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Nutritional Value, Bioactive Compounds, and Antioxidant Activity of Phak-liang (Gnetum gnemon Linn. var. tenerum Markgr.) in Surat Thani Province, Thailand

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Received: 3 April 2024 / Revised: 24 June 2024 / Accepted: 9 July 2024

Abstract

The nutritional value, bioactive compounds, and antioxidant activity of Phak-liang (Gnetum gnemon Linn. var. tenerum Markgr.) were investigated. The indigenous vegetables were collected from three districts; Muang, Bannasarn, and Khirirat Nikom in Surat Thani Province. The nutritional value of the samples was carried out according to the methods of AOAC. The moisture contents ranged from 84.72-87.52%, and the ash contents ranged from 0.78-1.07% fresh weight. The percentage of fiber, protein, fat, and carbohydrate in the leafy vegetables were in the range of 3.06-3.81, 3.44-4.68, 0.65-1.08 and 7.49-9.55 fresh weight, respectively. The samples were analyzed by ICP-OES for their mineral determinations. The amounts found in the samples, in descending order were: K > Ca > Mg > Mn > Na > Fe > Zn > Cu. Considering the amounts of minerals found in the vegetable and their recommended daily values by the Bureau of Nutrition (Ministry of Public Health, Thailand), the daily value (DV) percentage of Mn in the samples were relatively high (28.45-96.65%), whereas K, Mg, Ca, Fe, Cu and Zn contributed from 1.29-8.42% of the daily values. The vegetable had negligible amounts of Na (0.03 - 0.07% of the DV). The antioxidant capacities were analyzed using Ferric Reducing Antioxidant Power (FRAP) and 2,2-diphenyl-1-picrylhydrazyl (DPPH) assay. The results showed that antioxidant activity increased by the concentration of the ethanol extracts. The FRAP values of the extracts ranged from 89.01-160.70 μ M FeSO₄/100 g dry weight, and the IC₅₀ values calculated for DPPH radical scavenging activities of the extracts ranged from 54.37-60.57 mg/mL. The results suggested that Phak-liang is a potential source of natural phytochemicals and could be used for health promotion in the community.

Keywords: Nutritional value, Bioactive compounds, Antioxidant activity, Gnetum gnemon Linn. var. tenerum Markgr.

1. Introduction

Considerable attention has been focused on investigating the significant role of various edible plants recognized for their potential in mitigating chronic disease, particularly those abundant in antioxidants (Alam, Saqib, & Ashraf, 2017; Boeing et al., 2012; Pranprawit, 2019; Pranprawit, Heyes, Molan, & Kruger, 2015). A comprehensive review on the associations between the consumption of vegetables and fruits and the risk of several chronic diseases have been documented (Boeing et al., 2012). The findings of this review highlight that increasing the intake of vegetables and fruits from a range of botanical families, which provide sources of nutrients, dietary fiber and phytochemicals reduces the risk of hypertension, coronary heart disease, stroke, certain eye disease, dementia and osteoporosis. The presence of a wide range of bioactive compounds in fruits and vegetables, such as ascorbic acid, tocopherols, phenolic acids, flavonoids, carotenoids, and anthocyanin, has been suggested as the underlying cause for their beneficial effects. These substances possess antioxidant activities and play a significant role in promoting health and preventing chronic diseases, thereby enhancing the functionality

of these natural foods (Alfa & Arroo, 2019; Paran, Novack, Engelhard, & Hazan-Halevy, 2009; Zehiroglu & Sarikaya, 2019).

Phak-liang (*Gnetum gnemon* Linn. var. *tenerum* Markgr.) is an indigenous leafy vegetable in the southern region of Thailand. It has gained popularity as a consumable vegetable, with concurrent cultivation observed alongside rubber plantations, oil palm plantations, or fruit orchards. It is widely consumed in various forms, including raw consumption as leafy greens or as an ingredient in culinary preparations. Notably, the leafy vegetable stands out for its exceptional nutritional profile, exhibiting high levels of essential nutrients and bioactive compounds of considerable importance (Kongkachuichai, Charoensiri, Yakoh, Kringkasemsee, & Insung, 2015). Phak-liang vegetable possesses components including β -carotene, lutein, polyphenol, vitamin C, and vitamin E, which exhibit antioxidant properties that may contribute to anti-aging mechanisms and strengthen the immune system (Kongkachuichai et al., 2015). Nevertheless, the comprehensive investigation of the nutritional composition, bioactive compounds, and antioxidant properties of the leafy vegetable remains scarce. Thus, the objectives of this study were to determine the nutritional value of Phak-liang (*Gnetum gnemon* Linn. var. *tenerum* Markgr.) collected from Surat Thani Province, southern Thailand and to assess the bioactive compounds and antioxidant activities inherent in the vegetable extracts. This study endeavors to provide valuable insights into the nutritional composition of the indigenous leafy vegetable, thereby contributing to the existing body of knowledge in this field.

2. Materials and Methods

2.1 Sample collection and preparation

The vegetable samples were randomly collected from 9 different locations (Table 1) in the 3 districts i.e., Muang (S1, S2 and S3), Bannasarn (S4, S5 and S6), and Khirirat Nikom (S7, S8 and S9), Surat Thani Province, southern Thailand in December 2021 to February 2022. Four kilograms of the vegetable from each sampling location were taken to the laboratory in a cool box. The fresh vegetables were washed with tap water to remove dust or contaminant particles and rinsed with deionized water. The edible portion was separated and the deionized water was dried off at room temperature. The samples were dried in an oven at 45°C for 48 hours, ground to fine powder with a grinder. All homogenized samples were kept at 4°C until analysis.

Sampling site	Geographical coordinate	Area of collection
S1	9°00'57.9"N 99°21'56.0"E	Village No.10, Khun-Ta-Lay Sub-district, Muang District
S2	9°00'32.7"N 99°20'04.4"E	Village No.8, Khun-Ta-Lay Sub-district, Muang District
S 3	9°04'18.0"N 99°23'19.4"E	Village No.8, Ma-Kham-Tia Sub-district, Muang District
S4	8°56'24.5"N 99°21'45.4"E	Village No.1, Tung-Tao-Mai Sub-district, Bannasarn District
S5	8°55'51.9"N 99°22'03.5"E	Village No.5, Tung-Tao Sub-district, Bannasarn District
S 6	8°58'23.9"N 99°24'09.9"E	Village No.4, Tung-Tao-Mai Sub-district, Bannasarn District
S 7	9°03'02.8"N 99°00'38.0"E	Village No.4, Ban-Yang Sub-district, Khirirat Nikom District
S8	9°00'17.0"N 98°55'43.7"E	Village No.6, Tha-Kanhon Sub-district, Khirirat Nikom District
S 9	8°59'47.6"N 98°56'37.1"E	Village No.5, Yan-Yao Sub-district, Khirirat Nikom District

2.2 Proximate composition and nutritional analysis

Moisture content was determined by oven drying at a temperature of 100 ± 5 °C to a constant weight according to the AOAC method 950.46 (Latimer, 2016). Total ash content of the sample was determined by furnace incineration at 600 °C followed by the AOAC method 942.05 (Latimer, 2016). Total fiber content was carried out by digesting the samples with an acid and a base solution, then drying and combusting the residue according to the AOAC method 978. 10 (Latimer, 2016) Crude protein content was determined using a carbon/nitrogen determinator according to the combustion method of AOAC 990.03 (Latimer, 2016). The protein content was calculated from total nitrogen content multiplied by 6.25. Crude fats were extracted with hexane solvent in an automated fat extraction system (S306MK Gerhardt, Germany) according to AOAC method 922.06 (Latimer, 2016). Total carbohydrates were calculated by subtracting the sum of percentage of moisture, protein,

fat, and ash from 100%. Macroelements and microelements were analyzed by an inductively coupled plasma – optical emission spectrometer (ICP-OES, Prodigy 7, Teledyne Leeman Labs) after the samples were acid digested according to EPA Method 3052 (United States Environmental Protection Agency, 1996) using a microwave system.

2.3 Analysis of bioactive compounds and antioxidant activity 2.3.1 Preparation of crude extract of Phak-liang

Samples of dried Phak-liang were extracted using 95% ethanol. The preparation was done by soaking dried Phak-liang powder in 95% ethanol solvent in a ratio of 1g of powder to 5 mL of solvent and periodically shaking for 24 hours at room temperature. The extracts were then filtered twice through filter paper Whatman#1, and were

evaporated using a rotary evaporator at 45°C, then stored in a refrigerator at 4°C for further analysis.

2.3.2 Total phenolic content (TPC)

Total phenolics were measured using the Folin-Ciocalteu procedure (modified from the previous method of Singleton, Orthofer, and Lamuela-Raventós (1999) and Waterhouse (2002). Briefly, 200 μ L of extract was added to 200 μ L of Folin-Ciocalteu phenol reagent and 800 μ L of Milli-Q water, and allowed to react for 5 min. Subsequently, 2 mL of 7.5% (w/v) sodium carbonate solution and 1.6 mL of Milli-Q water were added and incubated for 30 min at room temperature. Absorbance of extracts were spectrophotometrically measured at 765 nm. A standard curve was prepared using different concentration (0-1,000 μ g/mL) of gallic acid solution. Total phenolics was calculated as mg gallic acid equivalent (GAE) per 100g of dry weight sample.

2.3.3 Ferric reducing antioxidant power (FRAP) assay

The FRAP assay was carried out according to the previous method of Benzie and Strain (1996) with slight modification. FRAP reagent consisted of 300 mmol/L acetate buffer (pH 3.6), 10 mmol/L TPTZ (2,4,6-tripyridyl-s-triazine) in 40 mmol/L hydrochloric acid, and 20 mmol/L ferric chloride (III) solution at the ratio of 10: 1: 1 (v/v/v), respectively. The sample extract (200 µL) was added to 1.8 mL of FRAP reagent and 2 mL of Milli-Q water, mixed well and incubated at 37°C for 30 min, then the absorbance was measured spectrophotometrically at 593 nm. A standard curve was prepared using various concentrations of FeSO₄.7H₂O. The antioxidant activity was calculated based on the ability of extracts to reduce ferric (III) iron to ferrous (II) iron, and results were expressed as micromole ferrous ion (II) equivalent per g of dry weight sample.

2.3.4 Scavenging of diphenyl-picrylhydrazyl (DPPH) radicals

The ability of the extract to scavenge DPPH-radical was determined using the previous method of Prior, Wu, & Schaich (2005) with slight modification. Briefly, 1 mL of sample extract was added to 2 mL of 0.2 mM DPPH in 95% ethanol and 1 mL of Milli-Q water, mixed well, and incubated in the dark at room temperature for 30 min to allow the reaction to progress to completion before reading the absorbance at 517 nm. The absorbance reading was compared to the standard curve of 0-100 μ g/mL of ascorbic acid. The scavenging activity of samples (% inhibition) at various concentrations was calculated and linear regression was performed to determine the concentration of samples that can scavenge DPPH-radical by 50% (IC₅₀).

2.4 Statistical data analysis

Descriptive statistics, including mean, standard deviation, and percentage were analyzed. The data were subjected to Analysis of Variance (ANOVA) to evaluate the variability, and Pearson's correlation coefficient (r) was used to determine linear relationships, with a significance level set at P < 0.05.

3. Results

3.1 Nutritional analysis

The vegetables were harvested at approximately 2-3 years of age, having been cultivated in sandy loam soil, and were subjected to the application of a balanced fertilizer formula consisting of either 15-15-15 or 16-16-16. The nutritional composition of Phak-liang vegetable was carried out on fresh weight basis and has shown in Table

2. The moisture contents of the vegetable were in the range of 84.72 - 87.52%, while the ash contents were ranged from 0.78 - 1.07%. The crude fiber, protein, fat, and carbohydrate contents were found between 3.06 - 3.81, 3.44 - 4.68, 0.65 - 1.08, and 7.49 - 9.55% fresh weight, respectively.

Sampling site	Moisture	Ash	Fiber	Protein	Fat	Carbohydrate
S1	87.52 ± 0.40	$0.85\pm0.00_2$	3.30 ± 0.05	3.44 ± 0.02	$0.70\pm0.00_3$	7.49 ± 0.41
S2	86.90 ± 0.33	$0.78\pm0.00_3$	3.66 ± 0.03	3.77 ± 0.01	0.68 ± 0.01	7.87 ± 0.33
S 3	86.30 ± 0.25	0.92 ± 0.01	3.18 ± 0.03	4.06 ± 0.01	$0.76\pm0.00_4$	7.96 ± 0.26
Mean±SD	86.91 ± 0.60	0.85 ± 0.06	3.38 ± 0.22	3.76 ± 0.27	0.71 ± 0.04	7.77 ± 0.36
S4	85.39 ± 0.19	1.07 ± 0.01	3.08 ± 0.07	4.68 ± 0.02	1.08 ± 0.01	7.78 ± 0.21
S5	84.76 ± 0.36	0.88 ± 0.01	3.06 ± 0.05	3.81 ± 0.01	1.00 ± 0.01	9.55 ± 0.36
S6	84.72 ± 0.92	0.96 ± 0.01	3.24 ± 0.07	4.52 ± 0.01	0.93 ± 0.01	8.85 ± 0.92
Mean±SD	84.96 ± 0.60	0.97 ± 0.08	3.12 ± 0.10	4.34 ± 0.40	1.00 ± 0.06	8.73 ± 0.92
S7	86.69 ± 0.34	$0.79\pm0.00_2$	3.18 ± 0.08	3.63 ± 0.01	$0.65\pm0.00_{\rm l}$	8.24 ± 0.34
S8	85.79 ± 0.76	$0.83\pm0.00_2$	3.81 ± 0.09	4.13 ± 0.02	$0.75\pm0.00_4$	8.50 ± 0.77
S9	85.91 ± 0.24	$1.00\pm0.00_3$	3.34 ± 0.05	4.24 ± 0.02	$0.66\pm0.00_3$	8.19 ± 0.22
Mean±SD	86.13 ± 0.60	0.87 ± 0.10	3.44 ± 0.29	4.00 ± 0.28	0.69 ± 0.05	8.31 ± 0.46

Table 2. Nutritional composition of Phak-liang vegetable (Gnetum gnemon) (%FW, n = 3).

3.2 Macroelements and microelements in the vegetable

Macroelements (Na, K, Mg, and Ca) and microelements (Fe, Cu, Zn, and Mn) were measured by an ICP-OES. The mean and standard deviation values of the elements in the vegetable are summarized in Table 3. The results showed that the amounts of macroelements, in descending order, were K, Ca, Mg, and Na with the range of 354.75-558.02, 34.37-64.18, 27.50-45.33, and 0.65-1.35 mg/100g fresh weight, respectively. In addition, the concentrations of microelements, in descending order, were Mn, Fe, Zn, and Cu, ranging from 1.76-5.18, 0.58-0.93, 0.39-0.52, and 0.05-0.08 mg/100g, respectively.

 Table 3. Macroelements and microelements present in Phak-liang vegetable (*Gnetum gnemon*) and percentage daily value.

Sampling	Mean \pm SD (mg/100g FW, n = 3)							
Site	Na	K	Mg	Ca	Fe	Cu	Zn	Mn
S1	1.35 ± 0.06	421.91 ± 11.27	43.57 ± 2.07	45.56 ± 1.57	0.58 ± 0.03	0.05 ± 0.01	0.43 ± 0.03	2.00 ± 0.15
S2	1.04 ± 0.04	354.75 ± 11.45	40.28 ± 1.37	59.39 ± 4.04	0.68 ± 0.03	0.08 ± 0.01	0.52 ± 0.03	2.86 ± 0.09
S 3	0.98 ± 0.03	440.12 ± 20.05	40.85 ± 1.64	59.92 ± 3.67	0.85 ± 0.06	0.07 ± 0.01	0.46 ± 0.03	3.05 ± 0.08
Mean±SD	1.12 ± 0.18	405.59 ± 41.00	41.57 ± 2.13	54.95 ± 7.60	0.70 ± 0.12	0.07 ± 0.01	0.47 ± 0.05	2.64 ± 0.49
S4	0.73 ± 0.06	558.02 ± 19.44	42.09 ± 1.75	43.32 ± 2.60	0.71 ± 0.07	0.05 ± 0.01	0.47 ± 0.03	1.93 ± 0.08
S5	1.09 ± 0.03	398.44 ± 12.72	36.95 ± 1.92	64.18 ± 3.67	0.93 ± 0.05	0.06 ± 0.01	0.43 ± 0.03	3.70 ± 0.20
S 6	0.65 ± 0.06	504.49 ± 15.17	40.00 ± 2.53	42.59 ± 2.97	0.65 ± 0.05	0.07 ± 0.01	0.39 ± 0.02	1.92 ± 0.08
Mean±SD	0.82 ± 0.21	486.98 ± 71.69	39.68 ± 2.88	50.03 ± 10.95	0.76 ± 0.14	0.06 ± 0.01	0.43 ± 0.04	2.52 ± 0.89
S 7	0.98 ± 0.05	442.49 ± 17.51	27.50 ± 2.35	34.37 ± 2.82	0.58 ± 0.02	0.05 ± 0.01	0.44 ± 0.05	1.76 ± 0.11
S 8	0.90 ± 0.05	475.10 ± 14.33	43.94 ± 2.82	58.59 ± 2.64	0.58 ± 0.03	0.05 ± 0.01	0.48 ± 0.04	1.92 ± 0.10
S9	1.05 ± 0.04	505.96 ± 18.37	45.33 ± 2.66	54.46 ± 2.55	0.70 ± 0.03	0.06 ± 0.01	0.49 ± 0.05	5.18 ± 0.43
Mean±SD	0.97 ± 0.07	474.51 ± 31.11	38.92 ± 8.88	49.14 ± 11.46	0.62 ± 0.06	0.06 ± 0.01	0.47 ± 0.04	2.95 ± 1.68
Recommended daily value,								
mg	910	2925	280	900	15	1.5	10	2.5
mg per								
serving*	0.26 - 0.61	148.06 - 246.21	10.67 - 20.25	13.40 - 29.32	0.23 - 0.42	0.02 - 0.04	0.16 - 0.24	0.71 - 2.42
Daily value,								
%**	0.03 - 0.07	5.06 - 8.42	3.81 - 7.23	1.49 - 3.26	1.56 - 2.79	1.29 - 2.41	1.59 - 2.37	28.45-96.65

* 1 serving size = 43 g fresh weight, based on the food exchange list that provides 25 kcal (Kongkachuichai et al., 2015)

** Daily value (%) = mineral amounts in the vegetable sample / recommended daily value x 100

3.3 Total phenolic content and antioxidant activity

3.3.1 Total phenolic content (TPC)

For the total phenolic content (TPC), each Phak-liang vegetable was extracted using 95% ethanol. The results are shown in Table 4. The TPC of ethanoic extract of Phak-liang vegetable collected from Muang district (S1-S3) ranged from 121.26 to 149.53 mg GAE/100 g dried weight, while these values were between 73.21 to

102.51 mg GAE/100 g dried weight in Phak-liang vegetable collected from Bannasarn district (S4-S6) and ranged from 109.15-127.79 mg GAE/100 g dried weight in Phak-liang vegetable collected from Khirirat Nikom district (S7-S9).

3.3.2 Ferric reducing antioxidant power (FRAP)

The antioxidant activity was determined using two commonly used methods: ferric reducing antioxidant power (FRAP), and scavenging of diphenyl-picrylhydrazyl (DPPH) radicals. In this study, ethanoic extract of Phak-liang vegetable collected from 3 different sites- Muang (S1-S3), Bannasarn (S4-S6), and Khirirat Nikom (S7-S9) in Surat Thani province had the reducing ability, which was measured by FRAP, in the range of 128.81-132.46, 89.01-101.59 and 131.02-160.70 μ M FeSO₄ per 100 g dried weight, respectively (Table 4).

3.3.3 Scavenging of diphenyl-picrylhydrazyl (DPPH) radicals

The ability of ethanoic extract of Phak-liang vegetable to scavenge DPPH-radicals increases with the concentration of the extract. The vegetable collected from Muang district (S1-S3) at the concentration of 100 mg/mL possess the ability to scavenge DPPH-radicals (Table 4) between 74.86% and 75.47% inhibition, whereas Phak-liang from Bannasarn (S4-S6) and Khirirat Nikom (S7-S9) exhibited inhibition in the range of 65.20% to 68.52%, and 71.30% to 72.71%, respectively. When IC₅₀ was calculated to assess the concentration of the extract required to scavenge 50% of the initial DPPH radicals. The lower the IC50 value, the more potent is the extract at scavenging DPPH and a higher antioxidant activity. It can be found that ethanoic extract of Phak-liang from Muang district (S1-S3), Bannasarn (S4-S6), and Khirirat Nikom (S7-S9) contained IC₅₀ values in the range of 54.368 \pm 0.911, 60.573 \pm 4.293 and 57.569 \pm 1.702 mg/mL, respectively. However, the scavenging ability of the extracts across all areas collected was significantly lower (P< 0.05) when compared to the standard ascorbic acid (0.022 \pm 0.000 mg/mL).

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Sampling site	Total phenolic content	FRAP value	DPPH	DPPH		
	(mg GAE/100g DW)	(µM FeSO4/100g DW)	(% inhibition)*	IC50 (mg/mL)		
S1	126.19 ± 3.57	128.81 ± 4.96	75.47	53.315		
S2	121.26 ± 1.67	132.46 ± 5.50	74.86	54.907		
S 3	149.53 ± 4.50	129.47 ± 7.67	75.33	54.880		
Mean±SD	132.33 ± 15.10	130.24 ± 1.94	75.22 ± 0.32	$54.368 \pm 0.911 **$		
S4	73.21 ± 1.56	89.01 ± 4.35	65.20	60.774		
S5	102.51 ± 1.99	101.59 ± 0.31	68.52	64.762		
S6	90.29 ± 2.14	93.03 ± 2.61	67.85	56.183		
Mean±SD	88.67 ± 14.72	94.55 ± 6.43	67.19 ± 1.76	$60.573 \pm 4.293 **$		
S7	127.79 ± 2.70	160.70 ± 2.80	72.10	59.096		
S8	109.15 ± 1.33	135.56 ± 5.33	71.30	55.735		
S9	121.33 ± 2.55	131.02 ± 1.86	72.71	57.877		
Mean±SD	119.42 ± 9.47	142.43 ± 15.99	72.04 ± 0.71	$57.569 \pm 1.702 **$		
Standard : Ascorbic	acid			0.022 ± 0.000		

Table 4. Total phenolic content, ferric reducing antioxidant power, and scavenging of DPPH-radical in ethanolic extract of Phak-liang vegetable (n = 3).

* Indicate scavenging activity of DPPH-radical of Phak-liang at the concentration of 100 mg/mL

**Indicate statistically significant differences from standard (ascorbic acid) at p<0.05

4. Discussion

Proximate composition and nutritional analyses including moisture, ash content, fiber, crude protein, crude fat, and carbohydrates, were carried out in this study. With the exception of fat content, these results were in agreement with a previous finding indicated that the moisture content, protein, fat and carbohydrate of Phak-liang vegetable collected from 6 Provinces in southern Thailand were 85.66, 4.23, 0.58, and 8.51% fresh weight, respectively (Kongkachuichai et al., 2015). Additionally, both macroelements (Na, K, Mg, and Ca) and microelements (Fe, Cu, Zn, and Mn) were examined. This revealed an alignment between the amounts of Mg and Ca in the present study and those of the examined vegetables collected from six provinces in the southern region of Thailand. Nevertheless, the concentrations of K and Fe in the vegetable were found to be comparatively higher in relation to the results obtained from the previous findings (Kongkachuichai et al., 2015). This could be

attributed to variations in the sources of the vegetables, which come from different cultivation areas with diverse soil conditions and agricultural management practices, such as the application of dissimilar chemical or organic fertilizers.

Research on mineral content in Phak-liang vegetable is relatively limited. Therefore, additional findings were presented by referencing studies conducted on other types of green leafy vegetables. A study reported the amounts of mineral in 15 leafy *Amaranthus* species., which indicated that Na concentrations were in consistent with the findings of this research. However, the concentrations of Ca, K, Mg, Cu, Fe and Zn present in the vegetables were relatively higher compared to this study, whereas Mn exhibited lower levels (Jiménez-Aguilar & Grusak, 2017). In addition, a study conducted on minerals in leafy vegetables of *Amaranthus blitum* reported that the levels of Mn were in agreement with the findings of this research, while the levels of Ca, Mg, Cu, Fe, and Zn were higher compared to the results obtained from this study. Conversely, the levels of K were found to be lower than those observed in this research (Sarker & Oba, 2020).

When considering the levels of essential and trace minerals present in the vegetable, a comparison was made with the recommended daily intake of minerals, which is expressed as a percentage daily value (Bureau of Nutrition, 2020) shown in Table 3. It was found that Mn in the samples had a high percentage daily value (28.45-96.65%), whereas the six minerals (K, Mg, Ca, Fe, Cu, and Zn) in the samples had relatively low percentage daily value (1.29-8.42%). In the case of Na, the percentage of the recommended daily intake is remarkably low (0.03-0.07%). Therefore, Phak-liang vegetable is considered a significant source of essential and trace minerals, especially for Mn.

Phak-liang vegetables are commonly found in local areas of the southern Thailand, however previous studies on bioactive compounds and antioxidant activity in Phak-liang vegetables are relatively limited. The study of Pichairat and Mahea (2014) investigated the phenolic content and antioxidant capacity of *Gnetum gnemon* Linn. var. *tenerum* Markgr. collected from Yantakhao district, Sikao district, Palian district and Nayong district in Trang province. The average total phenolic content in Phak-liang vegetables was 180.95 ± 2.84 mg GAE/100 g fresh weight, and the vegetables also contained antioxidant activity of 26.87 ± 0.40 and 17.08 ± 0.51 mg GAE/100 g fresh weight as measured by DPPH and FRAP assay, respectively. For the study with regards to indigenous vegetables in Surat Thani province, Yakoh, Weschasat, & Suwannachote (2017, 2018) analyzed the phenolic content and antioxidant activity by FRAP assay of Phak-liang vegetables collected from Muang district. In these two studies, total phenolic content of Phak-liang vegetables was compared between summer and rainy season, and found that in rainy season, Phak-liang vegetables had the TPC value of 279.23 ± 47.70 mg GAE/100 g, which exhibited the antioxidant activity (FRAP assay) in the average of $883.80 \pm 89.6 \,\mu$ mol TE/100g. While in summer season, the TPC value found in Phak-liang was increased to 584.73 ± 53.67 mg GAE/100 g and exhibited antioxidant activity of $430.83 \pm 203.20 \,\mu$ mol TE/100 g.

The total phenolic content of Phak-liang vegetables found in the study of Yakoh et al. (2018) is relatively higher than the results of TPC (132.33 \pm 15.10 mg GAE/100 g in Muang district) found in this study, which collected the samples in rainy season between December to February. The reason may be due to the difference in the preparation and extraction methods between the study. This study used the hot-air oven, which may cause to loss of some phenolic compounds during the drying process, and leading to lower TPC values than the study of Yakoh et al. (2018) which used the freeze drier. However, it should be noted in this study that among various indigenous vegetables, *Gnetum gnemon* Linn. var. *tenerum* Markgr. (Phak-liang) contained higher total phenolic compounds than *Musa acuminate* Colla (Hua-plee) (23.5 mg GAE/100 g) *Momordica charantia* Linn. (Marakee-nok) (69.5 mg GAE/100 g) found in Surat Thani province (Pranprawit, 2019), *Lasia spinosa* Thw. (Phaknaam) (50.14 mg GAE/100 g) and *Antidesma ghaesembills* Gaerth. (Mao-khai-pla) (64.03 mg GAE/100 g) in Trang province (Pichairat & Mahea, 2014). Furthermore, Phak-liang vegetable also had higher total phenolic compounds than some vegetables that are commonly consumed in daily life, such as cauliflower(81 mg GAE/100 g) cucumber (17 mg GAE/100 g) and carrot (16 mg GAE/100 g) (Isabelle et al., 2010).

The results of this study are consistent with previous reports (Charoenteeraboon et al., 2019; Pakdee, Poowanna, & Prathumtet, 2021; Pranprawit et al., 2015; Thonginla, Wanwimolruk, & Chuaybamroong, 2014) in finding a significant linear positive relationship between total phenolic content and antioxidant activity in various plants and vegetables. These findings clearly demonstrated that the phenolic compound such as flavonoids and

phenolic acid etc., which commonly found in green leafy vegetables including Phak-liang could contribute to their total antioxidant ability and health benefits. The study of Kato, Tokunaga, and Sakan (2009) found an abundance of stilbenoids (namely resveratrol), saponins, flavonoids, and tannins in seeds of *G. gnemon* collected from Indonesia. Moreover, young leaves of *G. gnemon* are an excellent source of polyphenols (253.45 \pm 5.68 mg GAE/100 g) (Kongkachuichai et al., 2015). In this study, there was a moderately high correlation between total phenolics and FRAP values (r = 0.748, p< 0.01), and a high positive correlation between total phenolics and DPPH values (r = 0.915, p< 0.01), suggesting that the phenolic compounds are likely to be the major contributors to the antioxidant activity, moreover, the phenolic concentration in Phak-liang extracts is more likely to exert a radical scavenging ability rather than the reducing ability. The results of this study suggested that Phak-liang vegetables, which is an indigenous plant in Southern Thailand, are a good source of phenolic compounds and effective antioxidants. Therefore, regular intake should be encouraged among consumers for health promotion in local community.

5. Conclusion

This research examined the leaf extracts of *Gnetum gnemon* Linn. var. *tenerum* Markgr. collected from 3 districts; Muang, Bannasarn, and Khirirat Nikom in Surat Thani province for their nutritional values, bioactive compounds and antioxidant activity. In general, Phak-liang vegetable contain considerable amounts of crude fiber (3.06 - 3.81% FW), protein (3.44 - 4.68% FW) and carbohydrate (7.49 - 9.55% FW). it is also considered a significant source of essential and trace minerals, such as K, Ca, Mg, especially for Mn which contributed from 28.45-96.65% DV recommended by the Bureau of Nutrition (Ministry of Public Health, Thailand). Furthermore, the ethanolic extract of Phak-liang vegetable showed high amounts of phenolic compounds (73.21-149.53 mg GAE/100 g DW) and antioxidant activity which analyzed by FRAP (89.01-160.70 µM FeSO4/100 g DW) and DPPH assay (54.37-60.57 mg/mL), and the phenolic content found in the extract of Phak-liang vegetable were positively related to their antioxidant activity.

The results of this study regarding the nutritional value, bioactive compound and antioxidant efficacy of this indigenous vegetable can be used for further development as a dietary supplement, and will also be useful for consumption recommendation in communities for the prevention and treatment of chronic diseases.

Acknowledgement

The Office of Thailand Science Research and Innovation and Suratthani Rajabhat University are acknowledged for financial support of this research.

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