Pharmacognostic and Phytochemical Evaluation of Hepatoprotective Plants

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Abstract :

Since the beginning of time, humans have used plants as a source of medicinal substances. Traditional medicinal plants are used in many indigenous medical systems to treat a variety of disorders in both humans and animals, and they are essential for healing due to presence of bioactive compounds such as primary and secondary metabolites. These Medicinal Plants have been used for antifungal, antibacterial, anti-inflammatory and hepatoprotective activities. The current manuscript gives Pharmacognostic, physicochemical & Phytochemical examination carried out on the crude drugs of some medicinal plants *i.e.* RCL, MOL, AIL, TCLS, CCL. The phytoconstituents like; Flavonoids, alkaloids, tannins, phenols, steroids, terpenoids, carbohydrates, glycosides, and proteins compounds were identified by phytochemical testing which will be helpful to carry out the investigation on liver diseases. Physicochemical parameters including moisture content (7.90, 7.42, 9.68, 5.93 and 17.8 % w/w), ash values (13,11.5, 11.5, 9 and 12.3 % w/w), extractive values such as ether soluble extractive value (11.2, 4.8, 14.6, 4, and 8% w/w), alcohol soluble extractive value (16, 9.6, 10.4, 5.6 and 6.4 % w/w) and water soluble extractive value (25.6, 28, 25.6, 30.4 and 24.8 % w/w) of powdered drug samples of above mentioned plants have been determined. These factors are helpful to differentiate between powdered materials. The current study is an effort to improve the data in terms of standardization by developing a few diagnostic indices for the identification of the plant, creation of a monograph and conducting further research on Ayurveda approach to medicine.

Keywords: *Standardization, Phytochemical, Microscopy; Morphology, Hepatoprotective*

INTRODUCTION:

As the population grows, so do serious health problems like cancer, heart disease, and respiratory disorders, as well as the prevalence of liver disease. Herbal medications are highly sought after in the developed world for the treatment of certain ailments because of their effectiveness, safety, and low occurrence of adverse effects. Ensuring the accuracy of the plant material's identification and quality control is crucial for maintaining its consistent quality, which in turn enhances its safety and effectiveness. Plant material is standardized using Pharmacognostic approaches that take into account its morphological, histological, physicochemical, and biochemical properties. So, the current research work was done for the Pharmacognostic, physicochemical and Phytochemical investigation of five hepatoprotective plants *i.e.* RCL, MOL, AIL, TCLS, CCL [1,2,3,4,5].

MATERIALS AND METHODS:

Collection and authentication of plant material

The fresh portions of selected plant materials required for the present study such as RC(Leaves), MO (Leaves), AI (Leaves), TC (Leaves and Stem), CC (Leaves) were received from surrounding areas of Latur and Osmanabad district, Maharashtra, India. Special precaution was taken to collect healthy plant materials and avoid foreign materials. Plant herbarium were prepared and sent for certification to Department of Botany, Botanical Survey of India (BSI) for their authentication. Further these plants were identified and authenticated by renowned botanist Prof.D.L.Shirodkar from the Department of Botany, BSI having authentication voucher number Ref.No.BSI/WRC/Iden.Cer./2021/3108210000452.

Chemical reagents

Analytical grade chemicals and solvents were employed in this investigation.

Extraction

The above collected and authenticated parts of selected plant materials were thoroughly cleaned with running tap water and then with distilled water; they were allowed to dry for some time at room temperature. The plant material was then ground into a coarse powder using an electronic grinder after being shade-dried for three to four weeks without any contamination. This dried plant material was placed in an airtight container after being packed. For physicochemical analysis utilizing Soxhlet apparatus, 100 g of Air-dried coarsely powdered plant materials was extracted through successive extraction method using variety of range of solvents, including petroleum ether, chloroform, ethyl acetate, and ethanol, in increasing order of polarity. Then rotary flash evaporator was used for concentration of extracts under reduced pressure andthe remainder of the substance was

dried over sodium sulphitein desiccator. After drying, the respective extracts were weighed and percentage yield were determined and stored in airtight container.[6,7,10,13.]

Pharmacognostic Studies

Morphology & Microscopy

Macroscopical and microscopical characters will help in the identification of the right variety and search for adulterants.

Macroscopic characteristics

The morphological characters of different plant parts can be served as diagnostic parameters. For morphological observationsfresh parts of above selected plant materials (approx. 6-20 cm in length) were used and Organoleptic features viz. color, odour, taste, shape, sizes were observed and evaluated botanically.[8,13.]

Microscopic characteristics

Microscopic evaluation was done by taking transverse sections of fresh parts of above selected plant materials and stained by safranine for lignified cells, mounted with glycerine and observed under a low power of compound microscope to study and confirm its histological characters[9,10,17.] Then powder microscopy was also carried out and their specific diagnostic characters were recorded.[11,12.]

Physiochemical Evaluation

The physicochemical parameters like ash values, extractive values and moisture content were performed according to the officinal methods in IP and WHO guidelines for quality of medicinal plant.[13,14,15,17.]

Phytochemical analysis

The qualitative phytochemical tests were carried out to identify the various primary and secondary metabolites present in the different extract of above selected plant materials.[6,8,9,16,17,18.]

RESULTS AND DISCUSSION:

Morphological Evaluation

RCL MOL AIL

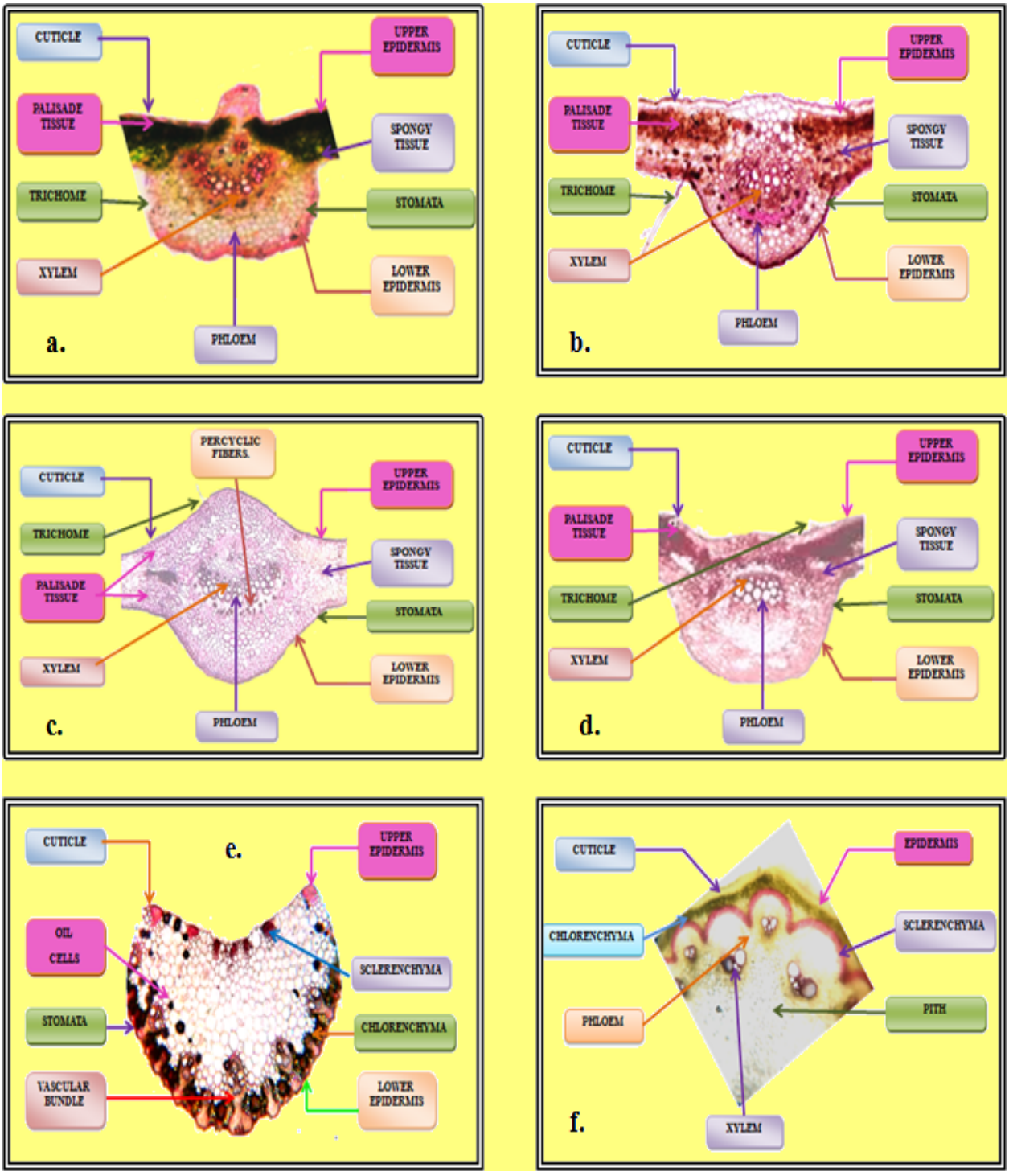
TCLS CCL

Photograph 1. Morphological Characters of Crude Drugs.

Table 1. Morphological Characters

SR. NO.	FEATURES	OBSERVATION				
		RC	MO	AI	TC	CC
1.	Color	Green or Reddish Green	Green	Dark Green	Dark Bright Green	Dark Green
2.	Odour	Odourless	Characteristic	Characteristic	Indistinct	Lemon Like
3.	Taste	Astringent	Slight	Bitter	Bitter	Bitter
4.	Size	2-7cm Long	2.5 Cm Long	2-6 Cm L, 1-3 Cm	10-12 Cm X 8-15 Cm	1- 2 Meter Long
5.	Shape	Lanceolate, elliptical	Rounded, elliptic	Lanceolate	Simple, ovate, heart Shaped	Linear, tapered to both ends
6.	Texture	Soft	Soft	Papery	Soft	Soft
7.	Margin	Serrate, dentate	Entire	Serrate	Entire not reflexed	Entire
8.	Leaf base	Symmetric or asymmetric	Symmetric	Oblique/Unequal/ Asymmetric	Cordate	Ligule
9.	Apex	Acute or acuminate	Emarginated	Acuminate	Acuminate	Acuminate
10.	Venation	Reticulate	Reticulate,	Reticulate	Reticulate	Parallel

Microscopic evaluation

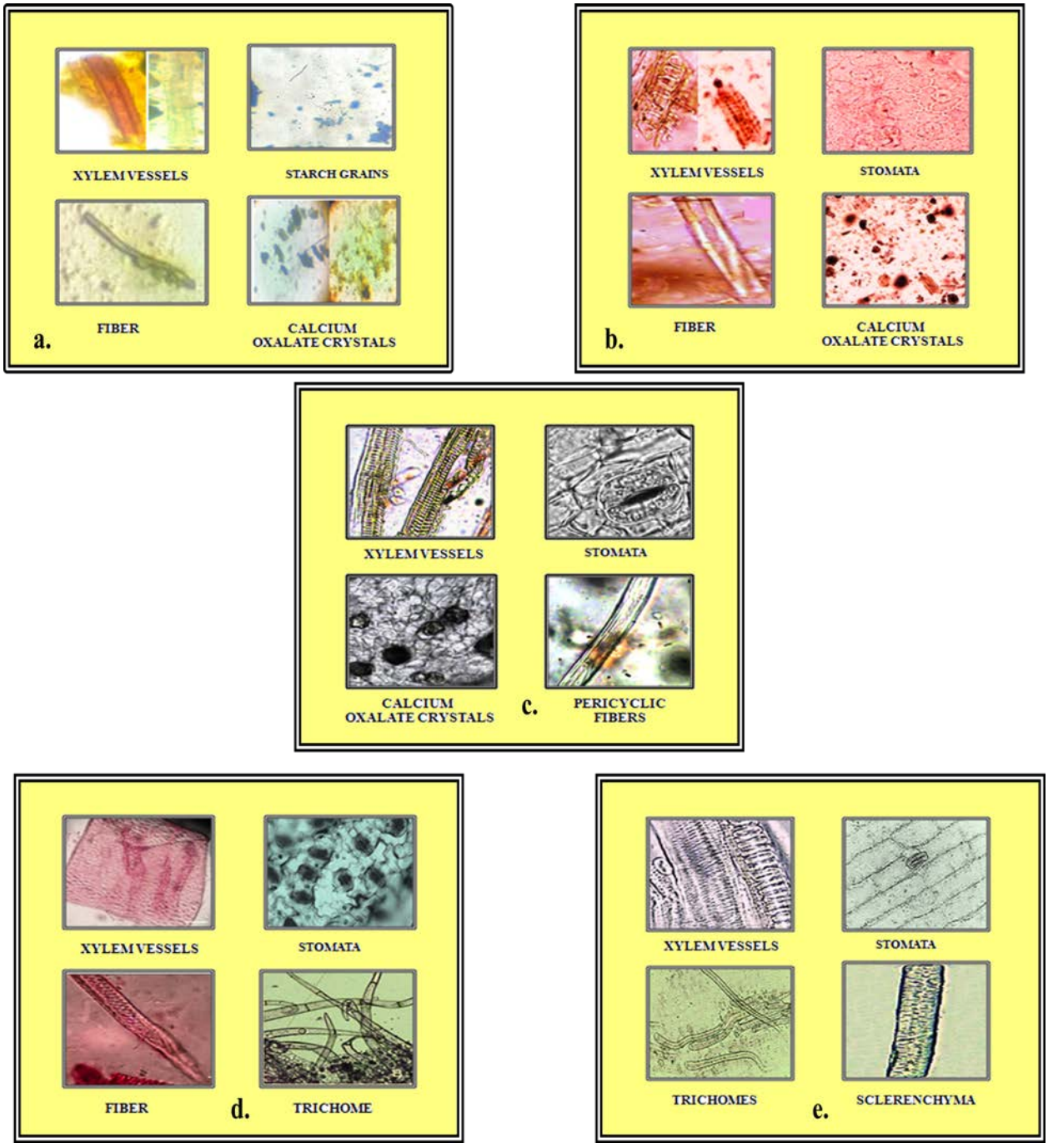


Photograph 2. Transverse sections of crude drugs.

a. *Ricinus communis* leaf **b. *Moringa oleifera* leaf** **c. *Azadirachta indica* leaf**

d. *Tinospora cordifolia* leaf **e. *Cymbopogon citratus* leaf** **f. *Tinospora cordifolia* stem**

Diagnostic Characters of Powdered Crude Drugs



Photograph 3. Powder microscopy of powdered crude drugs.

a. *Ricinus communis* **b. *Moringa oleifera*** **c. *Azadirachta indica***

d. *Tinospora cordifolia* **e. *Cymbopogon citratus***

Table 2. Microscopic characteristics of crude drugs

FEATURES	OBSERVATION				
	RC	MO	AI	TC	CC
Cuticle	Thin	Thin	Thick warty	Thin	Waxy
Epidermis	One layered Polygonal cells	One layered polygonal cells	One layered polygonal cells	One layered polygonal cells	V-shaped polygonal cells
Palisade tissue	Closely packed cells filled with chloroplast	2 to 3 layers of palisade cells	2 palisade rows in the upper and one row at lower surfaces	Closely packed cells	Closely packed cells
Trichomes	Covering trichomes	Unicellular trichomes with blunt ends	Unicellular covering Trichomes	Glandular Trichomes	Unicellular covering Trichomes
Stomata type	Paracytic	Anomocytic	Anomocytic	Anomocytic	Paracytic
Midrib vascular bundles	Vascular bundles with xylem, phloem and along with collenchymatous cells	Vascular bundles with xylem, phloem and along with collenchymatous cells	Arc shaped collateral type of vascular bundles with xylem, phloem and along with collenchymatous cells and percyclic fibers	Vascular bundles with xylem, phloem and along with collenchymatous cells	Vascular bundles with xylem, phloem and along with sclerenchymal sheaths and chlorenchyma cells
Calcium oxalate crystals	Spharaphide	Rosette shaped	Prism	-	-

Physiochemical Evaluation:

It is a crucial metric for determining whether crude drugs have been handled improperly or adulterated. The amount of moisture in the powdered form of RCL, MOL, AIL, TCLS, CCL is not extremely high, which would prevent the formation of bacteria, fungus, or yeast. The ash value was determined by using three different types of ash: total ash, water soluble ash, and acid insoluble ash. When assessing the purity of crude pharmaceuticals, such as whether or not they contain foreign inorganic materials like silica or metallic salts, one crucial criterion is total ash content. Sand's silica content is determined by acid insoluble ash, while water soluble ash determines the total quantity of ash soluble in water. The results were shown in (Table 3) which was less. Reduced levels of these three parameters suggest that there was less silica and inorganic materials in RCL, MOL, AIL, TCLS, CCL.The extractive value of crude powder was highest in water, alcohol and less in petroleum ether shown in (Table 3).

Table 3. Physiochemical Evaluation of Crude drugs

PARAMETER	DETERMINED VALUE				
	RCL	MOL	AIL	TCLS	CCL
(A) EXTRACTIVE VALUE(% w/w)					
Ether soluble	11.2	4.8	14.6	4	8
Alcohol soluble	16	9.6	10.4	5.6	6.4
Water soluble	25.6	28	25.6	30.4	24.8
(B) MOISTURE CONTENT(% w/w)					
Moisture content	7.90	7.42	9.68	5.93	17.8
(C) ASH VALUES(% w/w)					
Total ash	13	11.5	11.5	9	12.3
Acid insoluble ash	1.5	1.5	2	2.5	4.6
Water soluble ash	6	6	0.5	12.5	7.4

Phytochemical Evaluation

The qualitative Phytochemical screening results of the crude powder extract of RCL, MOL, AIL, TCLS, CCL was shown in (Table 4) and (Table 5).

Qualitative chemical examinations of various extracts above selected plant parts indicated the existence of flavonoids, phenolic and other significant phytochemicals in the ethanolic extract, and steroids in the pet ether extract respectively.

Table 4.Phytochemical Evaluation of Extract 1

PHYTO - CONSTITUENTS	OBSERVATION P.E. EXTRACT				
	RCL	MOL	AIL	TCLS	CCL
Carbohydrate	-	+	-	-	-
Glycoside	-	-	-	+	-
Protein	-	+	-	-	-
Steroids	++	++	++	++	++
Terpenoids	-	-	-	+	+
Tannins	-	+	-	-	-
Saponins	-	+	+	+	-
Phenols	-	+		-	-
Alkaloids	-	+	+	+	-
Flavonoids	-	+	+	+	-

Table 5. Phytochemical Evaluation of Extract 2

PHYTO - CONSTITUENTS	OBSERVATION (ETH EXTRACT)				
	RCL	MOL	AIL	TCLS	CCL
Carbohydrate	+	-	+	+	+
Glycoside	+	+	+	+	+
Protein	-	+	-	+	+
Steroids	-	-	-	-	-
Terpenoids	+	-	+	+	+
Tannins	+	-	+	+	+
Saponins	+	+	-	+	-
Phenols	++	++	++	++	++
Alkaloids	+	+	-	+	-
Flavonoids	++	++	++	++	++

Key: + = Present, -= Absent

CONCLUSION:

The goal of the current study was to establish guidelines that could be helpful in proving the authenticity of these plants. The standards that are covered here, macro, micro and physicochemical,, can be thought of as distinguishing factors to authenticate the drugs.

In this investigation, we discovered that the majority of biologically active phytochemicals were found in ethanolic extract of RCL, MOL, AIL, TCLS, CCL.The medicinal properties of above selected plant parts could be because of existence of above-mentioned phytochemicals so; these plants can be further processed for evaluation of their hepatoprotective activity in the form of suitable polyherbal formulation.

CONFLICT OF INTEREST:

No conflict of interest.

ABBREVATIONS:

RC - *Ricinus communis*

MO - *Moringa oleifera*

AI - *Azadirachta indica*

TC - *Tinospora cordifolia*

CC - *Cymbopogon citrates*

L - Leaves

LS - Leaves & Stem.

P.E. – Petroleum ether

ETH – Ethanol

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