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Pharmacognostical standardization of bark of *Thuja orientalis* Linn.

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Abstract

Thuja orientalis is a common ornamental evergreen tree that is originally native to Northwest China belonging to family Cupressaceae. The present study highlights the botanical as well as phytochemical studies including parameters such as macroscopic, microscopic characters, physicochemical evaluation and preliminary phytochemical studies of the bark. These observations will help in the botanical identification and standardization of the drug in the crude form and distinguish the drug from adulterants. These studies will be helpful in developing standards for quality, purity and sample identification of the plant. Preliminary phytochemical screening of extracts revealed the presence of glycosides, flavonoids, sterols, tannins, and carotenoids. The plant has exhibited extensively biological activities including anti-epileptic, anti-inflammatory, and hair growth-promoting, antiviral, anti-allergic, antibacterial, antioxidant, and antifungal activities.

Keywords: *Thuja orientalis*, Cupressaceae, Pharmacognostical, Physico-chemical parameters

Introduction

Thuja orientalis is a common ornamental evergreen tree, originally native to Northwest China belonging to family Cupressaceae. Plant is highly aromatic and resinous shrub widely cultivated in gardens located in temperate and semi-temperate areas. *Thuja orientalis* is hardy, large evergreen shrub or small to medium sized-tree rarely exceeding 20 m in nature. Shape is pyramidal, dense, but often exhibit, a more open and spreading form preferring moist, well-drained soil and full sun. The bark is grayish with brown highlights and has thin but deep furrows. Younger bark is a reddish-brown color and exfoliates in long, thin strips^[14].

Naturalized as an introduced species elsewhere in Asia: eastward to Korea and Japan; southward to northern India; and westward to northern Iran. The common name 'arbor-vitae' is from Latin, 'Tree of life', and is based on association with long life and vitality in Buddhist thought in China. This is probably based on the tree's unchanging evergreen nature in the cold dry climate of northwest China, and longevity; some of the larger specimens planted around Buddhist temples in China are said to be in excess of 1,000 years old. Although generally accepted as the only member of genus, included by botanists in the different classification. In older texts, *Platyclusus* was often included in *Thuja*, distantly related to genus. Differences from *Thuja* include distinct cones, wingless seeds, and almost scentless foliage. Later, genus name has been changed as *Biota orientalis* and latest as *Platyclusus orientalis*. But, it has been still known and sold as *Thuja orientalis* in market^[8].

Thuja orientalis leaves contain rhodoxanthin, amentofl-avone, hinokiflavone, quercetin, myricetin, carotene, xan-thophylls and ascorbic acid. The fruit and roots are strongly aromatic. Distillation of the dried roots yields an essential oil having the following properties- Sp.gr.200, 0.971[α]_D, -22.50n_D20, 1.5055: acid val, 2.1 and ester. Val. 26.27. After acetylation, 89.39; Carbonyls (as C₁₀H₁₆O), 5.65% and 50% in 7-8 vols of 95% alcohol^[10]. The composition of the oil is as follows: a new bicyclic sesquiterpene 51.10; 1-borneol, 17.10; bornyl acetate, 9.1; α-thujone and camphor, 5.6; and a new sesquiterpenic alcohol. The seed yields fatty oil showing composition as palmitic 5.28, stearic, 7.3; C₁₈ unsaturated acids, 1829 (linolenic, 44.6%); and C₂₀ un-saturated acids, 6.10%. The heartwood contains aroma- dendrin, taxifolin, widdrene, cedrol, thujopsadiene, dehydro-α-curcumene, β-isobiotol and Curcumenether. Thujone is a ketone and a monoterpene that occurs naturally in two diastereomeric forms: α-thujone and β-thujone. The fruit oil contained α-pinene (52.4%), 3-carene (14.2%), α-cedrol (6.5%) and-phellandrene (5.1%), the leaf oil contained α-pinene (21.9%), α-cedrol (20.3%), 3-carene (10.5%) and limonene (7.2%) as the main components^[12].

The root bark is used in the treatment of burns and scalds. The stems are used in the treatment of coughs, colds, dysentery and parasitic skin-diseases.

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The leaves are antibacterial, antipyretic, antitussive, astringent, diuretic, refrigerant and stomachic. Plant also used as Insecticidal, Molluscicidal and Nematicidal activity against different pests. Traditionally used in the treatment of cough, as a medicinal plant in various forms of traditional medicines like folk medicine, homeopathy for treatment of bronchial catarrh, enuresis, cystitis, psoriasis, uterine carcinomas, amenorrhea and rheumatism [9].

Materials and Methods:

Collection and identification of plant material:

The fresh plants of *Thuja orientalis* Linn were collected in the months of October from Hadapsar, Pune, Maharashtra, India, and authenticated by P. Lakshminarasimhan, Reference No. BSI/WRC/Cert./2015/ Botanical Survey of India, Pune.

Chemicals and Reagents

All the chemicals and reagents used for the study were of analytical grade and procedures were taken from official methods.

Macroscopical characters

Untreated sample was examined and studied for macroscopical characters such as colour, odour, taste, shape, size and fractures.

Microscopical characters

Thin transverse sections of the bark were cut and stained with phloroglucinol, concentrated hydrochloric acid and glycerine and observed under compound microscope [5, 6].

Powder studies:

For microscopical examination the powder was stained with phloroglucinol, concentrated hydrochloric acid and glycerin to study various anatomical features viz. oil globule, starch grain, calcium oxalate crystals, sieve tube and medullary rays [6].

Physico – chemical studies

The physico-chemical parameters are mainly used in judging the purity and quality of the drug. Ash values of a drug give an idea of the earthy matter or inorganic composition or other impurities. Extractive values give an idea about the chemical constituents present in the drug as well as useful in the determination of exhausted or adulterated drugs. The loss on drying reveals the percentage of moisture present in the drug. The dried plant material was subjected for determination of physicochemical parameters. The ash values such as total ash, water soluble ash, alcohol soluble ash and acid insoluble ash were determined according to standard procedures. The physicochemical parameters such as loss on drying, water-soluble extractive and alcohol-soluble extractive value were determined according to official methods for quality control of medicinal plants [3, 4].

Preliminary phytochemical screening

The dried bark was extracted with n-hexane, chloroform and ethanol. The behavior of powder with various chemical reagent and preliminary chemical tests for various extracts were also carried out according to the standard procedures [7].

Result

Macroscopical characters

A shrub or small tree, 9-10m tall, with a dense crown and thin, colour reddish brown. Bitter in taste; Characteristic

odour and cylindrical shape. Fracture short in outer bark and fibrous in inner part.

Microscopical characters

Microscopical studies are useful to establish the botanical identity for the valuable herbal drugs, and the basis for the identification and determination of adulterants. Cork is thick walled, flat, polygonal cells with reddish brown content; irregularly arranged cork cells are present. Phellogen, phelloderm is present below cork. Cortex is present beneath the inner cork cells. In cortex region essential oil globule present. Medullary rays are prominent multiseriate with thick walled parenchymatous cells. Oil cells are big, isolated

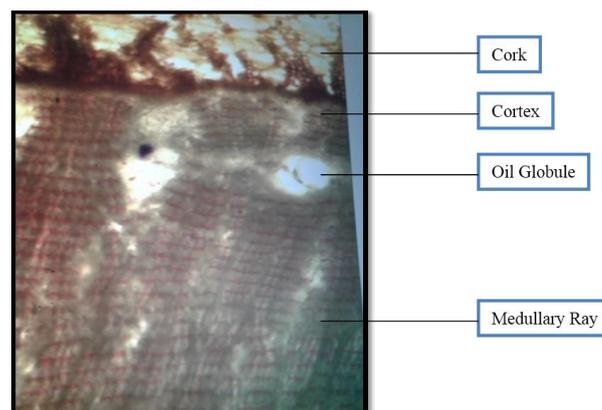


Fig 1: T.S of *Thuja orientalis* bark.

Powder studies

The powder characters of a drug are mainly used in the identification of the drug in the powdered form. Microscopical examination of the powder showed the presence of oil globule, starch grain, calcium oxalate crystals, sieve tube and medullary rays.

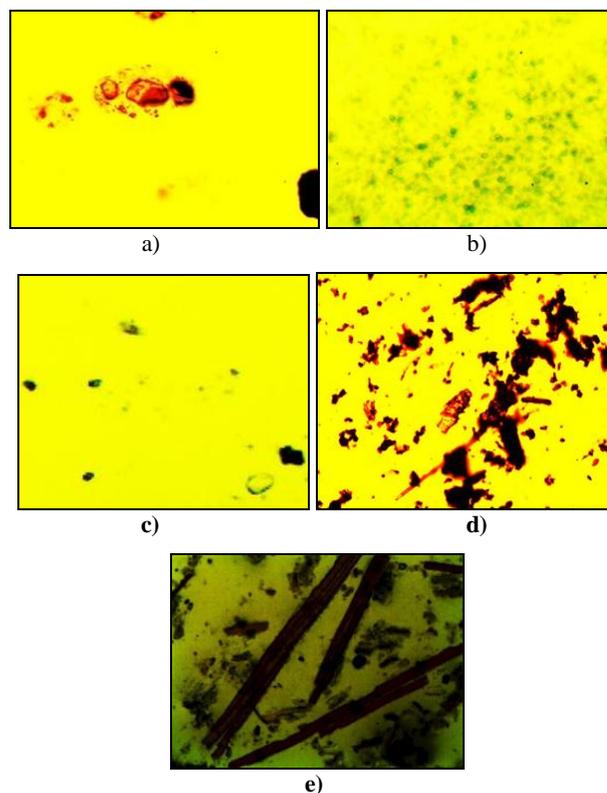


Fig 2: Microscopical characteristics of powdered drug:
a) Oil Globule b) Starch grain c) Calcium oxalate crystals d) Sieve tube e) Medullary Rays

Table 1: Characterization of Bark powder:

No.	Reagent	Observation	Inference
1	Powder + Phloroglucinol + Conc. HCL	Pink colour	Lignified Medullary ray present
2	Powder + Dil. Iodine solution	Blue colour	Starch present
3	Powder + Sudan red	Red coloured granules	Oil globules present
4	Powder + 60% H ₂ SO ₄	Needle shaped Ca-Sulphate crystals formation	Ca-Oxalate present

Preliminary phytochemical screening

The preliminary phytochemical screening was carried out on the extracts obtained after successive extraction with n-Hexane, chloroform and ethanol. The dried extracts were treated with chemical reagents for the detection of presence

and absence of phytoconstituents. The screening revealed the presence of different phytoconstituents like sterols, glycosides, alkaloids, flavonoids, tannins and carotenoids in various extracts as mentioned in Table no 2.

Table 2: Preliminary phytochemical screening of extracts

Test	n- Hexane	Chloroform Extract	Alcoholic Extract
1. Test for sterol: Salkowaski test	Present	-	-
2. Liebermann test	Present	-	-
3. Test for Glycoside: Molisch's test	-	-	Present
4. Test for Alkaloid: Dragendroff's test	-	Present	-
5. Test for Flavonoid: Shinoda test	-	-	Present
6. Test for Tannins: With FeCl ₃	-	-	Present
7. Test for Carotenoids: Carr-price reaction	Present	-	-

Physico – chemical studies

The physicochemical parameters such as total ash, water soluble ash, alcohol soluble ash, acid insoluble ash, loss on drying, water-soluble extractive value and alcohol-soluble extractive value were established mentioned in Table no 3. The ash values of the powdered bark revealed a high percentage of alcohol soluble ash. The results suggest that the powdered bark have high water soluble extractive value.

Table 3: Evaluation of Physical Parameters

Evaluation Parameter	Value (%w/w)
Total ash value	14.81
Water soluble ash value	5.56
Alcohol soluble ash value	11.70
Acid insoluble ash value	3.1
Loss on drying	7.61
Water-soluble extractive value	11.2
Alcohol-soluble extractive value	16.70

Extraction of phytoconstituents

The dried bark was extracted with n-hexane, chloroform, and ethanol. The extractive values by successive extraction method are mentioned in Table no.4 the percentage yield of ethanolic extract is more when compared to other extracts

Table 4: Evaluation of Extracts

Extract	Colour of Extract	Extractive value (% w/w)
n- Hexane	Dark Brown	6.5
Chloroform	Brown	5.6
Ethanol	Brown	13

Conclusion

The pharmacognostical standardization of express idea about identification, physical evaluation and monograph of plant. The present study on pharmacognostical characters of *Thuja orientalis* may be useful to supplement information with regard to identification and helpful in establishing the standardization criteria.

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