Journal of Tropical Biology and Conservation 20: 81–95, 2023 ISSN 1823-3902
E-ISSN 2550-1909

Research Article

Phenolic Content and Antioxidant Activity of Leaves and Rhizomes of *Etlingera coccinea* (Blume) S.Sakai and Nagam. (Zingiberaceae)

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Received 12 April 2023|Reviewed 13 July 2023|Accepted 01 August 2023|Published 15 October 2023
Doi: https://10.51200/jtbc.v20i.4648

ABSTRACT

Most of the Zingiberaceae species in the Philippines have been used as ethnomedicinal plants due to the benefits they possess. One of these species is *Etlingera coccinea* (Blume) S.Sakai and Nagam. (Zingiberaceae), a species with a variety of uses in Malaysia, Borneo, and Philippines. In this study, ethanolic extracts of the dry weight leaves and rhizomes of *E. coccinea* were used to determine its phenolic content and antioxidant activity. The total phenolic content and total antioxidant activity were determined using Folin-Ciocalteu and phosphomolybdenum methods, respectively. Data revealed that the total phenolic content of dry weight, expressed as milligram gallic acid equivalent per gram sample (mg GAE/g sample) recorded that the leaves (11.69 ± 0.47 mg GAE/g sample) have greater amount of phenolics than the rhizomes (0.58 ± 0.06 mg GAE/g sample). The total antioxidant activity (TAA), expressed as milligram ascorbic acid equivalent per gram sample (mg AAE/g sample), obtained higher activity in the leaves (12.76 ± 0.31 mg AAE/g sample) than the rhizomes (0.85 ± 0.12 mg AAE/g sample), and the reducing power, expressed as milligram gallic acid reducing power equivalent per gram sample (mg GRPE/g sample) also revealed higher activity for the leaves (9.37 ± 1.88 mg GRPE/g sample) compared to rhizomes (0.28 ± 0.07 mg GRPE/g sample). Based on the correlation analysis, a perfect positive linear relationship was observed among the TPC, TAA, and RP ($r=1$, $p<0.001$), which means that phenolic compounds significantly contribute to the antioxidant activities of the extracts of *E. coccinea*. These data imply that *E. coccinea* could be potentially used as a new source of natural antioxidant. Furthermore, this paper adds information on the habitat and ecology, phenology, and coloured photographs of this species for future related studies and conservation initiatives.

Keywords: ginger species; native species; total antioxidant activity; total phenolic content; reducing power

Introduction

Zingiberaceae is one the eight families in Order Zingiberales and is known as the ginger family. In the Philippines, members of this family have recently been the subject of scientific investigations from field expeditions (e.g., inventory, diversity, and assessment, etc.) to laboratory analyses (e.g., biochemical analyses). Determination of phenolic content and antioxidant activities of some ginger species, such as the works of Mabini & Barbosa (2018), Barbosa & Nueva (2019), Mendez et al. (2022, 2023) have been conducted in response to the perceived natural benefits the ginger species offer to humans as reported by local people living in the Mindanao Mountain ecosystems. It is imperative that several studies are conducted on this family in the country to further explore its potential and future use in medicines, condiments, and food (Mendez et al., 2023).

Phenolic compounds have recently received considerable attention because of their physiological functions including as antioxidants and free radical scavenging activities that are affected by quality and nutritional value (Govindarajah et al., 2017). The compounds stimulate the synthesis of endogenous antioxidant molecules in the cell (Côté et al., 2010). Phenolic compounds are important plant constituents with redox properties responsible for antioxidant activity (Soobarattee et al., 2005). Plant antioxidant molecules are mostly secondary metabolites (Maulana et al., 2019) and these plants produce these compounds as adaptive responses (Wang, 2012). Antioxidants stabilize or deactivate free radicals, often before that attack targets in biological cells (Nunes et al., 2012).

In Marilog District, the fruits of *E. coccinea* are eaten as a forest candy especially by children, and its rhizome and leaves are boiled to cure fever, cough, and muscle fatigue which were also done to other gingers in the area. In Sabah, Malaysia, *E. coccinea* has been used by various indigenous communities as a remedy for stomachache, food poisoning, and gastric problems (Poulsen, 2006). Its fruits are edible and the pith of the leafy shoot is used as a condiment (Lamb et al., 2013). The indigenous people especially the Kadazan-Dusun use this species to flavour local dishes and the *E. coccinea*-based products, such as ‘sambal tuhau’ (paste), ‘jeruk tuhau’ (pickles) and ‘serunding tuhau’ (floss) have been commercially produced for the local market (Jualang et al., 2015). However, the Philippine materials of *E. coccinea* have not been studied in any laboratory research in the country, and thus, this paper makes it the first study to evaluate its phenolic content and antioxidant activities in the Philippines.

Methodology

Entry Protocol

As a courtesy to the Barangay officials and local people in Brgy. Datu Salumay in Marilog District, Davao, Southern Philippines, a prior informed consent and a formal letter were personally submitted by the author at the Barangay Captain's office before the collection of samples at Mt. Malambo. The gratuitous permit of Dr. Victor B. Amoroso issued by the Department of Environment and Natural Resources (DENR) – Region XI was used as the collection permit in this study.

Place and Duration of the Study

The collection of specimens was carried out at the base of Mt. Malambo (7°22'40'' N 125°19'38" E) in Marilog District, Davao City in September 2021 – January 2022 at an elevationof 1090 masl.. Samples and voucher specimens were collected on September 25, 2021 and were brought to Central Mindanao University, Musuan, Bukidnon for processing. The determination of total phenolic content (TPC), total antioxidant activity (TAA), and reducing power (RP) of dry weight leaves and rhizomes of *E. coccinea* was conducted at the Natural Science Laboratory of Natural Science Research Center (NSRC) in Central Mindanao University after necessary permits were obtained from the authorities concerned.

Sample Collection, Preparation and Extraction

Prior to the collection of samples for laboratory analyses, ecological notes were obtained in the field by taking notes of the habitat of the populations and its habit following the method of Mendez et al. (2017), Acma & Mendez (2018), Mendez & Acma (2018). Plant associates near the populations of *E. coccinea* were also taken using field notebooks and pencil.

The leaves and rhizomes of *E. coccinea* were collected and placed separately inside labelled plastic bags with wet tissue paper to prevent dehydration (**Figure 1).** These samples were brought to Central Mindanao University for further processing. Leaf and rhizome samples were washed, and the earthy matters were removed prior to air-drying. Dried samples were powdered and stored until these were used. On the other hand, the vegetative and reproductive parts of *E. coccinea* were collected for voucher purposes. Flowers and fruits were preserved in 70% ethanol as spirit/pickled collection, and the herbarium specimens were dried. The herbarium specimens were then deposited at the Central Mindanao University Herbarium (CMUH).

Figure 1. Specimens of *Etlingera coccinea* (Blume) S.Sakai and Nagam. **A)** Fresh rhizome sample; **B)** Fresh leaf sample. Scale bar: A & B = 3 cm. *Photographs: N.P. Mendez (A&B).*

Extracts were prepared following the method of Padda & Picha (2008) with some modifications. The dried leaf and rhizome specimens were homogenized using a household blender and weighed in analytical balance (Mettler). The dried leaf and rhizome powder were extracted with absolute ethanol with a ratio of 1 g: 25.0 mL at room temperature (24°C). The mixtures were shaken for 1 hour in an orbital shaker (Premiere, USA) at 300 rpm and centrifuged (Lab Kits) at 5,000 rpm for 5 minutes. The resulting supernatants were collected in separate 15 mL conical tubes. Extracts were stored at 2–8°C and used in the succeeding analyses. The ethanolic leaf and rhizome extracts of *E. coccinea* were subjected for the total phenolic content, total antioxidant analysis, and reducing power determination in 96-well plate format colorimetric assays.

Total Phenolic Content

The TPC of the extracts was determined using the method described by Ainsworth & Gillespie (2007) with some modifications. Briefly, 200 µL of the extracts and 200 µL of 10% Folin-Ciocalteu reagent were transferred in a 2-mL centrifuge tube. The reaction mixture was set aside for 5 minutes and added with 800 µL of 10% sodium carbonate. The mixture was set aside at room temperature for 30 minutes and centrifuged at 11,000 rpm for 3 minutes. A 200 µL of the resulting solution was then transferred to the assigned microplate wells. The absorbance was determined at 750 nm against a blank (ethanol) using a microplate reader (Molecular Devices Spectramax® 250). Gallic acid (100 ppm) was used as the standard solution with concentration range of 0–200 ppm. A calibration curve was constructed by using the absorbance values obtained at various concentration of gallic acid. The TPC was determined and expressed as milligram gallic acid equivalent per gram sample (mg GAE/g sample) by

interpolating sample absorbance against the standard calibration curve using the formula below:

$$\text{Total Phenolic Content} = \left(\frac{\text{mg}}{\text{g sample}}\right) = \frac{\text{A}}{\text{B}}$$

where:

A = gallic acid concentration of the sample solution determined from the calibration curve (mg GAE/L)

B = the concentration of test solution (g/L, gram dried sample per L solution)

Total Antioxidant Activity

The TAA was determined using phosphomolybdenum method described by Prieto et al. (1999) with slight modifications. Briefly, 50 µL of extracts were placed in centrifuge tubes and diluted with 200 µL (1:1 ethanol: water). The solution was added with 600 µL of reagent solution (prepared by mixing equal amounts of 0.6 M sulfuric acid, 28 mM sodium phosphate, and 4 mM ammonium molybdate) and incubated at 95°C for 90 minutes. The samples were allowed to cool down at room temperature (24°C) and centrifuged at 11,000 rpm for 3 minutes. The absorbance of the supernatant was measured at 695 nm against ethanol using a microplate reader (Molecular Devices Spectramax® 250). Calibration curve was also prepared using 0–300 ppm ascorbic acid as standard. TAA was determined and expressed as milligram ascorbic acid equivalent per gram sample (mg AAE/g sample) by interpolating sample absorbance against the standard curve. The TAA was calculated using the following equation:

$$\text{Total Antioxidant Content} = \left(\frac{\text{mg}}{\text{g sample}}\right) = \frac{\text{A}}{\text{B}}$$

Where:

A = ascorbic acid concentration of the solution determined from the calibration curve (mg AAE/L)

B = concentration of the test solution (g/L, gram dried sample per L solution)

Reducing Power

The reducing power was determined by adapting the method described by Murugan & Iyer (2012) with some modifications. In a centrifuge tube containing 1 mL of extracts, 200 µL of 0.2 M phosphate buffer (pH 6.6) and 200 µL of 1% (w/v) solution of potassium ferricyanide were added. The mixture was incubated at 50°C for 30 minutes. After cooling to room temperature (24°C),

200 µL of 1% (w/v) trichloroacetic acid was added. The mixture was centrifuged for 3 minutes at 11,000 rpm. An aliquot of 200 µL of the supernatant was transferred to a 96-well plate and 20 µL of 1% (w/v) solution of ferric chloride was added. The absorbance was measured at 620 nm against a blank (ethanol) using a microplate spectrophotometer (Molecular Devices Spectramax® 250). Standard gallic acid with concentrations ranging from 0–300 ppm was used to establish a calibration curve. Sample concentration was determined by interpolating sample absorbance against the standard curve. The reducing power, expressed as milligram gallic acid reducing power equivalent per gram sample (mg GRPE/g sample) was calculated as follows:

$$\text{Reducing Power} = \left(\frac{\text{mg}}{\text{g sample}}\right) = \frac{\text{A}}{\text{B}}$$

where:
A = gallic acid concentration of the test solution determined from the calibration curve (mg GRPE/L)
B = concentration of the test solution (g/L, gram dried sample per L solution)

Statistical Analysis

The TPC, TAA, and RP analyses were done in triplicates, and the determination for each assay was carried out in three trials per replicate. Correlations among the TPC, TAA, and RP were obtained using Pearson's correlation coeeficient (*r*) at a 0.001 level of significance. The results of absorption measurement were separately put into Microsoft Excel to obtain a calibration curve of standard gallic acid solution (TPC and RP) and ascorbic acid solution (TAA) in the form of a graph of concentration versus absorption curve. All results obtained in the study were expressed as mean values ± SD (standard deviation).

Results and Discussion

Etlingera coccinea (Blume) S. Sakai & Nagam. (**Figure 2**)

Habitat and Ecology: On 16 July 2017, three populations of *E. coccinea* were found along the roadside at the foot of Mt. Malambo fronting the Bemwa farm in Marilog District, Davao City, Philippines. During a revisit to the area on 21 September 2021 to collect samples for biochemical analyses and collection of voucher specimens, seven populations were recorded and most of the individuals were fertile. The populations of *E. coccinea* naturally grow in a partial shade canopy in wet, slope lowland area, along the roadside where there is minimal contact with sunlight at an elevation of 1090 masl. Inflorescence of *E. coccinea*

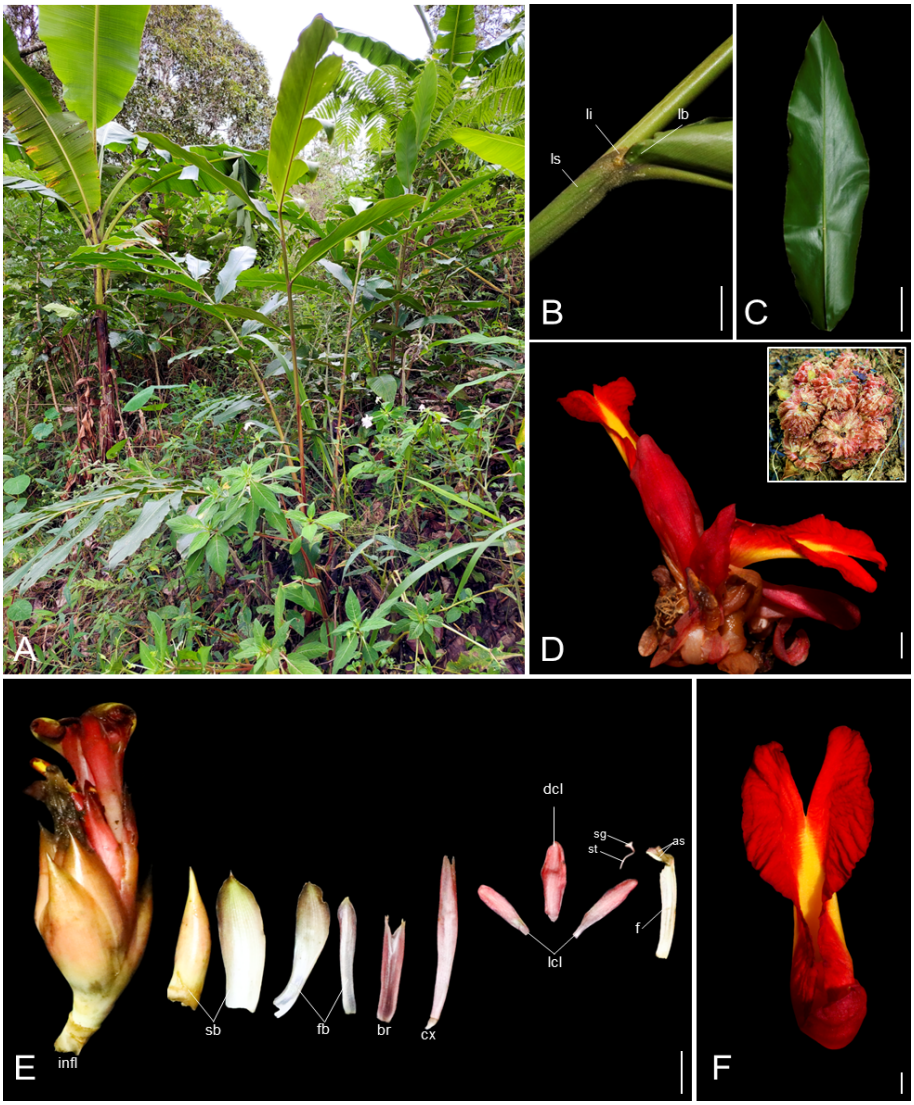

Figure 2. *Etlingera coccinea* (Blume) S.Sakai and Nagam. **A)** Habit; **B)** Leaf sheath (ls), ligule (li), and leaf base (lb); **C)** Lamina (adaxial surface); **D)** Inflorescence (inset: infructescence); **E)** Floral parts (infl – inflorescence, sb – sterile bracts, fb – fertile bracts, br – bracteole, cx – calyx, dcl –dorsal corolla lobe, lcl – lateral corolla lobes, sg – stigma, st – style, as – anther sacs, f – filament); **F)** Labellum (anterior view). Scale bars: 3 cm – B & C; 1 cm – D; 2 cm – E; 3 cm – F. *Photographs: N.P. Mendez (A-C and E), R.V.A. Docot (D & F).*

appeared at the ground level and showed four floral morphs during its anthesis. The species was found growing along with other Zingiberaceae species, such as *E. pubimarginata* (Elmer) A.D.Poulsen and *Zingiber* sp. (Zingiberaceae) and

associated with *Alocasia heterophylla* (C.Presl) Merr. (Araceae), *Musa textilis* Nées (Musaceae), *Mimosa pudica* L. (Fabaceae), *Impatiens platypetala* Lindl. (Balsaminaceae), and *Angiopteris evecta* (G.Forst.) Hoffm. (Marattiaceae).

Phenology: Flowering usually occurs from July to September, fruits not observed during the collection of samples.

Total Phenolic Content

The results of the TPC of the extracts were derived from the regression equation of a calibration curve (y = 0.0546 x 0.0658, R^2 = 0.9946) of gallic acid (0–200 mg/mL). The ethanolic extracts of *E. coccinea* revealed that the leaves (11.69 ± 0.47 mg GAE/g sample) exhibited higher phenolic content than the rhizomes (0.58 ± 0.06 mg GAE/g sample) (**Table 1**).

Table 1. Mean total phenolic content extraction yield of leaves and rhizomes of *E. coccinea*.

Plant parts	mg GAE/g sample
Leaves	11.69 ± 0.47
Rhizomes	0.58 ± 0.06

The values of phenolic content in this study varied slightly compared to the studies on other ginger species in the Philippines, *viz.*, Mabini & Barbosa (2018), Barbosa & Nueva (2019), Mendez et al. (2002, 2003) and previous study on phenolic content and antioxidant activity of *E. coccinea* collected from Brunei Darussalam by Shahid-Ud-Daula et al. (2015). The mean value of TPC in leaves (11.69 ± 0.47 mg GAE/g sample) in this study is relatively higher than the studies of Mabini & Barbosa (2018) with 0.55 mg GAE/g sample on methanolic extracts of *Etlingera philippinensis* (Ridl.) R.M.Sm., Barbosa & Nueva (2019) with 1.67 mg GAE/g sample on methanolic extracts of *Hornstedtia conoidea* Ridl., and Mendez et al. (2023) with 7.21 ± 0.33 mg GAE/g sample on ethanolic extracts of *E. philippinensis*. However, it is lower than the studies of Shahid-Ud-Daula et al. (2015) 13.49 ± 0.26 mg GAE/g sample on methanolic extracts of *E. coccinea*, Mendez et al. (2022) with 27.25 ± 0.72 mg GAE/g sample on ethanolic extracts of *E. pubimarginata* (Elmer) A.D.Poulsen and Mendez et al. (2023) with 13.20 ± 0.35 mg GAE/g sample on ethanolic extracts of *E. fimbriobracteata* (K.Schum.) R.M.Sm.

The mean value of TPC in rhizomes (0.58 ± 0.06mg GAE/g sample) of *E. coccinea* is higher than the studies of Mabini & Barbosa (2018) and Mendez et al. (2023) on *E. philippinensis* with 0.35 mg GAE/g sample and 0.46 ± 0.30 mg GAE/g sample. But this value is lower than the studies of Shahid-Ud-Daula et al. (2015)

with 7.94 ± 0.01 mg GAE/g sample, Barbosa & Nueva (2019) with 1.28 mg GAE/g sample on *H. conoidea*, Mendez et al. (2022) with 0.76 ± 0.11 mg GAE/g sample on *E. pubimarginata*, and Mendez et al. (2023) with 1.44 ± 0.04 mg GAE/g sample on *E. fimbriobracteata*. These slight variations in the mean values of TPC might be due to the presence of different amounts of sugars, carotenoids, ascorbic acid, duration, geographical variation or methods of extraction, which may alter the amount of phenolics (Burri et al., 2017).

Phenolic compounds contribute to antioxidant activity due to the arrangement of functional groups (hydroxyl) about its nuclear structure for hydrogen donation in order to stabilize radical molecules (Soobarattee et al., 2008; Alam et al., 2018). The total phenolic compounds also play an effective role in stabilizing lipid peroxidation (Yen et al., 1993). It is worth to note that the result of TPC depends on the type of solvent used (Cesoniene et al., 2012), degree of polymeration of phenolics, interaction of phenolics with other food constituents and formation of insoluble complex (Galvez et al., 2005).

Ethanol was used in this study since phenolic compounds are more soluble in polar organic solvents due to the presence of a hydroxyl group (Wang & Weller, 2005). The quantification of TPC in ethanolic extracts of leaves and rhizomes of *E. coccinea* which was determined by employing Folin-Ciocalteu method, is convenient, simple, and reproducible (Cirilo & Lemma, 2012; Danciu et al., 2015). The mechanism involved in this method is electron transfer in alkaline medium from phenolic compound to phosphomolybdic/phosphotungstic acid complexes to form blue complexes that are determined spectroscopically (Singleton et al., 1999).

Total Antioxidant Activity and Reducing Power

The results for the TAA of the ethanolic extracts of leaves and rhizomes of *E. coccinea* were calculated from a calibration graph which were linear over the calibration range with R^2 value of 0.9963 (y = 0.0149 x 0.0465) of L-ascorbic acid (0–100 mg/mL). The ethanolic extracts of *E. coccinea* revealed that the leaf extract (12.76 ± 0.31 mg AAE/g sample) had a higher activity compared to the rhizome extract (0.85 ± 0.12 mg AAE/g sample).

For the RP, results of ethanolic extracts of leaves and rhizomes of *E. coccinea* were derived from a calibration curve (y = 0.0505 x 0.5037, R^2 = 0.9975) of gallic acid (0–1000 mg/mL). The ethanolic extracts of *E. coccinea* revealed that the leaves (9.37 ± 1.88 mg GRPE/g sample) had a higher activity compared to the rhizomes (0.28 ± 0.07 mg GRPE/g sample) (**Table 2**).

Table 2. Mean total antioxidant activity and reducing power extraction yield of leaves and rhizomes of *E. coccinea*.

Plant parts	TAA (mg AAE/g dried sample)	Reducing Power (mg GRPE/g dried sample)
Leaves	12.76 ± 0.31	9.37 ± 1.88
Rhizomes	0.85 ± 0.12	0.28 ± 0.07

The mean value of TAA of the leaves (12.76 ± 0.31 mg AAE/g sample) of *E. coccinea* had a higher activity compared to the studies of Mabini & Barbosa (2018) with 0.79 mg AAE/g sample on *E. philippinensis*, Barbosa & Nueva (2019) with 4.67 mg AAE/g sample on *H. conoidea*, and Mendez et al. (2023) with 12.69 ± 0.36 on *E. fimbriobracteata*, but lower in the studies of Mendez et al. (2022) with 34.83 ± 0.49 mg AAE/g sample on *E. fimbriobracteata* and Mendez et al. (2023) with 12.69 ± 0.36 mg AAE /g sample on *E. philippinensis*. For the mean value of TAA of the rhizomes (0.85 ± 0.12 mg AAE /g sample), this study obtained higher activity than the studies of Mabini & Barbosa (2018) with 0.55 mg AAE /g sample on *E. philippinensis*, and Mendez et al. (2022) with 1.82 ± 0.09 mg AAE/g sample on *E. pubimarginata*, but lower than the study of Barbosa & Nueva (2019) with 2.03 mg AAE /g sample on *H. conoidea*, Mendez et al. (2023) with 1.82 ± 0.01 mg AAE /g sample and 1.38 ± 0.07 mg AAE /g sample on *E. fimbriobracteata* and *E. philippinensis*, respectively.

The reducing power of the leaves of *E. coccinea* (9.37 ± 1.88 mg GRPE/g sample) revealed to have a higher activity than the study of Mendez et al. (2022) with 24.83 ± 2.99 mg GRPE/g sample in *E. pubimarginata* and Mendez et al. (2023) with 7.53 ±0.80 mg GRPE/g sample in *E. philippinensis*, but lower in *E. fimbriobracteata* with 10.16 ± 2.18 mg GRPE/g sample in *E. fimbriobracteata*. In terms of the rhizome ethanolic extracts, *E. coccinea* (0.28 ± 0.07 mg GRPE/g sample) is lower compared to the study of Mendez et al. (2022) with 0.33 ± 0.10 mg GRPE/g sample on *E. pubimarginata* and Mendez et al. (2023) with 0.97 ± 0.18 mg GRPE/g sample on *E. fimbriobracteata*, but lower in *E. philippinensis* with 0.09 ± 0.09 in the study of Mendez et al. (2023).

The last several decades have seen increased research attention of potential phytochemicals from plants for therapeutic uses because many phytochemicals have been demonstrated to have antioxidant activities (Kairupan et al., 2019). The TAA of plant extracts might be due to the presence of polyphenols, which act as reductones by donating electrons and reacting with free radicals converting them to a more stable product and subsequently terminating free radical chain reaction (Gordon, 1990). Populations of *E. coccinea* were found at

the montane forests of Mt. Malambo in partially open forest, which supported Frankel & Berenbaum (1999) that foliage of tropical forest plants produced more antioxidants when exposed to elevated light conditions than the other plant parts. This answers why the leaves obtained the higher phenolic content and antioxidant activities than the rhizomes.

Correlation Analyses

The contribution of the phenolic compounds in the ethanolic extracts of the *E. coccinea* to the antioxidant activity was determined by Pearson's correlation coefficient. A perfect positive linear relationship was observed between TPC and TAA, TPC and RP, and TAA and RP ($r = 1$, $p < 0.001$) (**Table 3**). By comparing the correlation coefficients (R value), it is possible that phenolic compounds significantly contribute to the antioxidant activities of the plant's extracts.

Table 3. Pearson's correlation coefficients between TPC and TAA, TPC and RP, and TAA and RP.

Plant parts	Total phenolic content	Total antioxidant activity	Reducing Power
Total phenolic content	1	1**	1**
Total antioxidant activity	1**	1	1**
Reducing Power	1**	1**	1

**Correlation is significant at 0.001 level

The strong correlation between the results using the two methods of measuring TPC and antioxidant activity showed that phenol compounds largely contribute to the antioxidant activities of *E. coccinea* and, therefore, could play an important role in the beneficial effects of the plant. Several reports also showed a close relationship between TPC and antioxidant activity, since phenolic compounds serve as hydrogen-donating agents (Li and Jiang, 2007; Prasad et al., 2005; Yang et al., 2014). With the data of this paper, this study supported the claim of Stanković (2011) that the higher the phenolic content, the higher the antioxidant activity.

Conclusions and Recommendations

Higher TPC value was recorded higher in leaves (11.69 ± 0.47 mg GAE/g sample) than the rhizomes (0.58 ± 0.06 mg GAE/g sample). For the TAA, the leaves (12.76 ± 0.31 mg AAE/g sample) had a higher activity than rhizomes (0.85 ± 0.12 mg AAE/g sample) and the RP had a higher activity in the leaves (9.37 ± 1.88 mg GRPE/g sample) than the rhizomes (0.28 ± 0.07 mg GRPE/g sample). Overall, the leaves contributed higher phenolic content and antioxidant activity compared to rhizomes. Based on the correlation analysis, a perfect positive

linear relationship was observed among TPC, TAA, and RP. These imply that the high content of phenolic compounds contributes to the antioxidant activity of extracts of *E. coccinea*. As this is the first report of the phenolic content and antioxidant activity of *E. coccinea* using Philippine materials, this calls for thorough phytochemical analyses to be done to identify the active phenolic and antioxidant components of this ginger species.

Acknowledgments

The author would like to thank the Department of Environment and Natural Resources (DENR) – Region XI for the issuance of a gratuitous permit to Dr. Victor B. Amoroso which was used as the collection permit of this study and the barangay officials of Datu Salumay in Marilog District for allowing NPM to conduct a fieldwork at Mt. Malambo. Due acknowledgements are also given to Rainear A. Mendez and Angie Rose Villafranca-Tuba for helping in the conduct of the study and Romeo M. Tubongbanua, Jr. for the help in the collection of the specimens in field. The author is also thankful to the concerned authorities of CMU NSRC Natural Science Laboratory for the analysis of samples.

References

Acma FM, Mendez NP, Amoroso VB. 2020. Zingiberaceae of Marilog Forest Reserve, Southern Philippines: Its Richness and Endemism. *Asian Journal of Biodiversity, **11*** (1) : 76–90. doi: 10.7828/ajob.v11i1.1388.

Acma FM, Mendez NP. 2018. Pollen Morphology and Pollen Elemental Composition of selected Philippine Native Gingers in Tribe Alpinieae (Alpinioideae: Zingiberaceae). *Biological Forum – An International Journal, **10*** (1): 1–10.

Ainsworth EA, Gillespie KM. 2007. Estimation of total phenolic content and other oxidation substrates in pant tissues using Folin-Ciocalteu reagent. *Nature Publishing Group,* **2**: 875–877. doi: 10.1038/nprot.2007.102.

Alam MA, Syazwanie NF, Mahmod NH, Badaluddin NA, Mustafa KA, Alias N, Aslani F, Prodhan MA. 2018. Evaluation of antioxidant compounds, antioxidant activities and capsaicinoid compounds of Chili (*Capsicum* sp.) germplasms available in Malaysia. *Journal of Applied Research on Medicinal and Aromatic Plants,* **9**: 46–54. doi: 10.1016/j.jarmap.2018.02.001.

Barbosa GB, Nueva MCY. 2019. Antioxidant Activity and Phenolic Content of *Hornstedtia conoidea* (Zingiberaceae). *Asian Journal of Biological and Life Sciences, **8*** (1): 1–7. doi: 10.5530/ajbls.2019.8.1.

Burri SCM, Ekholm A, Hákansson Ā, Tornberg E, Rumpunen K. 2017. Antioxidant capacity and major phenol compounds of horticultural plant materials not usually used. *Journal of Functional Foods, **38***: 119–127. doi: 10106/j.jff.2017.09.003.

Cirillo G, Lemma F. 2012. *Antioxidant Polymers: Synthesis, Properties, and Applications.* Calabria: John Wiley & Sons. 510.

Côté J, Caillet S, Doyon G, Sylvain J-F, Lacroix M. 2010. Bioactive compounds in

cranberries and their biological properties. *Crit. Rev. Food Sci. Nutr.,* **50:** 666–679. doi: 10.1080/10408390903044107.

Cesoniene L, Daubaras R, Viskelis P, Sarkinas A. (2012). Determination of the total phenolic and anthocyanin contents and antimicrobial activity of Viburnum opulus Fruit juice. *Plant Foods for Human Nutrition* **67:** 256–261.

Danciu C, Vlaia L, Fetea F, Hancianu M, Coricovac DE, Ciurlea S A, Şoica CM, Marincu I, Vlaia V, Dehelean CA, Trandafirescu C. 2015. Evaluation of phenolic profile, antioxidant and anticancer potential of two main representants of Zingiberaceae family against B164A5 murine melanoma cells. *Biological Research, **48*** (1): 1–9. doi: 10.1186/0717-6287-48-1.

Frankel S, Berenbaum M. 1999. Effects of light regime on antioxidant content of foliage in a tropical forest community. *Biotropica, **31*** (3): 422–429. DOI: 10.1111/j.1744-7429.1999.tb00384.x.

Galvez CJ, Martin-Cordero P, Houghton AM. 2005. Antioxidant activity of methanol extracts obtained from plantago species. *Journal of Agricultural and Food Chemistry, **53:*** 1927–1933.

Gordon M. 1990. The mechanism of antioxidant action *in Vitro*. In Hudson BJF, ed. *Food. Antioxidants*, Springer, Berlin. 1–18. doi: 10.1007/978-94-009-0753-9_1.

Govindarajan R, Singh DP, Rawat AKS. 2007. High-performance liquid chromatographic method for the quantification of phenolics in „Chyavanprash" a potent Ayurvedic drug. *Journal of Pharmaceutical Biomedical Analysis, **43:*** 527–532.

Jualang AG, Nurul Humairah TA, Devina D, Hartinie M. 2015. *In vitro* shoot regeneration from rhizome bud of native ginger in Borneo, *Etlingera coccinea*. *Journal of Tropical Plant Physiology, **7***: 36–46.

Kairupan CF, Mantiri FR, Rumende RRH. 2019. Phytochemical Screening and Antioxidant Activity of Ethanol Extract of Leilem (*Clerodendrum minahassae* Teijsm. & Binn) as an Antihyperlipidemic and Antiatherosclerotic Agent. *IOP Conf. Series: Earth and Environmental Science, **217:*** 1–7. doi:10.1088/1755-1315/217/1/012016.

Lamb A, Goblik J, Ardiyani M, Poulsen AD. 2013. *A Guide to Gingers of Borneo.* Natural History Publications (Borneo), 144.

Li J, Jiang Y. (2007). Litchi flavonoids: isolation, identification, and biological activity. *Molecule, **12*** (4): 745–758. DOI: 10.3390/12040745.

Mabini JM, Barbosa GB. 2018. Antioxidant activity and phenolic content of the leaves and rhizomes of *Etlingera philippinensis* (Zingiberaceae). *Bulletin of Environment, Pharmacology and Life Sciences, **7*** (9)**:** 39–44.

Maulana, TI, Falah S,Andrianto D. 2019. Total phenolic content, total flavonoid content, and antioxidant activity of water and ethanol extract from Surian (*Toona sinensis*) leaves. In *IOP Conf. Series: Earth and Environmental Science* **299** (2019) 012021. doi:10.1088/1755-1315/299/1/012021.

Mendez NP, Mendez RA, Villafranca-Tuba ARA, Tubongbanua Jr. RM, Acma FM. 2023. Morphology, Phenolic Content, and Antioxidant Activity of *Etlingera fimbriobracteata* (K.Schum.) R.M.Sm. and *E. philippinensis* (Ridl.) R.M.Sm. (Zingiberaceae). *Philippine Journal of Science, **152*** (2): 653–664.

Mendez NP, Lumista HP, Lituañas CRM, Mendez RA, Villafranca-Tuba ARA, Acma FM. 2022. Phenolic Content and Antioxidant Activity of ethanol extracts of *Etlingera pubimarginata* (Zingiberaceae), including notes on its

Morphology. *Journal of Tropical Biology and Conservation, **19:*** 77–92. doi: https://doi.org/10.51200/jtbc.v19i.

Mendez NP, Acma FM. 2018. *In vitro* Studies on Pollen Viability, Pollen Germination and Pollen Tube Growth of *Hornstedtia conoidea* Ridl. – a Philippine endemic ginger species. *Journal of Tropical Life Science, **8*** (3): 279–288. doi: http://dx.doi.org/10.11594/jtls.08.03.10.

Mendez NP, Porquis HC, Sinamban EB, Acma FM. 2017. Comparative Pollen Viability and Pollen Tube Growth of two endemic Philippine *Etlingera* (Zingiberaceae, Alpinioideae). *Philippine Journal of Systematic Biology, **11 (2)**:* 1–9.

Murugan K, Iyer W. 2012. Antioxidant and antiproliferative activities of marine algae, *Gracilaria edulis* and *Enteromorpha lingulata* from Chenai coast. *International Journal of Cancer Research, 8* (1): 15–26. doi: 10.3923/ijcr.2012.15.26.

Padda MS, Picah DH. 2008. Quantification of phenolic acids and antioxidant activity in sweetpotato genotypes. *Scientia Horticulturae,* **119:** 17–20. doi: 10.1016/j.scientia.2008.07.008.

Pelser PB, Barcelona JF, Nickrent DL. (Eds.). 2011 onwards. *Co's Digital Flora of the Philippines.* Accessed at: www.philippineplants.org.

Poulsen AD. 2006. *Etlingera of Borneo.* Natural History Publications. Kota Kinabalu, Sabah. http://www.nhpborneo.com/book.

Prasad NK, Divakar S, Shivamurthy GR, Aradhya S. 2005. Isolation of a free radical scav¬enging antioxidant from water spinach (*Ipomoea aquatica* Forsk.) *Journal of the Science of Food and Agriculture, **85*** (9): 1461–1468. DOI: 10.1002/jsfa.2125

Prieto P, Pineda M, Aguilar M. 1999. Spectrophotometric quantitation of antioxidant capacity through the formation of a phosphomolybdenum complex: Specific application to the determination of vitamin E. *Analytical Biochemistry, **269*** (2): 337–341. doi: 10.1006/abio.1999.4019.

Shahid-Ud-Daula AFM, Kamariah AS, Lim LBL, Ahmad N. 2015. Phytochemical Screening, Antioxidant, and Antimicrobial Activities of Leaves, Stems, and Rhizomes of *Etlingera coccinea* (Blume) S.Sakai & Nagam. *International Journal of Pharmacognosy and Phytochemical Research, **7*** (5): 873–883.

Singleton VL, Orthofer R, Lamuela-Raventós RM. 1999. Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin-Ciocalteu reagent. *Methods in Enzymology, **299:*** 152–178. doi: 10.1016/S0076-6879(99)99017-1.

Soobrattee MA, Neergheen VS, Luximon-Ramma A, Aruoma OI, Bahorum OT. 2005. Phenolics as potential antioxidant therapeutic agents: mechanism and actions. *Mutation Research - Fundamental and Molecular Mechanisms, **579*** (1–2): 200–213. doi: 10.1016/j.mrfmmm.2005.03.023.

Soobrattee MA, Bahorun T, Neergheen VS, Googoolye K, Aruoma OI. 2008. Assessment of the content of phenolics and antioxidant actions of the Rubiaceae, Ebenaceae, Celastraceae, Erythroxylaceae and Sterculaceae families of Mauritian endemic plants. *Toxicology in Vitro, **22*** (1): 45–56. doi: 10.1016/j.tiv.2007.07.012.

Stanković MS. 2011. Total Phenolic Content, Flavonoid Concentration and Antioxidant Activity of *Marrubium peregrinum* L. extracts. *Kragujevac Journal of Science, **33:*** 63–72.

Wang DS. 2012. Secondary Metabolites from Plants. 1–52 pp.

Wang L, Weller CL. 2006. Recent advances in extraction of nutraceuticals from plants. *Trends Food Sci. Technol.*, ***17:*** 300–312. doi: 10.1016/j.tifs.2005.12.004.

Yang X, Yan F, Huang S, Fu C. 2014. Antioxidant activities of fractions from longan pericarps. *Food Science Technology*, ***34*** (2): 341–345. DOI: 10.1590/S0101-20612014005000034.

Yen GC, Duh PD, Tsai CL. 1993. Relationship between antioxidant activity and maturity of peanut hull. *Journal of Agricultural and Food Chemistry*, ***41:*** 67–70. doi: 10.1021/jf00025a015.