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Contributed Paper

Antioxidant Compounds and Activities in Selected Fresh and Blanched Vegetables from Northeastern Thailand

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ABSTRACT

This investigation aimed to identify the effect of blanching on 30 edible leaves of vegetables from northeastern Thailand on the total phenol content (TPC), total flavonoid content (TFC), 2, 2-diphenyl-1-picrylhydrazyl (DPPH), ferric reducing antioxidant potential (FRAP), and β-carotene bleaching activity. The results showed that blanching caused a significant ($p<0.05$) increase in TPC [fresh (0.39-0.85g/100g), blanched (0.60-1.84 g/100g)] and decrease in TPC [fresh (0.58-2.83 g/100g), blanched (0.27-2.58 g/100g)]. Interestingly, fresh and blanched *Oxystelma esculentum* reveal TPC and FRAP assay had high relationship ($R^2= 0.7423$, $R^2= 0.6908$). Blanching caused an increase or decrease in the antioxidant compounds and antioxidant activity depending on the specific vegetables. A highly positive correlation between TPC and FRAP in fresh and blanched vegetables was found. These results will be usefully when revising guidelines on the beneficial properties of local vegetables from the northeastern region of Thailand.

Keywords: antioxidant, vegetable, blanching

1. INTRODUCTION

Recently, phytochemicals in vegetables have received a great deal of attention focused on their disease preventing properties. Free radicals in humans might be the cause of various diseases, as they are involved in many organelle/cell function disorders including cardiovascular malfunctions, tissue injury, DNA damage tumor promotion, and especially cancer [1,2]. However, the harm caused by free radicals could be inhibited by antioxidant compounds. Many studies have suggested that antioxidants can block free radicals, protect cells against oxidative stress, which induces cell damage, and prevent diseases. These health benefits are attributed to the antioxidant activity derived from the phytochemicals present in vegetables [3].

Edible vegetables are widely produced in Thailand for both local consumption and non-local trade. Thailand is a country that presents great agro-ecological diversity and a large variety of vegetables. In addition,

traditional Thai foods are of interested to health-conscious consumers. Daily consumption of vegetables for disease risk reduction and to treat several chronic diseases is recommended by nutritionist [4]. Vitamins and phytochemicals, such as ascorbic acid, phenolic compounds, and fiber have been determined to be the bioactive compounds responsible for the antioxidant properties. Their presence in higher plants via in vitro experiments has been illustrated, which showed reduced oxidation damage by inhibiting or quenching free radicals and reactive oxygen molecule species. Their roles are similar to synthetic antioxidant structures [5].

Food processing methods involve mostly heating: mainly blanching, steaming or soaking, before consuming. Blanched vegetables are specially heated to modify the texture, color, flavor, nutritional value, and to inactivate enzymes for preservation. Generally in Thailand, most vegetables are actually blanched before being consumed in order to reduce or eliminate the bitterness of taste, make the vegetables clean, and maintain acid components [6]. Interestingly, many researches have reported that blanching caused many changes in the chemical composition of vegetables, such as their antioxidant capacity and total phenolic content [7]. For example, Sahlin et al. [8] showed that the blanching of tomatoes (*Lycopersicon esculentum*) caused a decrease in total phenolic content and antioxidant capacity.

People in each region of Thailand (northern, northeastern, central, or southern part) gather their own local vegetables to eat. Thai regional foods are often either served raw or cooked, generally blanched. There is limited information relating to the antioxidant compounds and antioxidant activities of these vegetables after blanching. Therefore, the objective was to determine the difference between fresh and blanched) selected vegetables from northeastern Thailand on antioxidant compounds and antioxidant activity.

2. MATERIALS AND METHODS

2.1 Vegetable Material

All 30 vegetables (Table 1) were obtained from four municipal fresh markets in different provinces (Loai, Roi Et, Nakhon Phanom, and Khonkean) of Thailand during April-August 2014. For each vegetable, one kilogram was selected from each market and divided into equal parts (200 grams) for ingredient investigation. They were identified from their botanical morphology (by Biology Department, Mahasarakham University), and the nomenclature that was used in the study is shown in Table 1.

2.2 Chemicals and Reagents

1, 1-Diphenyl-2-picrylhydrazyl (DPPH), 2, 4, 6-tris(2-pyridyl)-Striazine (TPTZ), Folin-Ciocalteu reagent, and gallic acid were obtained from Sigma Chemical Co. (St.Louis, MO,USA), and Catechin from Fluka (Neu-Ulm, Germany). All chemicals and reagents used in the study were of analytical grade.

2.3 Sample Preparation

The blanching processing was as follows: 100 g of the edible portion of the leaves were soaked in an aluminum pot containing 500 ml of boiling water for three min, left at room temperature for awhile and then they were placed in a hot air oven at 50 °C until dryness. All samples, after drying, had water contents below 10%. They were ground to a fine powder in a blender and kept in desiccators prior to extraction.

2.4 Extraction Method

The extraction method was performed on 0.5 g of leaf powder, which was soaked in flasks containing 10 ml ethanol overnight at room temperature. The suspension was filtrated with Whatman No.42 filter paper, and the residue was washed with ethanol. The insoluble residue was discarded. The filtrate was evaporated in a water bath at 50 °C to a final volume of 1 ml. The evaporated residues were used for the analysis of the antioxidant compounds and activity.

2.5 Determination of Total Phenolic Contents (TPC)

The amount of TPC in the samples was determined according to the Folin-Ciocalteu procedure [9]. Samples were mixed with 0.2 ml of 2 M Folin-Ciocalteu's reagent and 2 ml of 7.5% sodium carbonate; then each mixture was left for 30 min at room temperature. Absorbance was measured using a spectrophotometer at 725 nm. Gallic acid was used as the reference standard, and the results were expressed as g gallic acid equivalents.

2.6 Determination of Total Flavonoid Content (TFC)

The total flavonoid content was determined using the colorimetric method described by Bakar et al. [10]. Briefly, 0.5 ml of each sample was mixed with 2.25 ml of distilled water in a test tube, followed by the addition of 0.15 ml of 5% $NaNO_2$ solution. After 6 min, 0.3 ml of a 10% $AlCl_3 \cdot 6H_20$ solution was added and allowed to stand for another 5 min before 1.0 ml of 1 M NaOH was added. The mixture was mixed well with a vortex. The absorbance was measured immediately at 510 nm using a spectrophotometer. The results were expressed as mg catechin equivalents.

Table 1. Vegetables used in study.

Family name	Scientific name	Vernacular name	Traditional Uses
Acanthaceae	*Justicia gangetica* L.	Aom Sap	Body and blood tonic, eye nourishing
Amaranthaceae	*Alternanthera philoxeroides* Griseb.	PhakPed	Female blood tonic and mild laxative
Anacardiaceae	*Schinus terebinthifolius* Raddi	Matumsa-u	Antibacterial, antiviral, diuretic, digestive stimulant, and tonic
Apocynaceae	*Aganonerion polymorphum* Pierre	Somlom	Anti-tussive, expectorant, and use for muscle pain
Araceae	*Lasia spinosa* Thwaites	Nham	Anti-tussive and antipyretic
Asclepiadaceae	*Dregea volubilis* Benth. ex	PhakHuan Moo	Relieve fever and promote urination
Asclepiadaceae	*Oxystelma esculentum* (L.P.) Sm.	Jamukplalai	Anti-tussive and relieve throat pain
Asteraceae	*Acmella oleracea* (L.) R.K. Jansen	PhakCrard	To relieve toothache
Saururaceae	*Houttuynia cordata* Thunb.	PluKaow	Relieve fever, reducing swelling, and diuretic

Table 1. Continued.

Family name	Scientific name	Vernacular name	Traditional Uses
Brassicaceae	*Brassica juncea* (L.) Czen.	PhakKardHeen	To relive symptoms of diabetic cataract, anti-nociceptive, and anti-hyperglycemic
Compositae	*Pluchea indica* (L.) Less.	Khlu	Anti-hemorrhoids, astringent, antipyretic, astringent, and diuretic
Cucurbitaceae	*Sechium edule* Sw.	Ma Ra Wan	Diuretic and anti-inflammatory
Euphorbiaceae	*Sauropus androgynous* Roxb.	PhakWanBan	To relieve fever, urinary problems, and earache
Gentianaceae	*Fagraea fragrans* Roxb.	MunPla/Kankrao	To treat dysentery
Lamiaceae	*Coleus amboinicus* Lour.	Hu Sua	To relieve cough and appetizer
Lecythidaceae	*Careyas phaerica* Roxb.	Phak–Kradon	To relieve cough and astringent
Leguminosae	*Caesalpinia mimosoides* Lam.	Cha-Lueat/Kaya	Body tonic**,** appetizer, and anti-vertigo
Lemnaceae	*Wolffia globosa* (Roxb.)	Pham	Appetizer
Limnocharitaceae	*Limnocharis flava* Buchenau	Kun jong	Digestive tonic, restorative, and appetizer
Myrtaceae	*Syzygium gratum* Wall.	PhakMek	Treatment of dyspepsia, an dindigestion
Olacaceae	*Erythropalum scandens* Blume	PhakKeeNark	Anti-inflammatory
Passifloraceae	*Adenia viridiflora* Craib	PhakE-noon/ PhakSarb	-
Rhamnaceae	*Colubrina asiatica* (L.) Brongn.	PhakKarnToeng	Appetizer
Rubiaceae	*Paederia linearis* Hook	KruaTodMa	Carminative and to relieve headache
Rutaceae	*Feroniella lucida* Swingle.	Ma sang	Digestive tonic, carminative, and relieves flatulence
Scrophulariaceae	*Limnophila aromatica* Merr.	PhakKaYeang	Diuretic, muscle relaxant, and antispasmodic
Umbelliferae	*Anethum graveolens* L.	PhakChi Lao	Digestive tonic and carminative
Umbelliferae	*Oenanthe stolonifera* Wall.	PhakCheeLom	Expectorant and anti-asthmatic

Table 1. Continued.

Family name	Scientific name	Vernacular name	Traditional Uses
Umbelliferae	*Trachyspermum roxburghianum* H. Wolff	Sa Ngae	Digestive tonic, anti-flatulence, and to nourish the heart
Vitaceae	*Cissus quadrangularis* L.	Phetsangkhat	Anti-hemorrhoid and anti-hypertensive

10% $AlCl_3 \cdot 6H_20$ solution was added and allowed to stand for another 5 min before 1.0 ml of 1 M NaOH was added. The mixture was mixed well with a vortex. The absorbance was measured immediately at 510 nm using a spectrophotometer. The results were expressed as mg catechin equivalents.

2.7 Determination of Antioxidant Activity

- DPPH radical scavenging activity

Radical scavenging activity was determined using stable 2,2-diphenyl-1-picrylhydrazyl (DPPH) according to the modified method of Brand-Williams et al. [11]. In brief, the samples (0.2 ml) were mixed with a 0.20 mM DPPH ethanol solution for 2 ml. After incubation at room temperature in the dark for 30 min, the mixture was measured at the absorbance of 517 nm using a spectrophotometer. The standard curve was linear between 0.08 and 0.64 mM Trolox. The radical scavenging activity was calculated as a percentage of DPPH scavenging activity using the following equation: % scavenging activity $= 100 \times [1- (A_E/A_D)]$, where A_E is the absorbance of the DPPH solution with the extract added and A_D is the absorbance of the DPPH solution with nothing added.

- Ferric reducing antioxidant potential (FRAP) assay

The FRAP assay was conducted according to Benzie and Strain [12], with some modifications. The stock solutions included 300 mM acetate buffer (3.1 g $C_2H_3NaO_2 \cdot 3H_2O$ and 16 ml $C_2H_4O_2$) pH 3.6, 10 mM TPTZ (2,4,6-tripyridyl-s-triazine) solution in 40 mM HCl, and 20 mM $FeCl_3 \cdot 6H_2O$ solution. The fresh working FRAP solution was prepared by mixing 10 ml acetate buffer, 1 ml TPTZ solution, and 1 ml $FeCl_3 \cdot 6H_2O$ solution and then warming the solution to 37°C before use. The samples (0.15 ml) were allowed to react with of the FRAP solution (2.85 ml) for 30 min in the dark. Spectrophotometry readings of the coloured product (ferrous tripyridyltriazine complex) were then taken at 593 nm.

-β- Carotene bleaching activity

The determination of the antioxidant activity was performed according to Amin and Tan [13]. Antioxidant activity is expressed as the ability to delay the bleaching of a β-carotene/linoleic acid emulsion. To prepare the β-carotene emulsion, 0.1 ml of β-carotene solution (0.1 mg/ml in chloroform) was transferred to a beaker containing linoleic acid (20 µl) and Tween 40 (200 µl). The mixture was evaporated at 50°C for 10 minutes to remove the solvent, and distilled water (100 ml) was immediately added. The β-carotene emulsion (5.0 ml) was then transferred to a test tube containing the test sample (0.2 ml). The mixture was shaken and placed in a water bath at 50°C for 2 h before its absorbance was measured at 470 nm.

The antioxidant index was calculated in terms of β-carotene bleaching using the equation: Antioxidant index = $[1-(A_0 - A_t)/(A^o_0 - A^o_t)] \times 100\%$, where A_0 and $A^o_0$are the absorbance values measured at the initial incubation time for the samples and control, respectively, while A_t and A^o_t are the absorbance values measured in the samples or the standard and control at t = 120 min.

2.8 Statistical Analysis

The antioxidant compounds and antioxidant activity results statistically analysed by each values from experiments performed were triplicate and defined as mean ($\bar{x}$) ± standard deviation (SD). Mann-Whitney test was applied to interpreted statistically significance with p values at 0.05 cut of level.

3. RESULTS AND DISCUSSION

3.1 TPC

Phenolic compounds are phytochemicals found in all plants. These compounds are molecules that can act as antioxidants to prevent several diseases, such as heart disease, and lower the incidence of cancers and diabetes [3]. Not only the different types of plant have different TPCs, but also every plant extract contained a higher TPC than TFC, due to flavonoids being phenolic compounds [14].

The TPCs of the vegetables are displayed in Table 2. They were in the range of 0.39-2.83 and 0.27-2.83 g/100 g dry weight for the fresh and blanched vegetables, respectively. While fresh and blanched *Oxystelma esculentum* found highest TPC. The ethanol extracts were found between the TPC values for fresh and blanched vegetables. For instance blanched *Trachyspermum roxburghianum* (+54%) and *Lasia spinosa* (+45%) showed an increase in TPC ($p<0.05$), while blanched *Brassica juncea* (-63%) and *Careyas phaerica* (-74%) were found decrease ($p<0.05$) (Table 2). It was note that blanching exhibited different effects on different vegetables, with some vegetables found increase TPC while in others plant was decreased.

This result showed that 23% of vegetables had increased TPC after blanching. Our finding correlates with Turkmen et al. [15], who showed that TPC of pepper,squash, green beans, peas, leek, broccoli, and spinach increased after conventional cooking methods (boiling, steaming, or microwaving), and they illustrated that cooking modified the bound phenolic compounds to be free forms released into the cytosol. Interestingly, Hyo et al. [16] mentioned that phenolic compounds increased due to the release of phytochemicals. The thermal processing disrupts the cell membranes and cell walls which was release the soluble phenolic compound into cytosol [17]. Therefore, the blanching process was increased the amount of TPC.

Table 2. Antioxidant compounds of fresh and blanched vegetables.

Vegetable	TPC (g/100g)		TFC (mg/100g)	
	Fresh	Blanched	Fresh	Blanched
Justicia gangetica	0.58 ± 0.01*	0.60 ± 0.03*	245.47 ± 9.27*	179.42 ± 9.11*
Alternanthera philoxeroides	1.59 ± 0.02*	0.72 ± 0.02*	266.37 ± 7.03*	174.45 ± 1.12*
Schinus terebinthifolius	2.78 ± 0.17	2.79 ± 2.60	166.65 ± 4.80*	312.38 ± 9.75*
Aganonerion polymorphum	2.81 ± 0.02*	2.58 ± 0.01*	407.12 ± 7.83*	205.31 ± 6.40*
Lasia spinosa	0.39 ± 0.01*	0.71 ± 0.04*	59.70 ± 8.63	67.61 ± 5.12
Dregea volubillis	2.79 ± 0.06*	2.39 ± 0.04*	332.96 ± 3.04*	135.78 ± 8.79*
Oxystelma esculentum	2.83 ± 0.01	2.83 ± 0.02	2.21 ± 0.42	1.99 ± 0.89
Acmella oleracea	0.71 ± 0.14*	0.54 ± 0.03*	198.30 ± 8.63	198.53 ± 7.35

Table 2. Continued.

Vegetable	TPC (g/100g)		TFC (mg/100g)	
	Fresh	Blanched	Fresh	Blanched
Houttuynia cordata	1.27 ± 0.02*	0.63 ± 0.01*	87.39 ± 9.11	56.64 ± 7.83
Brassica juncea	1.26 ± 0.05*	0.47 ± 0.04*	225.78 ± 7.51*	126.06 ± 12.6*
Pluchea indica	0.59 ± 0.02*	0.86 ± 0.03*	120.75 ± 9.60	115.66 ± 9.11
Sechium edule	0.84 ± 0.08	0.90 ± 0.01	160.54 ± 4.48*	266.93 ± 7.20*
Sauropus androgynous	1.58 ± 0.01*	1.41 ± 0.05*	425.21 ± 9.11*	370.94 ± 2.08*
Fagraea fragrans	2.82 ± 0.01	2.82 ± 0.01	462.42 ± 7.51*	362.46 ± 4.96*
Coleus amboinicus	1.40 ± 0.16	1.32 ± 0.02	225.67 ± 3.84	226.23 ± 9.11
Careyas phaerica	2.14 ± 0.07*	0.56 ± 0.05*	26.34 ± 0.16*	10.63 ± 5.76*
Caesalpinia mimosoides	2.76 ± 0.01*	1.27 ± 0.01*	31.54 ± 4.64*	7.35 ± 1.12*
Wolffia globosa	1.24 ± 0.01*	0.77 ± 0.01*	252.23 ± 3.04*	202.04 ± 9.75*
Limnocharis flava	1.33 ± 0.01*	0.71 ± 0.03*	312.38 ± 7.83*	270.55 ± 4.00*
Syzygium gratum	2.83 ± 0.01*	2.66 ± 0.05*	296.10 ± 8.80*	131.49 ± 4.00*
Erythropalum scandens	0.75 ± 0.01*	0.62 ± 0.02*	60.26 ± 2.08*	56.42 ± 1.44*
Adenia viridiflora	1.31 ± 0.02*	0.75 ± 0.01*	135.22 ± 4.48*	40.14 ± 5.28*
Colubrina asiatica	1.42 ± 0.01*	0.85 ± 0.02*	289.20 ± 5.12*	59.24 ± 7.67*
Paederia linearis	0.48 ± 0.01*	0.34 ± 0.07*	87.73 ± 3.84	78.24 ± 5.44
Feroniella lucida	2.80 ± 0.01*	2.16 ± 0.05*	86.26 ± 7.84	80.16 ± 2.40
Limnophila aromatic	2.66 ± 0.01*	1.37 ± 0.01*	369.81 ± 9.75*	198.19 ± 9.75*
Anethum graveolens	1.65 ± 0.01*	1.03 ± 0.13*	234.26 ± 9.27*	196.50 ± 6.08*
Oenanthe stolonifera	0.60 ± 0.01*	0.83 ± 0.06*	202.71 ± 8.80*	436.86 ± 7.35*
Trachyspermum roxburghianum	0.85 ± 0.01*	1.84 ± 0.05*	170.27 ± 4.80	195.36 ± 7.67
Cissus quadrangularis	0.58 ± 0.01*	0.27 ± 0.01*	71.00 ± 0.32*	33.13 ± 0.48*

*significant at $\alpha < 0.05$ for Mann-Whitney test comparing fresh and blanched with values are expressed as mean ± SD of triplicate measurement.

On the contrary, other 80% vegetables decreased their TPC after blanching, which phenolic compounds being sensitive to heating process, vegetable should be blanched for a few minutes may have a significant loss of TPC which is leached into the boiling water [18]. Also the blanching causes solubilitization of the phenolic compounds and leads to a loss of TPC [19].

3.2 TFC

Flavonoids are one of the most common phenolic compounds and groups in plant tissues, and are often responsible, alongside carotenoids and chlorophylls, for color in plants [20]. TFCs had similar result as TPC, in which TFC depended on the type of vegetable. TFCs were in the ranges of 2.21-462.42 and 1.99-436.86 mg/100 g dry weight in fresh and blanched vegetables, respectively. Significant differences were detected among fresh and blanched vegetables. Fresh *Fagraea fragans* showed the highest TFC (462.42±7.51 mg/100g dry weight), while blanched *Oxystelma esculentum* had the lowest content (1.99±0.89 mg/100 g dry weight)(Table 2).However, the vegetables with lower flavonoid contents did not always have a lower phenolic content, as was evident for *Oxystelma esculentum* (2.21±0.42 mg/100 g dry weight), which had a lower TFC value compared with that of *Syzygium gratum* (296.10±8.80 mg/100

g dry weight), although the TPC was higher (2.83±0.01 g/100 g dry weight).

The results showed that blanching caused different effects on different vegetables, with some vegetables exhibiting increased TFC while in others it was decreased. The result is in agreement with Andrea et al. [21], who reported that compared to the raw, short and extended water-blanching times for leaves and fruits resulted in a strong increase in the contents of phenolic acids and flavonoids. Sakihama et al. [22] explained that flavonoids normally accumulate in the epidermal cells of plant organs, and the release of flavonoids and increased chemical extraction of these compounds could be induced by the blanching. Therefore, it is suggested that these flavonoids are released efficiently from complex plant tissues by blanching. In contrast, there were some vegetables with a decrease in TFC due to blanching. According to Oboh et al. [23], the TFC of the leaf of *Amaranthus cruentus* when blanched was lower than in the unprocessed, and they explained that during blanching some of the flavonoids would be leached into the water. Amin *et al*. [24] reported that heat treatment can induce significant changes in the phytochemicals, such as phenolic content and flavonoid content, which could be due to thermal degradation, diffusion, or/and leaching. Thus, this is a possible reason as to why flavonoids are leached into the blanching water and their breakdown during processing.

3.3 Antioxidant Activity

DPPH radical scavenging activity

The DPPH assay is used to investigate the antioxidant potential of extracts, and it is based on the scavenging of the stable DPPH by an antioxidant [25]. The percent inhibition (DPPH radical scavenging activity) of the vegetables covered a wide range from 1 in blanched *Limnocharis flava* to 88% in blanched *Careyas phaerica*. Whenever fresh *Alternanthera philo-xeroides* gave highest percent inhibition (58.06%) while blanched *Careya spaerica* gave lowest percent inhibition (87.58%). As observed from the result, about 30% of the vegetables had significantly ($p < 0.05$) increased ability when they were blanched. Meanwhile, about 43% had a significant ($p < 0.05$) decrease in antioxidant activity after blanching. Especially, *Sechium edule* (-93%) and *Aganonerion polymorphum* (-92%) had the greatest decline.

FRAP Assay

The ferric reducing properties of the vegetables are displayed in Table 3. The FRAP values were in the ranges of 29.33-143.70 and 20.60-143.67 mmol $FeSO_4$/100 g dry weight in fresh and blanched vegetables, respectively. The fresh and blanched *Oxystelma esculentum* had the highest FRAP values of 143.70 and 143.67 mmol $FeSO_4$/100 g dry weight (no difference at $p>0.05$), followed by fresh *Dregea volubillis* (143.43 mmol $FeSO_4$/100g dry weight) and fresh *Aganonerion polymorphum* (142.60 mmol $FeSO_4$/100 g dry weight).The results showed that approximately 27% were significantly ($p< 0.05$) increased after blanching. Interestingly, the blanched *Trachyspermum roxburghianum* had significantly ($p < 0.05$) increased FRAP values of about 65%.

β- Carotene bleaching activity

This method measures the antioxidant activity via the capability to delay the bleaching of β-carotene in a water/linoleic acid emulsion [26]. The results are summarized in Table 3. It was found that the extracts of the fresh and blanched vegetables exhibited antioxidant activity. Stronger activity is displayed by a higher antioxidant index. The extract of the fresh *Pluchea indica* showed the highest level of antioxidant activity, with an index of 84%, followed by fresh *Limnocharis flava* (70%) and fresh *Schinus terebinthifolius*

(67%). We found that the antioxidant indexes of 23 % of the blanched vegetables demonstrated higher antioxidant activity compared to the fresh vegetables. Meanwhile, the antioxidant indexes of *Aganonerion polymorphum*, *Houttuynia cordata* and *Wolffia globosa* were lost after blanching. About seven vegetables did not bleach β-carotene.

Table 3. Antioxidant activity of fresh vegetables and after thermal process.

Vegetable	DPPH (% scavenging activity)		FRAP (mmol $FeSO_4$/100 g dry weight)		β- Carotene bleaching activity (%)	
	Fresh	Blanched	Fresh	Blanched	Fresh	Blanched
Justicia gangetica	14.70 ± 0.02*	6.42 ± 0.72*	124.40 ± 1.41*	58.53 ± 0.94*	2.14 ± 0.77*	14.55 ± 3.06*
Alternanthera philoxeroides	58.06 ± 0.73*	26.62 ± 0.07*	108.80 ± 8.71*	48.73 ± 6.60*	4.64 ± 0.57	3.41 ± 0.82
Schinus terebinthifolius	48.14 ± 0.15*	22.73 ± 1.17*	141.20 ± 0.47*	141.63 ± 0.24*	66.87 ± 3.17*	34.06 ± 8.78*
Aganonerion polymorphum	23.99 ± 2.61*	1.99 ± 0.76*	143.23 ± 1.04*	142.60 ± 0.47*	18.89 + 8.32	ND
Lasia spinosa	44.02 ± 1.73	47.68 ± 0.58	29.33 ± 6.60*	40.63 ± 5.90*	ND	ND
Dregea volubillis	32.23 ± 0.88*	22.45 ± 0.95*	143.43 ± 0.24*	133.10 ± 5.89*	12.07 ± 4.82	25.39 ± 7.88
Oxystelma esculentum	6.67 ± 0.66*	17.22 ± 1.57*	143.70 ± 0.24	143.67 ± 0.00	12.07 ± 3.06	13.62 ± 0.04
Acmella oleracea	47.45 ± 0.33*	64.47 ± 0.78*	35.20 ± 4.24*	41.80 ± 6.60*	11.15 ± 7.44	34.37 ± 9.19
Houttuynia cordata	41.50 ± 2.19*	17.58 ± 0.46*	90.80 ± 6.60*	56.63 ± 9.19*	16.41 ± 5.25	ND
Brassica juncea	57.23 ± 2.16*	55.19 ± 9.75*	71.73 ± 6.13*	41.67 ± 7.54*	ND	ND
Pluchea indica	33.95 ± 8.35*	35.86 ± 1.04*	40.60 ± 1.89*	71.60 ± 8.01*	84.46 ± 8.32*	64.67 ± 9.20*
Sechium edule	51.02 ± 0.39*	3.57 ± 0.78*	60.43 ± 8.01*	105.90 ± 9.66*	41.80 ± 7.44*	23.22 ± 3.94*
Sauropus androgynous	43.77 ± 2.80*	32.26 ± 0.20*	121.63 ± 9.20*	108.87 ± 6.13*	ND	ND
Fagraea fragrans	2.82 ± 1.11*	7.19 ± 0.17*	142.03 ± 0.24*	142.40 ± 0.47*	ND	ND
Coleus amboinicus	2.52 ± 0.13*	20.89 ± 0.81*	126.50 ± 6.84	124.60 ± 8.49	15.17 ± 3.32*	3.41 ± 0.44*
Careya sphaerica	16.74 ± 1.50*	87.58 ± 0.78*	130.30 ± 4.95*	30.57 ± 2.60*	12.07 ± 3.67*	3.41 ± 1.19*
Caesalpinia mimosoides	21.32 ± 1.84	23.54 ± 1.15	140.93 ± 0.10*	96.30 ± 9.19*	62.85 ± 6.57*	27.74 ± 3.06*
Wolffia globosa	28.78 ± 1.83*	22.29 ± 1.24*	94.90 ± 8.25*	39.06 ± 7.07*	7.43 ± 0.44	-
Limnocharis flava	8.11 ± 1.24*	1.11 ± 0.78*	112.87 ± 6.28	43.70 ± 3.54	70.00 ± 7.88*	40.02 ± 0.88*
Syzygium gratum	52.84 ± 3.87*	33.85 ± 0.95*	143.54 ± 0.24*	142.90 ± 0.24*	12.70 ± 4.44	18.89 ± 3.94
Erythropalum scandens	30.29 ± 3.41*	18.35 ± 0.11*	49.53 ± 7.54	50.03 ± 9.20	50.46 ± 8.31	52.94 ± 2.19
Adenia viridiflora	55.45 ± 1.78*	17.52 ± 1.61*	111.30 ± 6.84*	44.90 ± 2.60*	ND	ND
Colubrina asiatica	39.84 ± 2.22*	5.50 ± 0.20*	97.33 ± 8.01*	69.37 ± 8.72*	47.68 ± 7.88	32.20 ± 6.18
Paederia linearis	10.52 ± 2.74*	3.79 ± 1.61*	51.33 ± 8.49*	20.60 ± 3.77*	ND	ND
Feroniella lucida	8.35 ± 2.15*	14.58 ± 0.26*	90.67 ± 9.90*	51.37 ± 2.12*	20.43 ± 7.44*	36.84 ± 2.19*
Limnophila aromatic	2.82 ± 1.11*	10.78 ± 0.17*	140.33 ± 1.41*	92.17 ± 2.59*	51.70 ± 7.44	30.65 ± 0.43
Anethum graveolens	52.35 ± 1.89	48.11 ± 0.06	115.13 ± 9.43*	101.33 ± 9.43*	ND	ND
Oenanthe stolonifera	48.01 ± 0.46*	33.90 ± 1.15*	61.87 ± 5.19*	83.00 ± 7.07*	11.46 ± 4.44	9.60 ± 3.32
Trachyspermum roxburghianum	28.76 ± 1.32	18.85 ± 0.17	43.97 ± 6.84*	124.33 ± 7.54*	7.43 ± 5.25	3.41 ± 2.19
Cissus quadrangularis	26.78 ± 0.46	34.48 ± 4.76	30.80 ± 8.96*	17.67 ± 8.01*	47.68 ± 6.57	10.53 ± 1.75

*significant at $\alpha < 0.05$ for Mann-Whitney test comparing fresh and blanched with values are expressed as mean ± SD of triplicate measurement

ND: Not detected

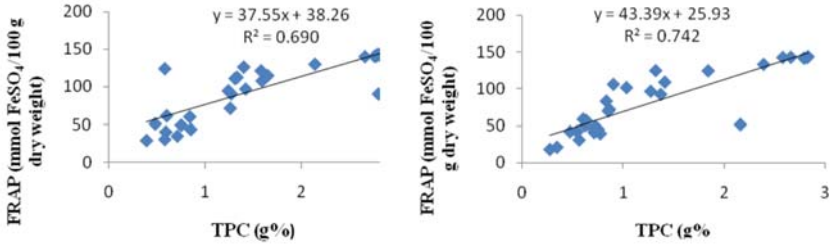

Figure 1. Correlation between antioxidant compounds (TPC) and antioxidant activity (FRAP) of (A) fresh and (B) blanched vegetables from northeastern Thailand.

Many other researchers have shown that blanching vegetables had different effects on their antioxidant activity. Some research foundthat blanched vegetable affected to antioxidant activity was significantly different, in which they exhibited to increase or decrease in antioxidant activity.

All the results obtained in this work depend on many factors, such as kind of vegetable, genotype, agronomic practices, maturity level at harvest, post-harvest storage, climate, and geographical location (northeastern Thailand) [27, 28]. In summary, increased antioxidant activity occurred due to the following:

(i) Blanching caused the cell wall to breakdown and release bound antioxidant compounds (TPC and TFC) from the vegetable matrix, which results in an increase in antioxidant activity [9, 17].

(ii) The release of bound phenolic compounds from the breakdown of cellular constituents and cell walls as well as being a result of polymerization, aglycosylation , and/ or oxidation of the phenolics [7, 10]. In the case of a decreased, it is assumed that it is possible the blanching leaches out the water-soluble phenolics into the water; in addition there was heat degradation of phenolic compounds during blanching. Especially, *Brassica juncea* and *Careyas phaerica* found decreased antioxidant activity after blanching might be two type of plant have thin cell structure.

We found that only TPC and antioxidant activity during blanching exhibited a high positive correlation by the FRAP assay. It was found in the blanched vegetables that the TPC and FRAP assays had the highest correlation (R^2= 0.7423), followed by the fresh (R^2= 0.6908) (Figure. 1). Not similarity of Pukumpuang et al. [29] study that showed flavonoids in *Eclipta prostrate* responsible for antioxidant activity.

This suggests that phenolic compounds are activated during the reduction of Fe^{3+} to Fe^{2+} and the formation of a colored Fe^{2+} complex [30]. The findings from this study assumed that phenolic compounds could be phytochemicals with antioxidant activity from the blanched vegetables.

4. CONCLUSIONS

In conclusion, the results from the 30 fresh and blanched vegetables from northeastern Thailand indicate that are a rich source of phytochemicals and have antioxidant activities. Blanching of the vegetables might increase the phenolic compounds that have antioxidant properties, while also improving the taste and cleanness of the vegetables [4]. Even though, some blanched vegetables have loss of dietary antioxidant. In addition, this study offers useful recommendations for consumers primarily about selecting vegetables that are sources of phytochemicals and cooking method for blanching might seem to be some type of plant sensible for heating with increasing antioxidant compound and activity.

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