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Pharmacognostic standardization and FT-IR analysis of various parts of *Sageretia thea*

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Key words: Fluorescence, phytochemical and FTIR analysis of various parts of *Sageretia thea*.

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Abstract

In the present study various parameters like fluorescence study, phytochemical screening analysis and FT-IR analysis of various parts of *Sageretia thea* (Osbeck) M.C.Johnston carried out for correct identification and detection of secondary metabolites and functional groups in these parts. The current study revealed that different plant material give different coloration when treated with various chemicals which act as a pharmacognostic parameters for identification and standardization of that drug from their adulterants. The phytochemical analysis showed the presence of carbohydrate, alkaloids, phenol, saponins, tannins, phytosterols, flavonoids and glycosides while the FT-IR analysis showed different functional groups in various part of the plant.

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Introduction

Pharmacognosy is the study of drugs derived from natural sources (plants and animals), which have active constituents capable of curing various ailments Okigbo *et al.* (2008). Today more than 50% drugs are obtained from plants in which 25% are contributed by higher plants Cragg and Newman (2005). Many flowering plants have the potency for curing different disease Newman *et al.* (2000) for, example leaves, root and seeds of *Datura lanosa* is used for skin ailments and various pains Bye (1991), *hyoscyamus niger* posses sedative and analgesic properties Duke, (1985). In broad since, Pharmacognosy is the study of different parameters of plants in which botanical dietary supplements, including herbal remedies are studied from biological, biochemical and economic point of view (Cardellina, 2002 and Tyler, (1999).

According to therapeutic point of view root and stem of *S.thea* has been used as folk medicine for hepatitis treatment in China (Xu *et al.,* 1994). Traditionally, its leaf has been used as a substitute for tea in Korea (Flora of China). Leaves water extracts of *S. thea* possessed strong antioxidative (Shin *et al.*, 2004) and moderate HIV-1 protease inhibitor activities (Cheol *et al.,* 2002). The HPLC and NMR characterization of *S. thea* leaves shows the presence of anthraquinone glycoside and flavonoid glycosides (Shen *et al.*, 1994). The ethyl acetate fractions of *S. thea* leaves showed high contents of phenolic compound and have the potential of strongest reactive oxygen species scavenging activity (Chung *et al.*, 2009). The extracted pigments from the edible fruit of *S. thea* showed good solubility in polar solvents while poor in alkaline solvents (Hong *et al.,* 2009). Different crude when viewed under UV light drugs show different fluorescence at different wavelengths, so different parts of *S. thea* also studied for fluorescence study to create a pharmacognostic parameter, Active constituents of medicinal plants are secondary metabolites which play important role in metabolism (Rajurkar and Damame, 1997), Kamboj & Saluja (2010) carried out the phytochemical analysis of *Bryophyllum pinnatum*. The FTIR spectroscopy is a simple and rapid analytical method used for detection of functional groups and highly polar bonds of the components, Sene *et al.,* 1994 carried out FTIR analysis of cell wall of five angiosperms members, Mariswamy *et al.,* (2012) carried out the FTIR analysis of *aerva lanata,* Kacurakova *et al.,* (2012) reported the FTIR spectroscopy of pectic polysaccharides and hemicelluloses compounds. The detection of the functional groups and phytochemical constituents is compulsory for the investigation of therapeutic potential of the drug. In the current study the fluorescence study, phytochemical screening test and FTIR spectroscopy of leaf, stem and root of *S. thea* is carried out for the correct identification and standardization of these drugs.

Material and method

Collection and preservation

Fresh specimens of *S. thea* collected from Dir, Khyber pakhtoonkhwa, Pakistan. The collected parts of the plant were cleaned, washed with tape water, separated and dried in shade. The dried specimens were next powdered by electric grinder and used for different tests i.e phytochemical screening test, FT-IR analysis and fluorescence analysis.

Powder drug florescence study

Powder drug of different parts of plant give different fluorescences in under ultraviolet radiation; therefore florescence evaluation is used for identification of plant and powder drugs (Jarald and Jarald, 2007).

Apparatus required

UV tube (366 nm), glass slide, dropper.

Chemicals required

NH_3 solution, Tape water , Picric acid, 50% HNO_3, Ethanol, n-hexan , 50% H_2SO_4, Methanol and 50% Hcl.

Procedure

The fluorescence analysis of dried powder of leaf, stem and root of *S. thea* was carried out by treating 1

gm dried powder of each part with different chemicals (50% Hcl, 50% H_2SO, 50% HNO_3, picric acid, untreated, methanol, tape water, NH_3 solution, ethanol and n-hexan) and each treated sample was observed under ordinary light and then under UV light of both long and short wave lengths (Brain and Turner, 1975; Evans, 2002).

Phytochemical screening test

Preparation of extract by using organic solvent

To derive extract from leaf, stem and root of *S. thea* 500 g of each sample were soaked separately in 3 liters of ethanol for 15 days and keep these on electric magnetic stirrer model no Corning PC-420D. The extract were then filtered and evaporated through rotary evaporator under reduced pressure. Each extract of the three samples were collected and used for qualitative analysis, as given below.

Qualitative analysis

For the detection of different secondary metabolites (carbohydrates, proteins, alkaloids, phytosterols, triterpenoids, phenols, tannins, saponins, flavonoids, fixed oil and volatile oil) various qualitative screening test were conducted for each part of *S. thea*.

FTIR Spectroscopy

The leaf, stem and root parts of *S. thea* were ground into fine powder by using electric grinder FT-IR spectrometer in the range of 4000-400cm^{-1} by employing standard KBr pellet technique.

Table 1. UV & Visible Fluorescence study of leaf, stem and root powder of S. thea with different chemical. reagents.

Powder Treatment	Leaf		Stem		Root	
	Visible light	**UV 366**	**Visible light**	**UV 366**	**Visible light**	**UV 366**
Untreated	Dull green	Dark green	Green	Light green	Yellow	Whitish
NH_3	Green	Black	Green	Dark green	Orange	Brown
Tape water	Dark green	Light green	Spring green	Grass green	Dry grass yellow	Pale yellow
Picric acid	Yellowish	Brown	Yellow	Brownish	Yellow	Yellow
50% HNO3	Yellow red	Brick red	Brick red	Dark brown	Brown	Reddish brown
Ethanol	Light Green	green	Yellowish	Spring green	Orange yellow	Yellow
50% HCL	Gray	Gray	Light red	Dark brown	Brick red	Light red
n-hexane	Green	Dark green	Light green	Grass green	Orange	whitish
50% H2SO4	Black	Black	Black	Brown black	Black	Dark black
Methanol	Light greenish	Light red	Yellowish	Dark Red	Light brown	Dark brown

Result and discussion

Florescence analysis

Different crude drugs when viewed under UV light show different fluorescence at different wavelengths. This is due to the presence of different chemical constituents in the drug. If the drugs do not show fluoresce it is first treated with different reagents which convert them into fluorescence derivatives. The fluorescence analysis of drugs is helpful for identification of crude drugs as well as for the detection of adulterations in whole or powder drugs (Reddy and Chaturvedi, 2010; Ansari *et al.*, 2006: Wallis, 1985). The florescence study the powder of leaf, stem and root of *S.thea* was carried out with different reagents under visible and ultra violet lights. The UV light (UV 366) was used for the florescence character of power drugs. The observations showed marked variation in coloration based on the part used, nature of reagents and wavelength light. The florescence study results are given in table (Table 1).

Table 2. Phytochemical study of leaf, stem and root of *Sageretia thea*.

S. No	Biomolecules	Leaves	Stem	Root
1	Carbohydrates	+	+	+
2	Phenols	+	+	+
3	Alkaloids	+	+	+
4	Saponins	+	+	-
5	Phytosterols andTriterpenes	+	+	+
6	Tannins	+	+	+
7	Flavonoids	+	+	+
8	Glycosides	+	+	+
9	Fixed oil	+	-	-

Many researchers have worked out fluorescence analysis of different medicinal plants like *Kirganelia reticulate* (Shruti *et al.,* 2011); *Polygonum cuspidatum* (Tian *et al.,* 2006) and *Hygrophila auriculata* (Hussain *et al.,* 2011). The results of present study have a great similarity with the findings of these workers.

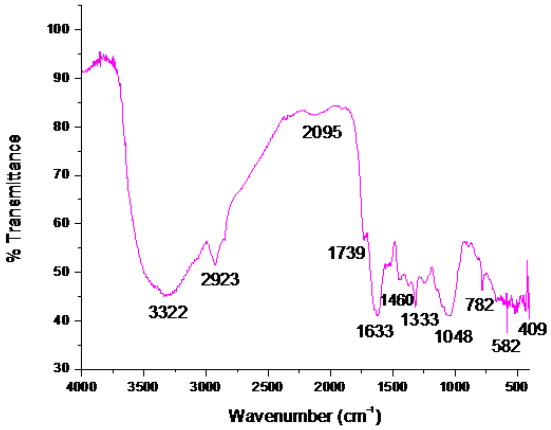

Fig. 1. FTIR spectrum of *Sageretia thea* leaves.

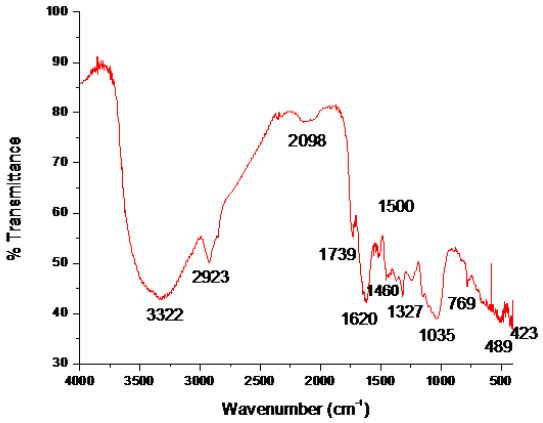

Fig. 2. FTIR spectrum of *Sageretia thea* Stem.

Phytochemical screening analysis of Sageretia thea

Each part of plant contains pharmacologically active compounds which are isolated from plant sources through a useful preliminary phytochemical screening method. In the present study, qualitative phytochemical screening was carried out using ethanolic extracts of leafs, stem and root of *S. thea*. The results are given in (Table 2). These results indicate that *S. thea* is a rich source of both primary (carbohydrate) and secondary metabolites. Saponins were present in leaves and stem but not detected in root. Alkaloids, Phytosterols and Triterpenes, Tannins, Flavonoids and Glycosides were detected in all the three parts while fixed oil were absent in all of these parts.

Some phytochemist carried out phytochemical analysis of various parts of plants like Rao *et al.*

(2011) carried out phytochemical screening of ethanolic extracts of *Tephrosia purpurea, Alternanthera sessilis*, and *Clitoria ternate* and detected flavonoids, phenols, alkaloids and tannins in the leaves of the plants. Menon and Latha (2011) worked out on different organic solvent extracts of *Coleus forskohlii* and reported a wide range of active compounds like saponins, protein, tannins, alkaloids, glycosides, carbohydrates, Phenols and cardiac glycosides.

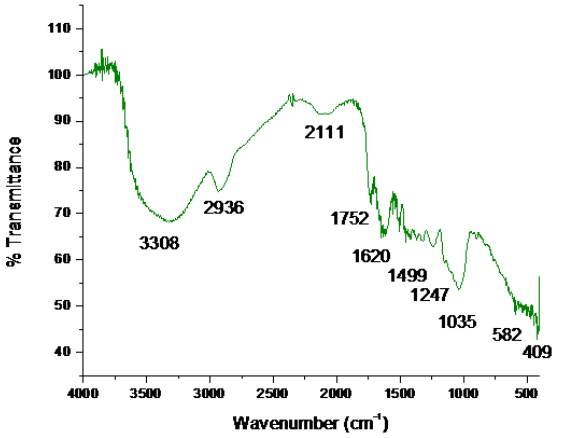

Fig. 3. FTIR spectrum of *Sageretia thea* root.

FTIR spectroscopy

Fourier transform infrared (FTIR) spectroscopy is a simple and high resolution method used for identification of different bonds and functional groups (Griffiths and Haseth, 1986). The FTIR spectrum was used for detection of functional groups based on the peak values in the region of infrared radiation. The leaves powder of *S. thea* was passed into FTIR and functional groups of the components were separated based on its peak. In the current study the FTIR analysis of *S.thea* leaf, stem and root confirmed the presence of amides, alkanes, bending water, lipids, alkenes, aromatic ring and chlorides compounds which shows peaks at range of 3305-3325, 2920-2937, 2050-2098, 1739-1769, 1617-1639, 1440-1505 and 405-4027 respectively (fig 1, 2, 3; table 3). Aldehydes shows peak at 765-789 only present in leaves and stem while phenyl compound shows peak at 582 present in leaf and root both. Ribose shows peak at 1048 present only in leaf. Similarly glucose shows peak at 1035 present in stem and root. Nitro compound, alcohol and alkyl halides shows peak at 1500, 1327 and 489 respectively only detected in stem. Carbohydrate present at 1247 with a high intensive peak only detected in root. Many other researchers also worked on FTIR analysis like Freeman *et al.,* 1994 and Wenning *et al.,* 2002. Similarly Wellner *et al.,* 1998, carried out the FTIR analysis for pectate and pectinate gel.

So our current results indicate that the presence of different functional groups in all the three parts of *S. thea* made it a perfects product for any kind of pharmaceutical application.

Table 3. FTIR Peaks values of leaf, stem and root of *Sageretia thea*.

LEAVES		STEM		ROOT	
Peak values	Functional groups	Peak values	Functional groups	Peak values	Functional groups
3322	Amines,amides	3322	Amines,amides	3308	Amines,amides
2923	Alkanes	2923	Alkanes	2936	Alkanes
2095	Rotating water	2098	Rotating water	2111	Binding water
1739	Lipids	1739	Lipids	1752	Lipids
1633	Alkenes	1620	Alkenes	1620	Alkenes
1460	Aromatic	1460	Aromatic	1499	Aromatic
1333	Alcohol	1500	Nitrocompoud	1247	Carbohydrates
1048	Ribose	1327	Alcohol	1035	Glucose
782	Aldehydes	1035	Glucose	582	Phenyl
582	Phenyl	769	Aldehydes	409	Chlorides
409	Chlorides	489	Alkyle halides		
		423	Chlorides		

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