Pharmacognostic evaluation of the rhizomes of *Curcuma zedoaria* Rosc.

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**ABSTRACT**

*Curcuma zedoaria* Rosc. (Family-Zingiberaceae), commonly known as ‘yellow zedoary’ is used in India system of medicine since time immemorial. The plant is found well in eastern Himalayas, Chittangang, Bengal, Kerala, Konkan and often cultivated throughout India. It is aromatic, pungent, bitter and useful in flatulent colic and debility of the digestive organs and also used as an ingredient in bitter tincture of zedoary and anti-periodic pills. A paste of rhizome is useful externally for cuts, wounds, itching and in sprains. A detailed phytochemical evaluation of its rhizome showed moisture content 83.22%, total ash 6.64%, acid insoluble ash 0.64%, alcohol soluble extractives 15.53%, water soluble extractives 18.96%, sugar 12.51% and starch 15.70%. A study of its volatile content also has been done indicating 2.8% of total volatile oil. These findings will be very useful for the identification of the species which may be useful to pharmaceutical industries for the quality control of the commercial samples.

**Key words:** *Curcuma zedoaria*, HPTLC, Pharmacognosy, Standardization.

**INTRODUCTION**

*Curcuma zedoaria* Rosc. (Zingiberaceae), commonly known as ‘yellow zedoary’ is aromatic, pungent, bitter and useful in flatulent colic and debility of the digestive organs and also used as an ingredient in bitter tincture of zedoary and anti-periodic pills, a paste of rhizome is useful externally for cuts, wounds, itching and in sprains.[1-4]

Rhizomes are employed in Asian and many other countries, including Brazil, for the treatment of several ailments, such as cervical cancer[5,4], hepatitis, inflammations[7] and dolorous processes.[9] Several studies have confirmed and also extended most of the mentioned popular uses of this plant. In this context, several workers were demonstrated its antifungal[9], antiulcer[10], antimutagenic[11], hepatoprotective[12] and cytotoxic[8] properties. It is well-documented that its main active principles are terpenoids, especially sesquiterpenoids[5,13,14] which also are produced by cultured cells.[15] *C. zedoaria* is also commonly used in medicine but with high starch content.[16]

Although the drug is fairly important and has good economics but no pharmacognostical work have been done in details. Therefore the present study had been done to document its detailed pharmacognostical information which will be utilized by the industries for the authentication and quality control of this drug.

**MATERIALS AND METHODS**

**Plant material**

The plant material was collected from field near Palghat area of Kerala, India, authenticated and lodged in Institute’s

Herbarium [LWG 221248, 1999] and the rhizomes were preserved in 70% ethyl alcohol for histological studies. Microtome sections were cut and stained with safranin and fast green and photographed with Nikon F70X camera.\textsuperscript{[19]}

**Physico-chemical and phytochemical assays**

Physico-chemical and phytochemical studies has been done from the shade dried powdered material according

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**Plate 1:** Macro and Microscopic characters of the rhizome of Curcuma zedoaria Rosc.

**Abbreviations**

ICO, Inner cortex; CK, Cork cells; IVB, Inner vascular bundle; OVB, Outer vascular bundle; CO, Cortex; FR, Fibre; ST, Starch; VS, Vessels; XY, Xylem; ED, Endodermis; PR, Pericycle.
to the recommended procedures.\textsuperscript{20-22} The behavior of the powdered drug with different chemical reagents was also studied as per methods described.\textsuperscript{23,24}

**RESULTS AND DISCUSSION**

**HPTLC studies**

The HPTLC analysis was carried out on precoated silica gel 60 F\textsubscript{254} Merk glass plates of 20 × 10 cm with the help of Camag Linomat- IV applicator and eluted the plate at room temperature in solvent system Chloroform:Ethanol:Acetic acid (95:5:0.1).

**Macroscopic characters of the rhizome**

The primary rhizome or rootstock is almost conical or top shaped. Attached to the primary rhizome are several sessile

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### Table 1: Fluorescence powder study of *C. zedoaria* rhizome

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Treatment</th>
<th>Day light</th>
<th>UV-254 nm</th>
<th>UV-366 nm</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Powder (P) as such</td>
<td>Yellowish Brown</td>
<td>Brown</td>
<td>Brown</td>
</tr>
<tr>
<td>2</td>
<td>P + Nitro-cellulose in amyl acetate</td>
<td>Florescent Yellow</td>
<td>Florescent Yellow</td>
<td>Yellow</td>
</tr>
<tr>
<td>3</td>
<td>P + N. NaOH in water</td>
<td>Brick Red</td>
<td>Dark Brown</td>
<td>Brown</td>
</tr>
<tr>
<td>4</td>
<td>P + 1N NaOH + Nitro-cellulose in acetate</td>
<td>Brick Red</td>
<td>Brown with Yellowish Tinge</td>
<td>Black</td>
</tr>
<tr>
<td>5</td>
<td>P + 1N HCl + Nitro-cellulose in amyl acetate</td>
<td>Florescent Yellow</td>
<td>Florescent Yellow</td>
<td>Yellow</td>
</tr>
<tr>
<td>6</td>
<td>P + 1N NaOH in Methanol</td>
<td>Brick Red</td>
<td>Brown with Yellowish Tinge</td>
<td>Black</td>
</tr>
<tr>
<td>7</td>
<td>P + 50% KOH</td>
<td>Brick Red</td>
<td>Brown with Yellowish Tinge</td>
<td>Black</td>
</tr>
<tr>
<td>8</td>
<td>P + 1N HCl</td>
<td>Brown</td>
<td>Light Brown</td>
<td>Black</td>
</tr>
<tr>
<td>9</td>
<td>P + 50% H\textsubscript{2}SO\textsubscript{4}</td>
<td>Black</td>
<td>Dark Brown</td>
<td>Black</td>
</tr>
<tr>
<td>10</td>
<td>P + 50% HNO\textsubscript{3}</td>
<td>Brown with Yellowish Tinge</td>
<td>Brown with Greenish Tinge</td>
<td>Brown with Violet Tinge</td>
</tr>
<tr>
<td>11</td>
<td>P + Conc. HNO\textsubscript{3}</td>
<td>Muddy Yellow</td>
<td>Brown with Greenish Tinge</td>
<td>Black</td>
</tr>
<tr>
<td>12</td>
<td>P + Acetic acid</td>
<td>Light Brown</td>
<td>Light Brown</td>
<td>Black</td>
</tr>
<tr>
<td>13</td>
<td>P + Conc. H\textsubscript{2}SO\textsubscript{4}</td>
<td>Black</td>
<td>Black</td>
<td>Black</td>
</tr>
<tr>
<td>14</td>
<td>P + Iodine water</td>
<td>Black with Greenish Tinge</td>
<td>Black with Greenish Tinge</td>
<td>Black</td>
</tr>
</tbody>
</table>

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**Figure 1:** Physicochemical values of *Curcuma zedoaria*
finger shaped lateral branches. The rhizome as well as its branches has an acrid or pungent taste and a distinct aromatic, camphoraceous smell. (Plate 1)

**Microscopic characters of the rhizome**
Epidermis, the outermost layer composed of rectangular tangentially elongated cells. In older rhizomes the epidermis is replaced by cork composed of 7 to 10 rows of typical rectangular to tangentially elongate thin walled cells. The ground tissue is differentiated into two regions, the outer cortex and the inner cortex by a distinct endodermis. The cortical ground tissue just beneath the cork contain yellowish contents i.e. curcumin. Almost all the parenchymatous cells forming the ground tissue are densely packed with starch grains. The starch grains are simple, comparatively big, flattened, rectangular or ovoid and possess a slight projection at one end. The striations on the grains are numerous. The endodermis is composed of a row of thin walled elongated cells with their radial walls slightly thickened. The cell layer within the endodermis also has tangentially elongated cells but narrower and some of them contain very small oblong starch grains. The cortical bundles as well as those within the stele are similar in structure. Bundles with a single vessel are very rare. Most of the bundles just within the endodermis are small. These contain only 2 to 5 xylem vessels. Each vascular bundle has got a sheath of small sized parenchyma cells, which completely encircle it. The cells forming the sheath, as is the case in the endodermal cells do not contain any starch (Plate 1).

**Quantitative microscopy**
On maceration, the vessels (744.939 × 15.829 μm) with annular and spiral thickenings are observed. Tracheids with bordered pits measuring 643.453 × 15.123 μm are also clearly discernable (Plate 1).

**Study powdered rhizome**
Powder light yellow; sweet, strong pungent, aromatic odour; shows fragments of storied cork, xylem vessels with reticulate thickenings, lignified xylem fibres, oil cells, patches of parenchymatous cells filled with starch grains which are oval-ellipsoidal, sometimes polygonal in shape, 10 to 60 μm, simple, hilum circular or a 2 to 5 rayed cleft, lamellae distinct and concentric (Plate 1).

The behavior of the powdered drug with different chemical reagents has been shown in the Table 1.

From the above studies rhizome can easily be differentiated on the basis of organoleptic characters for example the odour and taste of rhizome is quite characteristic and is
Table 2: Phytochemical screening of different extracts of *C. zedoaria* rhizome

<table>
<thead>
<tr>
<th>Extractive</th>
<th>Triterpenoids &amp; Steroids</th>
<th>Saponins</th>
<th>Flavanoids</th>
<th>Tannins</th>
<th>Reducing sugars</th>
<th>Resins</th>
<th>Glycosides</th>
<th>Alkaloids</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hexane</td>
<td>+</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Chloroform</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Acetone</td>
<td>+</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>+</td>
<td>–</td>
<td>+</td>
</tr>
<tr>
<td>Alcohol</td>
<td>–</td>
<td>+</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Water</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>+</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
</tbody>
</table>

Plate 2: HPTLC Profile of *Curcuma zedoaria* Rosc. and reference sample (Under UV-366)

1. Reference sample of Curcumin
2. HPTLC profile of *Curcuma zedoaria* Rosc. Rhizome

Abbreviations

*REF*, curcumin reference; CA, Curcuma zedoaria Rosc.

Physico-chemical studies
Physicochemical values viz. percentage of moisture, total ash, acid insoluble ash, alcohol and water-soluble extractives are observed. The total ash and acid insoluble ash, which are considered to be an important and useful parameter for detecting the presence of inorganic substances like silicate ion, it was found 6.64% and 0.64% respectively. Similarly the alcohol and water-soluble extractives, which are indicators of the total solvent soluble components, are 15.53% and 18.96% respectively. Likewise the essential oil, which is an important parameter for identification and authentication it was found to be 2.8% (Figure 1).

Successive Soxhlet extraction from non-polar to polar solvents viz. hexane, chloroform, acetone, alcohol and water were also carried out. It is interesting to note that C. zedoaria rhizome possessed an exceptionally high amount of acetone extractives i.e. 38.467%, which may be due to the higher percentage of curcumin which is purely soluble in acetone this is also comparable to amount of curcumin in C. longa (Figure 2). The preliminary phytochemical screening of different successive extractives is recorded in Table 2.

HPTLC studies
A densitometric HPTLC analysis was also performed for the development of characteristic fingerprint profile, which may be used as markers for quality evaluation and standardization of the drug. The HPTLC analysis was carried out on precoated silica gel 60 F254 Merck glass plates of 20 × 10 cm with the help of Camag Linomat- IV applicator and eluted the plate at room temperature in solvent system Chloroform:Ethanol:Acetic acid (95:5:0.1). The bands in the sample are obtained at Rf's 0.17, 0.43, 0.66, and 0.84, which can be used as identifying markers.

The Curcumin was identified at Rf 0.84. (Plate 2)

Heavy metal studies
The various heavy metals viz. Pb, Cd, Co, Mn, Cu, Zn and Hg concentrations was also estimated in the samples and all the metals are found within the permissible limits as prescribed by the WHO. (Figure 3)

Figure 3: Heavy Metal studies in Curcuma zedoaria rhizome
CONCLUSIONS

Thus on the basis of aforesaid studies, it can be concluded that the above parameters are very useful for the identification of the species which may be useful to pharmaceutical industries for the quality control of the commercial samples.

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REFERENCES