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Phenolics, Flavonoids and Antioxidant Activity of Vegetables as Thai Side Dish

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Abstract

The total phenolics, flavonoids and antioxidant activity of 21 selected kinds of Thai side dish vegetables were determined. The result showed that all kinds of Thai side dish vegetables contained phenolic and flavonoid compounds. The Thai side dish vegetables that contained the highest phenolic compound was Pak bung tai muang, 1.66±0.006 mgGAE/g. As Ma khua puang and Kra jeab kheaw contained the highest flavonoid compound (0.14±0.001 mgRE/g). In the part of antioxidant activity, Kha-on displayed the highest antioxidant activity with 91.99±1.36 %inhibit by DPPH assay and 1.65±0.03 mM $FeSO_4$ by FRAP assay method. Although this result revealed that Thai side dish vegetables may not high source of phenoloic and flavonoid compounds, they showed high efficiency of antioxidation power. It was suggested that Thai side dish vegetables would become new good source and natural of phenolic and flavonoid compounds because Thai people generally ate most of all vegetables as side dish in fresh cut and scald so phenolics and flavonoids were not destroyed from cooking process. In addition, they usually consumed Thai side dish fresh cut vegetables at least 100 gram/time/person which were benefit for the human health.

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Keywords: Antioxidant activity, Total phenolic content, Total flavonoid content, Thai Side Dish vegetables

1. Introduction

In every Thai native meals there are some kinds of vegetables or some styles of vegetables in set menu.

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Because vegetables can be cooked in difference styles of Thai food such as fried, boil and more. The most interesting vegetables of Thai people is fresh cut as side dish.[1],[2] Side dish means using vegetables in fresh cut for decreasing spicy taste of Thai food menu such as Namprick (a sauce of shrimp paste and chili) or Larb (pork mince with spicy Thai herb).[1],[2] It was shown the Thai side dish vegetables played the important role in Thai food.[3] Some kinds of vegetables, winged bean, yard long bean, and eggplant etc, can be consumed as side dish.[1],[2],[3] Thai people usually consumed large amount of varieties side dish fresh cut vegetables for 1 meal at least 100 gram per person per time.[4]

However, few researches have been an interest in important functional food components of Thai side dish vegetables. Therefore the study of such benefits of fresh vegetables is useful and important. Landrum and Bone [5] reported that vegetables and fruits were a good source of minerals, vitamins and phytochemicals. And two groups of phytochemical were phenolic and flavonoid compounds known as antioxidant agent.[5],[6] This agent was benefit for people health. Numerous researches have shown that high concentrations of phenolics and flavonoids were significantly associated with reduced risk of cardiovascular disease (CVD) through an improvement in vascular function and a modulation of inflammation.[3],[4],[7],[8] Moreover, they could protect cell and tissue damage from free radicals by potent antioxidant capacity.[6],[9],[10] Antioxidant activity study was one good way that could show the protective effects against cancer of antioxidant agent on free radicals. This essay showed reducing power and could measure the radical scavenging activity of antioxidants against free radicals.[7],[8] Therefore, the study of antioxidant capacity in fresh cut vegetables could claim direct effect of fresh cut vegetables on people health.

Although Thailand possesses a great edible plant biodiversity, there were not previous reports on their antioxidant activity. This research was carried out with an aim to evaluated study of antioxidant activity, total phenolics and flavonoid contents of selected vegetables as Thai side dish. It could be used as fundamental data for nutritionists or public health workers to recommend consumers to select appropriate types of Thai side dish vegetables for their health needs.

2. Material and Method

2.1. Plant materials

Twenty-one types of popular Thai side dish vegetables were selected for evaluation antioxidant capacity, total phenolic and total flavonoid compound contents. Fresh vegetables were purchased from the local market of Mahasarakham province, Thailand. All of vegetables were cleaned and trimmed of only edible portions before they were used and analysed. The common names, scientific names of these vegetables and edible portions were given in Table 1.

2.2. Determination of total phenolics

The total phenolic contents of all plant extracts were determined by using Folin-Ciocalteu reagent, based on the method described by Singlaton and Rossi(1965).[11] One millilitre of extract solution was added into a flask containing 9 ml of distilled water. The extract solution was thoroughly mixed with 1 ml of Folin-Ciocalteu's phenol reagent. After 5 min, 10 ml of 7% Na_2CO_3 was added. The mixture was further shaken and made up to 25 ml with the addition of 4 ml of distilled water. Then, the absorbance was measured at 750 nm after 90 min of incubation at room temperature using spectrophotometer. The total phenolic acid content was expressed as mg of gallic acid equivalents (GAE)/g fresh matter of vegetable (mg/g sample).

2.3. Determination of total flavonoids

Total flavonoid contents of all vegetable extracts were determined according as aluminium chloride colorimetric method described by Marinova et al. (2005).[12] Briefly, a 1 millilitre sample of the vegetable extract was pipetted into a flask containing 4 ml of distilled water and mixed with 0.3 ml 5% $NaNO_2$. After 5 min, 0.3 ml of 10% $AlCl_3$ was added. This mixture was mixed with 2 ml of 1 M NaOH after standing for another 6 min. Then, the mixture volume was adjusted to 10 ml by adding distilled water. The mixture was well mixed and the reaction mixture absorbance was measured at 510 nm. The results were expressed as mg of rutin equivalent (RE)/g fresh matter of vegetable.

2.4. Determination of total antioxidant activity as free radical scavenging activity(DPPH)

The antioxidant activity of all extracts was evaluated through free radical scavenging effect on 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical according to Akowuah et al. (2005).[13] Two ml of 0.1mM DPPH methanolic solution was added to 200 µl of sample extracts and 0.8 ml methanol. The reaction mixture was thoroughly mixed and kept in the dark at room temperature for 1 hr. The control was prepared by mixing 2 ml of DPPH and 1 ml methanol. The absorbance was recorded at 517 nm using microplate reader spectrophotometers. The estimation of total antioxidant activity in the vegetable extracts was carried out in triplicate. The data was expressed as the inhibition percentage of DPPH scavenging activity which was calculated using the following formula

$$\% \text{ inhibition of DPPH} = [\text{Abs control} - \text{Abs sample} / \text{Abs control}] \times 100$$

2.5. Determination of ferric reducing/antioxidant power assay (FRAP)

The FRAP assay was carried out according to the method of Benzie and Strain(1996).[14] FRAP reagent was prepared from acetate buffer (1.6 g sodium acetate and 8 ml acetic acid make up to 500 ml) (pH 3.6), 10 mM TPTZ solution in 40 mM HCL and 20 mM iron (III) chloride solution in proportion of 10:1:1 (v/v) respectively. The FRAP reagent was freshly made up daily. 50 µl sample extracts were added to a tube containing 1.5 ml of the warmed to 37 ^{o}C FRAP reagent and mixed well. The absorbance was measured at 593 nm using microplate reader spectrophotometers after 4 mins. The determinations were carried out in triplicate. Standard curves were made for each assay using iron (II) sulfate solution. The results were expressed as µmol of Fe (II) /100 g extract sample.

2.6. Statistical Analysis

This experiment was performed in triplicate by determination on three batches of each vegetable with completely randomized design. Data analyses of phenolic content, flavonoid content and antioxidant activity were performed by SPSS software version 13. A significant difference was considered statistically value at the confidence level of $p<0.05$.

3. Result and Discussion

In the present study, total phenolic contents, total flavonoid contents and antioxidant activity in twenty-one varieties of selected vegetables that people consumed as side dish from the market were determined. All selected vegetables might grow in all of seasons in Thailand. Thus, people could choose and buy for their meal around the year. Table 1 showed Thai common names and scientific names of selected vegetables. In

addition, the edible portions of vegetables and eating recipes that Thai people usually consumed were also shown in Table1. Due to difference in Thai native eating culture between other countries, it was important data that should be recorded. The selected varieties of vegetables that Thai people consumed in fresh cut form or they scald them for few minute in hot water before eating were presented in Table1. The objective of scald of some vegetables in Thai cuisine was decreasing some smell or increasing soft texture. Moreover, in traditional, the scald of vegetables used less time and was put a few salt to preserve their color.

Table 1. Thai common names, scientific names, edible portions and eating recipes of Thai side dish vegetables for analysis.

Thai common name	Scientific name	Edible portion	Eating recipes
Tua puu	*Psophocarpus tetragonolobus* Linn.	fruit	Fresh cut
Ma khua kai nok	*Solanum melongena L.*	fruit	Fresh cut
Ma khua puang	*Solanum torvum Sw.*	fruit	Fresh cut
Ma khua proh	*Solanum xanthocarpum* Schrad. & Wendl.	fruit	Fresh cut
Ma khua lai, Ma khua kang kob	*Solanum aculeatissimum Jacq*	fruit	Fresh cut
Pak kwang tung	*Brassica pekinensis*	leaf,flower	Fresh cut,scald
Pak bung thai khao	*Ipomcea aquatica Forsk.*	young stem	Fresh cut
Pak bung thai maung	*Ipomcea aquatica Forsk.*	young stem	Fresh cut
Pak kad khao	*Brassica chinensis* (L.) Jusl.	leaf	Fresh cut
Pak kad hom	*Lactuca sativa* (L.)	leaf	Fresh cut
Pak ka lum plee	*Brassica oleracea* L. var. capitata L.	leaf	Fresh cut,scald
Tua fak yaw	*Vigna sesquipedalis Koern.*	fruit	Fresh cut
Boub hom	*Luffa cylindrica* (L.) M.Roem.	fruit	Fresh cut,scald
Boub ngu	*Trichosanthes cucumerina* (L.)	fruit	Fresh cut,scald
Bai bua bok	*Centella asiatica* (L.) Urb.	leaf,stem	Fresh cut
Pak tum leung	*Coccinia grandis* (L.) Voigt	leaf,young stem	Scald
Bai cha plu	*Piper sarmentosum* Roxb.)	leaf	Fresh cut
Ma ra khee nok	*Momordica charantin* Linn	fruit	Fresh cut,scald
Kra jeab kheaw	*Abelmoschus esculentus* Linn.	fruit	Fresh cut,scald
Kha-on	*Alpinia nigra (Gaertn.)*B.L.Burtt	shoot	Fresh cut,scald
Pak chee lao	*Anethum graveolens* Linn.	leaf,stem	Fresh cut

The result showed that total phenolic contents in mgGAE/g and total flavonoid contents in mgRE/g in the twenty-one Thai side dish vegetables varied considerably. All selected kinds of Thai side dish vegetables contained phenolic compound and total phenolic contents ranged from 0.20±0.002 (Boub ngu) to 1.66±0. mgGAE/g (Pak bung thai maung) in different vegetable species (Table 2). Pak bung thai maung contained consistently the highest phenolic compound(1.66±0.006 mgGAE/g) and the lowest was observed in Boub ngu (0.20±0.002 mgGAE/g). And another part of this study, the total flavonoid contents in all kinds of Thai side dish vegetables varied widely between 0.04±0.001 mgRE/g(Ma khua lai and Pak ka lum plee) and 0.14±0.001(Ma khua puang and Kra jeab kheaw) (Table 2). The highest flavonoid content was found in Ma khua puang and Kra jeab keaw (0.14±0.001 mgRE/g), although the lowest flavonoid content was observed in Ma khua lai and Pak ka lum pree (0.04±0.001 mgRE/g).

The result of the antioxidant activity of the twenty-one Thai side dish vegetables by DPPH radical scavenging and FRAP assay was given in table 2. Kha-on displayed the highest antioxidant activity with 91.99±1.36 %inhibit by DPPH assay while the lowest antioxidant activity by DPPH assay was presented in

Buab ngu (4.47±0.37 %inhibit). In addition, the total antioxidant power was measured using ferric reducing FRAP assay. Among Thai side dish vegetables, Kha-on contained the highest antioxidant activity with 1.65±0.03 mM $FeSO_4$ by FRAP assay while the antioxidant activity by FRAP assay in Bai cha plu was the lowest (0.11±0.01 mM $FeSO_4$).

Table 2. Total phenolic contents, total flavonoid contents and Antioxidant activity (DPPH radical scavenging and FRAP) in Thai side dish vegetables.

Thai common name	Total phenolic content (mgGAE/g)	Total Flavonoid content (mgRE/g)	DPPH %inhibition	FRAP Value mM $FeSO_4$
Tua puu	0.41±0.004	0.07±0.002	59.70±1.06	0.63±0.01
Ma khua kai nok	1.21±0.001	0.08±0.005	64.19±1.93	0.71±0.02
Ma khua puang	0.65±0.011	0.14±0.001	57.89±1.32	0.70±0.02
Ma khua proh	0.73±0.011	0.08±0.001	49.03±1.40	0.86±0.01
Ma khua lai, Ma khua kang kob	1.02±0.017	0.04±0.001	81.86±0.70	1.19±0.01
Pak kwang tung	1.14±0.005	0.07±0.001	37.72±0.20	0.63±0.02
Pak bung tai khao	1.64±0.005	0.07±0.001	21.60±0.20	0.31±0.01
Pak bung tai maung	1.66±0.006	0.08±0.001	29.72±1.34	0.36±0.01
Pak kad khao	0.26±0.001	0.10±0.003	28.60±1.10	0.28±0.01
Pak kad hom	0.50±0.008	0.06±0.001	60.92±1.72	0.67±0.06
Pak ka lum plee	0.28±0.003	0.04±0.001	11.52±0.12	0.83±0.03
Tua fak yaw	1.12±0.004	0.07±0.001	55.63±1.24	0.62±0.20
Boub hom	0.23±0.007	0.08±0.001	18.04±0.40	0.54±0.02
Boub ngu	0.20±0.002	0.06±0.004	4.47±0.37	0.18±0.01
Bai bua bok	0.26±0.012	0.06±0.003	21.65±0.68	0.27±0.01
Pak tum leung	0.62±0.002	0.06±0.001	45.44±0.26	0.30±0.01
Bai cha plu	0.87±0.008	0.06±0.001	67.89±2.74	0.11±0.01
Ma ra khee nok	0.71±0.002	0.06±0.003	32.51±1.40	0.48±0.01
Kra jeab kheaw	0.99±0.002	0.14±0.001	81.51±0.88	0.70±0.02
Kha-on	0.83±0.015	0.07±0.001	91.99±1.36	1.65±0.03
Pak chee lao	1.62±0.020	0.05±0.001	51.32±0.80	0.38±0.01

Values were shown in mean ± SD of triplicate measurement.

4. Conclusions

The result of this study revealed important information that was useful for all people. The result showed that all selected kinds of Thai side dish vegetables contained phenolic and flavonoid compound. These vegetables may not be good sources of phenoloic and flavonoid compounds. However, the selected vegetables were rich in total antioxidant capacity values. It was suggested that Thai side dish vegetables would become new important sources and natural of phenolic and flavonoid compounds because Thai people generally ate most of all vegetables as side dish in fresh cut and scald so phenolics and flavonoids were not destroyed from cooking process. In addition, they usually consumed Thai side dish fresh cut vegetables at least 100 gram/time/person. Thai side dish vegetables are not only good sources of natural phenolics and flavonoids but also contain antioxidants. This study implies that these Thai side dish vegetables can be consumed as potential sources of natural antioxidants. In addition, the phenolic and flavonoid compounds of these

vegetables indicate considerable dietary and nutritional value. Therefore, this information promotes consuming natural meal and side dish vegetables than choosing food supplement that is very harmful and expensive.

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