Research Article

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PHARMACOGNOSTICAL AND PHYTOCHEMICAL EVALUATION OF LEAVES OF ERANDA (*RICINUS COMMUNIS* LINN)

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Received on: 12/08/17 Accepted on: 20/09/17

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DOI: 10.7897/2277-4343.085240

ABSTRACT

Eranda (*Ricinus communis* Linn.) is one of the most important drug used in Ayurveda therapeutics. It is used widely in different formulations in medical practice. The standardization regarding phytochemical and histological evaluation of *Ricinus communis* leaves was not at all mentioned in Ayurveda Pharmacopoeia of India (API). The present work attempts to establish the necessary pharmacognostic and phytochemical standards for evaluating the leaves of Eranda. Macroscopic, microscopic and phytochemical evaluation including physical parameters like moisture content, water and alcohol extractives, ash values etc., chemical parameters like TLC, HPTLC and quantitative analysis like detection of flavonoids, alkaloids etc. were done as per pharmacopoeial standards and the results were documented.

Keywords: *Ricinus communis* Linn, Eranda, TLC, HPTLC, flavonoids, alkaloids

INTRODUCTION

Eranda (*Ricinus communis* Linn.), of Euphorbiaceae family is an important drug mentioned in Ayurveda classics from Vedic period itself. It is used very commonly in rheumatic conditions, wounds, hydrocele etc.[1-3]. The plant considered probably a native of Africa, is found throughout the hotter parts of India and tolerates a wide range of climatic conditions[4]. While describing agryadravyas, Charaka considered Erandamoola as vrshya and vatahara[5].Susrutha mentioned Erandataila as best among oil purgatives[6].

The root and oil obtained from seeds are widely used in different formulations, but the leaves are used rarely. But the use of tender leaves and old leaves are mentioned in texts like Bhavaprakasa for some conditions like yakruthvikaras and medovriddhi[7]. But the phytochemical and microscopical evaluation of *Ricinus communis* leaves was not at all mentioned in API. So there is a need of standardized data for the establishment of a unique identification data. Nowadays Ayurveda gets more acceptance and popularity among the public due to its holistic approach. The drugs should be standardized by establishing quality parameters for the safe and effective use in clinical practice.

Pharmacognosy is an objective study of crude drugs from natural sources treated scientifically and it encompasses the knowledge of the history, distribution, cultivation, collection, processing for market and preservation, the study of sensory, physical, chemical and structural characters and uses of crude drugs[8]. It includes macroscopic, microscopic, phytochemical and pharmacological evaluation. Macroscopic evaluation refers to evaluation of drugs by size, shape, nature of outer and inner surfaces, type of fracture and organoleptic characteristics like colour, odour, taste, and consistency etc.[9]. Preliminary physicochemical evaluation is a step towards the genuinity and purity of the drug.

MATERIALS AND METHODS

Collection of plant material

The fresh samples of mature leaves of Eranda (*Ricinus communis* Linn) were collected from Kollam district, Kerala. After authentication by a taxonomist, herbarium was prepared and deposited in the department museum with a voucher specimen no.3654/2017. They were washed well and dried in shade. It was powdered and used for physicochemical evaluation as per the WHO guidelines and Ayurveda Pharmacopoeia of India[10,11].

Macroscopic evaluation

The general appearance of a crude drug indicates whether it is likely to comply with its prescribed standards.

Microscopic evaluation

Using a sharp blade, part of the leaf passing through the midrib is cut. A stained section is carefully transferred on a clean glass micro slide using thin brush. The slide is placed on a digital microscope for histological examination and direct images are taken at 2x, 4x, 10x and 40x magnifications. Materials used were glass slides, cover slips, digital microscope, glycerin and safranin stains.

Physical and physicochemical analysis

Dried leaves of Eranda were powdered and the phytochemical analysis was done. Reagents and apparatus used were Xylene, dilute Hydrochloric acid, Petroleum ether, Acetone, Acetic anhydride, concentrated Hydrochloric acid, concentrated sulphuric acid, magnesium ribbon, neutral ferric chloride, Benzene, Chloroform, Ethyl acetate, Potassium permanganate, Acetic acid, Fehling's solution, Sodium bicarbonate, Dragendorff's reagent, Ferric alum, Ethanol, Lead acetate, Sodium oxalate and distilled water, Dean and Stark's apparatus,

Clevenger apparatus, Soxhlet apparatus, silica crucible, Bunsen burner, round bottomed flask, measuring jars, beakers, conical flask, funnel, glass rods, watch glass, filter paper, electronic balance etc.

For TLC, maximum spots were obtained with mobile phase chloroform: petroleum ether: ethyl acetate (8:2:2). The spots were first viewed in visible light and then visualized in UV and iodine chamber.

For HPTLC also same solvent system was used. HPTLC was done with the instrument "Camag Linomat 5" using TLC silica gel 60 F_{254}. The chromatogram was created by using Camag Wincats software. As *Ricinus communis* plant mainly grows in wasteland near roads, railway station etc., there may be a chance to get contaminated by heavy metals. So the heavy metal analysis was done by Atomic Absorption Spectroscopy (Thermo electron corporation, M series AA spectrometer)

RESULTS

Results of macroscopic evaluation

Colour: Dark green above and pale beneath.
Odor: None
Taste: Bitter
Texture: Thin and delicate
Appearance: Long petiolate, stipulate, palmately veined, broad, nearly orbicular, 7-10 or more lobed. (Figure 1)
Apex: Acute or acuminate

Microscopic examination

Epidermis: Upper and lower epidermal layers consisting of a row of rectangular cells covered with cuticle. A few numbers of stomata present in the lower epidermal layer. (Figure 2)
Mesophyll: The mesophyll is differentiated into palisade and spongy tissues. The lamina represents one layer of palisade cells on the ventral side. Palisade tissue is dark green in colour and cylindrical in shape.
Loosely arranged polygonal to rectangular cells of spongy tissue containing chloroplasts are seen. In the midrib region, the ventral palisade and dorsal spongy cells are replaced by collenchyma cells.
Vascular bundles: The midrib consists of 2 sets of vascular bundles. One set on the dorsal region is the largest and arch shaped. Second set of vascular bundles which consists of 2 or more bundles arranged in a clustered manner, is on the ventral region. (Figure 3)
In all vascular bundles xylem towards the centre and phloem towards the periphery. Vascular bundles are surrounded by numerous layers of parenchyma. Calcium oxalate crystals are seen in this parenchymatous region. (Figure 4)

Physical evaluation

Results of quantitative parameters are described in table 1.

Qualitative analysis

The presence of different plant constituents determines the pharmacological action and 30% therapeutic potential of that plant. Testing for these phytoconstituents helps in determining the quality of the drug.

TLC & HPTLC

The Rf values of the spots under UV light and iodine chamber of thin layer chromatographic plates & HPTLC are given in table 3. Graphs indicating HPTLC profile are given in graphs 1 and 2.

AAS

Atomic Absorption Spectroscopy enables the quantitative measurement of the metal ion in the sample. Results are shown in table 5.

Table 1: Quantitative parameters

S.No	Experiment	Percentage
1	Moisture content (%)	Nil
2	Volatile oil content (%)	Nil
3	Total ash (%)	2
4	Water insoluble ash (%)	2
5	Acid insoluble ash (%)	2
6	Cold water soluble extractive (%)	10%
7	Alcohol soluble extractive (%)	7%
8	Fiber content (%)	8%
9	Sugar Reducing sugar (%) Total sugar (%)	 16.025 16.025

Table 2: Qualitative Analysis

S.No	Chemical constituent	
1	Phenols	+
2	Alkaloids	+
3	Flavonoids	-
4	Steroids	+

Table 3: Rf vales of spots obtained in TLC

Solvent system: chloroform: petroleum ether: Ethyl acetate (8:2:2)	No. of spots	Distance travelled by solvent front	Distance travelled by the spot	Rf value
After viewed under UV	4	15.5 cm	8 cm	1.938
			9.6 cm	1.615
			14.5 cm	1.069
			15.2 cm	1.02
After putting in iodine chamber	2	15.5 cm	14.5 cm	1.069
			15.2 cm	1.02

Table 4: Rf vales for peaks obtained in HPTLC

Peaks	Rf value start position	Rf value end position
1	0.42	0.49
2	0.51	0.60
3	0.67	0.77

Table 5: AAS Results

Metals	Sample drug (ppm)
Cadmium	0.0093
Lead	0.1432
Iron	0.36
Zinc	0.09

Figure 1: Eranda (*Ricinus communis* Linn)

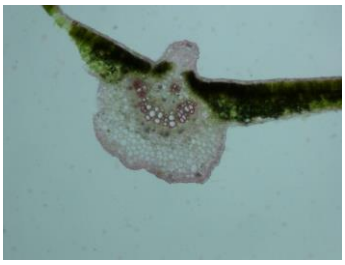


Figure 2: TS through midrib

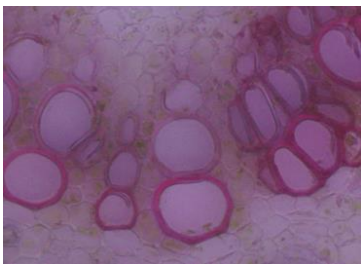


Figure 3: Vascular bundles

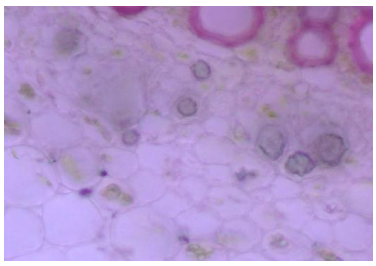
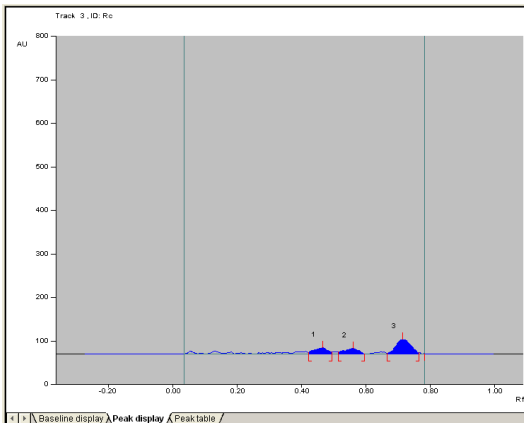


Figure 4: Calcium oxalate crystals

Graph 1: HPTLC profile of alcoholic extract of Eranda



Graph 2: HPTLC profile (peak display of the drug Eranda)

DISCUSSION

The evaluation of a crude drug is necessary due to biochemical variation in the drug, deterioration due to treatment and storage, substitution and adulteration. A drug must be standardized to establish its correct identity. So, the pharmacognostical evaluation including macroscopic and microscopic examination and physical and physico-chemical evaluation of leaves of Eranda (*Ricinus communis* Linn) were done for the documentation of data based on the results obtained.

Microscopic examination was done and the results obtained shows characteristic features like collateral vascular bundles arranged in 2 sets (One is arch shaped and other in clustered manner) and Calcium oxalate crystals. On physical evaluation total ash value, water insoluble ash and acid insoluble ash were found to be 2%. As ash value indicates the presence of foreign matter in the drug, it is important in the evaluation of purity of drugs. The water soluble extractives are more than alcohol soluble extractives, so more constituents are soluble in water. Fibre content and sugar is present. Qualitative analysis reveals the presence of phenols, alkaloids and steroids.

On Thin Layer Chromatography, 4 spots with Rf values 8, 9.6, 14.5 and 15.2 were obtained for the solvent system chloroform: petroleum ether: ethyl acetate (8:2:2). Rf values for peaks obtained in HPTLC were 0.49, 0.60 and 0.77 for the same solvent system. As AAS values are within permissible limits, the drug is not contaminated with heavy metals and it can be used safely in practice.

The data obtained can be documented and serve as a reference material for preparing monograph of the drug. It can be kept as a

source of document for authentication of the leaves of Eranda (*Ricinus communis* Linn).

CONCLUSION

Crude drugs provide essential intermediates for final synthesis of active compounds. The standardization of quality control of the crude drugs must be done to check its purity and genuineness. Physico- chemical parameters of leaves of Eranda (Ricinus communis Linn) are not mentioned in Ayurveda Pharmacopoeia of India (API). The leaves are mentioned in texts like Bhavaprakasa for treatment of some diseases. Physical and physico-chemical evaluation was done for the drug and obtained values were documented for further reference. More pre-clinical and clinical research can be done regarding the drug. The present study will be helpful for researchers and pharmaceutical industries as it serves as a reference document for the authentication of the leaves of *Ricinus communis* Linn.

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Cite this article as:

Sreekutty Jayan and Deepa M S. Pharmacognostical and phytochemical evaluation of leaves of Eranda (*Ricinus communis* Linn). Int. J. Res. Ayurveda Pharm. 2017;8(5):42-45 http://dx.doi.org/10.7897/2277-4343.085240

Source of support: Nil, Conflict of interest: None Declared