RESEARCH ARTICLE

Pharmacognostical, Physico-chemical and Phytochemical Evaluation of leaves of *Cassia tora* and *Cassia fistula*"

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Abstract

Cassia sp. is well known in Indian system of medicine for their huge medicinal properties. In the present study, pharmacognostical, physico-chemical and phytochemical characters of leaves of Cassia tora and Cassia fistula were studied. Microscopic and fluorescence characters of powder of the leaves were studied for identification and characterization of various features. Physicochemical parameters like Foreign Organic matter (*C. tora* 1.5% ± 0.005; C. fistula 1.0% ± 0.002), Total Moisture (*C. tora* - 0.7% ± 0.002; C. fistula -1.5% ± 0.07), Ash Values (total ash, acid insoluble ash, water soluble ash, sulfated ash) and Extractive values of leaves of both the plants were evaluated. Heavy Metals viz. As, Cd, Cr, Pb, Hg were found in permissible limit i.e. less than 0.01ppm in both the leaves of Cassia species. Zinc, an essential micronutrient, was also found in permissible limit (*C. tora* -23.67ppm; C. fistula- 5.07ppm). Phytochemical screening showed the presence of phenols, flavoniods, tannins, carbohydrates and triterpenoids in the polar solvent extracts of both the plants. TLC finger print profile was also studied by observing the developed and derivatized TLC plate under UV (254 and 366nm) light. The present study reveals specific characteristics of the particular plant materials that can have a significant use in identification of crude samples and quality assessment of raw product i.e. impurities and adulterations, which can serve as a reference for further investigations.

Keywords: *C. tora*, *C. fistula*, Pharmacognostical evaluation, Phytochemical screening, Physicochemistry.

INTRODUCTION

Cassia Linn. (Family – Caesalpiniaceae) is a large tropical genus with about 600 species of herbs, shrubs and trees; some of which are

widely distributed throughout the world especially in tropical countries and is abundantly available in India. Most of the plants of genus are well known in Indian system of medicine for their cathartic, purgative, antiparasitic, anti-helminthic, antifungal, antimicrobial, anti-inflammatory property etc.. Various plants of *Cassia sps.* are also used traditionally in the treatment of periodic fever and malaria in subtropical and tropical regions. Some plants of this genus are widely used as traditional medicine in Africa and India for the treatment of ulcers. Several of them yield timber, dyes, fodder, vegetables, edible fruits etc.. In some places seeds are used as substitute for coffee (Dave and Ledwani, 2012). Plants of *Cassia* genus are rich source of polyphenols, flavonoids, tannins, mucilage, polysaccharide, steroids, anthraquinone glycosides and derivatives of anthracene (Sanghi *et al.*, 2006). The anti-inflammatory activity of *Cassia* may be attributed to the flavonoid molecules present in them (Ganapaty *et al.*, 2002).

Cassia fistula Linn. (Indian Labernum), a semi-wild tree (20–30 ft.) also known as the Golden Shower, has become extensively distributed in various countries including Mauritius, India, South Africa, Mexico, China, etc. and is used as an ornamental tree for its beautiful bunches of yellow flowers (Mukhopadhyay *et al.,* 1998). Leaves contain free rhein (4,5-dihydroxyanthraquinone-2-carboxylic acid) and its glycosides – Sennosides A & B (Thirumal *et al.*, 2012). Leaves and flowers are both used as a purgative drug. Juice of leaves is useful as dressing for ringworm, relieving irritation and relief of dropsical swelling (Danish *et al.,* 2011).

Cassia tora is an annual herb also known as Wild Senna. In India, it occurs as waste land rainy season weed. *C. tora* leaves contain emodin, stigmasterol, β-sitosteral-β-D-glucoside, freindlen, palmitic, stearic acid, succinic, d-tartaric acids and derivatives of quercitrin. According to Ayurveda the leaves and seeds are acrid, laxative, antiperiodic, anthelmintic, ophthalmic, liver tonic, cardiotonic, expectorant etc. and they are useful in leprosy, ringworm, colic, dyspepsia, constipation, cough, bronchitis and cardiac disorders (Kawade and Vite, 2013).

The main objective of the present investigation is to study and compare pharmacognostical features and physiochemical constants along with phytochemical screening of the leaves of these two species as very less information is available on these parameters.

MATERIALS AND METHODS

The fresh leaves of *Cassia tora* (*C. tora*) and *Cassia fistula* (*C. fistula*) were collected from the suburban region of Maharashtra and they were identified and authenticated from the Blatter Herbarium, St. Xavier's College, Mumbai (Maharashtra). The leaves of both plants were separated from twigs and shade dried. Later they were crushed into coarse powder (sieve no. 10/44) and kept in properly labeled air tight containers. All chemicals used in assays were purchased from Sigma Chemical Co. (St. Louis, MO, USA) and Merck Co. (Santa Ana, CA, USA). All parameters studied under pharmacognostic features were followed from guidelines' of American Herbal Pharmacopoeia, AHP – Botanical Pharmacognosy, 2011; Khandelwal, 2008; Indian Pharmacopoeia, 2007; WHO, 1998 and British Pharmacopoeia, 1980.

Macroscopic evaluation:

The macroscopic evaluation of leaves of *C. tora* and *C. fistula* was done by observing the external characters like shape, size, texture, surface characteristics and fractured surface with the help of magnifying lens. The organoleptic features like colour, odour, taste, feel and fracture of the crude drug were observed with sensory organs and compared.

Microscopic evaluation:

Microscopical studies were carried out from transverse sections (T.S.) of fresh leaflets. The sections were doubled stained with saffranine and fast green. Mounted in 50% glycerine and

observed under Lawrence & Mayo monocular (LM–52-1602) and Phase contrast-trinocular microscope (LM–52-1802). Microphotographs of sections were taken for the identification of various tissues and their arrangement. Characteristic features of leaves of *C .tora* and *C. fistula* were noted for comparison.

Determination of Physicochemical Parameters:

Following physicochemical parameters were determined in coarsely powdered leaves of *Cassia species* as per standard procedures (Khandelwal, 2008; Evans, 2002 and WHO, 1998).

Determination of Foreign Organic Matter:

100–500 g of the crude samples were taken, weighed and spreaded in a thin layer. The foreign organic matter (FOM) was detected by the use of a lens (6x). Separated FOM weighed and percentage of presence was calculated.

Total Moisture content:

1g air-dried coarse powder of leaves of *C. tora* and *C. fistula* were weighed in previously tarred crucible and dried at 105°C in hot air oven and cooled. Total moisture was calculated with respect to difference on pre-dried and post dried weight of sample.

Ash Value:

Total ash, acid insoluble ash, water soluble ash and sulfated ash were determined in the powdered leaves of *C. tora* and *C. fistula* according to the standard procedure.These values are used for determining the quality and purity of the powdered form of crude drug.

Extractive values:

About 5 g of dried powdered leaves of *C. tora* and *C. fistula* were weighed and macerated for 24 hours with 100 ml of solvents (ethanol 95% and water) in a glass stopper flasks. The flasks were shaked frequently for six hours and allowed to stand for next 18 hours. Extracts were filtered; 25 ml of extract was transferred in tarred dish and evaporated to dryness on water bath. The dried extract was further kept in hot air oven at 105°C, cooled and weighed. The percentage of extractive values for different solvents was calculated.

Heavy Metal analysis:

The objective was to determine the essential and non-essential heavy metals and their amount in the leaves of plants *viz. C. tora* and *C. fistula*. Selected heavy metals *viz.* Mercury (Hg); Arsenic (As); Lead (Pb); Zinc (Zn); Chromium (Cr) and Cadmium (Cd) were analyzed from SAIF (Sophisticated Analytical Instrument Facility) Department of IIT, Powai (Maharashtra, India). The samples were digested by wet digestion method and analyzed by Inductively Coupled Plasma-Atomic Emission Spectrometry (ICP-AES) and amount of heavy metals were quantified in the leaves.

Foaming index:

1g powder of leaves of each plant was weighed and transferred to a conical flask containing 100 ml of boiling distilled water. Moderate boiling was maintained for 30 min., cooled and filtered into volumetric flask and the volume was made up to 100 ml with distilled water. The decoction was transferred into stopper test tubes in successive portions of 1 ml, 2 ml, 3 ml and up to 10 ml and volume was made up to 10 ml in each tube with distilled water. Tubes were shaken in length wise motion for 15 seconds and were allowed to stand. The height of foam in each tube was measured.

Swelling index:

1g of coarse powder of leaves of *C. tora* and *C. fistula* was taken in glass- stopper measuring cylinder. 25ml of water was added and shaked occasionally for 1 hour and kept for 3 hours. Swelling index was calculated by measuring the volume in ml occupied by the 1 g swollen drug.

pH Determination :

pH of extracts (5%) of leaves of *C. tora* and *C. fistula* was determined by a standard calibrated pH meter and values were recorded.

Fluorescence analysis of powdered drug :
Fluorescence powder drug analysis of the crude powder of *C. tora* and *C. fistula* was carried out in the UV (Ultra-Violet) light as per the method of Chase and Pratt (1949). The fluorescence patterns were obtained when the powdered drug reacted with different chemical reagents. The identification and comparison of the colors was done using the standard colour index chart.

Preliminary phytochemical screening:
Extracts of *C. tora* and *C. fistula* leaves were prepared in different solvents (Ethyl acetate, Chloroform, Acetone, Petroleum Ether, Methanol, Aqueous) by using condenser and subjected to preliminary phytochemical screening for the detection of various phytoconstituents such as alkaloids, glycosides, tannins and phenolic compounds, flavonoids, steroids, saponins, proteins, amino acids, carbohydrates and triterpenoids.

TLC profile of hydro alcoholic extract
The TLC profile of hydro-alcoholic extract was carried out by using Ethyl Acetate: Formic Acid: glacial Acetic Acid: Water (100 : 1.0 : 1.0: 28 v/v/v) solvent system (Wagner et al., 2009). The visualization of spot was done by observing the plate under UV light (both long and short) and after derivetizing with 10 % ethanoic NaOH spray reagent. Color for different spots was observed and recorded.

RESULTS AND DISCUSSION

Evaluation of different parameters *viz.* pharmacognostical features, physicochemical constants and phytochemical screening of the leaves of *C. tora* and *C. fistula* Linn. was carried out and discussed below:

Macroscopic evaluation:
Morphology is the pre-requisite variable for identification of any matter. Here, the leaves of *C. tora* and *C. fistula* Linn. were differentiated on the basis of size, shape, apex, base and presence or absence of gland and flexible spine (Table -1 and Fig.– 1.A).
The leaves of *C. fistula* have symmetric base without gland; whereas leaves of *C. tora* have asymmetric base with gland. In the *C. tora* main rachis has conical gland between the last two pairs of leaflets. Another differentiating character was presence of flexible spine on the dorsal surface near lowermost pair of leaflets in *C. tora* however it is absent in *C. fistula* (Cooke, 1967).

Microscopic Evaluation:
Microscopic evaluation of the plant is essential to identify the adulterants and for the correct identification of the plant. The results of microscopic evaluation are given below:-

Transverse section of leaflet:
T.S of leaflet through midrib and lamina showed dorsi-ventral structure and was covered by epidermis on both the surfaces. Upper and lower epidermis was further covered by cuticle. Below the epidermis, single layer of elongated palisade cells was observed followed by 3-4 layers of loosely arranged spongy parenchymatous cells in both the species of *Cassia*.

In *C. tora,* epidermis showed uni-to multicellular uniseriate trichomes with constricted uppermost cell. The midrib comprises of parenchymatous tissues embedding vascular tissues. Vascular tissues were randomly scattered in centre and surrounded by sclerenchymatous cells [**Fig. I (a–e**)]. Calcium oxalate crystals were observed in the centre of midrib within parenchymatous tissue (**Fig. I (f**).

In *Cassia fistula,* adaxial epidermal cells are mostly rectangular to square in shape and in some cases; it was polygonal [**Fig. II (g-l**)]. Non-glandular, unicellular trichomes were found on surface of epidermis. In the centre of midrib, vascular bundles were arranged in specific ring manner with radial clusters of xylem elements.
In both the species of *Cassia,* Paracytic stomata were present on both the surfaces of leaves with maximum stomata on abaxial or lower surface.

Table 1: Macroscopic description of Leaves.

Sr. No.	Parameters	Description	
		Cassia tora	***Cassia fistula***
1.	Arrangement	Paripinnate Compound	Paripinnate Compound
2.	Leaflets	3 pairs	4 – 8 pairs
3.	Apex	Obtuse to Slightly reduce	Acute
4.	Shape	Obovate – Oblong	Ovate – Oblong
5.	Margin	Entire	Entire
6.	Length	3 – 4 inch	9 –16 inch
7.	Colour	Green	Green
8.	Stipules	Long	Minute
9.	Base	Oblique /Asymmetrical	Symmetric
10.	Texture	Smooth	Coriaceous & Leathery
11.	Mid-rib	Biconvex and less prominent on either side	Biconvex and more prominent on lower side

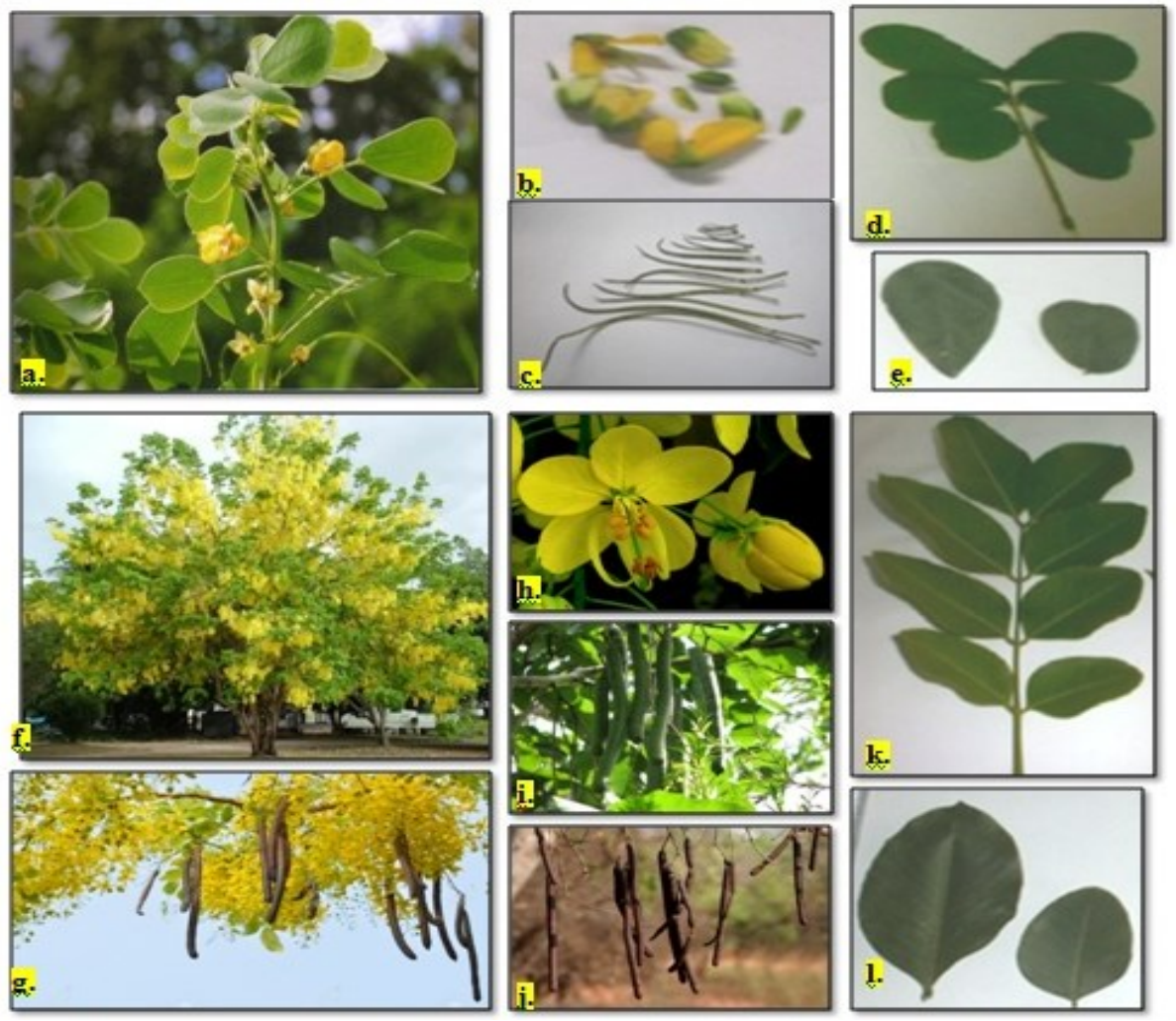
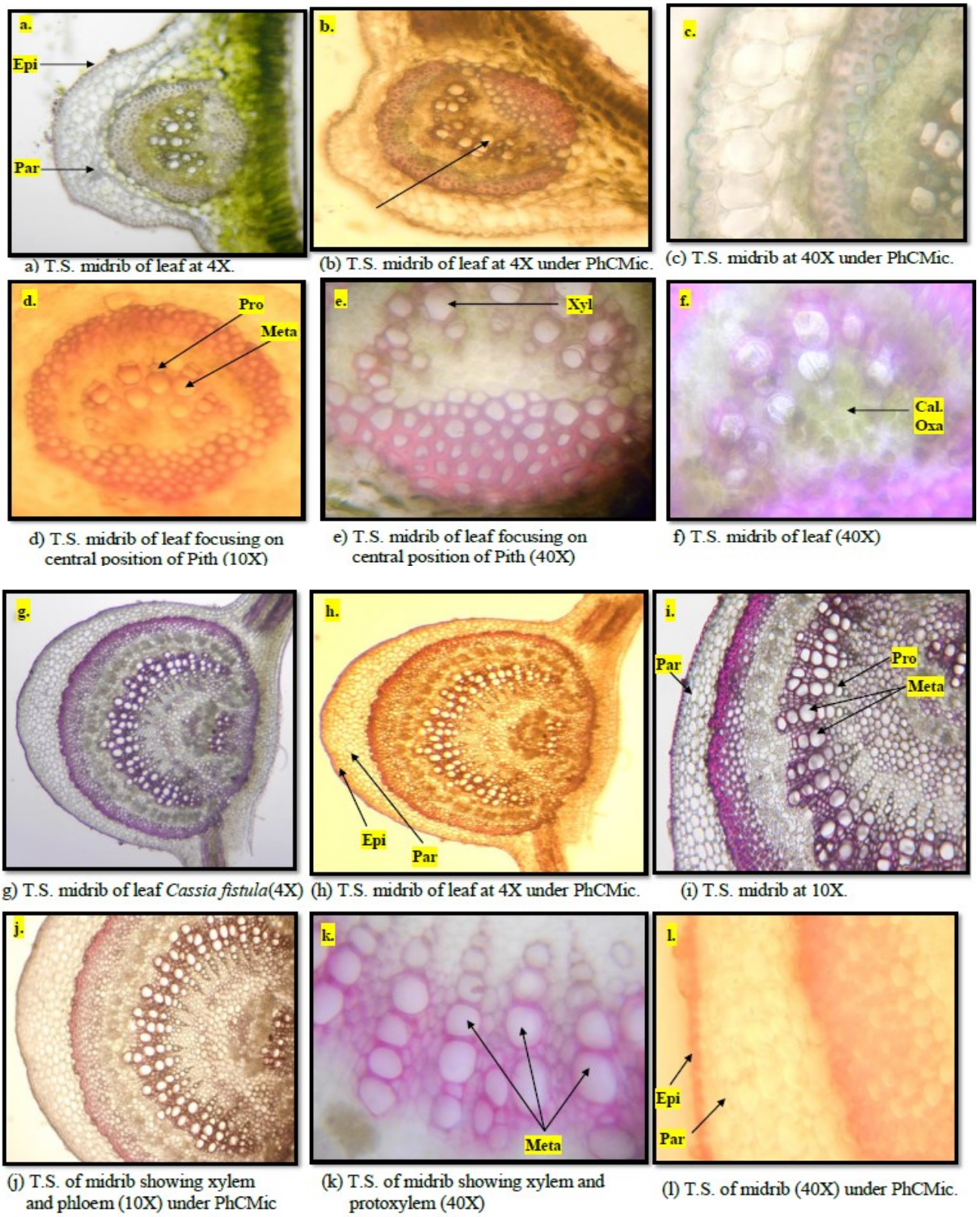


Fig. 1: Macromorphological photographs of *Cassia species.*

i) *Cassia tora* (a- e), a) An annual herb; b) Flower; c) Young pods d) Compound leaves e) Leaves (3-4 inches)

ii) *Cassia fistula* (f- l) f) Perinnial Tree; g) inflouresence with mature fruits;

h) Flower; i)Young Pods; j) Mature Pods; k) Compound leaves l) leaves (9-16 inches)



a) T.S. midrib of leaf at 4X. (b) T.S. midrib of leaf at 4X under PhCMic. (c) T.S. midrib at 40X under PhCMic.

d) T.S. midrib of leaf focusing on central position of Pith (10X) e) T.S. midrib of leaf focusing on central position of Pith (40X) f) T.S. midrib of leaf (40X)

g) T.S. midrib of leaf *Cassia fistula*(4X) (h) T.S. midrib of leaf at 4X under PhCMic. (i) T.S. midrib at 10X.

(j) T.S. of midrib showing xylem and phloem (10X) under PhCMic (k) T.S. of midrib showing xylem and protoxylem (40X) (l) T.S. of midrib (40X) under PhCMic.

Key – PhCMic – Phase contrast Microscope ; Epi – Epidermis ; Xyl – Xylem ; Par – Parenchyma ; Pro – Protoxylem ; Met – Metaxylem; Cal. Oxa – Calcium Oxalate Crystals.

Fig.1B: Photo- Micrograph of leaves of *Cassia sp.*: I. *Cassia tora* (a- f), II. *Cassia fistula* (g- l)

Powder analysis

The powders of both the species were analyzed for organoleptic characters and differences were observed (Fig. 2 and Table 2).

I. (a.) *Cassia tora* II. (b.) *Cassia fistula*

Figure 2: Photographs: Powders of leaves of *Cassia sps.* (Organoleptic Characters)

Table 2. Organoleptic Characters

S. N.	Characters of Powder	Observation	
		Cassia tora	***Cassia fustula***
1	Colour	Dill green	Yellowish green
2	Odour	Mild Aromatic	Mild pleasuent aromatic
3	Taste	Slightly Bitter	Slightly bitter
4	Size	Course	Fine
5	Miscellaneous	Cellulosic fibers	Fibres

Physicochemical evaluation

The evaluation of physico-chemical parameters, was carried out to ensure quality and purity of the crude sample (Table – 3).

Foreign organic matter (FOM)

The matter or part of the matter other than the crude drug which is not defined and described in the prescribed monograph of sample is known as foreign organic matter. High percentage of foreign organic matter is considered as a more deteriorating quality of drug or sample (Mukherjee, 2002). The content of FOM in selected samples was found in appraisable limit *i.e.* 1.5% ± 0.005 and 1.0% ± 0.002 for *C. tora* and *C. fistula* respectively (Table 3).

Total moisture content (TMC)

Total moisture content (Loss on drying) measures the amount of water and volatile matters or minerals *viz.* Cu, Fe, Pb, Hg, Ni, Zn in a sample when the sample is dried under specified conditions. Loss on drying is the loss of mass expressed as w/w. In both the samples, TMC values were found in negligible percentage *i.e.* 0.7 % ± 0.002 in *C. tora* and 1.5 % ± 0.07 in *C. fistula*.

Ash values

Ash values (Total ash, Acid Insoluble ash, Water soluble ash and Sulphated ash) of a drug give a relevant reference of the earthy matter or the inorganic composition and other impurities present along with the drug. The Total ash gives the information of physiological (plant tissues) and non-physiological ash (external matter adhering to plant surface) content in organic matter. Crude sample of *C. tora* leaves exhibited higher total ash content (17.35 % ± 0.003) than *C. fistula* (10.3% ± 0.05). Acid insoluble ash measures presence of amount of silica or silicates in the form sand or siliceous earth and in both the leaves of *Cassia,* it was found in permissible limit with negligible difference *i.e.* 2.20% ± 0.02 and 2.50% ± 0.04 for *C. tora* and *C. fistula* respectively (Table 3). The Water soluble ash is useful to determine inorganic content of ash of crude drug which is found to be soluble in water as this gives a useful indication of the quality of plant material. The powder of the *C. fistula* leaves was exhibited higher water soluble ash content (13.00% ± 0.033) than *C. tora* (9.06% ± 0.002). The Sulphated ash test is an analytical test for determining the content of inorganic impurities or residual matter in an organic substance which is not volatilized from a sample when the sample is ignited in the sulfuric acid. Sulphuric acid reacts with inorganic compounds and converted into their sulfates and preferably metal oxides. Total Sulfated Ash (%w/w) was found to be higher in *C. tora* (15.60 % ± 0.03) than *C. fistula* (7.06% ± 0.002).

Extractive values

The extractive values are primarily useful for the determination of exhausted or adulterated drug. The alcohol extractive (A.E.) values indicated the presence of polar constituents like phenols,

alkaloids, steroids, glycosides, flavonoids, etc. and the water extractive (W.E) values indicated the presence of sugar, acids and inorganic compounds (Indian Pharmacopoeia, 1996; Mukherjee, 2002). Both the extractive values [alcohol and water (%w/w)] were found to be maximum in *C. fistula* than in *C. tora* (Table-3). In both the plants, water extractive value was higher than alcohol extractive value. This signifies that the large amount of constituents of leaves were soluble in water than alcohol.

Heavy Metal analysis:

Contamination of medicinal plant material with heavy metals can be attributed to many causes including environmental pollution and traces of pesticides. Limit tests for such toxic metals are essential for herbal ingredients. In the examination of leaves of *C. tora* and *C. fistula*, all the selected heavy metals (Hg; As; Pb; Cr; Cd) were found to be less than 0.01ppm *i.e.* below permissible limit set by FAO/WHO for medicinal herbs and edible plants. Zinc (Zn) is also an essential trace element; plays an important role in various cell processes and WHO's recommended limit for Zn in medicinal plant is 50mg/kg *i.e*. 50ppm (Shah *et al.*, 2013; Jabeen *et al.*, 2010). In the leaves of *Cassia sps.*, Zn was found within the range of permissible limit *i.e*. in *C. tora* - 23.67 ppm and in *C. fistula* – 5.07 ppm.

Foaming and Swelling index

Many medicinal plant materials contain saponins that can cause persistent foam when an aqueous decoction is shaken. The foaming index measured the foaming ability of an aqueous decoction of plant materials and their extracts due to presence of saponin. In both of the species, it was found to be less than 100 ml. Swelling index gives an idea of the mucilage content in the crude drug**.** Powder of *Cassia species i.e*. *C. tora* and *C. fistula,* leaves showed no swelling index (Table-3).

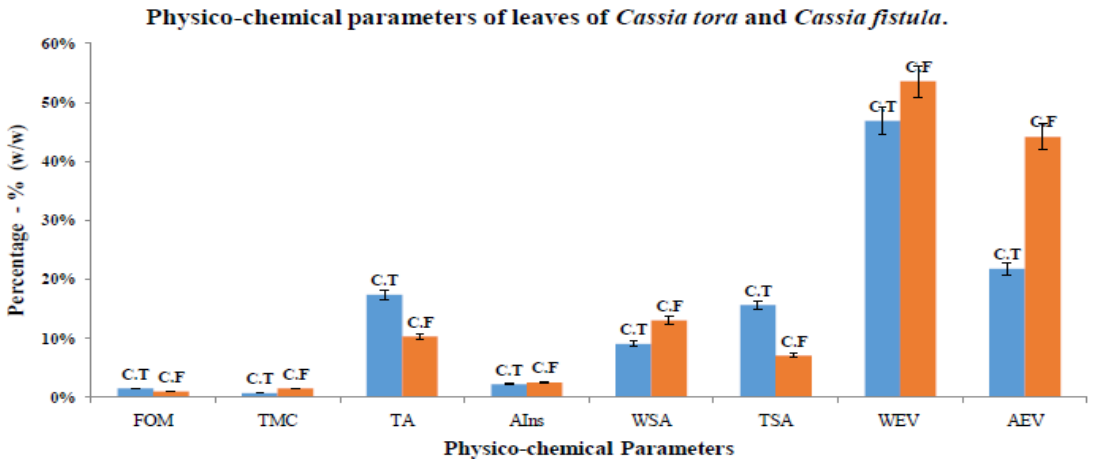
pH determination

pH refers to H^+ ion concentration in terms of acidity and basicity of the sample and its solvent. Leaves of *C. tora* and *C. fistula* extracts showed approximately same range of pH values *i.e.* 6 to 6.8 (Table-3).

Table 3: Physico-chemical parameters of leaves of *Cassia tora* and *Cassia fistula*.

S. No.	Parameters	*Cassia tora*	*Cassia fistula*
1.	Foreign organic matter **(%w/w)**	1.5% ± 0.005	1.0% ± 0.002
2.	Total Moisture Content **(%w/w)**	0.7 % ± 0.002	1.5 % ± 0.07
	Ash Values		
3.	Total Ash **(%w/w)**	17.35 % ± 0.003	10.3 % ± 0.05
4.	Acid Insoluble Ash **(%w/w)**	2.20 % ± 0.02	2.50 % ± 0.04
5.	Water soluble Ash **(%w/w)**	9.06 % ± 0.002	13.00 % ± 0.033
6.	Total Sulfated Ash **(%w/w)**	15.60 % ± 0.03	7.06 % ± 0.002
	Extractive Values		
7.	Water Extractive Value **(%w/w)**	46.88 % ± 0.05	53.6 % ± 0.2
8.	Alcohol Extractive Value **(%w/w)**	21.76 % ± 0.01	44.16 % ± 0.05
	Heavy Metal analysis (ppm)		
9.	Arsenic (As)	<0.01 ppm	<0.01 ppm
10.	Cadmium(Cd)	<0.01 ppm	<0.01 ppm
11.	Chromium(Cr)	<0.01 ppm	<0.01 ppm
12.	Lead (Pb)	<0.01 ppm	<0.01 ppm
13.	Mercury (Hg)	<0.01 ppm	<0.01 ppm
14.	Zinc (Zn)	23.67 ppm	5. 07 ppm
	Miscellaneous Parameters		
15.	Foaming index (ml)	< 100	< 100
16.	Swelling index	Nil	Nil

17.	pH (5 %)	6.5 ~ 6.8	6 ~ 6.8



Key:- **C.T.** *Cassia tora,* **C.F.:** *Cassia fistula;* **FOM:** Foreign organic matter, **TMC:** Total moisture contents **T.A.:** Total Ash; **AIns:** Acid insoluble Ash; **WAS:** Water soluble Ash; **TSA:** Total Sulfated Ash; **WEV:** Water Extractive Value; **AEV:** Alcohol Extractive Value

Fig. 3: Graphical representation of physicochemical parameters of *Cassia tora* and *Cassia fistula*

Table 4:Fluorescence Characters of the powdered leaves of *Cassia tora* and *Cassia fistula* under Ultra violet (UV) light

Sr. No.	Treatment	Fluorescence	
		C. tora	***C. fistula***
1	Powder mounted with nitrocellulose .	Greyish white	Greyish white
2	Powder treated with NaOH in methanol.	Green	Greenish Black
3	Powder treated with NaOH in water.	Yellow	Violet
4	Powder treated with NaOH in water dried and mounted with nitro cellulose.	Brown	Greenish Red
5	Powder treated with NaOH in methanol dried and mounted with nitro cellulose.	Yellowish green	Yellowish green
6	Powder treated with HCl	Grey	Bluish Black
7	Powder treated with HCl dried and mounted with nitro cellulose.	Greenish Yellow	Greenish Yellow
8	Powder treated with HNO_3 diluted with equal volume of water.	Blue	Grey

Table 5: Phytochemical screening of*Cassia tora* and *Cassia fistula* leaves

SOLVENTS	**E.A**		**Chloroform**		**Acetone**		**P.E**		**Methanol**		**Aqueous**	
Phytoconstituents	**C.T**	**C.F**	**C.T**	**C.F**	**C.T**	**C.F**	**C.T**	**C.F**	**C.T**	**C.F**	**C.T**	**C.F**
Alkaloids	–	+	–	–	–	+	+	+	+	+	+	+
Flavonoids	–	+	+	+	–	–	+	+	+	+	+	+
Glycosides	+	+	+	+	–	–	+	–	–	+	+	–
Anthraquinone	–	–	–	–	+	–	–	–	+	+	+	–
Phenols	–	+	–	–	–	+	+	+	+	–	+	+
Saponins	–	–	–	–	–	–	–	–	–	+	+	+
Steroids	+	+	+	–	+	-	+	–	+	–	+	–
Triterpenoids	–	+	–	–	–	+	–	+	–	+	+	–
Tannins	+	+	+	+	+	+	+	+	+	+	+	+
Carbohydrates	+	–	+	–	+	+	+	–	+	+	+	+
Proteins	–	+	–	–	–	-	–	–	+	+	+	+

Key: C.T : *Cassia tora*; C.R : *Cassia fistula* ; E.A : Ethyl Acetate; P.E : Petroleum ether.

'+' = Detected ; '–' = Not Detected.

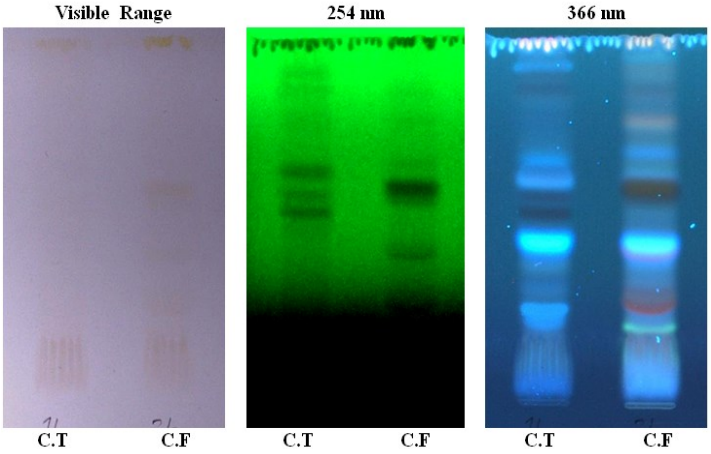


Fig. 4: Thin layer Chromatography of *Cassia sp.*

Fluorescence powder drug analysis:

Fluorescence analysis of powdered drug is another distinguishing parameter for identification of crude sample. When powdered drug and extracts were treated with different reagents, colour of the crude powder changed and when observed under UV light, they emitted various colored radiations or fluorescence (Table 4).

Preliminary phytochemical screening:

The preliminary phytochemical investigations of ethyl acetate, chloroform, acetone, petroleum ether, methanol and aqueous extracts of *Cassia tora* and *Cassia fistula* leaves were performed (Table-5). Maximum phyto-constituents were found in methanol and aqueous extracts of leaves of *C. tora* and *C. fistula* and showed the prominent presence of major secondary metabolites like flavonoids, tannins, phenolic compounds, anthraquinone, triterpenes and carbohydrates. Whereas non-polar solvent extracts such as ethyl acetate, chloroform, acetone and petroleum ether showed minimum phytocompounds. The presence of various phytoconstituents may help to develop therapeutic activity of both the species of *Cassia*.

TLC finger printing profile:

The TLC profile of hydro-alcoholic extracts of *Cassia sps.* leaves was established and carried out in solvent system [Ethyl Acetate : Formic Acid : Glacial Acetic Acid : Water (Wagner *et al.;* 2007)]. In developed TLC plates, number of prominent bands were observed at short (254 nm) and long (366 nm) UV wavelength, indicating the presence of various types active phyto-compounds in sample. TLC plates were further derivatized for confirmation of phytocontituents (Fig. 4).

CONCLUSION

Standardization is an essential measurement for ensuring the quality control of any herbal drugs. The pharmacognostic features (morphology, microscopy), physicochemical constants (foreign organic matter, total moisture, ash values, extractive values, heavy metals quantification, index values, pH etc.) and phytochemical screening are some of the integral parameters for the standardization of any crude or herbal drug. The pharmacognostical evaluation and physico-chemical parameters, may help in differentiation of *Cassia sps. viz. Cassia tora* and *Cassia fistula*, based on their morphology, anatomy and physico-chemical characters. Phytochemical screening gave qualitative data of various

phytoconstituents that further can be harvested for characterization of bioactive compounds. All the above information may act as reference for correct identification and authentication of plant materials (*C. tora* and *C. fistula*). Moreover, these investigations will help in standardization and evaluation of quality (*i.e.* impurities and adulterant identification) of crude sample which further can be useful in formulation of herbal or medicinal drug. Thus the present comparative study is significant for standardization and quality assessment of crude drug.

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