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Pharmaceutical Standardization

Phytochemical evaluation of the wild and cultivated varieties of *Eranda Mula* (Roots of *Ricinus communis* Linn.)

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Abstract

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In Ayurveda, the roots of *Eranda* (*Ricinus communis* Linn.) are used in the treatment *Amavata* (rheumatism), *Sotha* (inflammation), *Katisula* (backache), *Udararoga* (disease of abdomen), *Jwara* (fever), etc, Due to high demand, root of the cultivated variety is mainly used in place of wild. But, a comparative phytochemical profile of both varieties is not available till date. Considering this, a preliminary study has been done to ensure basic phytochemical profile of both the varieties. Preliminary physicochemical parameters, phytochemical screening, quantitative estimation of alkaloid, high-performance thin layer chromatography (HPTLC), and heavy metal analysis were carried-out in the study. Analysis of physicochemical data reveals no significant difference in between both varieties of roots, while alkaloid was found to be more in cultivated variety (0.34%) than wild one (0.15%). Though, the analytical profiles are almost identical, except the quantity of alkaloid; inferences should be made through well designed pharmacological and clinical studies.

Key words: Alkaloid, Eranda, high-performance thin layer chromatography, Ricinus communis

Introduction

Ricinus communis Linn (Euphorbiaceae) commonly known as *Eranda* in Ayurveda is a soft-wooded small tree wide spread throughout tropics and warm temperate regions of the world. In the Indian system of medicine, the leaf, root, and seed oil of this plant have been used for the treatment of inflammation and liver disorders.^[1] In Ayurveda, the roots of *Eranda* are used in the treatment of *Amavata* (rheumatism), *Sotha* (inflammation), *Katisula* (backache), *Udararoga* (diseases of abdomen), *Jwara* (fever), etc.^[2] Its roots have also been highlighted for its Vrishya (aphrodisiac) and Vatahara actions by Acharya Charaka.^[3] This plant also possesses hepatoprotective,^[4,5] anti-diabetic,^[6] laxative,^[7] anti-fertility,^[8] anti-inflammatory and free radical scavenging activities.^[9]

Due to high demand, roots of the cultivated variety are mainly used instead of wild. But, a comparative phytochemical profile of both varieties is not available till date. Hence, to ensure quality of both varieties, phytochemical evaluations of both the varieties was undertaken.

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Materials and Methods

Collection of drug

Fresh roots of wild and cultivated varieties of *R.communis* Linn. after proper identification were collected from the adjacent area of Jamnagar, Gujarat, with the help of taxonomist. Specimen herbarium of both varieties for wild (No. 1490) and cultivated (No. 1491) were preserved in the Pharmacognosy Laboratory, IPGT and RA, Gujarat Ayurved University, Jamnagar for further reference. The obtained roots were shade dried and made into coarse powder with the help of mechanical grinder and preserved in a glass container for future studies [Figure 1].

Physicochemical study

Moisture content, ash values (total ash, acid insoluble ash), and extractive values (alcohol soluble extractive, water soluble extractive) were determined by following standard analytical procedures.^[10,11]

Preliminary phytochemical screening

Five grams coarse powder of the roots was subjected for extraction with methanol (100 ml), keeping it for overnight with initial occasional shaking up to 6 h. and then set aside. After 24 h, it was filtered and alcoholic extract was collected. Similarly, water extract was prepared. Both the extracts were evaporated to dryness. The dried extracts were weighed, and percentage yield was calculated. The extracts were used for preliminary phyto-chemical screening with a set of various chemical tests viz., Dragendorff's Mayer's, Hager's, and Wagner's tests for alkaloids; ferric chloride, lead acetate, potassium dichromate, and dilute iodine tests for tannins and phenolics; and foam test for saponin glycosides.^[12]

Table 1: Physicochemical analysis of *Eranda Mula*:Wild and cultivated varieties

Parameters	Wild variety	Cultivated variety
Foreign matter	Nil	Nil
Loss on drying % w/w	6.48	7.43
Total ash content % w/w	5.17	5.18
Acid insoluble ash % w/w	0.94	1.04
Water soluble extractive value % w/v	11.02	10.11
Alcohol soluble extractive value % w/v	22.56	17.92
pH value	5.48	5.32

Table 2: Preliminary phytochemical profiles of Eranda Mula: Wild and cultivated varieties

Chemical	Wild variety	Cultivated variety	
Alkaloids	+	+	
Cynogenic glycoside	+	+	
Cardiotonic glycoside	-	-	
Phenols	-	-	
Flavanoid	+	+	
Terpenoids	+	+	
Protein	-	-	
Resin	-	-	
Tannin	+	+	
Carbohydrate	+	+	
Saponin	+	+	

+: Present, -: Absent

High-performance thin layer chromatography

High-performance thin layer chromatography (HPTL \overline{C}) was carried out by the standard methods.^[13]

Chloroform extract of cultivated variety was labeled as track 1, while the wild variety as track 2. The solvent System used in the study was Toluene:Ethyl acetate:dimethylamine (7:2:2).

Chromatographic conditions

Application mode: Camag Linomate V; Development chamber: Camag Twin trough chamber; Plates: Pre-coated silica gel GF254 plates; Chamber saturation: 30 min; Development time: 30 min; Development distance: 7 cm; Scanner: Camag scanner II; Detection: Deuterium lamp and mercury lamp; Photo-documentation: Camag reprostar; Data system: Win cats software; Drying device: Oven and was visualized under 254 nm and 366 nm.

Quantitative estimation of alkaloid

The samples were estimated quantitatively for total alkaloid content by gravimetric method.^[14]

Heavy-metal analysis

Heavy metal analysis of the root powder of both varieties, for arsenic, lead, mercury, and cadmium, by following standard procedure,^[15] was carried out at Analytical Testing Laboratory, Konark Research Foundation, Daman.

Results and Discussion

Physico-chemical analysis

Results of physicochemical analysis, qualitative tests, and R_f values of HPTLC in wild and cultivated varieties of *R.communis* Linn. are mentioned in Tables 1-3, respectively. Alkaloid percentage was estimated as 0.34% in cultivated, and 0.15% in wild variety. Results of heavy metal analysis for arsenic, lead, mercury, and cadmium are mentioned in Table 4.

Analysis of physicochemical data [Table 1] reveals absence of foreign matter in both samples. Moisture content in wild

Table 3: R, values of chloroform extracts of wild and cultivated variety of <i>Eranda Mula</i>							
	No. of spots	Cultivated variety	No. of spots	Wild variety			
254 nm	5	0.13, 0.20, 0.35, 0.37, 0.43	3	0.13, 0.18, 0.35			
366 nm	5	0.13, 0.18, 0.25, 0.30, 0.34	5	0.13, 0.17, 0.24, 0.29, 0.35			

Table 4: Heavy metal analysis of wild and cultivated variety of Eranda Mula								
Sample name	Element	Wave length	Instrument detection limit (ppm)	Results	Permissible limits			
Wild variety	Lead (Pb)	217.0	0.001	BDL	3 ppm			
	Arsenic (As)	193.7	0.02	BDL	1 ppm			
	Mercury (Hg)	253.7	0.0005	BDL	0.3 ppm			
	Cadmium (Cd)	228.8	0.001	BDL	10 ppm			
Cultivated variety	Lead (Pb)	217.0	0.001	BDL	3 ppm			
	Arsenic (As)	193.7	0.02	BDL	1 ppm			
	Mercury (Hg)	253.7	0.0005	BDL	0.3 ppm			
	Cadmium (Cd)	228.8	0.001	BDL	10 ppm			

ppm: Parts per million, BDL: Below detection limit

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Figure 1: Raw and crushed roots of wild and cultivated variety

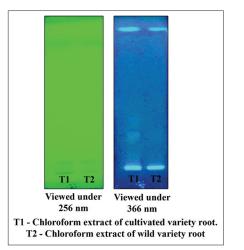


Figure 2: Photograph of high-performance thin layer chromatography

variety (6.48% w/w) is less than cultivated variety (7.43% w/w). The difference in total ash, acid insoluble ash, water soluble extractive value and pH in between the samples is insignificant. Alcohol soluble extractive is comparatively higher in wild variety (22.56% w/v) than cultivated variety (17.92% w/v).

The root of cultivated and wild varieties of *R.communis* exhibits almost similar phytochemical profile indicating presence of alkaloids, cyanogenic glycosides, flavonoids, terpenoids, tannin, carbohydrates, and saponin [Table 2]. Quantitatively, tannin was found to be more in cultivated variety (0.34%) than the wild one (0.15%).

Chloroform extract of wild variety showed 03 spots and cultivated variety showed 05 spots under 254 nm, among them R_f values 0.13 and 0.35 are similar in both samples. In 366 nm, chloroform extract of both varieties showed 05 spots, among them one R_f value 0.13 is similar in both samples. Other R_f values are nearer to each other, indicating that the components present in both varietiesmay be similar [Figure 2; Table 3]. *In situ*Ultra Violet spectral comparison graph also shows chemical similarity at 0.08 R_f 0.15 R_f and 0.36 R_f values [Figure 3].

Heavy metal analysis of both varieties shows that the samples are free from heavy metal contamination and the observations are under the prescribed limits for heavy metals.^[16][Table 4].

Conclusion

Both cultivated and wild varieties have a chemical similarity except higher percentage of alkaloid in cultivated than wild variety. HPTLC finger printing shows similarities in wild and cultivated varieties. Heavy metals were within prescribed limit in both the varieties. Though, the analytical profiles are almost identical, except the quantity of alkaloid; inferences for clinical use should be made through well designed pharmacological and for clinical studies.

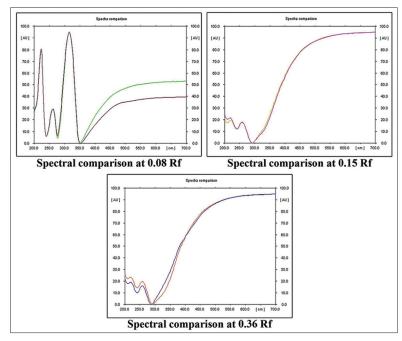


Figure 3: In-situ UV spectral comparison grap

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हिन्दी सारांश

वन्य एवं कृषिज एरण्डमूल के गुणधर्मों का तुलनात्मक अध्ययन

दोशी कृनाल, आचार्य रबिनारायण, वी. जे. शुक्ल, रेणुका, कोमल खानपरा

आयुर्वेद में एरंड मूल का उपयोग आमवात, शोथ, कटिशूल, उदररोग, ज्वर आदि मे बताया गया है। मूलो को वन्य एवं खेतों से एकत्रित किया जाता है। एरंड मूल की ज्यादा मांग होने के कारण वन्य जाति के मूलों के स्थान पर खेती की जाति के मूलो का मुख्यतः उपयोग किया जाता है।प्रस्तुत शोधपत्र मे दोनों जाति के मूलो का तुलनात्मक अध्ययन भौतिक परीक्षण, प्राथमीक रासायणिक परीक्षण, आल्कलोईड का प्रमाण?अभ्यास ओर भारी धातुओ का विश्लेषण किया गया है। रासायनिक एवं भौतिक विश्लेषण के आधार पर दोनो जाति के मूलो में परस्पर भिन्नता नही है लेकिन आल्कोहोल मे घुलनशील निष्कर्ष मूल्य का प्रमाण वन्य जाति मे २२.५६% खेति की जाति १७.९२% की तुलनामे ज्यादा पाया गया। आल्कलोइड का प्रमाण खेति की जाति में 0.३४% ओर वन्य जाति मे २२.५६% पाया गया। अभ्यास का परिणाम दर्शाता है की दोनो मूलों को ३६६ पा के अन्तर्गत निरीक्षण करने से दोनो मूलों मे ५ बिन्दु दिखाई देते है लेकिन ३५६पा के अन्तर्गत निरीक्षण करने से वन्य जाति मे ३ बिन्दु ओर खेति की जाति मे ५ बिन्दु दिखाई देते है। दोनो जाति के मूलो में रासायनिक साम्यता है लेकिन खेती की जाति में आल्कलोइडका प्रमाण ज्यादा है। भारी धातु के विष्लेषण के परिणाम मे भी दोनों जाति के मूलों मे साम्यता दिखाई देती है। फिर भी यह अनुमान भेषजगुण विज्ञान और चिकित्सकीय परिक्षण के द्वारा भी करना चाहिए।