

Rapid Communication

Antioxidant and tyrosinase inhibition properties of leaves and rhizomes of ginger species

E.W.C. Chan, Y.Y. Lim^{*}, L.F. Wong, F.S. Lianto, S.K. Wong,
K.K. Lim, C.E. Joe, T.Y. Lim

School of Arts and Sciences, Monash University Sunway Campus, Bandar Sunway, 46150 Petaling Jaya, Selangor, Malaysia

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Abstract

Total phenolic content (TPC) and ascorbic acid equivalent antioxidant capacity (AEAC) of leaves of 26 ginger species belonging to nine genera and three tribes were screened. For 14 species, TPC and AEAC of rhizomes were also assessed. Ferrous ion-chelating (FIC) abilities of leaves and rhizomes of eight species were compared. Leaves of five species of *Etilingera* were analysed for tyrosinase inhibition activity. Of the 26 species, leaves of *Etilingera* species had the highest TPC and AEAC. Eleven of the 14 species had significantly higher TPC and/or AEAC in leaves than in rhizomes. Values of leaves of *Etilingera elatior* and *Etilingera maingayi* were seven to eight times higher than those of rhizomes. In terms of FIC ability, six of the eight species clearly showed higher values in leaves than in rhizomes. The most outstanding was the FIC value of *Alpinia galanga* leaves which was more than 20 times higher than that of rhizomes. Of the five species of *Etilingera*, leaves of *E. elatior* displayed the strongest tyrosinase inhibition activity, followed by leaves of *Etilingera fulgens* and *E. maingayi*. Values of their inhibition activity were significantly higher than or comparable to the positive control. Besides promising tyrosinase inhibition ability, leaves of these three *Etilingera* species also have high antioxidant activity and antibacterial properties.

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Keywords: Zingiberaceae; Leaves; Rhizomes; Total phenolic content; Antioxidant activity; Tyrosinase inhibition activity

1. Introduction

Rhizomes of ginger plants (family Zingiberaceae) have been widely used as spices or condiments (Larsen, Ibrahim, Khaw, & Saw, 1999). Rhizomes are eaten raw or cooked as vegetables and used for flavouring food. Major commercially cultivated species are *Zingiber officinale*, *Curcuma longa*, and *Alpinia galanga*. As traditional medicine, rhizomes of ginger plants are consumed by women during ailment, illness and confinement. Rhizomes are also taken as carminatives for relieving flatulence.

Leaves of ginger plants have also been used for food flavouring and in traditional medicine. In Malaysia, leaves of *C. longa* are used to wrap fish before steaming or baking

(Larsen et al., 1999). Leaves of *Kaempferia galanga* and *C. longa* are ingredients of curries. Some tribal natives in Malaysia flavour their wild meat and fish dishes with leaves of *Elettariopsis slahmong* (Lim, 2003). In Thailand, its leaves are eaten as salad. Despite their repulsive stinkbug odour, leaves of *E. slahmong* are considered a delicacy. Traditionally, leaves of *Elettariopsis latiflora* have been used to relieve flatulence, to improve appetite and as an antidote to poisons. In Okinawa, Japan, leaves of *Alpinia zerumbet* are sold as herbal tea, and are commonly used to flavour noodles and to wrap rice cakes. The hypotensive, diuretic, and anti-ulcerogenic properties of tea from *A. zerumbet* leaves have been reported (Mpalantinos, de Moura, Parente, & Kuster, 1998). Leaves of *Etilingera elatior*, mixed with other aromatic herbs, are used by *post-partum* women for bathing to remove body odour (Ibrahim & Setyowati, 1999). They are also used for cleaning wounds. Leaves of

^{*} Corresponding author. Tel.: +60 3 55146103; fax: +60 3 55146099.
E-mail address: Lim.Yau.Yan@artsci.monash.edu.my (Y.Y. Lim).

Kaempferia rotunda and *K. galanga* are eaten fresh or cooked as vegetables, and used as cosmetic powder and as food flavouring agents (Ibrahim, 1999). In Peninsular Malaysia, boiled leaves of *Hedychium* species are eaten for indigestion (Ibrahim, 2001). Leaves are sometimes eaten with betel nut to ease abdominal pain. In Thailand, boiled leaves of *Hedychium coronarium* are applied to relieve stiff and sore joints.

Past studies on the antioxidant properties of ginger species were confined to rhizomes (Habsah et al., 2000; Jitoe et al., 1992; Zaeoung, Plubrukarn, & Keawpradub, 2005). Rhizomes of gingers have been reported to have tyrosinase inhibition properties (Lee, Kim, Kim, Heo, & Kim, 1997). Skin-lightening cosmeceutical products were recently developed from rhizomes of gingers (Rozanida, Nurul Izza, Mohd Helme, & Zanariah, 2006). Although leaves of ginger species have been used for food flavouring and in traditional medicine, little research has been done on their antioxidant and tyrosinase inhibition properties.

In our present study, phenolic contents and radical-scavenging activities of leaves of 26 ginger species were screened. For 14 species, antioxidant properties of rhizomes were assessed. For eight species, metal ion-chelating abilities of leaves and rhizomes were also compared. Leaves of five species of *Etingera* were analysed for tyrosinase inhibition activity. This study represents the most comprehensive study, where antioxidant properties of leaves and rhizomes of ginger species were systematically compared, and tyrosinase inhibition properties of leaves of *Etingera* species were analysed.

2. Materials and methods

2.1. Plant materials

Locations where species were sampled for leaves and rhizomes are listed in Table 1. Voucher specimens of ginger plants studied were deposited in the herbaria of the Forest Research Institute Malaysia (FRIM) and Monash University Sunway Campus (MUSC), Malaysia.

2.2. Chemicals and instruments

Folin–Ciocalteu's phenol reagent (Fluka, 2N), gallic acid (Fluka, 98%), and anhydrous sodium carbonate (Fluka, 99%) were used for TPC analysis. 1,1-Diphenyl-2-picrylhydrazyl (Sigma, 90%) was used for DPPH radical-scavenging assay. Ferrozine (Acros Organics, 98%) and ferrous sulphate heptahydrate (HmbG chemicals) were used for FIC assay. L-DOPA (Sigma), mushroom tyrosinase (Sigma), and DMSO (Fisher Scientific) were used for assessing tyrosinase inhibition. Absorbance was measured with an Anthelie Advanced 5 Secoman UV–vis spectrophotometer for TPC and antioxidant activity, and with a BIOTEK PowerWave XS Microplate scanning spectrophotometer for tyrosinase inhibition activity.

Table 1
Locations of sampling leaves and rhizomes of ginger species

Species	Tribe	Location of sampling
<i>Alpinia galanga</i>	Alpineae	Bukit Maluri, Kepong, KL
<i>A. malaccensis</i>		FRIM, Kepong, Selangor
<i>A. purpurata</i>		SUC, Sunway, Selangor
<i>A. zerumbet</i>		Janda Baik, Pahang
<i>A. zerumbet</i> 'Variegata'		FRIM, Kepong, Selangor
<i>Boesenbergia rotunda</i>	Hedychieae	Sungai Buluh, Selangor
<i>Curcuma aeruginosa</i>	Hedychieae	Damansara Utama, Selangor
<i>C. longa</i>		FRIM, Kepong, Selangor
<i>C. mangga</i>		Damansara Utama, Selangor
<i>C. zanthorrhiza</i>		Damansara Utama, Selangor
<i>Elettariopsis latiflora</i>	Alpineae	FRIM, Kepong, Selangor
<i>E. slahmong</i>		FRIM, Kepong, Selangor
<i>E. smithiae</i>		Janda Baik, Pahang
<i>Etingera elatior</i>	Alpineae	Janda Baik, Pahang
<i>E. fulgens</i>		Janda Baik, Pahang
<i>E. littoralis</i>		Genting Highlands, Pahang
<i>E. maingayi</i>		Janda Baik, Pahang
<i>E. rubrostriata</i>		Ulu Gombak, Selangor
<i>Hedychium coronarium</i>	Hedychieae	Lake Gardens, KL
<i>Kaempferia galanga</i>	Hedychieae	Damansara Utama, Selangor
<i>K. pulchra</i>		Sungai Buluh, Selangor
<i>K. rotunda</i>		Bukit Maluri, Kepong, KL
<i>Scaphochlamys kunstleri</i>	Hedychieae	FRIM, Kepong, Selangor
<i>Zingiber officinale</i>	Zingibereae	Bukit Maluri, Kepong, KL
<i>Z. ottensii</i>		Bukit Maluri, Kepong, KL
<i>Z. spectabile</i>		FRIM, Kepong, Selangor

Abbreviations: FRIM, Forest Research Institute Malaysia; SUC, Sunway University College; KL, Kuala Lumpur.

2.3. Extraction of plant samples

For antioxidant analysis, fresh leaves and rhizomes (1 g) were powdered with liquid nitrogen in a mortar and extracted using methanol (50 ml), with continuous swirling for 1 h at room temperature using an orbital shaker. Extracts were filtered under suction and stored at -20°C for further use. For tyrosinase inhibition, fresh leaves (10 g) were extracted three times using methanol (100 ml). Methanol was removed by drying at 35°C in a rotary evaporator prior to storage at -20°C . Analysis of methanol extracts for antioxidant and tyrosinase inhibition properties was done in triplicate.

2.4. Total phenolic content

Total phenolic content (TPC) of extracts was determined using the Folin–Ciocalteu assay reported by Kähkönen et al. (1999). Samples (300 μl in triplicate) were introduced into test tubes, followed by 1.5 ml of Folin–Ciocalteu's reagent (10 times dilution) and 1.2 ml of sodium carbonate (7.5% w/v). The tubes were allowed to stand for 30 min before absorbance at 765 nm was measured. TPC was expressed as gallic acid equivalents (GAE) in mg per

100 g fresh material. The calibration equation for gallic acid was $y = 0.0111x - 0.0148$ ($R^2 = 0.9998$).

2.5. DPPH radical-scavenging activity

The 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical-scavenging assay reported by Miliauskas, Venskutonis, and van Beek (2004) was adopted with modifications. Different dilutions of the extract (1 ml; triplicate) were added to 2 ml of DPPH \cdot (5.9 mg/100 ml methanol). Absorbance was measured at 517 nm after 30 min. Radical-scavenging ability was calculated as IC₅₀ and expressed as ascorbic acid equivalent antioxidant capacity (AEAC) in mg ascorbic acid/100 g (Leong & Shui, 2002) as follows:

$$\text{AEAC (mg AA/100 g)} = \text{IC}_{50(\text{ascorbate})} / \text{IC}_{50(\text{sample})} \times 10^5$$

The IC₅₀ of ascorbic acid used for calculation of AEAC was 0.00387 mg/ml.

2.6. Ferrous ion-chelating ability

The ferrous ion-chelating (FIC) assay reported by Singh and Rajini (2004) was adopted. Solutions of 2 mM FeSO₄ and 5 mM ferrozine were diluted 20 times. FeSO₄ (1 ml) was mixed with different dilutions of extract (1 ml), followed by ferrozine (1 ml). Absorbance was measured at 562 nm after 10 min. The ability of extracts to chelate ferrous ions was calculated as follows:

$$\text{Chelating effect\%} = (1 - A_{\text{sample}}/A_{\text{control}}) \times 100$$

2.7. Tyrosinase inhibition

Tyrosinase inhibition was determined using the modified dopachrome method with L-DOPA as substrate (Masuda, Yamashita, Takeda, & Yonemori, 2005). Assays were conducted in a 96-well microtitre plate and a plate reader was used to measure absorbance at 475 nm with 700 nm as reference. Samples were dissolved in 50% DMSO. Each well contained 40 μ l of sample with 80 μ l of phosphate buffer (0.1 M, pH 6.8), 40 μ l of tyrosinase (31 units/ml) and 40 μ l of L-DOPA (2.5 mM). Each sample was accompanied by a blank that had all the components except L-DOPA. Results were compared with a control consisting of 50% DMSO in place of sample. The percentage tyrosinase inhibition was calculated as follows:

$$(A_{\text{control}} - A_{\text{sample}}) / A_{\text{control}} \times 100\%$$

3. Results and discussion

3.1. Description of plant species

Leaves of the 26 ginger species screened for antioxidant properties belong to nine genera and three tribes (Table 1). The tribes and genera are Alpineae (*Alpinia*, *Elettariopsis* and *Etilingera*), Hedychieae (*Boesenbergia*, *Curcuma*, *He-*

dychium, *Kaempferia*, and *Scaphochlamys*), and Zingibereae (*Zingiber*). Alpineae species are medium- to large-sized forest plants of which *Etilingera* is the largest (Larsen et al., 1999). Zingibereae species are medium-sized plants and Hedychieae species are small- to medium-sized herbs.

3.2. Antioxidant properties of leaves

TPC and radical-scavenging activities of methanol extracts of leaves were assessed using the Folin–Ciocalteu and DPPH radical-scavenging assays, and expressed in mg GAE/100 g and mg AA/100 g, respectively.

Of the 26 ginger species screened, leaves of *Etilingera* species had the highest TPC and AEAC. Values ranged from 2390 mg GAE/100 g and 2280 mg AA/100 g in *E. elatior* to 1110 mg GAE/100 g and 963 mg AA/100 g in *Etilingera maingayi*, respectively (Table 2). Ranking was in the order: *E. elatior* \approx *Etilingera rubrostriata* \approx *Etilingera littoralis* > *Etilingera fulgens* \approx *E. maingayi* in terms of TPC and *E. elatior* \approx *E. rubrostriata* > *E. littoralis* > *E. fulgens* \approx *E. maingayi* in terms of AEAC. Among the *Alpinia* species, leaves of *A. zerumbet* also showed high TPC and AEAC, with

Table 2
Total phenolic content (TPC) and ascorbic acid equivalent antioxidant capacity (AEAC) of leaves of 26 ginger species (fresh weight)

Species	TPC (mg GAE/100 g)	AEAC (mg AA/100 g)
<i>Alpinia zerumbet</i>	1990 \pm 62a	2180 \pm 42a
<i>A. purpurata</i>	1190 \pm 174b	1100 \pm 113b
<i>A. zerumbet</i> 'Variegata'	1150 \pm 41b	1250 \pm 184b
<i>A. malaccensis</i>	744 \pm 61c	800 \pm 62c
<i>A. galanga</i>	392 \pm 50d	90 \pm 36d
<i>Boesenbergia rotunda</i>	260 \pm 8	157 \pm 2
<i>Curcuma zanthorrhiza</i>	503 \pm 57a	287 \pm 39a
<i>C. aeruginosa</i>	282 \pm 78b	140 \pm 47b
<i>C. mangga</i>	275 \pm 36b	118 \pm 11b
<i>C. longa</i>	230 \pm 19b	113 \pm 18b
<i>Elettariopsis latiflora</i>	423 \pm 26a	395 \pm 27a
<i>E. slahmong</i>	346 \pm 45b	269 \pm 67b
<i>E. smithiae</i>	303 \pm 18b	147 \pm 21c
<i>Etilingera elatior</i>	2390 \pm 329a	2280 \pm 778a
<i>E. rubrostriata</i>	2250 \pm 113a	2290 \pm 118a
<i>E. littoralis</i>	2150 \pm 94a	1990 \pm 87b
<i>E. fulgens</i>	1280 \pm 144b	845 \pm 158c
<i>E. maingayi</i>	1110 \pm 93b	963 \pm 169c
<i>Hedychium coronarium</i>	820 \pm 55	814 \pm 116
<i>Kaempferia galanga</i>	146 \pm 9a	77 \pm 7a
<i>K. rotunda</i>	140 \pm 48ab	46 \pm 15b
<i>K. pulchra</i>	112 \pm 9b	30 \pm 3b
<i>Scaphochlamys kunstleri</i>	203 \pm 21	171 \pm 33
<i>Zingiber officinale</i>	291 \pm 18a	96 \pm 7a
<i>Z. spectabile</i>	242 \pm 7b	121 \pm 24a
<i>Z. ottensii</i>	162 \pm 13c	52 \pm 6b

Values of TPC and AEAC are means \pm SD ($n = 3$). For each column, values followed by the same letter (a–d) are not statistically different at $P < 0.05$ as measured by the Tukey HSD test. ANOVA compares values of leaves of species in each genus and does not apply between genera.

values of 1990 mg GAE/100 g and 2180 mg AA/100 g, respectively. Leaves of the commercially cultivated *A. galanga* had the lowest values of 392 mg GAE/100 g and 90 mg AA/100 g, respectively. Although most *Etilingera* species and some *Alpinia* species displayed high phenolic content and radical-scavenging activity, species of *Elettariopsis*, which also belong to the tribe Alpineae, had much lower values, ranging from 303 to 423 mg GAE/100 g and 147–395 mg AA/100 g, respectively.

TPC and AEAC values of leaves of genera belonging to the tribes Hedychieae and Zingibereae were comparatively lower (Table 2). They include species such as *Boesenbergia rotunda*, *Curcuma aeruginosa*, *C. longa*, *Curcuma mangga*, *Curcuma zanthorrhiza*, *H. coronarium*, *K. galanga*, *K. rotunda*, and *Zingiber ottensii* which are used in food flavouring and traditional medicine. Among these species, *H. coronarium* had the highest TPC and AEAC with values of 820 mg GAE/100 g and 814 mg AA/100 g, respectively. Species of *Kaempferia* had very low phenolic content and radical-scavenging activity with values ranging from 112 to 146 mg GAE/100 g and 30–77 mg AA/100 g, respectively. Leaves of *Kaempferia pulchra* exhibited the lowest values.

Foliage of tropical forest plants produces more antioxidants when exposed to elevated light conditions (Frankel & Berenbaum, 1999). Plants growing along the seashore, which receive much sunlight, have efficient antioxidant properties to prevent oxidative damage (Masuda et al., 1999). These observations may also apply to species of *Etilingera*, which have the highest leaf TPC and AEAC. *Etilingera* species are the largest of the ginger plants and can grow up to 6 m in height (Khaw, 2001). They grow in gaps of disturbed forest and are continually exposed to direct sunlight. *Alpinia* species with high TPC and AEAC are medium-sized to large forest plants (Larsen et al., 1999). The other genera are small- to medium-sized herbs. Among the various tribes and genera of gingers, there appears to be a positive correlation between the phenolic content and radical-scavenging activity of leaves with plant size and site conditions. Larger ginger plants growing in exposed forest sites have greater antioxidant properties than have smaller plants growing in shaded sites.

3.3. Antioxidant properties of leaves and rhizomes

TPC and antioxidant activity of methanol extracts of leaves and rhizomes of 14 species from the same plant/location were also assessed for comparison purposes. Results in Table 3 show that leaves of *E. elatior* and *E. maingayi* which had the highest TPC and AEAC were seven to eight times higher than those of rhizomes. Other species with leaves having significantly higher TPC and AEAC than rhizomes were *C. aeruginosa*, *C. mangga*, *C. zanthorrhiza*, *K. galanga*, and *Scaphochlamys kunstleri*. Species with higher TPC or AEAC were *A. galanga*, *B. rotunda*, *E. slahmong*, and *Z. officinale*. This would mean that about 80% of the species had significantly higher TPC and/or AEAC in

Table 3

Total phenolic content (TPC) and ascorbic acid equivalent antioxidant capacity (AEAC) of leaves (L) and rhizomes (R) of 14 ginger species (fresh weight)

Species	Part	TPC (mg GAE/100 g)	AEAC (mg AA/100 g)
<i>Alpinia galanga</i>	L	392 ± 50a	90 ± 36a
	R	214 ± 20b	168 ± 13b
<i>A. malaccensis</i>	L	744 ± 61a	800 ± 62a
	R	564 ± 209a	745 ± 342a
<i>Boesenbergia rotunda</i>	L	260 ± 8a	157 ± 2a
	R	197 ± 50a	89 ± 7b
<i>Curcuma aeruginosa</i>	L	282 ± 78a	140 ± 47a
	R	145 ± 31b	55 ± 11b
<i>C. longa</i>	L	230 ± 19a	113 ± 18a
	R	534 ± 205b	390 ± 127b
<i>C. mangga</i>	L	275 ± 36a	118 ± 11a
	R	112 ± 21b	33 ± 1b
<i>C. zanthorrhiza</i>	L	503 ± 57a	287 ± 39a
	R	250 ± 52b	134 ± 21b
<i>Elettariopsis slahmong</i>	L	346 ± 45a	269 ± 67a
	R	219 ± 57b	197 ± 76a
<i>Etilingera elatior</i>	L	2390 ± 329a	2280 ± 778a
	R	326 ± 76b	295 ± 96b
<i>E. maingayi</i>	L	1110 ± 93a	963 ± 169a
	R	160 ± 52b	122 ± 53b
<i>Kaempferia galanga</i>	L	146 ± 9a	77 ± 7a
	R	57 ± 1b	17 ± 1b
<i>Scaphochlamys kunstleri</i>	L	203 ± 21a	171 ± 33a
	R	73 ± 3b	14 ± 2b
<i>Zingiber officinale</i>	L	291 ± 18a	96 ± 7a
	R	157 ± 18b	84 ± 3a
<i>Z. spectabile</i>	L	242 ± 7a	121 ± 24a
	R	157 ± 100a	124 ± 109a

Values of TPC and AEAC are means ± SD ($n = 3$). For each column, values followed by the same letter (a–b) are not statistically different at $P < 0.05$ as measured by the Tukey HSD test. ANOVA compares values of leaves and rhizomes of each species and does not apply between species.

leaves than in rhizomes. Exceptions were AEAC of *A. galanga*, and TPC and AEAC of *C. longa* where rhizomes showed significantly higher values than did leaves. TPC and AEAC of leaves and rhizomes of *Alpinia malaccensis* and *Zingiber spectabile* were comparable. Values were generally more variable between rhizomes than between leaves of a species, as evident in *A. malaccensis*, *C. longa*, and *Z. spectabile*.

Analysis of metal ion-chelating properties showed that six of the eight species studied clearly displayed higher FIC ability in leaves than in rhizomes. The species were *C. longa*, *K. galanga*, *Alpinia galanga*, *E. elatior*, *Zingiber spectabile*, and *E. maingayi*. (Figs. 1a and b, and 2a). FIC values of leaves and rhizomes of *C. zanthorrhiza* were comparable (Fig. 2b). At lower extract concentration, leaves of *S. kunstleri* showed lower values but, at higher concentration, values were comparable. Of particular interest is *C.*

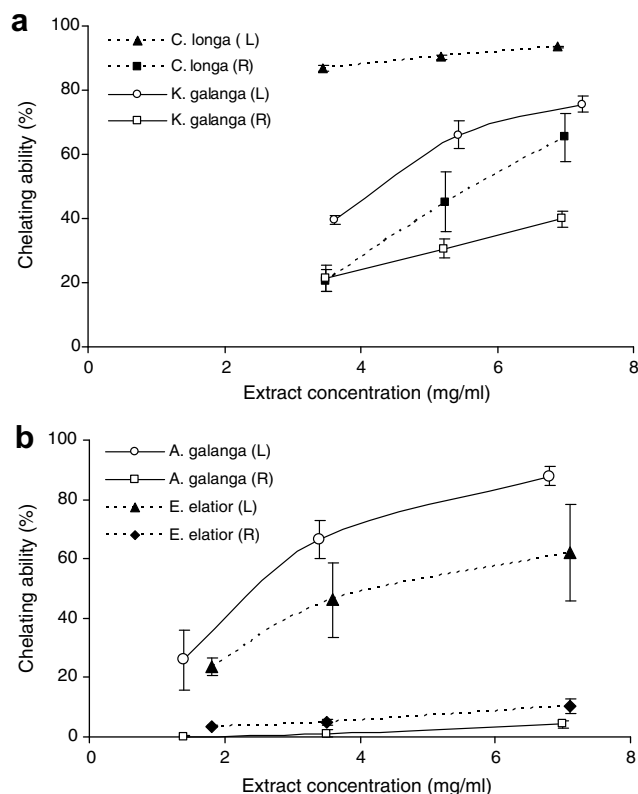


Fig. 1. Ferrous ion-chelating (FIC) ability of leaves (L) and rhizomes (R) of *Curcuma longa*, *Kaempferia galanga*, *Alpinia galanga*, and *Etlingera elatior* (fresh weight).

longa where TPC and AEAC were significantly higher in rhizomes (Table 3), but the FIC ability was higher in leaves (Fig. 1a). In the case of *Z. spectabile*, although TPC and AEAC were comparable (Table 3), FIC value of leaves was higher than that of rhizomes (Fig. 2a). The most outstanding was the FIC value of *A. galanga* leaves which was more than 20 times higher than that of rhizomes (Fig. 1b).

There are few studies comparing the antioxidant properties of leaves and rhizomes of ginger species. Essential oils from leaves of *Aframomum giganteum* had higher antioxidant activity than had those from rhizomes (Agnaniet, Menut, & Bessière, 2004). Leaves of *A. zerumbet* showed higher inhibition of β -carotene oxidation and radical-scavenging activity than did rhizomes (Elzaawely, Xuan, & Tawata, 2007). Contrary to our results, higher phenolic content and antioxidant activity have been reported in rhizomes than in leaves of *Z. officinale* (Katsube et al., 2004). These studies involved one or two ginger species and it is not known whether their comparisons were based on plant samples from the same or different locations. Our present study is probably the first where the phenolic content, radical-scavenging activities and metal ion-chelating abilities of leaves and rhizomes of ginger species from the same plant/location were systematically compared.

Antioxidants are secondary metabolites produced by plants to protect against oxidative damage by free radicals

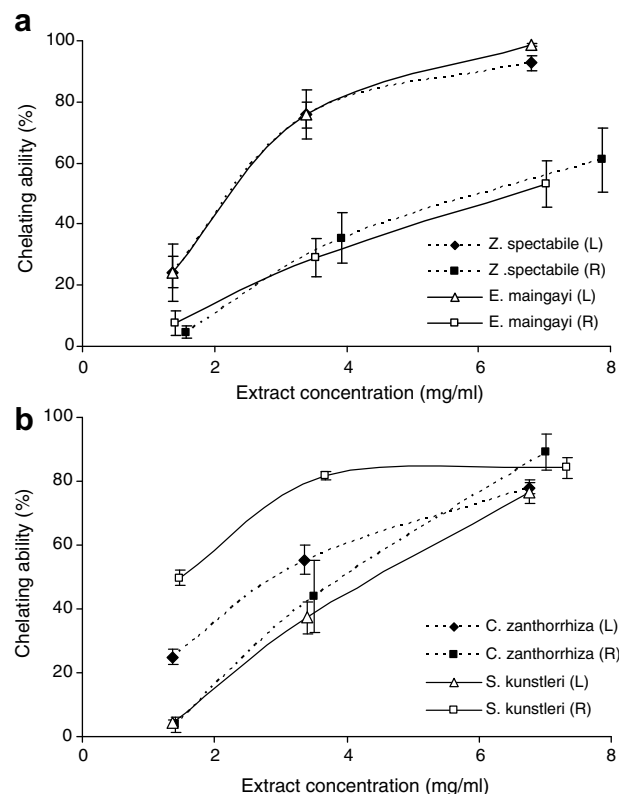


Fig. 2. Ferrous ion-chelating (FIC) ability of leaves (L) and rhizomes (R) of *Zingiber spectabile*, *Etlingera maingayi*, *Curcuma zanthorrhiza*, and *Scaphochlamys kunstleri* (fresh weight).

(Larson, 1988). In the family Zingiberaceae, it is generally believed that antioxidants produced by the plant are transported to the rhizomes where they are accumulated. This implies that rhizomes would have higher antioxidant activity than other plant parts. However, results of this study showed that this might not be true as the majority of the species studied had significantly higher phenolic content and antioxidant activity in leaves than in rhizomes. Similar observations have been made by Herrmann (1988), who reported much greater concentrations of flavones and flavonols in leaves of vegetables which are exposed to sunlight. Only trace amounts were found in unexposed parts below the soil surface which include roots and rhizomes. This could explain why leaves have significantly higher phenolic contents and antioxidant activities than have rhizomes in ginger plants.

3.4. Tyrosinase inhibition activity of leaves of *Etlingera*

With outstanding leaf TPC and AEAC, methanol extracts of leaves of five *Etlingera* species were analysed for tyrosinase inhibition activity using the modified dopachrome method with L-DOPA as the substrate. Leaves of *Hibiscus tiliaceus* were chosen as positive control as they displayed the highest tyrosinase inhibition activity among 39 tropical plant species screened by Masuda et al. (2005).

Table 4
Tyrosinase inhibition activity of leaves of five *Etilingera* species (fresh weight)

Species	Tyrosinase inhibition % (0.5 mg/ml of extract)
<i>Etilingera elatior</i>	55.2 ± 3.1a
<i>E. fulgens</i>	49.3 ± 6.5ab
<i>E. maingayi</i>	42.6 ± 4.2b
<i>E. rubrostriata</i>	29.5 ± 4.0c
<i>E. littoralis</i>	22.0 ± 5.2c
<i>Hibiscus tiliaceus</i>	43.9 ± 4.6b

Results are means ± SD ($n = 3$). Values followed by the same letter (a–c) are not statistically different at $P < 0.05$ as measured by the Tukey HSD test. Leaves of *Hibiscus tiliaceus* were used as positive control.

Tyrosinase inhibition activity was strongest in leaves of *E. elatior* (55.2%), which was significantly higher than the positive control (43.9%) (Table 4). Inhibition activities of leaves of *E. fulgens* (49.3%) and *E. maingayi* (42.6%) were comparable. Activities of leaves of *E. rubrostriata* (29.5%) and *E. littoralis* (22.0%) were significantly lower. This would mean that three out of five *Etilingera* species studied had activity values that were significantly higher or comparable to the positive control.

Masuda et al. (2005) observed that seashore plant species, which are exposed to full sunlight, possess strong antioxidant activity and high tyrosinase inhibition ability. Findings from this study agree with this observation. Compared with species of other genera, *Etilingera* species had outstanding TPC and AEAC with *E. elatior* having the highest values. In our earlier study (Chan, Lim, & Omar, 2007), leaves of *E. maingayi* had the highest FIC ability and lipid peroxidation inhibition activity, and leaves of *E. fulgens* had high FIC ability. Leaves of *E. elatior*, *E. maingayi*, and *E. fulgens* also showed inhibition of all Gram-positive bacteria of *Bacillus cereus*, *Micrococcus luteus*, and *Staphylococcus aureus* tested. In our present study, leaves of these three *Etilingera* species displayed high tyrosinase inhibition activity. This would mean that, besides promising tyrosinase inhibition ability, they also have high antioxidant activity and antibacterial properties.

4. Conclusion

Of the 26 ginger species screened, leaves of *Etilingera* species had the highest TPC and AEAC. Eleven of the 14 species showed significantly higher phenolic content and/or antioxidant activities in leaves than in rhizomes. Values of leaves of *E. elatior*, and *E. maingayi* were seven to eight times higher than those of rhizomes. Six of the eight species clearly displayed higher FIC ability in leaves than in rhizomes. The FIC value of *A. galanga* leaves was more than 20 times higher than that of rhizomes. Three species of *Etilingera* displayed tyrosinase inhibition activity that was significantly higher or comparable to the positive control. With high tyrosinase inhibition, antioxidant activity, and antibacterial properties, leaves of these *Etilingera* species can be developed into skin-lightening products and natural preservatives to inhibit food spoilage.

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