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ATLAS OF MACRO-MICROSCOPY OF RAW DRUG SOLD AS MUSTA – *CYPERUS ROTUNDUS* (L.)

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ABSTRACT: The quest for eternal health has made mankind to intensively research the nature itself. Nature has a rich source of medicinal plants, the proper use of which is necessary to maintain health. According to WHO, almost 80% of the population of developing countries has utmost faith in traditional medicines, of which plants are the important source. WHO recommends macroscopic and microscopic studies of the herb should be the first step in authentication, which is necessary to ensure the quality of any medicinal product. *Cyperus rotundus* (L.), a medicinal plant belonging to the family of the Cyperaceae, grows all over India. In the present study macroscopic and microscopic characterization of the rhizomes of Musta was done to develop an atlas of diagnostic characters by the standard methodology of macro-microscopic characterization of herbal raw drugs. Macroscopical and organoleptic characters as well as the microscopic features of Musta rhizomes were documented. A self-explanatory atlas of photomicrographs was prepared to aid rapid identification of raw drug called Musta. The atlas will serve as standard reference for identification and distinguishing Musta from its substitutes and adulterants.

INTRODUCTION: The quest for eternal health has made mankind to intensively research the nature itself. Though the medicines obtained from medicinal plants are effective in various ailments the standardization of drugs remains an uphill task. Authentication should be the primary criteria of any research using plants, which will help to ensure the quality of any medicinal product¹.

Pharmacognostic characters of herbs play an important role since particular macro-microscopic features are unique for each plant. WHO recommends macroscopic and microscopic studies of the herbs should be the first step to identify the botanical source before doing any research on plants².

Cyperus rotundus, commonly called Musta is a medicinal plant belonging to the family of the Cyperaceae. It grows all over India up to 2000 meters altitude, especially on the banks of streams, rivers. It appears among Indian, Chinese and Japanese natural drugs used as home remedy³.

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This plant grows well in tropical, subtropical and temperate regions⁴ and it is an invasive weed which significantly reduces the crop yield⁵⁻⁶.

In Indian system of medicine, its rhizomes are used in the treatment of several clinical conditions like candidiasis⁷, diabetes⁸, diarrhea⁹, malaria¹⁰, dysmenorrhea and menstrual irregularities¹¹.

It is also known for its cytoprotective¹², anti-mutagenic¹³, antioxidant¹⁴, anti-inflammatory, antipyretic and analgesic¹⁵ properties. Even though the rhizome of this plant has got tremendous medicinal properties, there is very minimum information on proper identification of the same. The present communication deals with the detailed macroscopic and microscopic features of the rhizome of *C. rotundus* as whole rhizomes as well as powdered form.

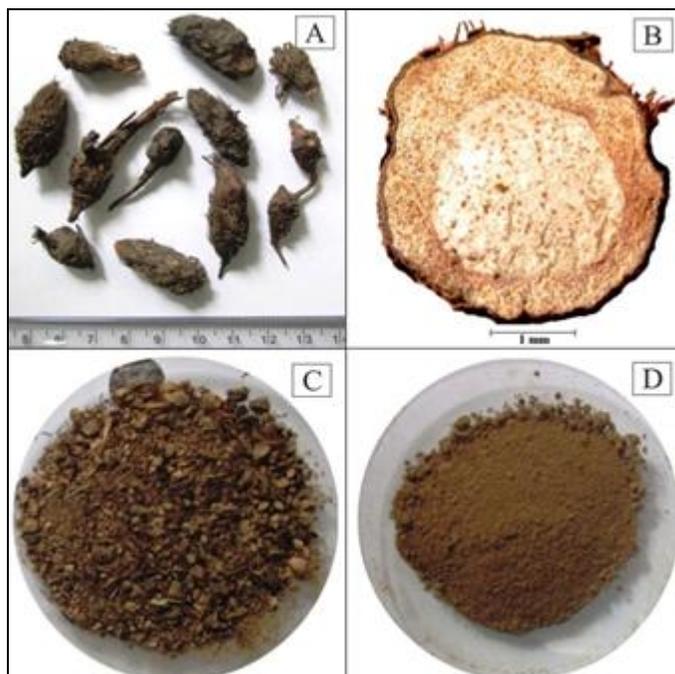
MATERIALS AND METHODS: The dried rhizomes of *C. rotundus* were collected from the local Ayurvedic pharmacy in Mangalore. The specimen (No. 11110101) is being stored in the Pharmacognosy Department of SDM Centre for Research in Ayurveda and Allied Sciences, Laxminarayana Nagar, Kuthpady, Udupi for future reference. The dried rhizomes were cleaned, coarsely powdered and preserved in air tight container for further studies. Dried whole rhizomes were used to study the external appearance, color, odor and taste. Coarse and fine powders are also prepared to record the organoleptic characters¹⁶.

Few fully matured rhizomes were preserved in fixative solution FAA (Formalin-5ml + Acetic acid-5ml + 70% Ethyl alcohol-90ml) for more than 48 hours. The preserved specimens were cut into thin transverse section using sharp blade¹⁷. The sections were stained with safranin as per standard methodology¹⁸. The selected diagnostic characters of the transverse section were photographed under suitable magnification using camera (Zeiss AxioCam ERc 5s) attached to trinocular microscope (Zeiss Axio Lab.A1) and the micro-measurements were taken using the pre-calibrated scale available in the software (AxioVision Rel. 4.8). A pinch of powder was warmed with drops of chloral hydrate on a microscopic slide and mounted in glycerin. Slides observed under microscope and diagnostic characters were observed and photographed using Zeiss AXIO trinocular microscope¹⁹.

RESULTS:

Macroscopy: *C. rotundus* rhizomes are bluntly conical with number of wiry and tough slender roots, often attached to one another by a thin and tough connective. Each rhizome is tumucate, varies in size and thickness, crowned with remains of stem and leaves forming a hairy to scaly covering.

The length of the rhizome is 1.5 to 3cms and diameter is 0.8 to 1.6 cms. Its stolons are elongated and about 1.5 to 3.5cms long. Externally the rhizome is dark brown or black in color and internally creamish yellow; odor- fragrant, taste- slightly pungent (**Figure 1A**). Fracture tough, mealy, shows dots of stelar vascular bundles and a distinct endodermal margin (**Figure 1B**). Coarse and fine powders are coffee brown in color, slightly pungent in taste, odor fragrant (**Figure 1C-D**).

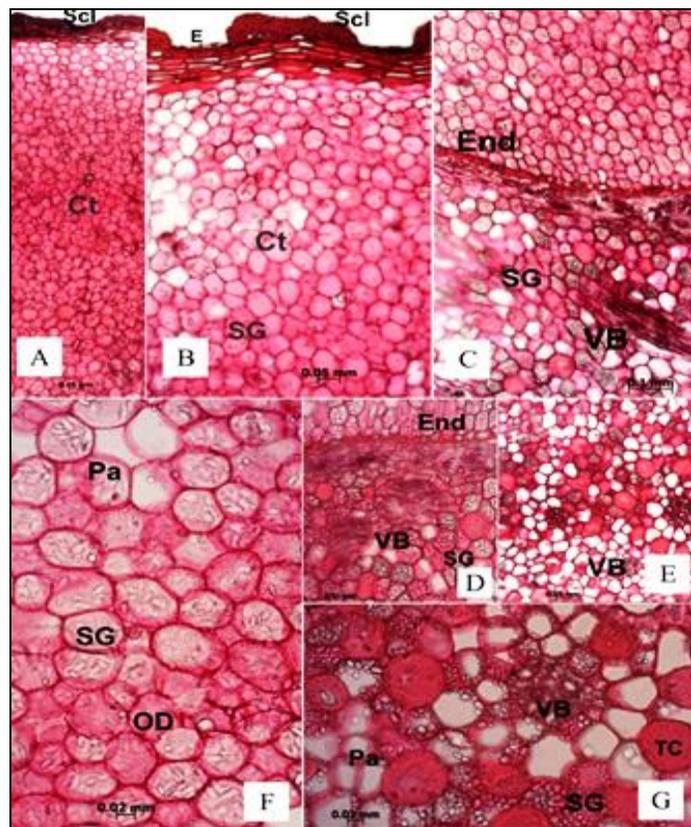


A. Dried rhizome **B.** Transversely cut surface of rhizome **C.** Coarse powder **D.** Fine powder

FIGURE 1: MACROSCOPY OF RHIZOME OF *CYPERUS ROTUNDUS*

Microscopy: Transversely cut surface of rhizome shows a line of demarcation of endodermis separating cortical portion from the central ground tissue. Few remnants of scale leaves are seen at the few places of the periphery (**Fig. 1B**). Detailed transverse section of rhizome shows single layered epidermis with brown colored pigment, often peeling off during maturity, leaving behind 2-6 layers of

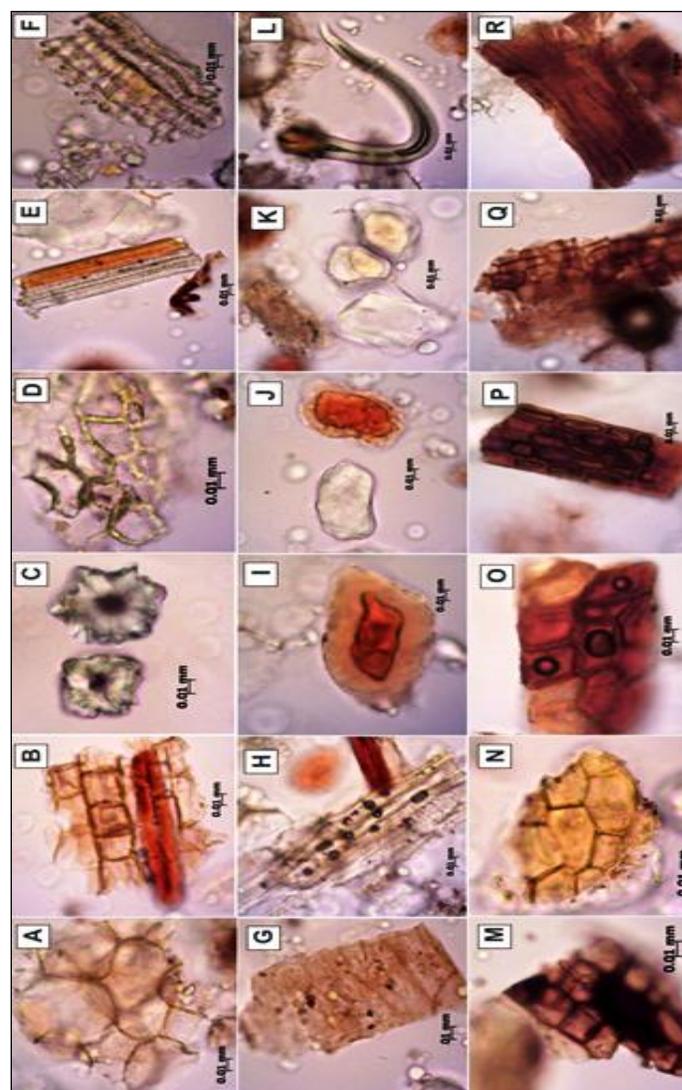
suberized sclerenchymatous cap-like patches filled with brown pigment; epidermis and outer sclerenchymatous layers are followed by 4 to 6 layers of radially elongated thick-walled cork-like cells which ends in compact outermost cortical parenchyma cells with no intercellular spaces; ground tissue of cortex consists of circular to oval, thin-walled, parenchymatous cells with small intercellular spaces; endoderm is distinct surrounding the wide central zone beneath endodermis, the stelar region is composed of circular to oval, thin-walled, parenchymatous cells with intercellular spaces, numerous collateral, closed, vascular bundles surrounded by bundle sheath of fibres, scattered in this region; vessels spiral to simple pitted; simple round, oval to elongated starch grains, a number of pigmented cells filled with reddish-brown oleo-resin content, present throughout the cortex and stele (Figure 2).



A. Transverse section
 B. Outer region enlarged
 C. Inner region enlarged
 D. Endodermis
 E. Inner region
 F. Cortex showing starch grains
 G. Portion enlarged to show vascular bundle
 TC - Tannin cell
 Ct - Cortex
 E - Epidermis
 End - Endodermis
 OD - Oil drops
 Pa - Parenchyma
 Scl - Sclerenchyma
 SG - Starch grains
 VB - Vascular bundle

FIGURE 2: MICROSCOPY OF RHIZOME OF *CYPERUS ROTUNDUS*

Powder microscopy: Creamish-brown; shows reddish-brown cells, spiral and simple pitted vessels; fibre-like, closely packed sclerified cells, narrow tracheids with pitted thickness and thick-walled trichomes from the remnants of leaves simple, thick-walled rounded to elongated parenchyma with brown content of tannin or pale oleo-resin, fragment of endodermis with pitted wall in surface view; round, oval to elongated starch grains and few rosette crystals of calcium oxalate (Figure 3).



A. Cortical parenchyma; B. Parenchyma with content cell; C. Rosette crystals; D. Endodermis layer; E. Fibre and pitted tracheid; F. Vessel fragment and starch grains; G. Parenchyma in surface view; H. Pitted tracheids; I. Thick-walled parenchyma; J. Parenchyma with content; K. Parenchyma with oleo-resin; L. Thick-walled trichome; M. Epidermis in surface view; N. Cortical parenchyma; O. Thick walled parenchyma; P. Hypodermal layer; Q. Transversely cut epidermis; R. Sclereidal fibres

FIGURE 3: POWDER MICROSCOPY OF *CYPERUS ROTUNDUS*

CONCLUSION: In the present study, we analyzed the macroscopic and microscopic characters of the dried rhizomes of *C. rotundus*. The results obtained will help to identify the correct species since no such detailed photographic atlas is available for exact picture of the characteristics.

This study will serve as a reference for identification and distinguishing rhizomes of *Cyperus sp* commercially available and to differentiate them from their substitute and adulterants. Detailed study on chemical analysis will be essential to acquire significant results.

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