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New Report on Antioxidant 'Xanthone' in Some Members of Zingiberaceae

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Abstract
The present investigation is centered on the evaluation of antioxidant activity and detection of antioxidants in different plant parts of some selected members of Zingiberaceae such as *Alpinia nutans* (L.)*, Amomum subulatum* Roxb., *Costus speciosus* (Koenig) Sm.*, Hedychium spicatum* Buch. Ham.*, Elettaria cardamomum* Maton., and *Curcuma longa* (L.). Antioxidant activity was determined by an Assay of POD (peroxidase) activity (EC-1.11.1.7). High phenolic content, higher ascorbic acid content, and high antioxidant activity (POD) in different plant parts of all the selected members were obtained from*Alpinia nutans* rhizome, *Alpinia nutans* leaf, and *Costus speciosus*root respectively. An attempt has also been made to extract xanthones to confirm the presence of more antioxidant compounds, due to the high antioxidant potential of Zingiberaceae members. Todetect xanthone, methanolic extracts of the plant parts were scanned in the wavelength ranging from 200-800 nm using a Simadzu UV-1800 Series Spectrophotometer, and the characteristic peaks were detected. However, the resulting peaks were observed in the 200-400 nm wavelength. *Amomum, Hedychium,* and *Costus* were noted to contain xanthone. This is the first report on the presence of xanthone and the spectrophotometric method for its detection in these plants.

Keywords: antioxidant activity, xanthone, antioxidant, POD, Ascorbic acid, Zingibers.

Introduction

In Plants, the deleterious consequences of oxidative stress are inhibited or prevented by natural antioxidants which are present in all plants. Plants contain free radical scavengers like polyphenols, flavonoids, and phenolic compounds (Saha & Verma, 2015)Ascorbic acid, also known as vitamin C, is one of the potent naturally occurring antioxidants in a biological system (Evans &Omaye, 2017).The antioxidant property of ascorbic acid is attributed to its ability to reduce potentially damaging reactive oxygen species, forming, resonance-stabilized and relatively stable ascorbate free radicals(Buettner, 1993).Plant polyphenols have potent antioxidant properties that's why have the ability to

prevent various oxidative stress-associated diseases such as cancer (Taghipour et al., 2017).Therefore, identifying and extracting phenolic compounds from different plants has become a major area of health- and medical-related research.
Xanthones arealso a kind of natural product with a polyphenolic structure and of special interest for having many pharmacological effects, such as monoamine oxidase (MAO) inhibition, antitumor, antibacterial, antioxidant, antifungal, anticancer,and anti-inflammatory properties (Negi et al., 2013).It is also reported as an anti-cancer compound (Shan et al., 2011).
The medicinal properties of the rhizomes of Zingiberaceous plants have been widely discussed and accepted worldwide. These plants contain many essential oils which have been reported for their potential antioxidant, anti-inflammatory, and antimicrobial properties (Julie & Ernest, 2012).
Hence, the experimental work presented in this research paper focuses on the screening of some selected members of Zingiberaceae for potential antioxidant activity as well as the detection of antioxidantsin various plant parts. It was expected that through phytochemical evaluation of selected plants,if some new or alternative source of antioxidants is found out,the selected plants may become more useful for humankind.

Materials and Methods

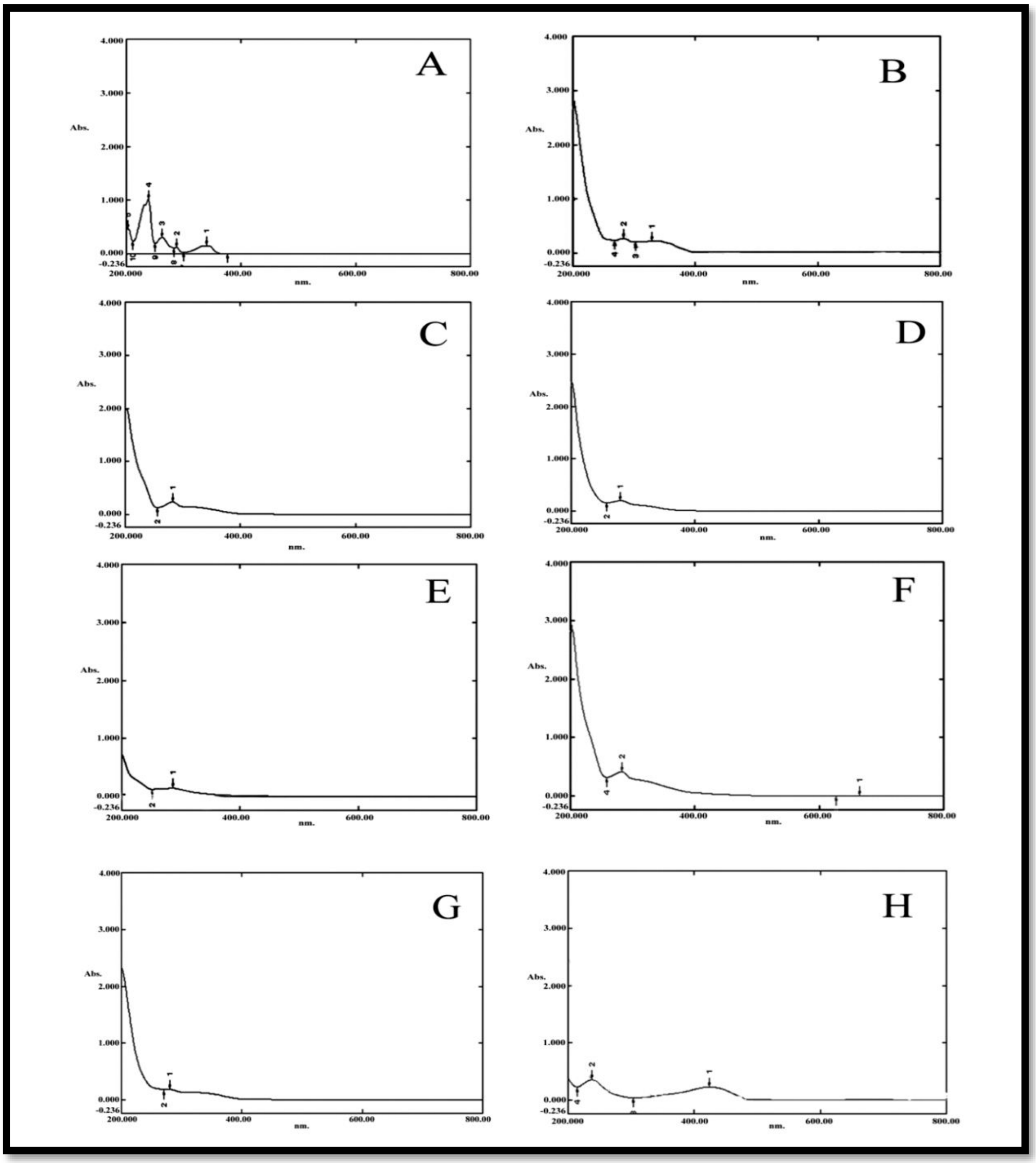
In the present study members screened for the presence of antioxidant compounds were*Alpinia nutans, Costus speciosus,Hedychium spicatum, Zingiber officinale, Elettaria cardamomum, Amomum subulatum*and *Curcuma longa*were selected for the present investigation. The plants have already been nurtured in the Botanical Garden of C.C.S. University Meerut, Uttar Pradesh, India due to their aesthetic, ethical, and medicinal value. Different plant parts such as root, rhizome, shoot, and seed of all the selected members were washed thoroughly with running tap water to remove the entire soil particle. The samples were cut into small pieces, oven-dried at 60°C, ground into fine powder, and stored in airtight polythene bags until use.
Estimation of total phenolic content, ascorbic acid, andPOD activity was carried out by well-defined protocol of (Bray & Thorpe, 1954), (Shukla et al 1979), and (Maehly& Chance 1954) respectively. The standardized protocol of (Liu et al 2010),opted for the extraction of xanthone.
Further analysis for the detection of xanthone was carried outby scanning thesamples' methanolic extract via a spectrophotometer. We find out that the spectrophotometer is a sophisticated instrument for qualitative as well as quantitativeanalysis of xanthone. A Spectrophotometer for the detection of xanthone is used for the first time in this investigation.

Results

The total phenolic content of selected plant materials ranges from 1.8-24.38mg cinnamic acid eq/g Dwt (dry weight). The ascorbic acid content and POD activity range from 3.15-54.49mg/g Fwt (fresh weight) and 0.27-25.55ΔA475/min/g Fwt respectively (Table 1 and 2).

Comparison of absorption spectra of standard and sample revealed the presence of xanthone or some polyphenolic compounds in leaf and rhizome of *Alpinia* (Figure 1 B and C), leaf and rhizome of *Amomum* (Figure 1 F and G), rhizome of *Hedychium* (Figure 1 H) and rhizome and seed of *Costus* (Figure 1 D and E). This is not found elsewhere in the literature. No peak was observed in the *Curcuma* leaf, *Curcuma* rhizome, and *Costus* leaf.

Figure 1 (A-H)Spectrophotometric screening for xanthone in plant parts of selected members along with authentic marker (Standard) A- Standard, B- *Alpinia* leaf, C- Alpinia rhizome, D- *Costus* rhizome, E- *Coustus* seed, F- *Amomum* leaf, G- *Amomum* rhizome, H- *Hydechym* rhizome


A
B
C
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Abs.
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200.000
400.00
600.00
800.00
nm.

Table- 1 Total phenolic content in different plant parts

S.No	PLANT NAME	PHENOLIC CONTENT, (mg cinnamic acid eq/g Dwt) PLANT PART			
		LEAF	RHIZOME	ROOT	SEED
1	*Costus speciosus*	6.21± 0.46	7.29± 0.79	9.39± 0.65	4.82± 3.50
2	*Curcuma longa*	7.21± 0.18	4.7±0.51	23.37± 1.29	NA
3	*Hedychiumspicatum*	8.93± 0.78	1.8± 0.14	11.95± 2.06	NA
4	*Alpinia nutans*	18.35± 0.54	24.38± 0.92	16.06± 0.69	NA
5	*Elettaria cardamomum*	6.25± 0.25	15.23± 0.84	13.02± 0.98	NA
6	*Amomum subulatum*	7.34± 0.61	17.58± 0.96	15.87± 0.37	NA

NA= Not available

To confirm the presence of more antioxidant compounds due to the high antioxidant potential of members of Zingiberaceae, xanthones were tested using authentic xanthone from Sigma-Aldrich because xanthone is a polyphenolic compound (Sima et al., 2016),which exhibits antioxidant activities as reported by several workers (Cruz et al., 2017; Blanco-Ayala et al., 2013). Spectrophotometric scanning of xanthone was conducted at a wavelength range of 200–400 nm to observe absorption spectra of standard xanthone at different wavelengths.

Table- 2 Ascorbic acid and POD activity in different plant parts

Plants	Plant Parts	Ascorbic acid (mg/g Fwt)	POD Activity (ΔA475/min/g Fwt)
Alpinia	Leaf	54.49 ± 1.36	7.49
	Stem	12.99 ± 1.19	1.38
	Rhizome	10.24 ± 0.10	0.27
	Root	54.68 ± 0.15	1.11
Costus	Stem	ND	ND
	Rhizome	7.6 ± 0.16	0.83
	Root	8.12 ± 0.04	25.55
	Seed	11.28 ±0.24	6.38
Hedychium	Rhizome	4.07 ± 0.54	1.38
	Root	9.59 ± 0.89	5.55
Curcuma	Rhizome	3.15 ± 0.67	1.94

	Root	4.08 ± 0.31	4.16
Zingiber	Rhizome	19.88 ± 0.01	8.88

ND= Not detected

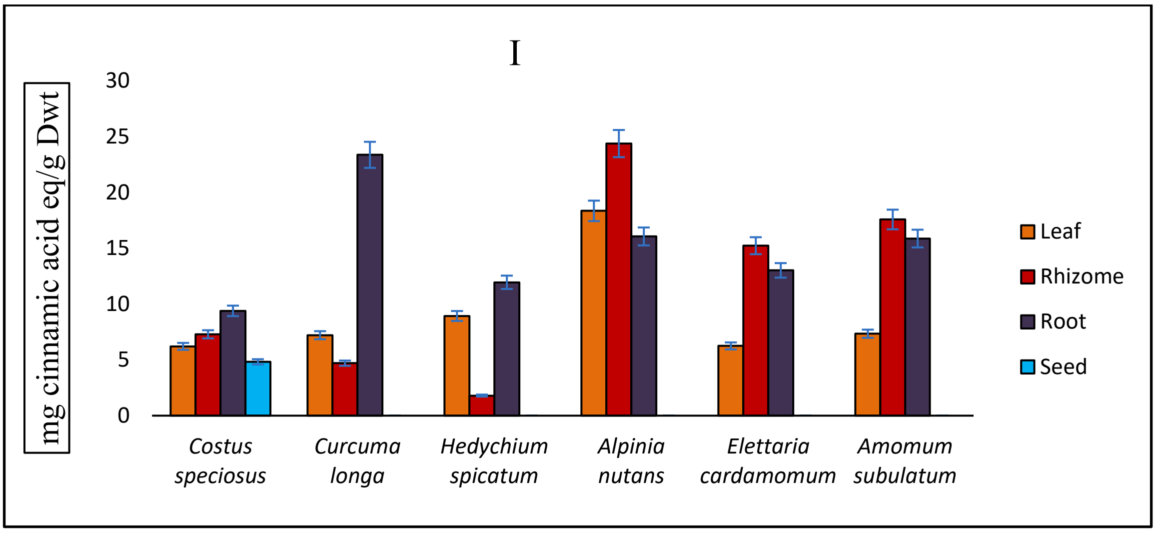
The attribution of the peaks to xanthones was unambiguous, as xanthones present characteristic UV spectra with four bands of decreasing intensity (Liu et al., 2010), However, in our investigation authentic marker exhibited five peaks instead of four with decreasing intensity between 200-400 nm regions of the absorption spectrum (Figure 1A). (Liu et al., 2010), reported that UV spectra of all the xanthone glycosides revealed maximum absorbance near 260 nm. However,we observed that standard xanthone exhibited maximum absorbance at 238.5 nm followed by 202.5 nm **(Table 3).**

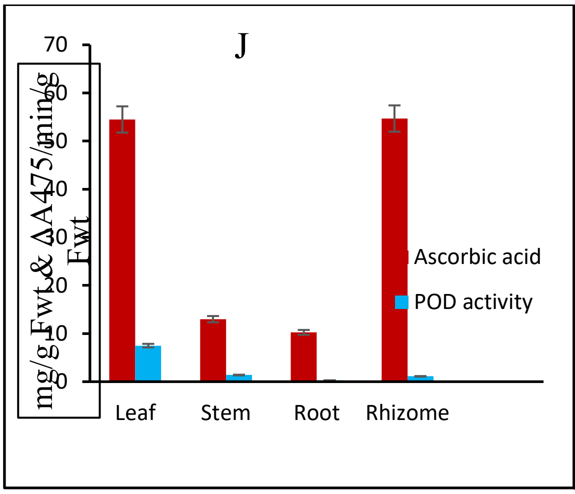
Table- 3 Absorption spectra of standard xanthone at a different wavelength

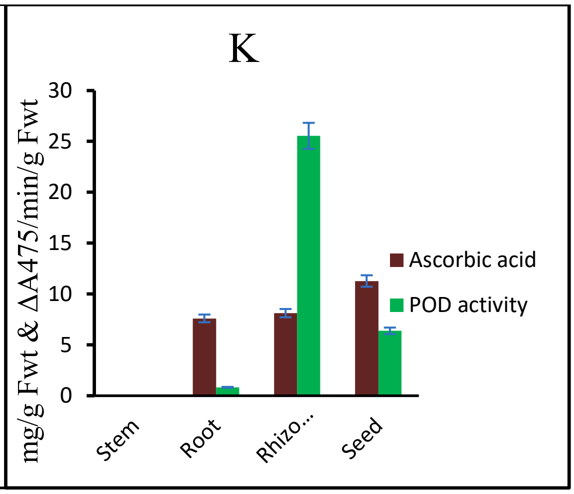
No.	P/V	Wavelength	Abs.
1	↑	339.500	0.153
2	↑	287.500	0.123
3	↑	262.500	0.304
4	↑	238.500	1.017
5	↑	202.500	0.460
6	↓	376.000	-0.003
7	↓	300.000	0.024
8	↓	283.000	0.109
9	↓	250.500	0.197
10	↓	211.500	0.233

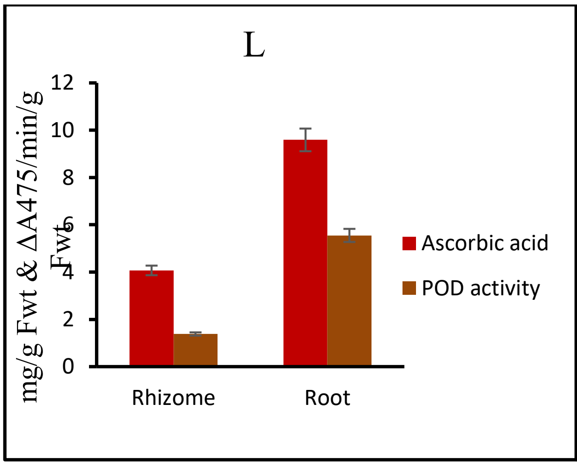
P=Peak, V=Valley, Upward, and downward arrows show peak and valley respectively

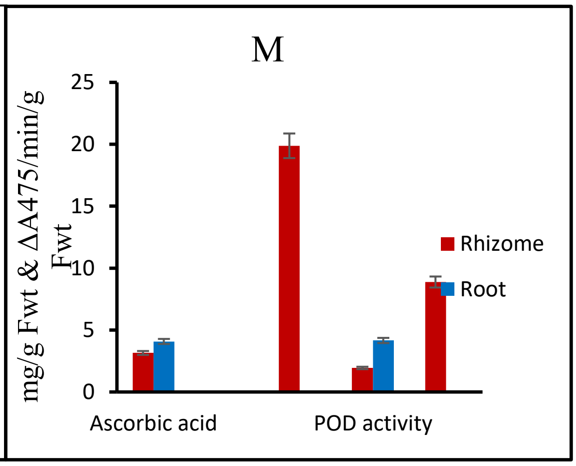
Figure(I-M)graphical representation of Phenolic content, Ascorbic acid, and POD activity in different plant species of Zingiberaceae (I- Phenolic content in different plant parts of members of Zingiberaceae. J- Ascorbic acid and POD activity in different parts of Alpinia, K- Ascorbic acid and POD activity in different parts of Costus, L- Ascorbic acid and POD activity in different parts of Hedychium, M- Ascorbic acid and POD activity in different parts of Curcuma and Zinger.











Discussion

In Alpinia, a leaf with higher antioxidant potential appears to be a better medicinal material than a rhizome, stem, or root. In *Alpinia,* higher phenolic content in stem and rhizome are accompanied by low antioxidant activity (POD) and ascorbic acid content indicating the phenolics to be carrying out the function of defense alone, not the antioxidants, as observed during the study of(Schmidt et al., 2014) where the phenolic compound content increased by more than 100% with solid-state fermentation of rice bran with the *Rhizopus oryzae* fungus and the phenolic extracts of fermented rice bran inhibited DPPH and peroxidase activity. In the other members of Zingiberaceae studied, with only storage parts available, higher phenolic content accompanied by higher ascorbic acid and antioxidant activity (POD) indicating phenolics, ascorbic acid, and antioxidant activity to contribute towards overall antioxidant potential rather than defense of the plant parts. As reported by (Zhang, 2015) phenolic compounds function as antioxidants. Accordingly, we also observed the same relation between POD activity and phenolic compounds and /or antioxidants (Ascorbic acid) as given away in **Tables 1 and 2.**

Conclusion

The potential antioxidant activity as well as the high phenolic content of these selected plantsrevealed that components responsible for the antioxidant activity could be due to the presence of xanthone or some polyphenolic compound.Standardization of the spectrophotometric method for the detection of xanthone will be helpful for quick and cost-effective analysis without using modern techniques.Although further research work for confirmation of xanthone or some other polyphenolic compound using TLC, HPTLC, and FTIR techniques is in progress.

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