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Assessment of Elements in *Curcuma caesia* Rhizome through Various Instrumentation Techniques

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ABSTRACT

Background: *Curcuma caesia* Roxb. is a zingiberaceae plant and their rhizomes have been used for various medicinal properties based on the presence of alkaloid, glycosides, essential oils and vitamin. But, the studies on medicinal properties of rhizomes of the plant in inorganic component are very limited. The trace elements and certain elements present in medicinal plants are important for safety of herbal drugs and also to help in establishing the structure in phytochemistry and chelating therapy. The present study emphasises to investigate the presence of nutrient and trace elements in the rhizome of the plant quantitatively, as deficiency or excessiveness leads to various health issues.

Methods: *Curcuma caesia* rhizomes were collected from Thoubal District, Manipur, North-East India during the month of November-December, 2017. Rhizomes powder was investigated for the determination of various elements by instrumental techniques like, Flame atomic absorption spectroscopy (FAAS), Inductively coupled plasma mass spectrometer (ICP-MS), Scanning electron microscopy (SEM), Energy dispersive X-ray (EDX) and Carbon - Hydrogen - Nitrogen - Sulphur (CHNS).

Result: After comparison of the various results given by FAAS, ICP-MS, SEM, EDX and CHNS Techniques, 17 elements consisting of both major and minor elements *viz*. C (7.705 wt.%), H (1.22wt.%), N (4.41 wt.%), O (36.97 at. %), Na (5.5 µg/mL), K (15.8 µg/mL), Mg (1.3 µg/mL), Fe (0.53 µg/mL), Ca (0.46 µg/mL), Co (<2 ng/mL), V (<2 ng/mL), Cr (6 ng/mL), Ni (30 ng/mL), As (<5 ng/mL), Mn (82 ng/mL), Cu (42 ng/mL) and Pb (10 ng/mL) have been determined in *Curcuma caesia* rhizome (µg/mL means micro gram per millilitre while ng/mL means nano gram per millilitre). A comprehensive elemental analysis data *Curcuma caesia* rhizome of such kind is not yet reposted earlier. The study will be very helpful in further study of the medicinal values of the rhizome.

Key words: *Curcuma caesia*, Medicinal value, Perennial herb, Rhizome, Trace elements.

INTRODUCTION

Medicinal plants are considered as the backbone of traditional medicine (Mazumdar *et al*., 2008). Nowadays, herbal-based medicine and the herbal-based drug industries are playing major role in international market (Sharma *et al*., 2008). In India, about 45,000 different plant species are available, out of which about 15,000-20,000 plants have been reported to possess good medicinal properties but only about 7,000-7,500 are being used by traditional practitioners (Kataky *et al*., 2010).

Plants belonging to zingiberaceae have huge medicinal properties besides it is used in food, perfume and in dyes (Riao *et al*., 2006; Chen *et al*., 2007; Nelson *et al*., 2017; Kumar *et al*., 2013). The plants consist of about 52 genera and more than 1600 species (Pitopang *et al*., 2018). They are grown in tropical and subtropical regions of the world. A few commercially important plant species in Zingiberaceae are ginger (*Zingiber officinale* Rosc.), turmeric (*Curcuma longa* L.), kasturi turmeric (*Curcuma aromatica* Salisb.), mango ginger (*Curcuma amada* Roxb.), large cardamom (*Amomum subulatum* Roxb.), *Aframomum* spp., *Kaempferia* spp., *etc*. One of the interesting characteristics of ziniberaceae is that they have the varied coloured rhizomes viz. blue, pale yellow, deep yellow, greenish blue, pink, colourless and combination of these and accordingly, their significances to the mankind are also different.

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Medicinally, the rhizomes of the plant have been used for its antifungal activity, smooth muscle relaxant, anti-asthmatic activity, antioxidant activity, analgesic activity, loco motor depressant, anticonvulsant, anxiolytic, CNS depressant activity, anti-ulcer activity and many other activities (Arya *et al*., 2017). The plant is widely cultivated in many countries: Ceylon, Belgium, Indonesia and France. Also, the plant is found in many parts of India: North- East states, Papi hills of East Godavari, Andhra Pradesh and North hill

forest of Sikkim (Sasikumar, 2005). The plant is native to North-East and Central India (Ravindran *et al*., 2007).

The studies of medicinal properties of the plant based on its organic component, *viz*. alkaloids, glycosides, essential oils, vitamins, other active components have been found extensive. But, assessment of medicinal property based on nutrient content is scanty. Various elements play vital roles in combating different human diseases (Prasad, 1993). Elements in small doses present in medicinal plants which have both therapeutics and prophylactic properties are known as trace elements (Hutchinson *et al*., 1963). They perform various vital metabolic reactions in the plants and animals (Suzanne, 2007). They not only play significant part in formulation of herbal drug but also in establishing structure and chelating therapy (Taylor, 1975; Burton, 1976). Excess or deficiency of the elements such as Fe, Cu, Co, Ni, Zn, Mg, Mn, Mo, Cr, V, Li, Se, F and I may also disturb normal biochemical functions of the body (Iyengar,1989; Burton,1976;Binita ,2018; Rupesh ,2018; Natalia *et al*., 2017; Silvy, 2019). Therefore, the present study was designed to investigate nutrients and trace elements present in the *Curcuma caesia* rhizome as a comprehensive and quantitative manner so that the rhizome can be used for various herbal formulations.

MATERIALS AND METHODS

Collection of rhizomes

The *Curcuma caesia* plant was identified by comparing with the voucher specimen (IBSD/-19), deposited in Institute of Bio-resources and Sustainable Development, Takyelpat, Imphal. The rhizomes of the plant were collected from Thoubal District, Manipur, North-East India during the month of November-December 2017 (Fig 1). The collected rhizomes were washed thoroughly with running tape water, followed by washing with double distilled water. The rhizomes were then sliced into small pieces and shade air dried at room temperature. The dried rhizomes were ground into powder using an electrical grinder and then stored for further use.

Sample preparation for analysis

Sample preparation and elemental analysis have been carried out in the analytical division of Bhabha Atomic Research Centre, Mumbai, India in the year, 2018-19.



Fig 1: (a) *Curcuma caesia* young plant, May 2017 (b) *Curcuma caesia* leaves with characteristic purple midrib, (c) Wither flower, June 2017, (d) A complete Plant's parts of *Curcuma caesia*, (e) Collection of rhizomes, (f) T.S. view of Rhizome, (g) Shed air dry of rhizomes, (h) Powder rhizome and (i) Rhizome solution.

1 gm. of the rhizome powder sample was added in 10 mL of ultra-pure nitric acid in a volumetric flask and warmed to dissolve it. Then, the final volume of the solution was made as 100 mL by adding deionized water. From this solution, FAAS and ICP-MS studies had been performed. SEM-EDAX study was performed on pellets of 1 cm diameter prepared from the rhizome powder under hydraulic pressure. And, about 200 mg each of dried rhizome powder was used to perform CHNS and XRD studies.

Characterization methods

Flame atomic absorption spectroscopy (FAAS) of GBC 906AA AAS unit with deuterium-arc background correction was used to detect the following elements Fe, Mg, Na, K, Ca down to ppm (mg/ml). The air-acetylene flame and nanopure water (18.3 Mega ohm) as diluent were used in this estimation.

ICP-MS (Inductively coupled plasma mass spectrometer) of model VG PQ Ex Cell, VG Elemental, UK, was used for determination of elements *viz.* Cu, Pb, Cr, Mn, Ni, Co, As and V down to ppb (ng/ml),

$$\text{Relative standard deviation (RSD)} = \frac{SD \times 100}{x}$$

Here x is in terms of ppm or ppb as mean. RSD values for FAAS and ICP-MS techniques in this study were calculated in the range of 2-5% and 5-10%, respectively.

SEM (scanning electron microscopy) images were recorded using model TESCAN VEGA3 and EDX (energy dispersive x-ray) analysis of elements present in the sample was also analysed by the same instrument.

Thermo-Fischer Flash EA 1112 Series CHNS Analyzer was used for quantitative determination of the following elements C, N, H and S.

XRD (X-ray diffraction) data of the dried powder sample was recorded using PAN analytical powder X-ray diffractometer of Ni-Filtered Cu-Kα (1.5405 Å) at 40 kV and 30 mA.

Results and Discussion

Using FAAS and ICP-MS analytical techniques, it is found that *Curcuma caesia* rhizome powder has the presence of the elements *viz.* Na, K, Fe, Mg and Ca in ppm level and Cr, Mn, Ni, Co, Cu, Pb, As and V in ppb level (Table 1).

In the scanning electron microscope (SEM), a high energy electron beam with a few keV targets surface of pellet of rhizome powder. After that, electrons interact with surface of pellet. In this way, backscattered electrons, secondary electrons, Auger electrons, as well as characteristic x-rays of elements present in surface of pellet are produced (Kumari *et al.*, 2012). SEM image and EDX (Energy dispersive X-ray analysis) spectrum of rhizome are shown in Fig 2(a, b). EDX data are given in Table 1. This study suggests that the contents of C and O are high and the amount of C is about 1.7 times that of O. Elements such as Si, Cl, Zr, Mo are also detected but are less than 1 at.% and so, they are not counted to be present in the sample which is the limitation of this instrument. The technique cannot provide the exact contents of light elements (Z<10) because the intensity of X-ray characteristics lines is low. CHNS analysis data indicates the presence of the elements: C, H and N in the sample (Table 1). Here, the weight percentages of the elements were expressed with respect to the mass of sample taken. The present study revealed that the three elements *viz.* C, H and N accounts 47.17 wt. % in the sample. The XRD pattern of the dried rhizome is shown in (Fig 2c.) It shows a broad peak with maximum at $2\theta = 23^{\circ}$ indicating the amorphous nature.

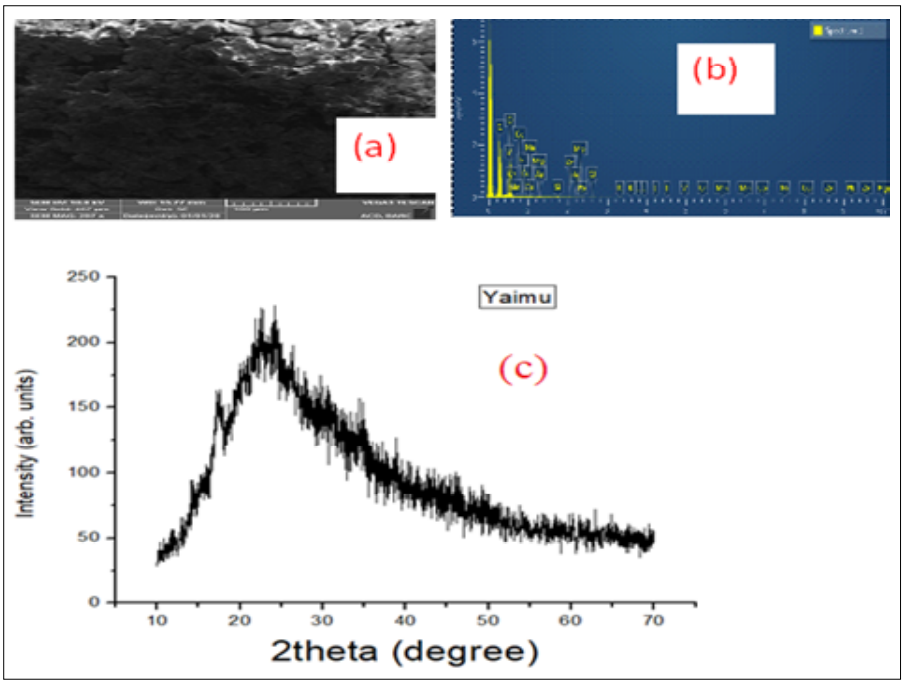

Fig 2: (a) SEM image of rhizome powder, (b) EDX spectrum of rhizome powder and (c) XRD pattern of rhizome powder.

In the present study, SEM-EDX technique determines the presence of the elements (in atomic %) C (62.74), O (36.97), Na (0.07), Si (0.09), Mg (0.04), K (0.02), Cl (0.01), Zr (0.02), Mo (0.01), As (0.03). In another study, the technique determined the presence of elements (in atomic %) C (63.7), O (24.2), Mg (0.6), K (2.8), Zr (5.2), Cl (0.1) and Mo (3.4) (Tamrakar *et al*., 2019) in the same plant species. It is inferred that the same plant species possesses different elemental contents depending on places where plants are collected. It indicates a medicinal plant could have different therapeutic values. The varied elemental content of the plant sample could be due to different locations in the world (latitude and longitude), climate, season, cultivation practices, fertilizer application, stress during growth or maturity, harvesting time, stage of maturity, storage, extraction and analysis methods (Dosoky, 2015; Sanghamitra *et al*., 2015; Srinivasan *et al*., 2016; Burt, 2004). Additionally, the present study indicates the presence of toxic elements such as Pb (10 ng/ml), As (<5 ng/ml); however, their presence is negligible (Bowen, 1979) which may not cause any adverse side effect, apart from its medicinal values. The present study covers a wider range of elemental analysis and determines a greater number of health beneficial elements in the sample than the reported ones. The study concludes the presence of C, O, H and N as major elements and Na, Mg, K, Fe, Ca, V, Cr, Mn, Co, Ni, Cu, As, Pb as minor elements (Fig 3) in the *Curcuma caesia* rhizome.

In this study, different instrumental techniques *viz*. FAAS, ICP-MS, SEM-EDX and CHNS have been used to investigate the presence of various nutritional and trace

Table 1: Elemental analysis of *Curcuma caesia* rhizome and its comparison to elsewhere studies.

Elements	*Curcuma caesia* (our study)				*Curcuma caesia* (reported)	
	FAAS	ICP-MS	SEM-EDX	CHNS	SEM EDX (Tamrakar *et al*., 2019)	PIXE (Dosoky, 2015)
	µg/mL	ng/mL	at.%	wt.% per mass of sample	at.%	Concentrations of elements are in ppm except for those in %.
C	-	-	62.74	7.705	63.7	-
H	-	-	-	1.22	-	-
N	-	-	-	4.41	-	-
S	-	-	-	0	-	-
O	-	-	36.97	-	24.2	-
Na	5.5	-	0.07	-	-	-
Si	-	-	0.09	-	-	-
Mg	1.3	-	0.04	-	0.6	-
K	15.8	-	0.02	-	2.8	1.62%
V	-	<2	0.00	-	-	-
Cr	-	6	0.00	-	-	-
Mn	-	82	0.00	-	-	215
Fe	0.53	-	0.00	-	-	27
Co	-	<2	0.00	-	-	-
Ni	-	30	0.00	-	-	-
Cu	-	42	0.00	-	-	3.4
Zr	-	-	0.00	-	5.2	-
Cl	-	-	0.01	-	0.1	-
Sb	-	-	0.00	-	-	-
Rb	-	-	0.00	-	-	8.2
Zn	-	-	0.00	-	-	153.5
Zr	-	-	0.02	-	-	-
Mo	-	-	0.01	-	-	-
As	-	<5	0.03	-	-	-
Hg	-	-	0.00	-	-	-
Se	-	-	0.00	-	-	-
Pb	-	10	0.00	-	-	-
Ca	0.46	-	0.00	-	-	0.069%
Mo	-	-	0.01	-	3.4	-

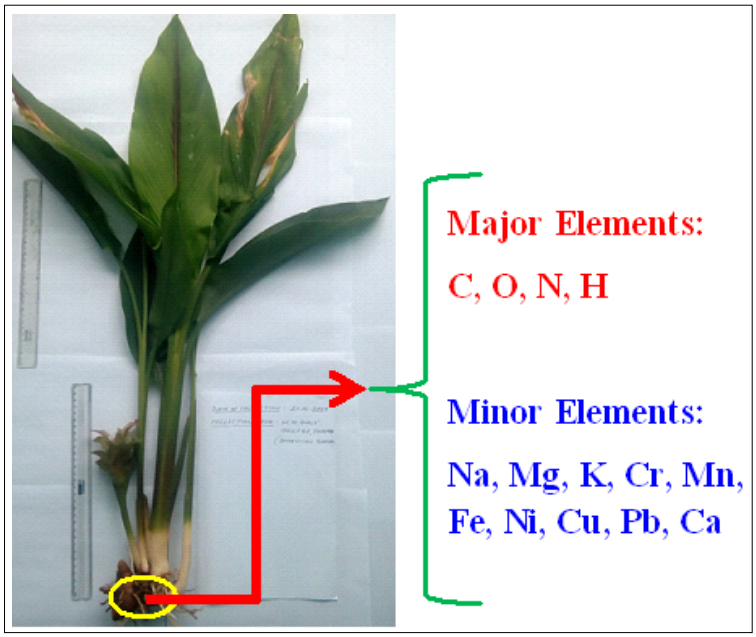

Fig 3: Major and minor elements present in the rhizome of *Curcuma caesia* Roxb.

elements in the sample as sensitivities of these techniques are different. FAAS techniques measures the presence of elements in µg/mL (ppm) level, ICPS technique measures down up to ppb label (part per billion) while SEM-EDX measures in terms of atomic per cent. It cannot measure the presence of elements in the sample below 1 atomic per cent.

CONCLUSION

Out of 17 elements determined in the rhizome of *Curcuma caesia* plant, nutrients and trace elements altogether comprises of 13 elements. The elements are Na, K, Mg, Fe, Ca, Co, V, Cr, Ni, As, Mn, Cu and Pb. All the trace elements found in the rhizomes were within the permissible limits of FAO and WHO. Therefore, the rhizomes can be considered as potential source for nutritional as well as herbal formulations.

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Conflict of interest

The authors declare no competing financial interest.

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