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Pharmacognostical Profiling of *Jasminum grandiflorum* Linn. leaves

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Abstract

To evaluate the pharmacognostic characters including macroscopic, microscopic and physico-chemical characters of the leaves of therapeutically important plant *Jasminum grandiflorum* Linn.

Fresh and dried leaf samples were studied macroscopically and microscopically. Preliminary phytochemical screening and physciochemical studies were performed by following WHO recommended guidelines for the standardization of the leaves.

The detailed microscopy revealed the presence of multicellular trichomes, xylem tissue, phloem tissue, collenchyma, spongy parenchyma and palisade cells. Physico-chemical parameters such as extractive values, ash values, foreign matter, loss on drying, volatile oil content, swelling index, foaming index, crude fibre content, fluorescent behaviour, microbial contamination, aflatoxin content, heavy metal profile, pH values of drug solution were also determined. Preliminary phytochemical screening showed the presence of carbohydrate, terpenoids, steroids, saponins, flavonoids, tannins & phenolics compounds.

Various pharmacognostic characters observed in this study can help in the identification and standardization of leaves of *Jasminum grandiflorum* Linn.

Keywords: *Jasminum grandiflorum,* Pharmacognostic character, Aflatoxin content, Heavy metal analysis, Microbial contamination.

Introduction

Over the last decade, due to the limitations associated with synthetic pharmaceutical products; the avenues have been opened for Green Medicine which is considered to be safe, more accessible and affordable too[1]. Since crude plant drugs form the basis for the manufacture of numerous medicinal preparations, accurate determination of drug identity forms an essential aspect of its study. It becomes extremely important to make an effort towards standardization of the plant material. The process of standardization can be achieved by stepwise pharmacognostic studies [2]. This study helps in identification and authentication of the plant material. It is estimated that about 25% of all modern medicine are directly or indirectly derived from higher plants. Jasminum is a genus of shrubs and vines in the olive family Oleaceae with about 200 species throughout the world, out of which around 40 species are reported to be growing in India [3].

Jasminum grandiflorum Linn. commonly known as Chameli; one of the species of jasminum is native to Asia, China, Afghanistan, Persia, India, Philippines, Myanmar and Sri Lanka. Its various parts such as the leaf, stem, bark, flower and root are very useful and important in pharmaceutical industry and have been reported to possess medicinal values. Traditionally leaves are used in dysmenorrhoea, fixing loose teeth, leprosy, ottorrhoea, otalgia, odontalgia, skin diseases, strangury, ulcerative stomatitis, ulcers, wound and corns [4, 5, 6]

The plant is reported to possess anti-viral, anti-microbial, anti-inflammatory, anti-acne, wound healing, spasmolytic, anti-ulcer and anti-oxidant activities [7-13]. The leaves contains sambacin I-III, oleacein, isoquercitrin, ursolic acid, 2-(3, 4-dihydroxyphenyl) ethanol [14], oleanolic acid, 2"-epifraxamoside, demethyl-2"–epifraxamoside, jasminanhydride, 3, 4-dihydroxy benzoic acid,2-hydroxy-30,40 di- hydroxyacetophenone [15], indoleoxygenase [16], resin, salicylic acid and jasminine.

Due to numerous medicinal uses attributed to this plant, it becomes enormously important to interpret morphological and anatomical descriptions of crude drugs as well as characteristic features of drugs of commercial significance. So, in order to establish the pharmacognostical, and physcio-chemical characters of leaves of the plant, the present study was undertaken, which would assist in standardization and guarantee quality, purity and identification of crude drug sample.

Materials and Methods

Plant collection and authentication

Leaves were collected from healthy plants of *Jasminum grandiflorum* Linn. from the medicinal garden of Hindu College of Pharmacy, Sonepat (Haryana). Herbarium so prepared was authenticated by Dr. H. B. Singh (Scientist F and Head, Raw Materials Herbarium and Museum, NISCAIR, New Delhi) under voucher specimen no. NISCAIR/RHMD/Consult/-2010-11/1627/225 dated January 5, 2011 and specimen was deposited in the department. The plant material was dried under shade by spreading as single layer and then coarsely powdered.

Macroscopy

Untreated sample was examined under diffused day light and colour of sample was recorded. The powder was rubbed slowly between fingers and odour was examined. Taste of the powder was also checked. Surface of material was touched to determine whether it was soft or hard [17, 18].

Microscopy

Thin, free hand transverse sections of fresh leaves were made with the help of sharp blade and cleared with chloral hydrate solution. The sections were stained with phloroglucinol and conc. hydrochloric acid and mounted in glycerin. These were observed under compound microscope and photographed [19, 20].

The powder microscopy was carried out after passing the powdered drug through #60. The powder so obtained was treated with chloral hydrate solution and stained with phloroglucinol and conc. hydrochloric acid and mounted in glycerin. This was observed under compound microscope and photographed [19, 20].

Physico-chemical parameters

Extractive values and successive extractive values of *J. grandiflorum* leaves powder were determined according to standard procedures using petroleum ether (60-80 C), chloroform, ethanol and water. Total ash, water soluble ash and acid insoluble ash values were studied according to standard procedures [21-26].

Preliminary phytochemical analysis of successive extract of leaves extracts was performed according to standard procedure [20, 25, 26].

Fluorescence analysis was conducted according to standard procedure [27-28].

Quantitative studies

Loss on drying, foaming index, swelling index, volatile oil content, crude fibre content, aflatoxin content, microbial contamination, heavy metal analysis of powdered leaves and pH values of 1% w/w and 10% w/w powder in water were determined as per WHO guidelines [21-26].

Results and Discussion

Macroscopy

Leaves are compound and variable in shape usually ovate to somewhat elliptic in shape. The apex is acute, margin entire, base asymmetrical, 1-1.6cm (length) by 0.5-1.5cm (breadth) as shown in Plate 1. Leaflets are 7-11 and the terminal one is larger than the rest but not very markedly so. The lower surface is comparatively rough with prominent midrib and pinnate venation. The fresh leaf is green in color with characteristic odor and slight bitter taste.

Microscopy

A transverse section of the leaflet shows a central midrib with lamina on both sides passing. It shows the following structure as shown in Plate 2.

Both the upper and lower epidermis are covered with thick striated cuticle. Multicellular thick walled warty trichomes are present only on upper epidermis. Paracytic stomata are present only on the lower epidermis.

Below the upper epidermis, two layers of rod shaped palisade parenchyma are present. The 2nd row of palisade cells underneath epidermis in the midrib region is ill developed.

Palisade layer is followed by three to five rows of spongy parenchyma. This layer has several vascular strands encircled by parenchymatous sheath.

Midrib region is deeply convexed on lower side (abaxial surface) while its upper surface (adaxial surface) is only slightly depressed. Midrib region has large conjoint collateral vascular bundle in the centre having about ten radially arranged xylem strands in the centre. The parenchymatous phloem is external to xylem. The phloem is encircled by sclerenchymatous sheath. Below this vascular bundle are present. Five to six rows of thick walled collenchymatous cells are also present. Rachis is concavo-convex in shape.

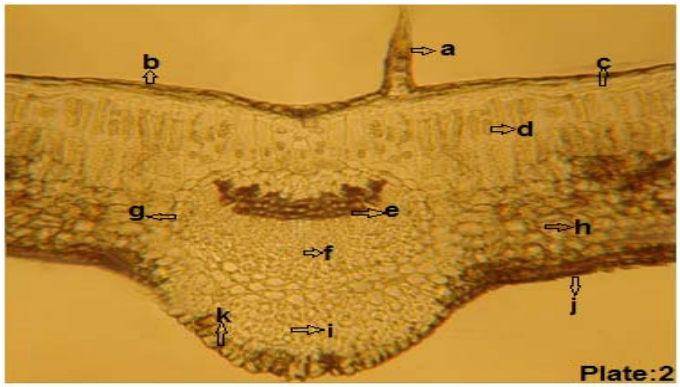

Plate 2: a: Trichome, b: Upper cuticle, c: Upper epidermis, d: Palisade cell, e: Xylem, f: Phloem, g: Sclerenchymatous sheath, h: Spongy parenchyma, i: Collenchyma, j: Lower cuticle, k: Lower epidermis.

Powder Microscopy

The dried leaf powder was green in color with characteristic odor and slightly bitter taste. Microscopy of the powder revealed the presence of double celled uniseriate trichome with pointed end and reticulately lignified xylem vessels. The surface view showed the presence of paracytic stomata and palisade cells with circular appearance as depiciated in Plate 3(a-d).

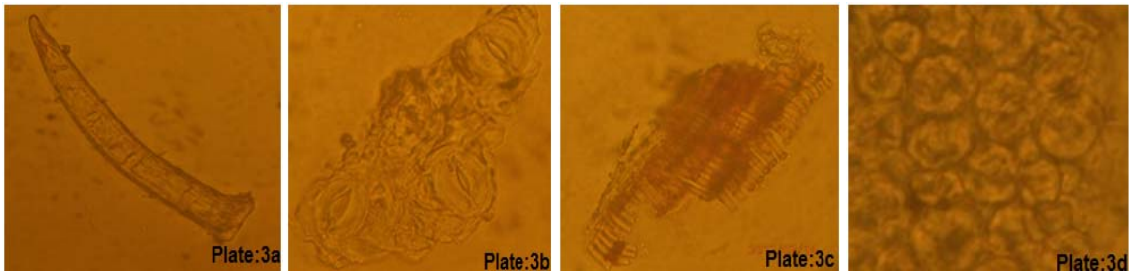

Plate 3: 3a: Covering trichome, 3b: Paracytic stomata, 3c: Xylem vessel, 3d: Palisade cell.

Qualatative Analysis

The ethanol soluble and water soluble extractive values were found to be 15.91% and 16.86% w/w respectively. The leaves yielded successive extractive values of 1.33%, 2.93%, 14.67% and 15.26% w/w with petroleum ether (60-80 C), chloroform, ethanol and water respectively. The total ash value of the crude drug was found to be 9.07% w/w while water soluble ash and acid insoluble ash values were determined as 2.65% and 1.05% w/w, respectively. The preliminary phytochemical examination showed the presence of steroids, saponins, flavonoids, tannins & phenolic compounds. The fluorescence behaviour of the powder of leaves, moistened with solvents and chemical reagents; under UV (long and short) and normal day light is given in Table 1.

Table 1: Fluorescence analysis of Leaves of *J. grandiflorum*

Treatment of Powder with	Observation under		
	Visible light	UV light	
		254 nm	356 nm
As such	Blackish green	Bluish black	Dark green
1N HCl	Brownish black	Black	Black
1N H_2SO_4	Light brown	Greenish dark brown	Dull greenish brown
1N HNO_3	Blackish brown	Greenish black	Dark brown
5% $FeCl_3$(Alc.)	Black	Black	Black
5% $FeCl_3$(Aq.)	Greenish black	Yellowish dark blue	Black
1N NaOH(Alc.)	Black	Black	Black
1N NaOH(Aq.)	Redish brown	Greenish black	Greenish black
1% nitrocellulose in amyl acetate	Greenish black	Greenish black	Greenish black
1N NaOH(Alc.) +1% nitrocellulose in amyl acetate	Black	Greenish black	Black
1N NaOH(Aq.) +1% nitrocellulose in amyl acetate	Redish black	Greenish redish brown	Yellowish dark green
1N HCl +1% nitrocellulose in amyl acetate	Blackish brown	Dark blue	Dark green

Quantitative Studies

Loss on drying content and crude fibre content was determined to be 5.83% and 7.09% w/w respectively. The drug was devoid of volatile oil content and foaming index was found to be less than 100. The swelling index of crude drug was also found to be nil. Aflatoxin content and microbial contamination of leaves powder were confirmed to be within limits as shown in Table 2. Heavy metal analysis revealed that each element was present within specified limits as per Ayurvedic Pharmacopoeia of India as shown in Table 3. The pH values of 1% and 10% w/w drug solutions were found to be 7.1 and 6.2, respectively.

Table 2: Aflatoxin and Microbial Contamination Test

Parameter	Value	Specified limit
Total bacterial count	72 c.f.u./g	$1 X 10^5$ c.f.u./g
Total yeast/mould count	Nil	$1 X 10^3$ c.f.u/g
E. coli	Nil	Nil
Salmonella sp.	Nil	Nil
S. aureus	Nil	Nil
P. aeruginosa	Nil	Nil
Aflatoxin B_1	Absent	0.5 ppm
Aflatoxin B_2	Absent	0.1 ppm
Aflatoxin G_1	Absent	0.5 ppm
Aflatoxin G_2	Absent	0.1 ppm

Table 3: Heavy Metal Content

Heavy metal	Result (ppm)	Specified limit (ppm)
Arsenic	Nil	3.00
Cadmium	0.09	0.30
Lead	1.20	10.0
Mercury	Nil	1.00

Discussion

For the utilization of medicinal plant as a biosource,/ or To utilize a plant as a biosource or herbal drug, correct identification and quality assurance of the starting materials is an essential prerequisite. Therefore the evaluation of a crude drug is an integral part for establishing the correct identification of a plant material to ensure reproducible quality of herbal medicine which will contribute to its safety and efficacy. Before including any crude drug in herbal pharmacopoeia, its pharmacognostic parameters and standards must be established [29].

In the present investigation the macroscopic, microscopic and physiochemical studies of *J. grandiflorum* leaves was carried out. The results of macroscopic study might be valuable for distinguishing it from its substitutes and adulterants.

Microscopic evaluation allows more conscientious examination of crude drug and enables to identify the organized structural features such as epidermis, trichomes, parenchymatous cells.

Studies of physico-chemical parameters can serve as a valuable source of information and are usually used in judging the purity and quality of the drug. Loss on drying for *J. grandiflorum* leaves was nearly 6%. It signifies the considerable amount of moisture in leaves. The moisture content of a drug should be minimized in order to prevent decomposition of crude drug either due to chemical change(s) or due to microbial contamination.

The extractive values in different solvents give us information about the chemical nature of the constituents present in the plant whereas ash value tells us regarding contamination of drug with sand and soil. These values are important quantitative standards as it is useful in determining authenticity and purity of drugs [30].

The fluorescence behaviour of powdered drugs plays a vital role in the determination of quality and purity of the drug material. Fluorescence is the phenomenon exhibited by various chemical constituents present in the plant material. Some constituents show fluorescence in the visible range of daylight. The ultra violet light produces fluorescence in many natural product (e.g. alkaloids like berberine), which is not visible in day light. If the substances themselves are not fluorescent, these may often be converted into fluorescent derivatives or decomposition products by treating with different reagents. Hence, some crude drugs are often assessed qualitatively in this way and it is an important parameter of pharmacognostical evaluation [31]. The result of fluorescence analysis of leaf powder showed their characteristic fluorescent color in different organic and inorganic solvents.

The preliminary phytochemical analysis revealed the presence of various secondary plant metabolites in the leaves which are known to possess various therapeutic values in medical sciences. The

microbes usually contaminate medicinal plants; which may arise during cultivation, harvesting, processing and storage. Hence the total microbial load represents the care taken during these procedures.

Microbes are mainly represented by bacteria and fungi. The fungi can produce aflatoxin; some of which can be potentially dangerous. Even the bacteria can lead to exo or endo toxins. The presence of microorganism coupled with moisture can lead to enhanced enzymatic activity hence transforming some of the active constituents to other metabolites which may be less or non potent. All this necessitate the control of microbial contamination with in prescribed limits of pharmacopeia [32].

The medicinal plants accumulate lead, cadmimum, mercury and arsenic from their environment. The heavy metals are health perilous for humans and animals. Hence their content in medicinal plant is controlled with in prescribed limits of pharmacopeia [33].

In conclusion, the parameters which are reported here will be used as a diagnostic tool for standardization of this medicinal plant. Further studies are in progress on these leaves in order to isolate, identify, characterize and elucidate the structure of bioactive compounds along with exploration of their pharmacological activity.

Authors Contribution

AM, SS, AP designed and planned the study. AM carried out experimental work. SS participated in identification and collection of plant material. AP participated in analysis of heavy metal, aflatoxin and microbial contamination. AM drafted and revised the manuscript. All authors read and approved the final manuscript.

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