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# PHARMACOGNOSTIC EVALUATION OF *OSYRIS QUADRIPARTITA* SALZ. EX DECNE

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**ABSTRACT**

The comprehend assess the pharmacognostic characteristics of an important ethnomedicinal plant, *O. quadripartita* Salz. ex Decne. The pharmacognostic evaluation was executein terms of taxonomic, macroscopic, microscopic, fluorescence analysis, and phytochemical analysis. The *Osyaris* members heartwood majorly used to make furniture and perfumes. The *Osyris* plant members are known for fragrance and volatile oil constituents. The microscopic features of stem bark were observed as cork, cortex, phloem fibers, phloem medullary rays, and scleridesin the transverse section (T.S.) and longitudinal section (L.S.). The microscopy of stem bark powder observed the presence of cortex cells, sieve tubes, calcium oxalate crystals, and lignified fibers. The initial screening of phytochemical constituents of stem bark crude plant extracts indicate the presence of carbohydrates, proteins, alkaloids, glycosides, flavonoids, saponins, tannins & phenolic in different polar and non-polar solvent systems such as petroleum ether, chloroform, acetone, methanol, and water. Many pharmacognostic characters observed during the study, it's help in the standardization of *O. quadripartite*.



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## Introduction

In the world about 3000 to 3150 plant species used as medicinal pourpouse form ancient times [1]. Various diferent types secondary metabolites recorded in plant kingdom such as alkaloids, tannins, phenols saponins flavonoids etc., this phytochemicls used agints many diseases [2, 3].Crude drug discovery from natural medicinal plant resources these natural products very useful for to treatment of various disorders or diseases [4, 5]. *O. quadripartita* is a dry deciduous forest of family Santalaceae family and is commonly known as a wild tea plant and African sandalwood. Making the strong red-colored dye from the stem bark. The bark and heartwood of *O. quadripartite* can be easily adulterated with low-grade materials such as heartwood (African sandalwood) and strong red-colored dye [6, 7]. *O. quadripartita* comes under the least concern IUCN category [8]. The heartwood of this plant is faintly fragrant so, this plant is used as adulterating sandalwood in the commercial market. The stem bark is important for tanning the leather. The bark is used as a tonic, aromatic oils used in different products for manufacturing expensive perfumes, rare soaps, quality lotions, and sweet-smelling candles [9, 10]. In Folk, Indian, and other traditional medicine systems, bark and leaves are used by tribal peoples this plant leaves uses as making herbal tea [11, 12]. It is used as curing various diseases such as antihelmintic, leishmaniases like leprosy diseases, abnormalities, kidney disease, forage, hygienic/perfuming, eye pain, cold, anaphylactic shock, epilepsy, circumcision wound, toothache, tonsillitis, abdominal pain, and urine problem [13, 14]. It's also used for the treatment of cancer or tumor [15, 16], breast cancer [15, 17], anthrax and pasturolosis [18], Jaundice [19], wild edible fruit [20, 21], stomacache, malaria, cough, swelling and anti-corona virus disease (COVID-19) [22, 23], reduce inflammation and powerful emetic [24, 25], diarrhea [26], rabies [27], wound healing plants [28, 29], anti-ulcer activity [30, 31], mental problems [32], mastitis and poor motheringand antiviral activity [33]. This adulteration can be avoided by means of various evaluation variable such as

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microscopic as well as macroscopic evaluation. Macroscopic, microscopic, and chemical profiling evaluation techniques are important tools for the authentification of plant based crude drugs and the study of powdered drugs. Standardize of this plant part as per Indian Pharmacopoeia [34, 35]. It is significantly important characters such as morphological and anatomical characters, which is useful for drugs and their adulterants of commercial significance. The significant setup of the macroscopicaly, microscopically, and Pharmacognostic parameters of stem bark of the medicinal plant, will help in regularized, which can help quality control, identification, and purity of the sample [36].

## Materials and Methods

*Chemicals*
The analytical grade chemical used for staning of the microscopic parts of the studied medicinal plant of E. Merck Limited India and Hi-Media Laboratories, Mumbai, India used for analysis of sample.

*Botanical Parameter*
*Taxonomy & Morphology:* Identification and classification of plants using different Floras and other available literature [37].

*Collection Plant Materials*
The fresh stem bark part of *O. quadripartite* medicinal plant, were collected from different regions of Junar taluka, Pune districts of Maharashtra (India). The plant materials were taxonomically identification, authenticated by BSI, Pune reference numberis BSIS000001612. The plant materials dried in shade until all the water get evaporated and plant part became well dried for grinding. After drying, the plant materials were crush using mechanical blender (Mixer) into fine powder. This dried sample make fine powder and use for phytochemical and Pharmacognostic analysis.

*Pharmacognostic Studies- Microscopic Characters*
Plant anatomy Microscopical characters were studied by hand section of the stem *Osyris quadripartite* specimen. Wherever sections stained with safranin, fastgreen and iodine to identify and detection of the living tissues, dead tissues and starch granuels [38, 39].

*Microscopic Examination of Powder Sample by Using Maceration Technique*
Study of stem bark elements such as vessels, cortex cells, sieve tubes etc. Use a maceration technique by Jeffery's fluid [38, 39].

*Sensory Evaluation*
Various sensory variable done of the plant sample such as odour, taste, colour, size and shape.

*Physico- Chemical Parameters- Extractives Values*
Shade dried stem bark powder of *O. quadripartite* was subjected to constantly extraction with cold decoction for water only and hot extraction.

*Ash Values*
Water soluble ash, acid insoluble ash value and total ash value calculated by using plant sample **Table 2**.

*Powder Microscopy*
Take crude powder sample dissolved in water, then takes drop of this sample on slide, then put a coverslip and observed under light microscope.

*Fluorescence Analysis*
Take crude powder sample treated various chemicals then observe under visible light and U.V light **Table 4** [40].

*Method of Extraction*
The powder of shade dried stem bark of *O. quadripartita* was subjected to continuous extraction with cold decoction for water only and maceration hot extraction used differnet organic solvents such as petroleum ether, chloroform, acetone & methanol [40].

*Phytochemicals Qualitative Tests*
Followings phytochemicals qualitative tests used for analysis of *O. quadripartita*plant crude extracts, there are test for carbohydrates, amino acids, proteins, alkaloids, tannins glycosides, phenolic compounds, flavonoids, saponins & steroids [40].

## Results and Discussion

*Sensory Evaluation*

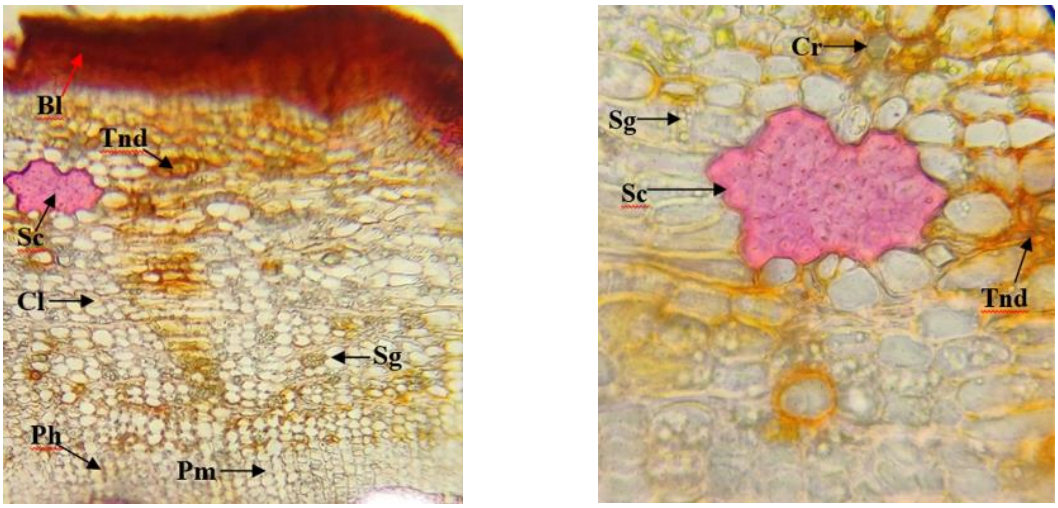
The Sensory evaluation of the stem bark showed dark brown color on the upper surface and a light brown color on the lower surface. The shade dried stem bark was dark brown in color, with a bitter taste, unpleasant in odor, and coarse texture [38].

*Macroscopic Evaluation*

The morphological and taxonomy characters are much branched shrubs; young branches sharply triangular. Leaves subsessile, coriaceous, elliptic–oblong, apex mucronate. Flowers yellowish–green, axillary; male flowers in long pedunculate, shortly pedicellate, 5–10–flowered umbels, the peduncles of bisexual flowers carrying 1–3 flowers and elongating in fruit, perianth of 3 triangular lobes. The dried stem bark thickness was about 3.37 mm [37].

*Microscopic Evaluation*

A superficial and strongly thickened cork layer was observed in the species. The cortex cells are arranged in compact form. The pericycle is a part of ground tissue. Ground tissue's primary function is mechanical support. Pericyclic fiber is sclerenchyma in nature. The phloem is stratified into soft and hard portions. In which hard portions are nothing but primary phloem fibers, proto-phloem fibers, and secondary phloem fibers. The various cluster of crystalline structures deposition was present in the phloem cells **Figure 1** [39, 41].




a) b)

**Figure 1.** a and b) Stem bark anatomy of *O. quadripartita*
Bl= Barklayer, Cl=Cortex layer, Ph = phloem, Pm= Phloem medullary layer, Sc= Sclerides, Cr= Crystalliferous cells, Tnd= Tannin deposition, Sg= Starch granules

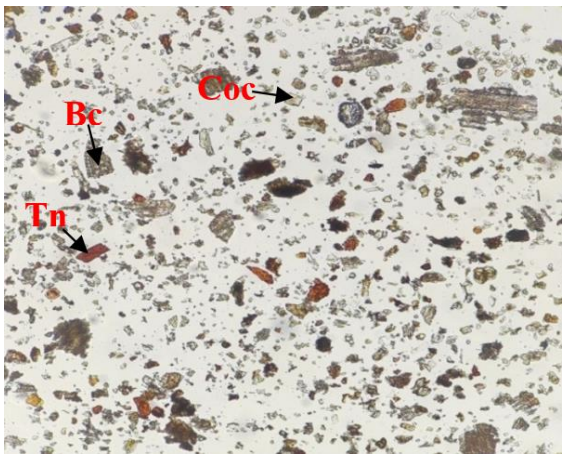



**Figure 2.** Microscopic observation of stem bark *O. quadripartita* Coc= Calcium oxalate crystals, Tn= Tannin, Bc= Bark cells

*Physical Evaluation*

The physical standards, such as ash values, and extractive values will be helpful to identify the standardness of the superior quality drugs, secondary drug, and drug adulteration from the powdered plant materials. It will maintain significant quality control of the drug obtained from medicinal plant sources. The facts collected from the crude bark powder ash values and extractive values are helpful for the quality control of drugs. Physical evaluation results are water-soluble ash 44.33%; acid

insoluble ash 21.66%; total ash 32.00 %; alcohol soluble extractives 97.6 % and water-soluble extractives 89.2 % w/w values were observed in dried stem bark of *O. quadripartita*. The results were mentioned in **Table 1** [40].

**Table 1.** Physical parameters of stem bark of *O. quadripartita*

| Sr. No. | Parameters | Determined value* (% w/w) |
|---|---|---|
| | **Ash values** | |
| 1 | Total ash | 32.00 |
| | Acid insoluble ash | 21.66 |
| | Water soluble ash | 44.33 |
| | **Extractive values** | |
| 2 | Alcohol soluble extractives | 6.2 |
| | Water soluble extractives | 11.8 |

* Mean value of three readings

**Table 2.** Stem bark average extractive values of *O. quadripartita*

| Sr No. | Solvent used | Colour appeared | Average percentage yield (% w/w on dry weight basis) |
|---|---|---|---|
| 1. | Petroleum ether | Light brown | 2.4 |
| 2. | Chloroform | Coffee brown | 3.6 |
| 3. | Acetone | Dark brown | 9.4 |
| 4. | Methanol | Dark brown | 7.2 |

*Average Extractive Values (% w/w)*

During the experimental work of consecutively solvent extraction, the overall percentage yields obtained, were purposeful as petroleum ether (94.6 %), chloroform (92.2 %), acetone (85.4 %), and methanol (97.0 %) w/w. The crude dried stem bark powder color developed during the successive extraction process as noted in **Table 2**.

*Fluorescence Analysis*

The fluorescence analysis results obtained were mentioned in **Table 3**.

**Table 3.** Fluorescence analysis of *O. quadripartita* stems bark powder.

| Sr. No. | Test | Visible Light | UV light |
|---|---|---|---|
| 1. | Powder as such | Cinnamon brown | Pecan brown |
| 2. | Powder + 1N NaOH (aqueous) | Brown | Greenish brown |
| 3. | Powder + 1N NaOH (alcoholic) | Brown | Greenish black |
| 4. | Powder +50%$HNO_3$ | Yellowish brown | Yellowish green |
| 5. | Powder + 50 % $H_2SO_4$ | Dark brown | Mehndi green |
| 6. | Powder + 1N HCl | Brown | Coffee brown |
| 7. | Powder + 5% KOH | Dark brown | Greenish black |
| 8. | Powder +Methanol | Brown | Greenish brown |
| 9. | Powder + Petroleum ether | Light brown | Ceramic brown |
| 10. | Powder + Chloroform | Brown | Coffee brown |

*Powder Microscopy*

Powdered stem bark of *O. quadripartita*under compound light microscopic observation showed tannin and cortex cells, calcium oxalate crystals. The calcium oxalate crystals of cubical type were observed in **Figure 2**.

*Preliminary Phytochemical Screening of O. Quadripartita Stembark*

The ethnomedicinal therapeutic properties of medicinal plants presence of several secondary metabolites and chemical components like alkaloids, glycosides, flavonoids, saponins, tannins & phenolics. This preliminary analysis of phytochemical tests helpful to the detection of active principles of chemical components and may accelerate to novel drug discovery in the pharmacognosy and pharmacology field. The preliminary crude extract of phytochemical screening leads to

revealing the existence of carbohydrates, proteins, alkaloids, glycosides, flavonoids, saponins, tannins & phenolics present in the dried stem bark crude extracts of *O. quadripartita* .The results noted in the **Table 4** [40].

**Table 4.** Qualitative tests for preliminary phytochemical extracts of *O. quadripartite*

| Phytochemical Constituents | Tests | Results | | | | |
|---|---|---|---|---|---|---|
| | | Petroleum ether | Chloroform | Acetone | Methanol | Water |
| Carbohydrates | Benedict's test | + | + | + | + | + |
| | Barfoed'stest | - | - | + | + | + |
| Starch | Iodine test | - | - | - | - | - |
| Proteins & amino acids | Xanthoproteic test | + | + | - | - | + |
| | Millon's test | + | + | - | - | - |
| Alkaloids | Mayer's test | + | + | - | - | - |
| | Wagner's test | - | + | + | + | + |
| Glycosides | Legal's test | - | - | + | + | + |
| | Borntrager's test | - | - | + | + | + |
| Tannins & phenolic compounds | Ferric chloride test | - | - | + | + | + |
| | Potassium dichromate test | + | + | - | - | - |
| Flavonoids | Concentrated H2SO4 test | - | + | - | - | + |
| | 10% sodium chloride test | - | + | - | - | - |
| Saponins | Foam test | + | + | + | + | + |

**Legend**: (-) absent, (+) present

## Conclusion

A study on morphological and anatomical characters of *O. quadripartita*, significant anatomical features helpful to maintain its quality control. The morphological, and anatomical characters of this plant, are the primary steps to establishing its botanical excellence. According to WHO rules, botanical standards set a proposed for the determination of the plant based herbal drugs. The pharmacognostic parameters is supportive for the future recognition, identification, and authentification of in the medicinal plant-based industrial componies such as herbal, cosmetics, and pharmaceutical. The physical quality analysis of stem bark plant material, like ash values (water soluble ash, acid insoluble ash, and total ash), and extractive values will be useful for recognition, identification, and standardization of the dried stem bark powdered of medicinal plant material. The scientific study developed standard quality data used for the quality control of the various medicinal preparations, were helpfulin the future for drug development. The ash values as well as extractive values are very much helpful for the standardization of crude drugs. The physicochemical standards of this plant can be separated from other related species**.** The dried stem bark constituents can be incorporate as macroscopic and microscopically standards as per Indian Pharmacopoeia. This medicinal plant in dried stem bark has some secondary metabolites reported like alkaloids, glycosides, saponins, flavonoids, tannins and phenolic. The calcium oxalate crystals of cubical type observed in the cortex region of stem bark. Thus this preliminary analysis of phytochemical tests will use in the find out of the active essential chemical components and later may guide to novel drug finding. These novel pharmacologically active principal chemical components will be used for curing various diseases and abnormalities**.**

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