lipase, amylase, alkaline phosphatase, and acid phosphatase (Table 5).

X-ray fluorescence spectrophotometry data suggested that the plant is rich in minerals, especially Fe, Ca, K, Mn, Mg, Zn, Cu, Na, P, and S (Table 6). From these data, it can be deduced

Table 5 – Nutritional value and biochemical composition of *Cassia occidentalis*.

S. No.	Parameters	Content
1	Energy value (kcal)	34.44
2	Crude fiber (mg/g)	5.69
3	Free amino acids (mg/g)	1.52
4	Carbohydrate (mg/g)	1.38
5	Protein (mg/g)	0.49
6	Total fat (mg/g)	0.03
7	Cholesterol (mg/g)	0.03
8	Thiamine (µg/g)	6.9
9	Niacin (µg/g)	12.6
10	Riboflavin (µg/g)	71.5
11	Catalase (µg/g)	9.8
12	Lipase (µg/g)	13.6
13	Amylase (µg/g)	10.8
14	Alkaline phosphatase (mg/g)	0.41
15	Acid phosphatase (mg/g)	0.21

that *C. occidentalis* has a high content of Fe and so can be used in the treatment of anemia. Deficiency of calcium and phosphorous leads to the classic bone symptoms associated with rickets, such as bowlegs, knock knees, curvature of the spine, and pelvic and thoracic deformities. Magnesium plays an important role in the structure and function of the human body. Iron, zinc, copper, and manganese play important roles in the improvement of the antioxidant system. The positive impact of zinc supplementation on the growth of some stunted children, and on the prevalence of selected childhood diseases such as diarrhea, suggests that zinc deficiency is likely to be a significant public health problem, especially in developing countries [56,57]. According to Food and Agricultural Organization's (FAO's) food balance data, it has been calculated that about 20% of the world's population can be at a risk of zinc deficiency with an average daily intake of <70 µg/d [58]. These findings stimulate the on-farm cultivation of *C. occidentalis* in a large scale to relieve the iron and zinc deficiencies in local community.

The suggested concentration of lead (Pb) in plant species is 2–6 mg/L [59]. Lead has carcinogenic properties, it impairs both the respiratory and the digestive systems, and it suppresses the immune system. It is particularly harmful in

Table 6 – Mineral composition and heavy metal content of *Cassia occidentalis*.

S. No.	Minerals	Content
1	Fe (%)	11.036
2	Ca (%)	2.69
3	Mn (%)	2.39
4	K (%)	2.36
5	Mg (%)	1.54
6	Zn (%)	1.24
7	Cu (%)	0.74
8	Na (%)	0.58
9	P (%)	0.54
10	S (%)	0.29
11	Pb (ppm)	<0.005
12	Hg (ppm)	<0.005
13	Cd (ppm)	<0.005

children, damaging their intelligence and the nervous systems [60]. The presently investigated plant materials have chances of lead contamination from soil, water, and atmosphere, and hence a high level of Pb can be accumulated. As per the World Health Organization Guidelines, the limit for lead is 10 ppm, cadmium 0.3 ppm, and mercury 1 ppm [33]. The atomic absorption spectrophotometric investigation suggested that the selected plant material has meager quantities of Pb, Cd, and Hg, which further clarifies its use as a safe nontoxic food supplement.

GC–MS analysis of the hexane extract of *C. occidentalis* revealed the presence of volatile phytochemical compounds such as nonanoic acid, dodecanoic acid, tetradecanoic acid, n-hexadecanoic acid, 10-octadecenoic acid methyl ester, 9,12-octadecadienoic acid, and oleic acid 3-hydroxypropyl ester (Figure 1A and Table 7). GC–MS of the chloroform extract revealed the presence of phytochemical compounds such as hexanoic acid, octanoic acid, n-decanoic acid, 9-oxononanoic acid, dodecanoic acid, 1,6-anhydro- α -D-galactofuranose, 3-ethyl-2-hydroxy-2-cyclopenten-1-one, tetradecanoic acid, n-hexadecanoic acid, and phytol (Figure 1B and Table 8). Phytoconstituents such as dodecanoic acid, tetradecanoic acid, and n-hexadecanoic acid were commonly found in both

hexane and chloroform extracts of *C. occidentalis*. These compounds were also detected in the GC–MS analysis of surface of clay tea pots [61].

HPTLC fingerprinting of the plant is presented in Figure 2 with Rf values under 254 nm and 366 nm, which confirms the presence of different types of phytochemical compounds in the aqueous extract of the aerial part of *C. occidentalis* (Table 9). Four bands of dark green color were observed with Rf values of 0.10, 0.21, 0.80, and 0.86 at 254 nm, while 12 bands of red (Rf values 0.14 and 0.80), pink (Rf value 0.19), green (Rf values 0.32 and 0.46), fluorescence blue (Rf values 0.65 and 0.74), yellow (Rf values 0.77 and 0.88), brown (Rf values 0.83 and 0.92), and fluorescence green (Rf value 0.97) colors were noted at 366 nm. The presence of such bands of different colors and Rf values indicated the occurrence of different types of phytochemicals in the extract of *C. occidentalis*.

3.1. Conclusion

Plant samples are safe and effective in treating various ailments without serious side effects. However, the lacuna existing in the herbal industry is the lack of standardization. Hence, attempts were made in the present work to determine

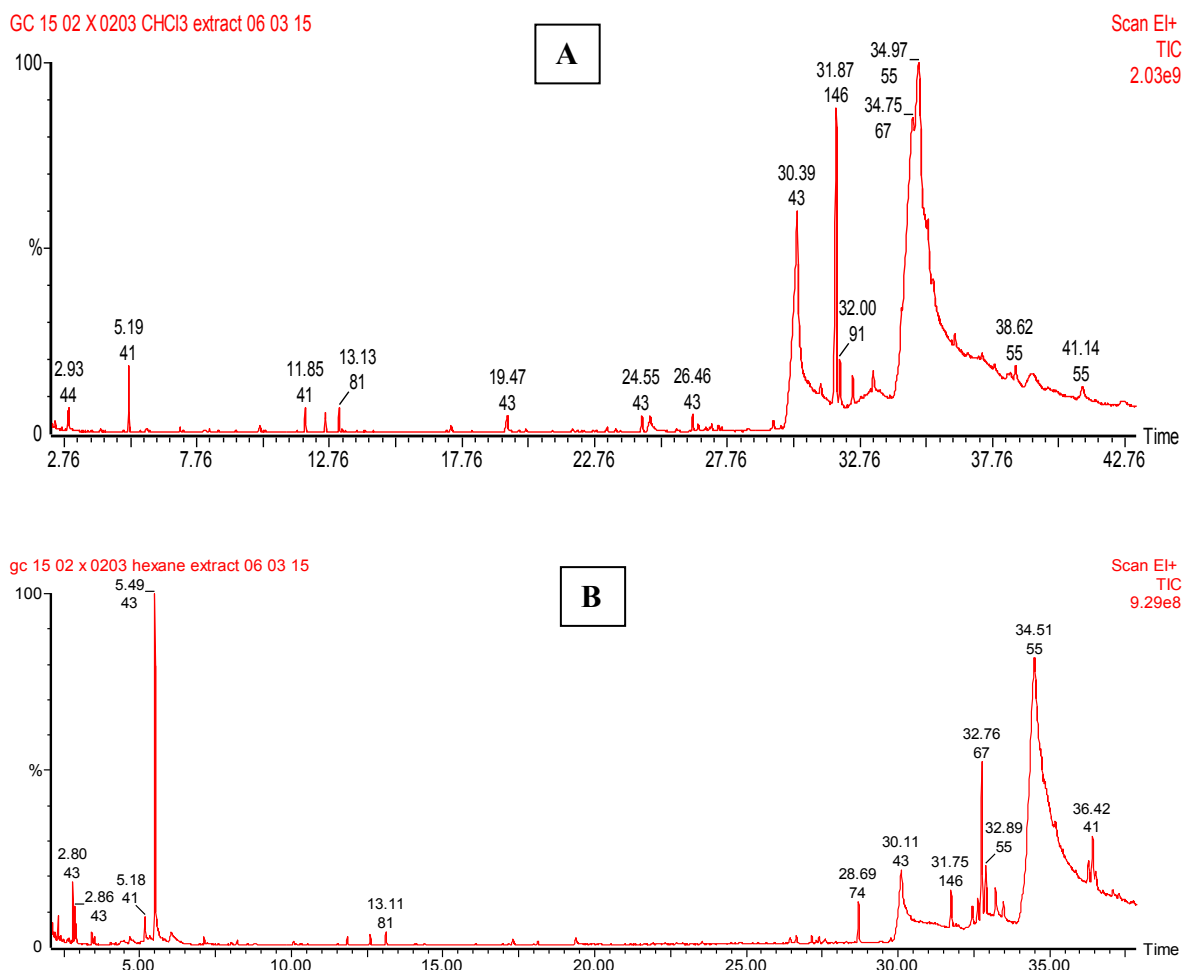


Figure 1 – GC–MS chromatogram of (A) chloroform and (B) hexane extracts of *Cassia occidentalis*. GC–MS = gas chromatography–mass spectrometry.

Table 7 – GC–MS profile of hexane extract of *Cassia occidentalis*.

S. No.	Peak name	Retention time (min)	Peak area	% Peak area
1	Nonanoic acid	10.06	502,244	0.1584
2	Dodecanoic acid	19.38	1,149,147	0.3625
3	Tetradecanoic acid	24.79	515,242	0.1625
4	n-Hexadecanoic acid	30.11	25,483,818	8.0380
5	10-Octadecenoic acid methyl ester	32.89	13,129,060	4.1411
6	9,12-Octadecadienoic acid	34.51	171,295,296	54.0291
7	Oleic acid 3-hydroxypropyl ester	36.52	2,453,627	0.7739

GC–MS = gas chromatography–mass spectrometry.

Table 8 – GC–MS profile of chloroform extract of *Cassia occidentalis*.

S. No.	Peak name	Retention time (min)	Peak area	% Peak area
1	Hexanoic acid	5.86	1,692,489	0.3081
2	Octanoic acid	10.14	1,974,734	0.3595
3	n-Decanoic acid	14.43	373,347	0.0680
4	9-Oxononanoic acid	17.35	2,940,956	0.5354
5	Dodecanoic acid	19.47	7,191,673	1.3092
6	1,6-Anhydro- α -D-galactofuranose	21.67	58,710	0.0107
7	3-ethyl-2-hydroxy-2-cyclopenten-1-one	22.36	482,644	0.0879
8	Tetradecanoic acid	24.85	10,046,852	1.8289
9	n-Hexadecanoic acid	30.39	152,382,432	27.7395
10	Phytol	33.27	5,594,367	1.0184

GC–MS = gas chromatography–mass spectrometry.

the physical properties and chemical composition of an Indian medicinal plant *C. occidentalis* L., and also to evaluate its nutritional potential so that it can be used as a herbal supplement for treating various diseases and disorders. The findings of the present study revealed that the selected plant has high levels of energy value, crude fibers, vitamins, and minerals, especially iron and zinc, with antioxidant enzymes. Hence, *C. occidentalis* can be used as a safe, nutritious, and

Table 9 – HPTLC fingerprinting profile of *Cassia occidentalis*.

S. No.	Rf values		Color of the spot	
	254 nm	366 nm	254 nm	366 nm
1	0.10	0.14	Dark green	Red
2	0.21	0.19	Dark green	Pink
3	0.80	0.23	Dark green	Red
4	0.86	0.32	Dark green	Green
5	—	0.46	—	Green
6	—	0.65	—	Fluorescence blue
7	—	0.74	—	Fluorescence blue
8	—	0.77	—	Yellow
9	—	0.83	—	Brown
10	—	0.88	—	Yellow
11	—	0.92	—	Brown
12	—	0.97	—	Fluorescence green

HPTLC = high-performance thin layer chromatography.

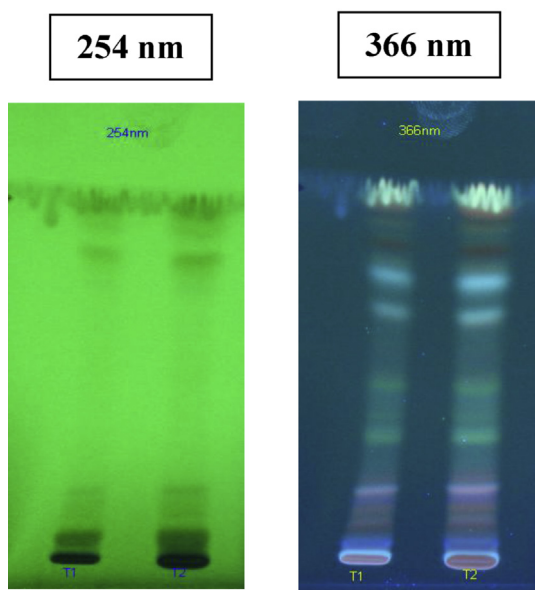


Figure 2 – HPTLC fingerprinting profile of *Cassia occidentalis*. HPTLC = high-performance thin layer chromatography.

medicinally active food supplement. The sample has many favorable physicochemical characteristics such as high ash content, water solubility, and extract yield. Physicochemical properties determined in the present study will be useful in the identification and authentication of this plant material and can be used as quality control parameters. The presence of certain phytochemical constituents was confirmed by qualitative, quantitative, GC–MS, and HPTLC analyses.

Conflicts of interest

All authors declare no conflicts of interest.

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