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PHARMACOGNOSTIC AND PHYTOCHEMICAL INVESTIGATION OF POTENTIALLY IMPORTANT PLANTS OF WESTERN GHATS, INDIA

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ABSTRACT: Objective: The Indian traditional system of medicine, namely Ayurveda and Siddha, are primarily based on the use of the whole plant or different parts of plant singly or in combination to make multifactorial formulations. The use of plant drugs, however, demands correct identification of species and the characterization of phytoconstituents. **Materials and Methods:** The plants reported for their wound healing property in the Western Ghats by tribal community viz. *Semecarpus anacardium* L. (Bibba), *Argemone mexicana* L. (Firangi Dhotara), *Cocculus hirsutus* L. (Vasanvel), *Woodfordia fruticosa* Kurz. (Dhatki) were identified, authenticated and processed. The plant materials were subjected to morphological and microscopical evaluations. The proximate analysis (moisture content, ash values, extractive values and foreign organic matters) of powdered drugs were carried out. The powdered plant materials were extracted by a Soxhlet extraction process using different solvents. The concentrated extracts were subjected to preliminary phytochemical investigation. **Results:** The *Semecarpus anacardium* L. shows less moisture (fruits; 8.73 ± 0.95) and high inorganic content (leaves; 17.50 ± 0.92) as compared to other powdered drugs. The *Argemone mexicana* L. (35.68 ± 2.92), *Cocculus hirsutus* L. (41.53 ± 3.08) and *Woodfordia fruticosa* K. (29.26 ± 1.75 and 24.06 ± 3.01) shows high extractive values in methanolic extract whereas *Semecarpus anacardium* L. (fruits; 34.61 ± 0.75 and leaves 37.69 ± 1.86) shows in ethanolic extract. The least foreign organic matters (0.76 ± 0.45) were found in *Woodfordia fruticosa* K. flowers. The alkaloids, carbohydrates, steroids, glycosides, flavonoids and phenolic compounds were found in ethanolic and methanolic extracts. **Conclusion:** The data thus obtained by standardization of plant materials can be used during the preparation of polyherbal formulations to increase stability and efficacy.

INTRODUCTION: The importance of medicinal plant in drug development is known to humans and they have been using them for the treatment of different diseases since the beginning of human life ¹.

The medicinal plants are available abundantly for human beings and animals from thousands of years due to the presence of phytochemical constituents.

India is recognized as an emporium of medicinal plants because of the presence of over 45,000 plant species in different bioclimatic zones. In India, about 90% of the herbs and medicinal plants used in industries are collected from wild sources ². The traditional folk treatment from wild plants has always guided researchers to search for novel medications to develop healthy life for humans and animals ¹.

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The Indian traditional system of medicine, namely Ayurveda and Siddha, is primarily based on the use of whole plant or different parts of the plant singly or in combination to make multifactorial formulations. The renowned interest of plant-based drugs or medicines is because of easy availability, economic and less or no side effects³. Nowadays, the demand for plant-based drugs is increased, which ultimately leads to the chances of substitution or adulteration. The therapeutic efficacy of medicinal plants always depends upon the quality and quantity of chemical constituents. The use of plant drugs, however, demands correct identification of species and the characterization of phytoconstituents. The misuse of herbal medicine or natural products starts with wrong identification, which can be solved by pharmacognostic studies comprising authentication, processing and standardization of medicinal plants⁴.

The process of standardization can be achieved by stepwise pharmacognostic studies, which are an essential measure of quality. Most of the research in 'Pharmacognosy' consists of the identification and processing of controversial plants species through morphological, phytochemical and physicochemical analysis³.

Despite the available modern techniques, ordinary light microscopy is still the most common method for primary authentication and has been universally used in the authentication of herbal medicines in India and many other countries because of its virtues of the requirement of a small amount of sample, fast speed and economy⁴.

According to the World Health Organization, the macroscopic and microscopic description of a medicinal plant is the simplest and cheapest step towards establishing the identity and degree of purity of such materials and should be carried out before any tests are undertaken⁵. Unlike taxonomic identification, the pharmacognostic study includes parameters which help in identifying adulteration in dry powder form also. Therefore pharmacognostic studies will ensure reproducible quality of herbal products which will lead to safety and efficacy of natural products³. It will also provide helping hands for the industrialists for manufacturing of superior herbal based formulations⁶. The objective of the present research work was to study the

various pharmacognostic properties of therapeutically important plants of Western Ghats, India, *i.e.* *Semecarpus anacardium* L. (Bibba), *Argemone mexicana* L. (Firangi Dhotara), *Cocculus hirsutus* L. (Vasanvel), *Woodfordia fruticosa* Kurz. (Dhatki). These plants are used in traditional and folk medicinal practices and reported by the tribal or local community for their wound healing potential^{7,8}. Present research work deals with pharmacognostic evaluation and standardization (macroscopical, microscopical and physicochemical) of research above plants, which will help their further utilization in the preparation of polyherbal formulation to explore its wound healing property.

MATERIALS AND METHODS:

Procurement, Authentication and Processing of Plant Materials:⁹ The research plants *viz.* *Semecarpus anacardium* L. (Anacardiaceae), *Argemone mexicana* L. (Papaveraceae), *Cocculus hirsutus* L. (Menispermaceae), *Woodfordia fruticosa* Kurz. (Lythraceae) were collected from Western Ghats, India and deposited to Botanical Survey of India (B.S.I.), Pune (Maharashtra, India) for identification and authentication (Reference number BSI/WRC/Tech./2013/SND-1 Dated 06/12/2013; BSI/WRC/Tech./2013/JRB-01 Dated 27/11/2013; BSI/WRC/Tech./2013/GVG-01 Dated 31/12/2013; BSI/WRC/Tech./2013/GG-01 Dated 31/12/2013, respectively). The identified and authenticated plant materials were processed to remove adhered dirt and toxic components, dried and pulverized by the use of laboratory blender (REMI) to get coarse powder by passing through sieve no. 40 and retained on sieve no. 60.

Standardization of Plant Materials: The powdered crude drugs and freshly collected plant materials were subjected to stepwise pharmacognostic studies. The standardization of plant materials was carried out based on qualitative (morphological and microscopical) and quantitative (proximate) evaluations.

Morphological and Microscopical Evaluation:^{10, 11, 12} The freshly collected plant parts were evaluated qualitatively for morphological and microscopical characteristics. The characteristics *viz.* size, shape, apex, margin, color, odor, taste and extra features, *etc.* were used for morphological

evaluations. The microscopical characteristics were identified by dissecting plant parts.

Proximate Evaluations: ^{5, 9, 11, 13} The powdered crude drugs were analyzed quantitatively for different physicochemical parameters viz. moisture content, ash values, extractive values, foreign organic matters as per the standard procedures.

Preparation of Test Extracts: ^{11, 14} The powdered plant materials were packed in Soxhlet apparatus and then successively extracted with series solvents; petroleum ether, chloroform, ethanol and methanol (40 cycles each). The aqueous extract was obtained by maceration of remaining marc for seven days. After complete extraction extracts were filtered and concentrated for further studies at

reduced pressure and temperature in a rotary evaporator.

Preliminary Phytochemical Evaluation: The concentrated tests extracts were subjected to preliminary phytochemical investigation for the detection of various secondary metabolites such as tannins, saponins, sterols, triterpenes, alkaloids, flavonoids, protein/amino acids and carbohydrates and glycosides ^{9, 11}.

RESULTS AND DISCUSSION: The plants for investigation were collected from different regions of Western Ghats, India **Table 1**. Detoxification or purification process involves the conversion of the poisonous drug into beneficial, non-poisonous, or non-toxic one's **Table 2**.

TABLE 1: DETAILS OF COLLECTION OF PLANT SPECIES AND PLANT PARTS

S. no.	Common/Local Names of Plant	Plant Parts	Place of Collection
1	Bhallataka/Bibba	Fruits and leaves	Kanersar, Tal- Khed, Pune, Maharashtra, India
2	Mexican Prickly Poppy/ Firangi Dhotara	Whole plant	Wagholi, Tal- Haveli, Pune, Maharashtra, India
3	Broom Creeper/Vasanvel	Whole plant	Pirangut, Tal- Mulasi, Pune, Maharashtra, India
4	Red Bell Bush/Dhatki	Leaves and flowers	Pirangut, Tal- Mulasi, Pune, Maharashtra, India

TABLE 2: DETAILS OF DETOXIFICATION OF COLLECTED PLANT MATERIALS

S. no.	Plant Species	Plant Part	Method of Detoxification	Significance of Detoxification
1	<i>Semecarpus anacardium</i> L.	Fruits	The thalamus portions of the collected fruits were removed and kept in Gomutra (for 7 days) and then washed with water. The fruits were then shifted to a jute bag containing brick gravels (for 3 days), rubbed thoroughly and dried ^{13, 15}	The toxic compounds like Bhilawanol and Anacardic acids present in tarry oil of pericarp of the fruits were removed
		Leaves	The collected leaves of the plant were washed with distilled water	The dust, foreign particles, debris, etc. were removed.
2	<i>Argemone mexicana</i> L.	Whole plant	The collected whole plant materials were washed with distilled water and lime-water	The dust, earthy matters, foreign particles, debris, etc. were removed
3	<i>Cocculus hirsutus</i> L.	Whole plant		
4	<i>Woodfordia fruticosa</i> Kurz.	Leaves	The collected leaves and flowers were washed with distilled water	The dust, foreign particles, debris, etc. were removed
		Flowers		





The processing of collected plant materials and parts was performed to detoxify indigenous toxic ingredients, to improve the purity of the drug, to reduce the drying time, to prevent damage from mold and to achieve desired therapeutic efficacy of the drug ¹⁵. The excessive moisture of the collected plant materials was removed by the drying process. These dried drugs were coarsely powdered by using a mechanical grinder and stored in airtight containers separately with appropriate labeling until they were used for further evaluations. The current pharmacognostical methods in identification of crude drugs are based on their

morphological, microscopical or histological studies and chemical analysis. The morphological or macroscopical characteristics of the plant materials would be beneficial for identification and standardization on primary basis ^{5, 11}.

The plant parts were evaluated for morphological as well as for organoleptic properties **Table 3**. The morphological and organoleptic characters of crude drugs were not always sufficient for identification and standardization of herbal drug. Therefore, crude or fresh parts of plant materials were confirmed by their histological evaluation ^{5, 11}.

The microscopic analysis can provide supporting evidence, which, when combined with other analytical parameters for standardization and evaluation of herbal drugs.

TABLE 3: MORPHOLOGICAL CHARACTERISTICS OF COLLECTED PLANT MATERIALS

S. no.	Plant Species	Collected Plant Part	Description
1	<i>Semecarpus anacardium</i> Linn.		Fruits are dark or light green, ovoid and smooth lustrous, about 2.5 cm long, 2–3 cm broad and shining black when ripe. Leaves are stiff, large and light to dark green. It is obovate-oblong in shape which having rounded blunt apex and symmetrical base. The texture is leathery
2	<i>Argemone mexicana</i> Linn.		It is a prickly, hairless, branching herb of height 1-4 feet with milky yellow juice and showy yellow flowers at the end of branches. Leaves are whitish green, sessile, with thistle-like shape and prickles on both surface, margins somewhat the teeth tipped with a prickle
3	<i>Cocculus hirsutus</i> Linn.		It is a perennial scandent shrub of height 2-3m above ground. Leaves are light green, oblong-ovate, variable in shape, 3 to 5 veined from the base with yellowish velvety hairs. Flowers are unisexual in axillary clusters, sepals densely hairy, fruit ellipsoid, fleshy and purple-blue on ripening
4	<i>Woodfordia fruticosa</i> Kurz.		Leaves are dark, dull, narrow, pointed and grow straightly from the branches, opposite or in whorls of three. They are harsh but paler underneath. Flowers are brilliant red, arranged in dense axillary paniculate cymose clusters, with short glandular pubescent Pedicels

The special microscopical and chemomicroscopical characteristics of freshly collected or crude plant materials were carried out using thin sections **Fig. 1-4**.

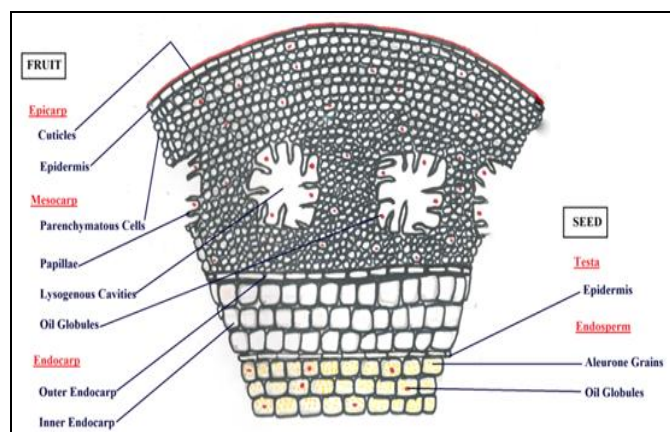


FIG. 1: TRANSVERSE SECTION OF FRUIT OF SEMECARPUS ANACARDIUM LINN.

The T. S. of the fruit of *Semecarpus anacardium* Linn. showed pericarp which comprises three

layers of parenchymatous cells, *i.e.* epicarp, mesocarp, and endocarp. Epicarp showed the presence of epidermis made up of radially elongated, single layered cells of parenchyma. Epidermal cells were covered with a thin layer of cuticles (Red color with Sudan red solution). Mesocarp comprises numerous layers (about 30-40 layers) of small and sub globular parenchymatous cells immediately below to the epicarp. The parenchyma of mesocarp consists of special characteristics *viz.* oil globules (Red color with Sudan red solution), papillae cells, lysogenous cavities, *etc.*

Rosette calcium oxalate crystals were found scattered in parenchymatous cells. Endocarp cells of fruit were differentiated into two layers of parenchyma *viz.* outermost and innermost endocarp layer. The outermost layer was shorter and thick than that of the innermost layer which comprises of somewhat elongated cells. The innermost layer was

big, thick, and fashioned with very radially elongated cells. Below to the endocarp, the cells of seed were observed, which comprises cotyledons with endosperm, aleurone grains (Yellow color with alcoholic picric acid) and fixed oil (Red color with Sudan red solution).

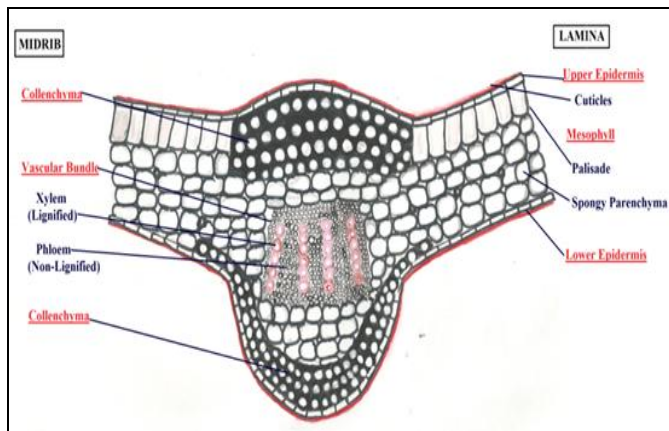


FIG. 2: TRANSVERSE SECTION OF LEAF OF ARGEMONE MEXICANA LINN.

The microscopical studies of the leaf of *Argemone mexicana* Linn. showed midrib and lamina. Midrib consists of upper and lower epidermises, which were biconvex in outline. Below to the upper epidermis and above to the lower epidermis few layers of rounded, thick-walled, cellulosic collenchymatous cells were observed. Most of the part of midrib occupied with arc-shaped vascular bundle. The vascular bundle consists of lignified xylem vessels (Pink color with phloroglucinol and conc. HCl) and few layers of small, rounded, non-lignified phloem parenchyma.

The lamina (Dorsiventral) of the leaf consists of the upper epidermis, mesophyll and lower epidermis. The epidermises were surrounded by thick cuticles (Red color with Sudan Red solution). The upper epidermis consists of thin-walled, single layered, polygonal, tangentially elongated parenchymatous cell with diacytic stomata. The lower epidermis was the same as that of the upper epidermis. No trichomes were found on both epidermises. Mesophyll comprises of radially elongated, compactly arranged, single layered palisade cells. Mesophyll also consists of few layers of rectangular or rounded shaped spongy parenchyma cells, which were loosely arranged with intracellular spaces. The microscopical characteristics of the leaf of *Cocculus hirsutus* Linn. were almost the same as that of the leaf of

Argemone mexicana Linn. The few special and differential microscopical characteristics of the leaf were identified.



FIG. 3: TRANSVERSE SECTION OF LEAF OF COCCULUS HIRSUTUS LINN.

The hemispherical (Crescent) vascular bundle covered by sclerenchymatous bundle sheath was observed. The stomata were absent on upper epidermis, but lower epidermis consists of numerous anomocytic stomata. Upper and lower epidermis both carry few long, ribbon-shaped, unicellular covering trichomes which were tapering at the apex and mesophyll comprises spongy parenchyma with starch grains (Blue color with iodine solution).

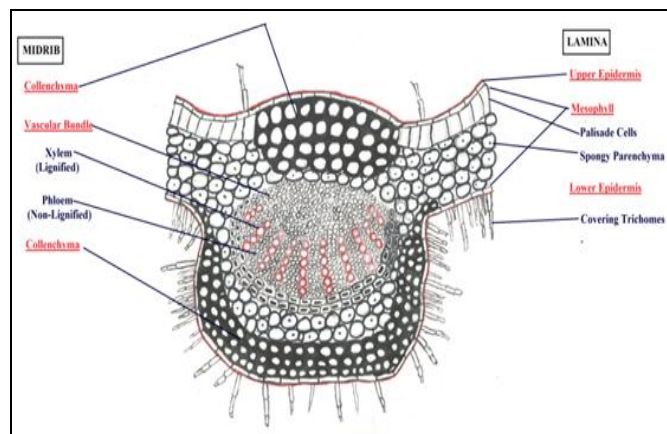


FIG. 4: TRANSVERSE SECTION OF LEAF OF WOODFORDIA FRUTICOSA KURZ.

The microscopy of the leaf of *Woodfordia fruticosa* Kurz. revealed the presence of arc-shaped bi-collateral vascular bundle in the center. The numerous simple, curved and unicellular to multicellular trichomes were observed on lower epidermis. Below upper epidermis, mesophyll comprises of a single layer of radially elongated, compactly arranged, cylindrical shaped palisade

cells containing chlorophyll pigments were observed. Mesophyll consists of spongy parenchyma with few simple, oval starch grains

(Blue color with iodine solution) and cluster crystals of calcium oxalate (Insoluble in acetic acid and soluble in sulphuric acid).

TABLE 4: MOISTURE CONTENT OF POWDERED PLANT MATERIALS

S. no.	Parameters	Values in Mean % w/w \pm S.D					
		<i>Semecarpus anacardium</i> L.		<i>Argemone mexicana</i> L.	<i>Cocculus hirsutus</i> L.	<i>Woodfordia fruticosa</i> K.	
		Fruits	Leaves	Whole Plant	Whole Plant	Leaves	Flowers
1	Moisture Content	8.73 \pm 0.95	15.45 \pm 0.00	12.99 \pm 1.05	15.76 \pm 0.20	16.75 \pm 0.73	9.46 \pm 1.01
2	Total Ash	7.54 \pm 0.21	17.50 \pm 0.92	9.54 \pm 0.311	13.86 \pm 0.61	8.16 \pm 0.34	10.72 \pm 0.91
3	Acid Insoluble Ash	0.8 \pm 0.10	2.77 \pm 0.55	0.90 \pm 0.055	0.86 \pm 0.07	1.11 \pm 0.23	2.85 \pm 0.30
4	Water Soluble Ash	1.33 \pm 0.17	11.29 \pm 2.53	7.41 \pm 1.07	7.49 \pm 0.19	5.27 \pm 0.25	5.90 \pm 0.24
5	Ethanol Soluble Extractive	34.61 \pm 0.75	37.69 \pm 1.86	28.03 \pm 2.89	27.62 \pm 1.95	26.21 \pm 2.21	20.21 \pm 2.48
6	Methanol Soluble Extractive	31.08 \pm 2.15	31.14 \pm 3.62	35.68 \pm 2.92	41.53 \pm 3.08	29.26 \pm 1.75	24.06 \pm 3.01
7	Chloroform Soluble Extractive	4.40 \pm 1.00	5.40 \pm 0.99	8.02 \pm 2.17	6.06 \pm 0.81	3.71 \pm 0.87	7.93 \pm 1.89
8	Water Soluble Extractive	18.09 \pm 2.60	20.26 \pm 2.74	14.95 \pm 2.01	21.50 \pm 3.12	17.73 \pm 1.52	14.03 \pm 1.81
9	Foreign Organic Matters	2.87 \pm 0.94	5.61 \pm 0.58	8.66 \pm 1.43	6.85 \pm 1.57	6.75 \pm 1.98	0.76 \pm 0.45

The proximate analysis involves the determination of physicochemical attributes of the drug to determine its identity, quality and purity. Results of the proximate analysis are shown in **Table 4**. The determination of moisture content gives an idea about stability and susceptibility of crude drug toward bacterial growth. The values of moisture content of *Semecarpus anacardium* L. (leaves), *Argemone mexicana* L. (whole plant), *Cocculus hirsutus* L. (whole plant) and *Woodfordia fruticosa* K. (leaves) was found to be 15.45 \pm 0.00, 12.99 \pm 1.05, 15.76 \pm 0.20 and 16.75 \pm 0.73 respectively.

Ash value indicates the earthy matter or the inorganic composition viz. carbonates, phosphates, silicates and other impurities present along with the drug¹¹. The inorganic content was found to be high in the powdered drug of *Semecarpus anacardium* L. (leaves) and *Cocculus hirsutus* L. (whole plant) viz. 17.50 \pm 0.92 and 13.86 \pm 0.61 respectively. Ash insoluble in hydrochloric acid is the residue obtained after extracting the total ash with hydrochloric acid. This acid-insoluble ash value particularly indicates contamination with siliceous materials like earth or sand¹¹. The value of acid insoluble ash of *Semecarpus anacardium* L. (leaves; 2.77 \pm 0.55) and *Woodfordia fruticosa* K. (leaves; 1.11 \pm 0.23 and flowers; 2.85 \pm 0.29) was found to be more. Water soluble ash is the part of total ash content soluble in water and a good indicator of either previous extraction of water-soluble salts in the drug¹¹.

The water-soluble salt content was found to be high in the powdered drug of *Semecarpus anacardium* L., *Argemone mexicana* L. and *Cocculus hirsutus*

L. viz. 11.29 \pm 2.53 (leaves), 7.41 \pm 1.07 (whole plant) and 7.49 \pm 0.19 (whole plant) respectively. The extractive value indicates amount of phytoconstituents present in the sample crude drug and helps in the determination of exhausted or adulterated drug species¹¹.

The extractive values of methanolic extract of *Argemone mexicana* L. (35.68 \pm 2.92), *Cocculus hirsutus* L. (41.53 \pm 3.08) and *Woodfordia fruticosa* K. (29.26 \pm 1.75 and 24.06 \pm 3.01) were found to be more whereas, extractive values of *Semecarpus anacardium* L. were high in ethanolic extract of fruit (34.61 \pm 0.75) and leaves (37.69 \pm 1.86). The determination of foreign organic matters signifies the contamination of plant parts with insect, molds or other animals. The high content of foreign organic matters indicates that plant material is not appropriate for further study¹¹. The foreign organic matters of whole plant *Argemone mexicana* Linn. (8.66 \pm 1.43), leaves of *Semecarpus anacardium* Linn. (5.61 \pm 0.58), whole plant of *Cocculus hirsutus* Linn. (6.85 \pm 1.57) and leaves of *Woodfordia fruticosa* Kurz. (6.75 \pm 1.98) was found to be high.

The polar and non-polar solvents were used to isolate phytoconstituents of powdered crude drugs. The continuous Soxhlet extraction method was used to prepare test extracts. The extracts thus obtained were dried and utilized for preliminary phytochemical screening.

Preliminary phytochemical evaluation of the concentrated test extracts was carried out through

various qualitative chemical tests for identification of phytoconstituents like alkaloids, glycosides, phenolic compounds, steroids, carbohydrate, fats, proteins, amino acids *etc.* **Table 5.**

TABLE 5: PRELIMINARY PHYTOCHEMICAL EVALUATION OF TEST EXTRACTS

S. no.	Solvents Chemical Test	Pet. Ether 40-60 °C				Chloroform				Ethanol				Methanol				Water			
		S A	A M	C H	W F	S A	A M	C H	W F	S A	A M	C H	W F	S A	A M	C H	W F	S A	A M	C H	WF
1	Test for Alkaloids																				
a)	Mayer's Test	-	+	+	-	-	-	+	-	-	+	+	+	-	-	-	-	-	-	-	+
b)	Hager's Test	-	+	+	-	-	-	+	-	-	+	+	+	-	+	+	-	-	-	-	+
c)	Wagner's Test	-	+	+	-	-	-	+	-	-	+	+	+	+	-	-	-	-	-	-	+
2	Test for Carbohydrate																				
A	General Test																				
a)	Molisch's Test	-	-	-	-	-	-	-	+	+	-	+	+	+	-	+	+	+	+	+	+
B	Test for Reducing Sugar																				
b)	Fehling Test	-	-	-	-	-	-	-	+	+	-	-	+	+	-	-	+	+	+	-	+
c)	Benedict's Test	-	-	-	-	-	-	-	+	+	-	-	+	+	-	-	+	+	+	-	+
3	Test for Steroids																				
a)	Salkowski Reaction	-	+	+	+	+	+	+	+	+	-	-	+	+	-	+	-	-	+	-	-
b)	LiebermanBurc hards Test	-	+	+	+	+	+	+	+	+	-	-	+	+	-	+	-	-	+	-	-
4	Test for Glycosides																				
A	Test for Cardiac Glycoside																				
a)	Baljet's Test	-	-	-	-	-	-	-	-	+	-	+	+	+	-	+	+	+	+	-	+
b)	Keller-killiani Test	-	-	-	-	-	-	-	-	+	-	+	+	+	-	+	+	+	+	-	+
B	Test for Anthraquinone Glycosides																				
a)	Brontrager's Test	-	-	+	+	-	-	+	+	-	-	-	-	-	-	-	-	-	-	-	-
C	Test for Saponin Glycosides																				
a)	Foam Test	-	-	-	-	-	-	-	-	+	+	-	+	+	+	-	+	+	+	-	+
5	Test for Flavonoids																				
a)	Lead Acetate Test	-	-	-	-	-	-	-	-	+	+	+	+	+	+	+	+	+	-	+	+
b)	Shinoda Test	-	-	-	-	-	-	-	-	+	+	+	+	+	+	+	+	+	-	+	+
6	Test for Fat and Oils																				
a)	Solubility Test	+	-	+	+	+	-	+	+	-	-	-	-	-	-	-	-	-	-	-	-
b)	Filter Paper Test	+	-	+	+	+	-	+	+	-	-	-	-	-	-	-	-	-	-	-	-
7	Test for Tannin and Phenolic Compound																				
a)	Ferric Chloride Solution Test	-	-	-	-	-	-	-	+	+	+	+	+	+	+	+	+	+	-	+	+
b)	Lead Acetate Solution Test	-	-	-	-	-	-	+	-	+	+	+	+	+	+	+	+	+	-	+	+
c)	Bromine Water Test	-	-	-	-	-	-	+	+	+	+	+	+	+	+	+	+	+	-	+	+
8	Test for Protein																				
a)	Biuret Test	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-	+
9	Test for Amino Acid																				
a)	Ninhydrin Test	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-	+
10	Test for Gums and Mucilage's																				
a)	Hydrolyse Test (Gums)	-	-	-	-	+	-	-	-	+	-	-	+	+	-	-	+	+	+	-	-
b)	Ruthenium Red Test (Mucilage)	-	-	-	-	+	-	-	-	+	-	-	+	+	-	-	+	+	+	-	-

"+" Indicates presence and "-" Indicates absence of active constituents

The preliminary phytochemical investigation reveals that ethanolic and methanolic extracts of *Semecarpus anacardium* L. were consists of carbohydrates, steroids, cardiac and saponin glycosides, flavonoids, tannins and phenolic compounds. All extracts of *Semecarpus anacardium* L. were found to be devoid of alkaloids, proteins and amino acids. Whereas gums and mucilages were found to be present in all test extracts of *Semecarpus anacardium* L. except petroleum ether extract. The ethanolic extract of *Argemone mexicana* L. was found to contain alkaloids, saponin glycosides, flavonoids, tannins and phenolic compounds.

However, methanolic extract was found to contain saponin glycosides, flavonoids and phenolics. The fats and oils were found to be absent in all test extracts of *Argemone mexicana* L. whereas petroleum ether extract was found to contain only alkaloids and steroids. The methanolic extract of *Cocculus hirsutus* L. was found to contain carbohydrates, steroids, cardiac glycosides, phenolics like tannins and flavonoids. The Phytoconstituents like alkaloids, carbohydrates, cardiac glycosides, flavonoids and tannins and phenolic compounds were found to be present in ethanolic extract. Whereas petroleum ether and chloroform extracts revealed alkaloids, steroids, fats and oils. The aqueous extract of *Cocculus hirsutus* L. contains only phenolic compounds and carbohydrates. The proteins, amino acids and gums and mucilages were found to be absent in all test extracts of *Cocculus hirsutus* L.

CONCLUSION: The pharmacognostic studies of herbal drugs will help in the development of Pharmacopoeial standards and their further exploration in various Ayurvedic or polyherbal formulations.

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