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## Preliminary pharmacognostic standardization of leaves of *Camellia sinensis* leaf (Theaceae)

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### Abstract

*Camellia sinensis*, being belonging *Theaceae* family, is present more India less than China. A study on *Camellia sinensis* leaves sample extracted the air-dried leaves powder with different solvents like *n*-hexane, petroleum ether, chloroform (61°C), ethanol (78°C) and distilled water. Preliminary phytochemical analysis was done measurement of various parameters like extractive value, (moisture content) loss on drying, and quantitative estimation of total ash, and acid insoluble ash, and water-soluble ash value may be helpful for identification of tea leaves powdered drug. The fully grown leaves are bright green colour smooth and leathery, young leaves are pubescent. The estimation of powder microscopy presence in the parenchyma, calcium oxalate, vascular traces, upper epidermis and lower epidermis. These studied recommended that the examined pharmacognostic and phytochemical parameters are of whole great value in quality control, formulation development, and biological activities of *camellia sinensis*.

**Keywords:** *Camellia sinensis*, phytochemical, extractive value, ash value, microscopy

### Introduction

The green tea is obtain from plant *C. sinensis* belongs to the *Theaceae* family. The tea is mainly consumed drink after water in the world. It is cultivated in at least 30 countries around the all world. The major tea producers are Southeast Asia including China, India, Japan, Taiwan, Sri Lanka, and Indonesia, and central African countries. Trials across India, China and common in multiple indigenous medicine systems such as Ayurveda, Unani and Homoeopathy are commonly used in remedial plants [2]. The tea leaves shrub classified in the *Theaceae* family with two *Camellia sinensis* varieties mainly *Camellia sinensis* var. *sinensis* and *Camellia sinensis* var. *assamica*. For contemporary pharmaceutical compounds, the use of leaves, extracts, pure and isolated compounds from natural sources has always given an organization. There are mainly three kinds of tea according to the level of fermentation. They are classified as non-fermented (green and white tea), semi-fermented (oolong tea) and fermented (black tea). The green tea is recognized as 'non-fermented' tea and includes more catechins (20-30 % of the dry weight) than black tea. Catechins are *in vitro* and *in vivo* strong antioxidants [15] and antimicrobial activity [1, 16], anticholinestrase activity [17], reproductive system [18]. The results indicate a higher concentration of phenolic compound in non-fermented tea. These results specify that antioxidant activity demonstrated have no direct relationship with the anticandidal activity. *Camellia sinensis* that prevent cancer cells survival [12], antiviral activity of catechins from green tea against FCV [13].

### Materials and Methods

#### Collection and identification of plant material

The tea fresh leaves were collected from the *Camellia sinensis* (family *Theaceae*) plant growing in the tea gardens of Palampur, Kangra, Himachal Pradesh, India in the September and identified by an expert taxonomist in Department of Taxonomy & Pharmacognosy, Central Institute of Medicinal and Aromatic Plants. The plant were authenticated (No. CIMAP/Bot-Pharm./2018/08) and the leaves were washed with running water and air-dried for pharmacognostic evaluation, including examination of morphological and microscopical features and some preliminary phytochemical determination were then explored.

#### Instrumentation and techniques

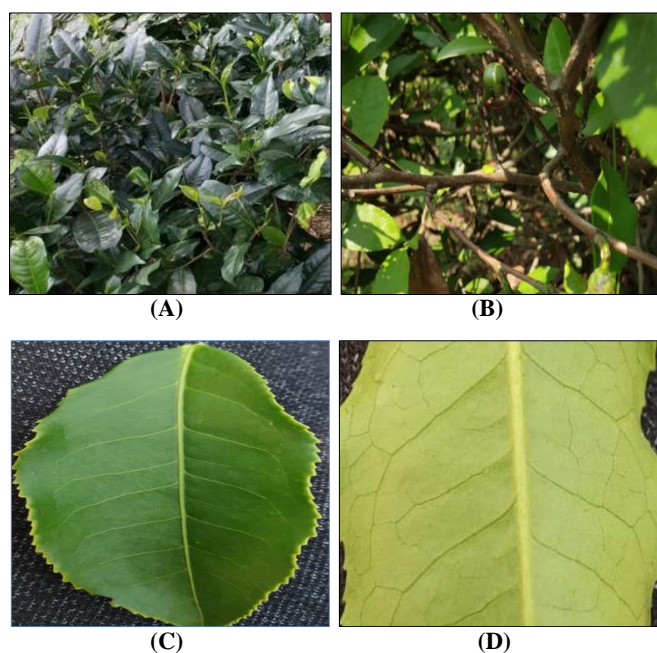
The leaf specimens were set and embedded in paraffin block, followed by dehydration, infiltration, sectioning, ultimately staining and photographing of the sections. Photography was done by using a *RADICAL® RXL-4T* Biological Research Trinocular Microscopes. Using *SYSTRONICS*, the powder leaves in different solvent were examined under normal daylight and UV-light (254 nm).

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The total ash, water-soluble ash, and acid-insoluble ash substance determined by using standard analysis methods as described in the Indian Pharmacopoeia 1966. The powdered leaves drug was performed as physicochemical constants and phytochemical screening carried out [19].

### Morphological characteristics of leaf

*C. sinensis*, a associate of *Theaceae* family is an entire green tree that attains a height of 10 – 15 meter in the wild and 0.6 - 1.5 meter when cultivated. The odour of leaves characteristics and bitter in taste. The leaves are bright green colour, short stalked, coriaceous, alternating, lanceolate [9], serrate margin. The mature leaves are gloomy green colour, smooth and leathery while young leaves are pubescent. The Flowers are white fragrant, 2.5 - 4 cm in diameter, found in solitary or in clusters of 2 or 4. Flowers bear various stamens with pale yellow anther and turn out dark-brown red capsules. The tea fruit is a three capsule compressed, smooth, rounded trigonous, solitary seed in each, the mass of a small nut [3].



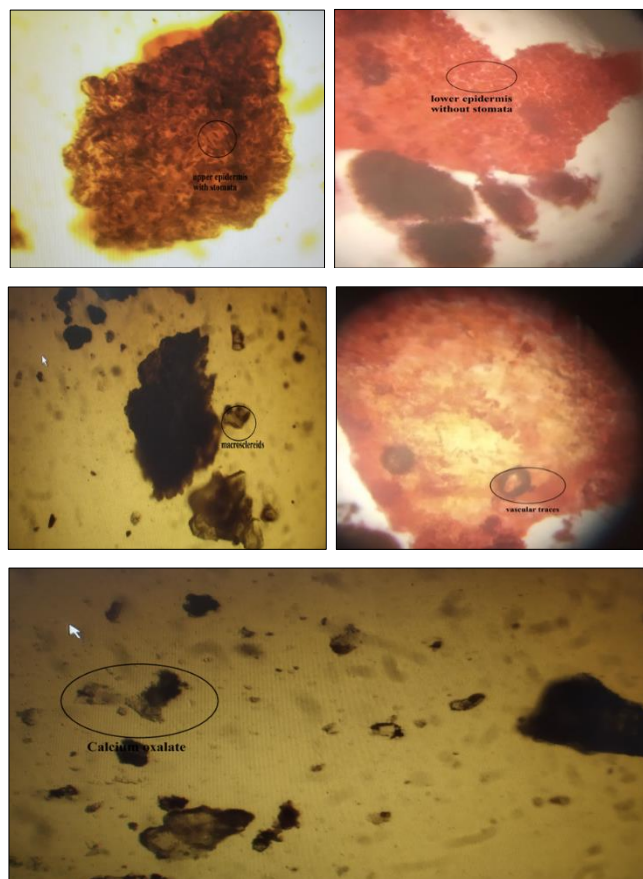
**Fig 1:** (A) Twig with oppositely arranged sessile leaves; (B) Seeds arranged on leaves (C) Broadly elliptical, pubescent when young and glabrous on maturity; (D) Portion of the midrib, lamina showing venation pattern

### Powder microscopical characteristics of leaf

Paraffin embedded leaves of *C. sinensis* were powder microscopy [21] with help of rotary microtome. Powder shows simple, multicellular, sharp or blunt ended, warty or smooth lower epidermal without stomata and upper epidermal with stomata cells leaf showing sinuous walls, and upper with equitably straight walls macrosclereids with globular head 3 or 4 cells, thick-walled and thin-walled fibres with sharp ends; cork tissue, fragments of vessel elements, prismatic and rosette and sandy crystal of calcium oxalate, vascular traces and palisade parenchyma scattered as such present [8].

**Table 4:** Preliminary Phytochemical screening of powdered Leaf of *C. sinensis*

| Phytochemical test | Ethanolic extract | Petroleum-ether | Chloroform |
|--------------------|-------------------|-----------------|------------|
| Carbohydrates      | +                 | +               | +          |
| Alkaloids          | +                 | -               | +          |
| Flavonoids         | +                 | +               | -          |
| Amino acids        | +                 | -               | +          |
| Glycosides         | -                 | +               | -          |



**Fig 2:** Powder microscopy of *Camellia sinensis*

## Results

### Physicochemical analysis

The microscopic and phytochemical studies are essential to authenticate this Ayurvedic preparation. Considering this requirement, powdered microscopy, preliminary phytochemical tests [9], physicochemical parameters i.e. ash value, extractive value, moisture content of a drug given an idea of the earthy inorganic or earthy matter composition and other impurities present all along with the drug.

**Table 1:** Ash value of powdered Leaf of *C. sinensis*

| Type of Ash        | Ash value (% w/w) |
|--------------------|-------------------|
| Total ash          | 6                 |
| Acid-insoluble ash | 3                 |
| Water-soluble ash  | 3.5               |

**Table 2:** Extractive value of powdered Leaf of *C. sinensis*

| Type of Extract                  | Extractive value (% w/w) |
|----------------------------------|--------------------------|
| Water soluble extractive value   | 0.20                     |
| Alcohol soluble extractive value | 0.15                     |

**Table 3:** Moisture Content value of powdered Leaf of *C. sinensis*

| Name of Plant            | Loss on drying (% w/w) |
|--------------------------|------------------------|
| <i>Camellia sinensis</i> | 6.43                   |

|                                |   |   |   |
|--------------------------------|---|---|---|
| Steroids and sterols           | + | + | + |
| Tannins and Phenolic compounds | + | + | - |
| Fixed oils and Fatty acids     | - | - | - |
| Terpenoids                     | + | - | - |

“+” = Presence of compound, “-” = Absence of compound

## Discussion

The macroscopical and microscopic analysis of leaves of tea, the quantitative estimation of leaves constants, ash values, and preliminary phytochemical screening of the leaves powder would be of appreciable use within the identification this drug. Empirical data regarding healthful plants play an important role in initial health care and has great potential for the invention of latest seasonal medicine. These findings could also be helpful to supplement existing information with relevancy the identification and standardization of tea, even in the powdered form of the plant drug, to evolve it from substitutes and adulterants. These studies additionally recommended that the discovered pharmacognostic and physicochemical parameters are of large values in internal control and formulation improvement. All in all, the present studies can be valuable to supplement data with observe to its recognizable proof and, institutionalization, and completing more research and authenticate its use the Ayurvedic System of medicine.

## Conclusion

The various specific pharmacognostic and physicochemical analysis of *Camellia sinensis* can be used as diagnostic tools for the correct identification of the plant drug and also to detect the authenticity, adulteration of this medicinally helpful plant and presence in various bioactive compounds.

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