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PHARMACOGNOSTIC, PHYTOCHEMICAL AND HPTLC PROFILE OF *THEV DHANTU* [PETIOLE OF *COLOCASIA ESCULENTA* (LINN.) SCHOTT.]

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ABSTRACT

Introduction: Ayurveda always emphasizes on utility of local flora in treatment. It also advices to know about the plant completely with respect to its identity, property and various therapeutic benefits including safety before prescribing it to a patient. **Methods:** *Colocasia esculenta* (Linn.) Schott. belonging to the family Araceae is believed to be one of the earliest available plants common in Udupi, called *Thev dhantu* in Tulu language and used very frequently in traditional medicine to treat Otalgia, Otorrhoea, Adenitis, Internal haemorrhage, Hepatomegaly etc., thus proper identification is very important. **Results:** The Pharmacognostical study will help to standardize its identification and some of the important diagnostic features of the plant. Preliminary phytochemical studies revealed the presence of several phyto-constituents which helps in characterization of the drug. **Discussion:** The study will help the practitioners to get the genuine drug and best outcome in their practice.

KEYWORDS: *Colocasia esculenta* (Linn.) Schott., Pharmacognostic study, Preliminary phytochemical investigations, Standardization.

INTRODUCTION

Treating diseases by using locally available natural resources can be drawn from the pre-vedic period itself. It further developed in the vedic period, where in the names and usage of around 100 plants were mentioned. This knowledge continued and found its utmost development in the *Samhita* period (Classical literature), where in it was systematically documented. Though the details of medicinal plants mentioned in *Samhita* are limited compared to that of *Nighantu kala* (Lexicon), they believed every plant is medicinal and the protocol of adding a drug into *Ayurveda* materia medica is elaborated by quoting about collecting the information from cowherds, shepherds and other persons who are well versed with names as well as forms of the plant.[1] As the time passed the knowledge was propagated to the society and more over the common people indigenously developed their own way of treating the disease using Flora in and around the vicinity of their practice.

Colocasia esculenta (Linn.) Schott. belonging to the family Araceae is one such therapeutically potent drug which is believed to be one of the earliest available plants common in Udupi,[2] called *Thev dhantu* in Tulu language, *Kesu* in Kannada, *Pindaluka*, *Aluki* in Sanskrit, is an annual herbaceous plant with long history of usage in traditional medicine in several countries across the world, especially in tropical and subtropical regions. The herb has been known since ancient times for its curative properties and has been utilized for treatment of various ailments such as Otalgia, Otorrhoea, Adenitis, Asthma, Arthritis, Internal haemorrhage, Hepatomegaly, Neurological disorders, Skin disorders etc. In the recent times its extract is proved to be having Anti-bacterial, Anti-fungal, Anti-inflammatory, Analgesic, Anti-hepatotoxic, Anti-microbial activity.[3-6] It is a good source of Provitamin A, Vitamin C, Calcium, Phosphorus, Iron etc.[7], [8] and hence the demand for the drug is increased.

The petiole of *Colocasia esculenta* (Linn.) Schott. can be easily confused with *Alocasia* and *Xanthosoma* species and possibility of getting adulterated if the supply of crude drug is inadequate, hence Pharmacognostic study is the initial step to confirm the identity and to assess the quality and purity of crude drug. The adulteration of the crude drug can be prevented by means of its evaluation like macro-microscopic study. Microscopy is an indispensable tool for authentication of crude drug. Setting up a standard of pharmacognostic,

morphological and microscopical characters of petiole of *Colocasia esculenta* (Linn.) Schott. will enhance standardization, which can promise quality, purity and identity of samples. Preliminary phytochemical studies helps in characterization of the drug which in future will help the practitioners to get the genuine drug and best outcome in their practice. The present study will provide the precise information in respect of identification as well as its characterization.

MATERIALS AND METHODS

Sample collection

The plant source was identified in the field of Katapadi, Udupi District, Karnataka, India with the help of regional flora (Flora of South Kanara), collected in the month of March 2018, authenticated from an expert botanist and a voucher specimen was deposited in the Dept. of Pharmaceutical chemistry and Pharmacognosy, S. D. M. Centre for Research in Ayurveda and Allied Sciences, Udupi, Karnataka, India with Sample number as 18032402. The fresh plant was collected and used for macro-microscopy.

Macroscopic evaluation

The external features of the test samples were documented using Canon IXUS digital camera. The macroscopic features were compared to local flora for authentication.

Microscopic evaluation

Sample was preserved in fixative solution. The fixative used was FAA (Formalin-5ml + Acetic acid-5ml + 70% Ethyl alcohol-90ml). The materials were left in FAA for more than 48 hours. The preserved specimens were cut into thin transverse section using a sharp blade and the sections were stained with saffranine. The slides were also stained with iodine in potassium iodide for detection of starch. Transverse sections were photographed using Zeiss AXIO trinocular microscope attached with Zeiss AxioCam camera under bright field light. Magnifications of the figures are indicated by the scale-bars.

Physico-chemical analysis

Organoleptic examination and physico-chemical studies, viz., total ash, water-soluble ash, acid-insoluble ash, water and alcohol soluble extractive, loss on drying at 105°C as per standardized methods were carried out.

Preliminary phytochemical analysis

Tests for alkaloids, carbohydrates, steroids, saponins, tannins, flavonoids, phenol, coumarins, triterpenoids, carboxylic acid, resin and quinine were done.[9]

High Performance Thin Layer Chromatography

1g of Petiole of *Colocasia esculenta* (Linn.) Schott. was suspended in 10 ml of alcohol. 3, 6 and 9µl of the above extract was applied on a pre-coated silica gel F254 on aluminum plates to a band width of 7 mm using Linomat 5 TLC applicator. The plate was developed in Toluene: Ethyl acetate (7.0: 1.0). The developed plates were visualized under short UV, long UV and then derivatised with vanillin sulphuric acid and scanned under UV 254nm, 366nm and 620nm (After derivatisation). Rf, colour of the spots and densitometric scan were recorded.

RESULTS

Vernacular names[10]

Sanskrit	*Aluki, Alukam, Alupam, Pindaluka, Pindalu*
Kannada	*Kesu, Kesave dantu*
English	Taro, Cocoyam, Eddo
Hindi	*Aruvi, Kacchu, Pattarveliya*
Tulu	*Thev, Sev*
Tamil	*Sempu, shamakkilangu*
Telugu	*Chamadumpa, Chamagadda, Chamakura*
Malayalam	*Chempu, Chempakizhanna*

Taxonomical position

Kingdom	Plantae
Clade	Angiosperms
Clade	Monocots
Order	Alismatales
Family	Araceae
Genus	*Colocasia*
Species	***C. esculenta***

Morphology

Colocasia esculenta (L.) Schott Syn. *C. antiquorum* Schott belonging to the family Araceae is a large herb; stems slightly swollen at the base of the leaf-sheaths, arising from a hard tapering rhizome. Tubers up to 15cm diameter. Leaves up to 50 x 30cm, ovate to suborbicular-cordate, apex rounded and usually apiculate, basal sinus triangular, dark green, sometimes clouded with black; Petiole up to 100cm long, lamina thinly coriaceous, peltate-

ovate, cordate at the base, up to 50cm long, rarely longer with a triangular sinus cut one-third to half way to petiole, with a dull, not polished surface above, paler and coloured beneath, but rarely glaucous, green or violet. Peduncle much shorter than the petiole up to 50cm long; spathe c.30cm long, tube oblong, green; limb yellow to orange, narrowly lanceolate, caudate-acuminate, never widely open, curved slightly backwards in flower; spadix much shorter than the spathe, rather slender up to 20cm long. Female inflorescence as long as the sterile male inflorescence.

Organoleptic characters

Petiole characters

Colour: Violet

Taste: Mucilaginous, Slightly bitter, pungent and irritant

Odour: nothing specific

Touch: Smooth

Macroscopic characters

Petiole erect, with a triangular sinus cut one-third to half way to petiole, with a dull violet, not polished surface above, paler or colored beneath, but rarely glaucous.(Fig 1).

Figure 1a. Leaf and Petiole of *Colocasia esculenta* (L.) Schott.

Figure 1b. Petiole enlarged.

Figure 1: Macroscopy of petiole of *Colocasia esculenta* (L.) Schott.

Microscopic characters of petiole

It has epidermis followed by collenchyma next to which there are pericyclic fibres inner to this lies an arrangement of vascular bundles and vascular strands. Ground tissue consists of starch grains and acicular raphides. (Fig 2).

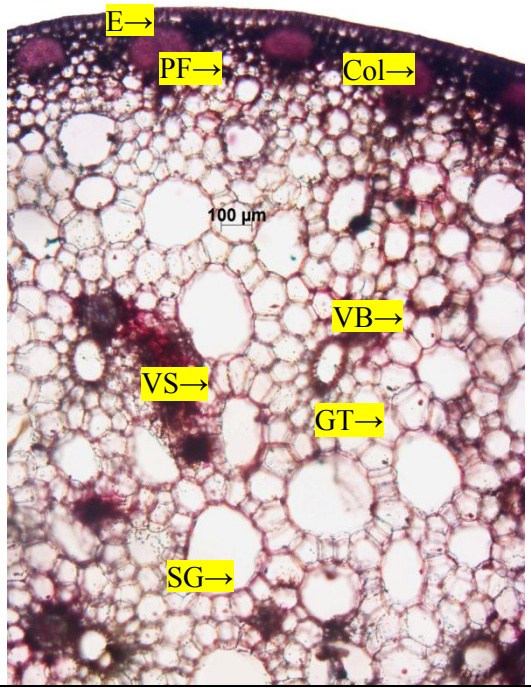


Fig 2a. Petiole of *Colocasia esculenta* (L.) Schott.

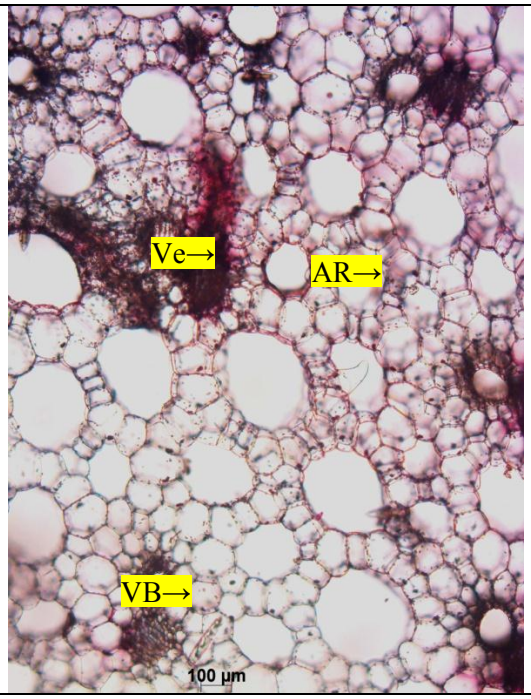


Fig 2b. Inner portion enlarged.

Figure 2: Microscopy of Petiole of *Colocasia esculenta* (L.) Schott.

AR – acicular raphides; Col – collenchyma; E – epidermis; GT – ground tissue; PF – pericyclic fibres; SG –starch grains; Ve – vessels; VB – vascular bundles; VS – vascular strands.

Physico-chemical tests

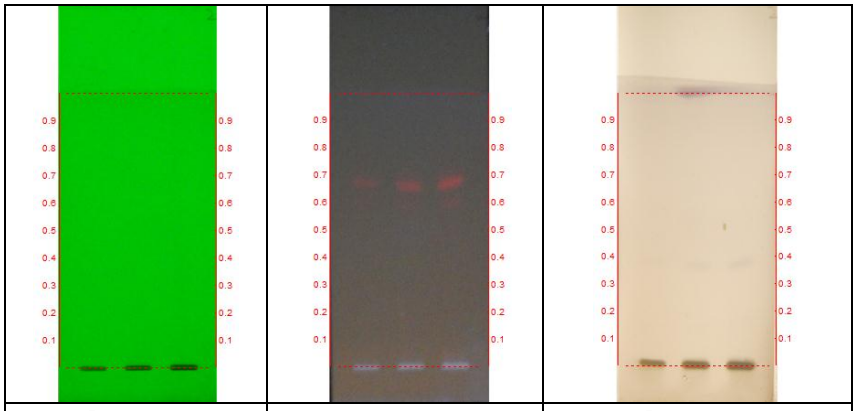
Results in (Average ± SEM) of Loss on drying of the petiole was found to be 17.15%, Total ash 19.51±0.24, acid insoluble ash 0.15±0.05, water soluble ash 15.16±0.24 alcohol soluble extractive value 7.37±0.04 and water soluble extractive value 43.34±1.18.

Preliminary phytochemical tests

Phytochemical tests carried out showed the presence of alkaloid, carbohydrate, tannins and coumarins.

HPTLC

HPTLC fingerprint was carried out using the alcoholic extract of Petiole of *Colocasia esculenta* (L.) Schott *Swarasa* (expressed juice of the petiole). The photo documentation shows at short UV, no bands were present. At long UV, 2 bands were evident at Rf values of 0.58 and 0.66, both fluorescent with red colour. After derivatisation with vanillin sulphuric acid under white light presence of 1 spot was seen corresponding to Rf of 0.37, purple in colour. (Fig.3).



Short UV	Long UV	Post derivatisation

Track 1: *Colocasia esculenta* (L.) Schott *Swarasa* - 3µl

Track 2: *Colocasia esculenta* (L.) Schott *Swarasa* - 6µl

Track 3*: Colocasia esculenta* (L.) Schott *Swarasa* - 9µl

Solvent system - Toluene: Ethyl acetate (7.0: 10)

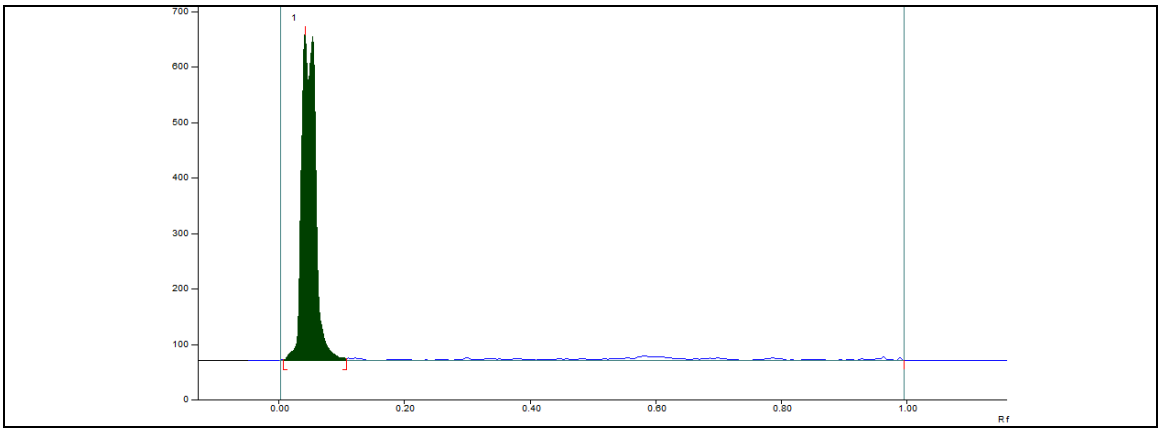
Figure 3: HPTLC photo documentation of ethanol extract of sample of Petiole of *Colocasia esculenta* (L.) Schott *Swarasa.*

Table 1: Rf values of Petiole of *Colocasia esculenta* (L.) Schott *swarasa.*

Short UV	Long UV	Post derivatisation
-	-	0.37 (Purple)
-	0.58 (F. red)	-
-	0.66 (F. red)	-

***F - fluorescent**

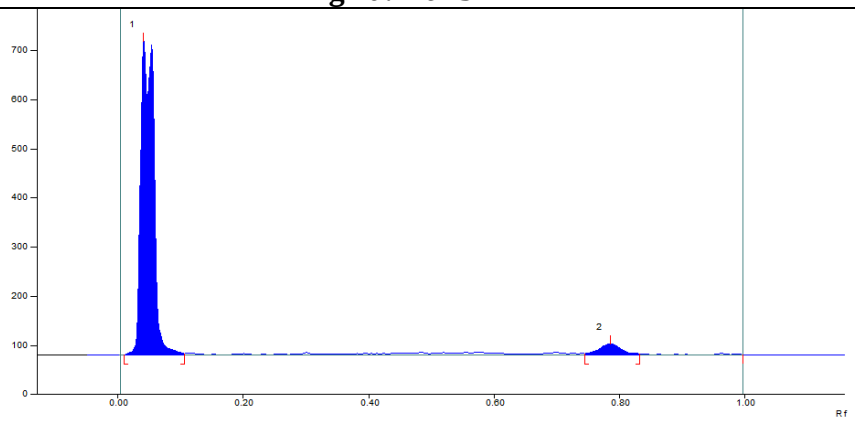
The densitometric scan at 254nm showed 1 peak corresponding to the phyto constituents present which was 0.04(100.00%). At 366nm 2 peaks were seen which were 0.04(94.21%) and 0.79(5.79%). At 620nm 3 peaks were seen which were 0.07(80.06%), 0.24% (3.31%) and 0.47(16.64%) (Fig. 4).



Track 3, ID: Colocasia esculenta swarasa

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.01 Rf	1.3 AU	0.04 Rf	586.8 AU	100.00 %	0.11 Rf	3.7 AU	9473.2 AU	100.00 %

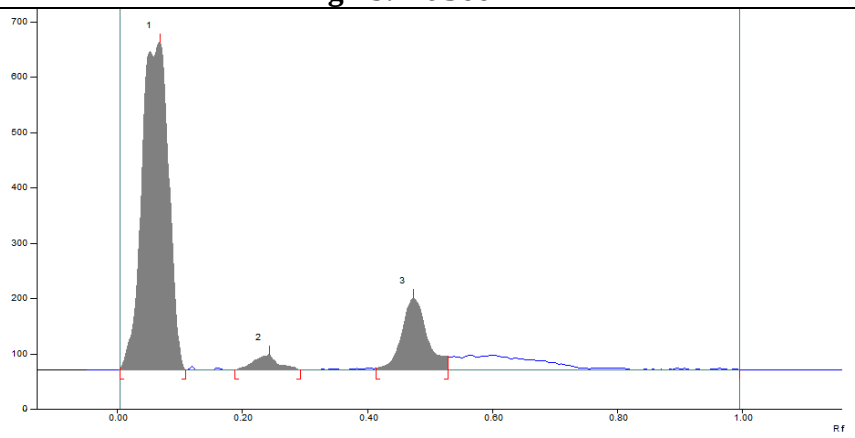
Fig 4a. At 254nm



Track 3, ID: Colocasia esculenta swarasa

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.01 Rf	0.3 AU	0.04 Rf	637.8 AU	96.55 %	0.11 Rf	2.6 AU	9605.8 AU	94.21 %
2	0.75 Rf	2.7 AU	0.79 Rf	22.8 AU	3.45 %	0.83 Rf	2.6 AU	590.2 AU	5.79 %

Fig 4b. At 366nm



Track 3, ID: Colocasia esculenta swarasa

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.00 Rf	3.1 AU	0.07 Rf	591.7 AU	79.00 %	0.11 Rf	0.2 AU	18156.9 AU	80.06 %
2	0.19 Rf	0.4 AU	0.24 Rf	27.5 AU	3.67 %	0.29 Rf	0.1 AU	750.2 AU	3.31 %
3	0.41 Rf	2.9 AU	0.47 Rf	129.9 AU	17.34 %	0.53 Rf	24.1 AU	3773.3 AU	16.64 %

Fig 4c. At 620nm

Figure 4: Densitometric scan of the sample *Colocasia esculenta* (L.) Schott *swarasa.*

DISCUSSION

The pharmacognostical study is one of the major criteria for identification of plant drugs. The incidences of harmful nature of synthetic drugs which are regarded as harmful to human beings and environment are ever-increasing. The advantage of natural drugs is their easy availability, economic and less or no harmful effects but the disadvantage is that they are victims of adulteration. The more effective the natural drug is, more is its demand and the

chances of the non-availability increases. The natural drug source is easily adulterated with low grade material these days to meet the growing demand. Therapeutic efficiency of medicinal plants are highly influenced upon the quality and quantity of chemical constituents.

This misuse of natural drug source begins with wrong identification. Generally the common error that happens is due to one common vernacular name that is given to two or more entirely different species. All these dilemmas can now be solved by pharmacognostic studies of medicinal plants. Pharmacognostic studies ensures plant identity, lays down standardization parameters which will help in preventing adulteration. Thus it will help in authentication of the plants and assures reproducible quality of herbal products which will lead to safety and efficacy of finished products. The preliminary phytochemical screening showed the presence of alkaloid, carbohydrate, tannins and coumarins.

Alkaloids are the eminent groups of phytoconstituents obtained from natural drug sources. They have varied and significant physiological effects on living beings. The medicinal properties are also quite diverse. It is an excellent analgesic. They have a wide range of pharmacological activities including analgesic, antibacterial, stimulant, antifungal, anti-inflammatory, anti-oxidant and antimicrobial activities.[11]

Plants produce and store several carbohydrates, most of which are regarded as primary metabolites. Plants like *Colocasia esculenta* (Linn.) Schott rich in mucilage are used in herbal medicine to treat cough, inflammation and to improve digestion.[12] Tannins are strong anti-oxidants, with anti-inflammatory, anti-parasitic, antibacterial and antifungal activities. Several medicinal plants are used internally and externally to treat inflammation and infection. Tannins are antiseptic on skin and mucus membrane, they are used as healing agents in inflammation.[13] Coumarins are known to be effective anti-inflammatory drugs and it aims against cell-adhesion molecules, thus very important in inflammatory responses. Coumarin has clinical medical value by itself, as an oedema modifier. Coumarin derivatives have been shown to possess antimicrobial and anti-oxidant properties. Studies have also shown that they exhibit anti-fungicidal property. Coumarins show anticipated therapeutic effect of anticoagulation by regulating blood fluidity and eliminating toxic effect of bleeding.[14]

The above phytochemicals present in the plant justifies its therapeutic value as to be a better analgesic, anti-inflammatory, antifungal, anti-oxidant drug as the action of the drug is directly reliant on its chemical composition.

CONCLUSION

The results of the pharmacognostical, physico-chemical analysis and HPTLC profiling helps in standardization with respect to its identity, purity and genuinity of the herbal material. Preliminary phytochemical screening showed the presence of alkaloid, carbohydrate, tannins and coumarins. With this sufficient data the drug can be taken up for studying it as per guidelines of *Ayurveda* and in future will help the practitioners to get the genuine drug and best outcome in their practice. The present study provides the precise information in respect of identification as well as its characterization.

SOURCE OF SUPPORT: Nil.

CONFLICT OF INTEREST: None declared.

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