Xanthopyll, lycopene, β-carotene and antioxidant activity of selected Thai fruits

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ABSTRACT: Xantophyll, lycopene, β-carotent content and antioxidant activity of 11 types of Thai fruit were studied. The result shown that the highest average of Xanthophyll was found in cantaloupe with (1.31±0.07 mg/100g edible portion); Lycopene content in Tang-Mo was found highest with (131.00±1.72 mg/100g edible portion); and β-carotene content in Ma-moung was found highest with (127.12±2.12 mg/100g edible portion). The antioxidant activity the eleven selected Thai tropical fruit was undertaken by DPPH radical scavenging and FRAP assay. The results shows that Mayom displayed the highest antioxidant activity by DPPH assay, with 94.33±0.60% inhibi; and Mayom displayed the highest antioxidant activity by FRAP assay, with 4.19±0.01 mM FeSO4. There are two important factors were found in the study, first one is that all types of Thai tropical fruits were selected for the study contain Xantophyll, Lycopene and β-carotene content so people can choose them as a favorite ones to eat. Second one is antioxidant activity efficiency is high in all the fruits selected for the study. People can choose to consume any kind of selected Thai tropical fruit in local market about 3-5 serving/day for getting appropriate carotenoid agent that have the effective antioxidant activity to maintain their health.

Keywords: Xanthophyll, Lycopene, Beta-carotene, Antioxidant activity, Thai selected fruit

Introduction

It is well established that free radicals are one of the greatest threats for health because they can oxidize and injure cell, damage the DNA, thereby creates the seed for disease. When a cell's DNA changes, the cell becomes mutated. It grows and reproduces abnormally and quickly. There are many free radical generators and external toxins in the day-to-day environment such as cigarette smoke and air pollution, etc. (Devasagayam et al., 2004). Therefore, it is found that consuming antioxidant agent from food is good way to health body against with free radicals. There are many kinds of antioxidant agent in food such as phenolic compound, flavonoid compound, and more.

Carotenoid is the one group of antioxidant agent in food. Landrum and Bone (2001); Richard et al. (2009) normally carotenoid is known as fruit organic pigment. But in fact, carotenoid is an important bioactive compound with high antioxidant activity. Many literatures have reported that there are many health benefits from xanthophyll, lycopene and Beta-carotene, as powerful antioxidants. Several epidemiological studies demonstrated that consumption of fruit and vegetable with rich of xanthophyll, lycopene and Beta-carotene lower the risk of cancer, cardiovascular disease. Although xanthophyll, lycopene and beta carotene are more benefits for human health, the best way to get high xanthophyll, lycopene and beta carotene is from natural foods, such as fruits

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and vegetables (Micozi, 1998; Enrique et al., 2010). Some reports reveal that high consumption of xanthophyll, lycopene and beta carotene as supplement is a risk of carotenoid toxicity like a carotenedermia (Kubola et al., 2011; Nhung et al., 2010). A tell-tale sign of excessive consumption of beta-carotene is a yellowish discoloration of the skin, most often occurring in the palms of the hands and soles of the feet.

Thailand is known as a land of fruits. But the fruits variety that you can consume around year has no more variety. In summer season is quite less of fruits. So the study of fruit that can consume all year is important because free radicals destroy body health every day. The study of seeking rich source of xanthophyll, lycopene and Beta carotene are out of season which less choices of fruits is very important. Therefore, the objective of this study was to select Thai tropical fruits that have in summer season and to determine their xanthophyll, lycopene, beta carotent and to confirm antioxidation power by DPPH and FRAP Method. That will get useful information about benefit of summer season selected Thai tropical fruits for nutritionist to recommend people to consume fruits for their health.

Material and Method

Sample

Eleven popular Thai fruits were selected for evaluation of antioxidant content viz., Xanthophyll, Lycopene, Beta-carotene contents. All the fruits selected and used in this study were purchased from the local market at Mahasarakham province, Thailand. All fruits were cleaned and trimmed of only edible portion before they were used for analysis. The Thai common name, common name and scientific name of these fruits and edible peroidortion are given in **Table 1**.

Extraction and determination of xanthophyll lycopene and Beta-carotene content

Five grams of fresh fruit as edible portion were crushed and then extracted using solvents (chloroform: methanol (2:1v/v)), Approximately 5 grams of well-ground dried samples was extracted with 50.0 ml of each solvent and stored at room temperature and evaporated under reduced pressure at 25°C. The liquid phase was filtered and washed 3 times with saturated sodium chloride solution. The organic layer was taken and dehydrated with anhydrous sodium sulphate and evaporated under reduced pressure at 25°C. The residue was dissolved in 10 ml solution of dichloromethane (DCM) and MeOH(6:4). The contents of xanthophyll, lycopene and beta-carotene were quantified by high pressure liquid chromatography(HPLC), this method is referred to Nhung et al. (2010). The RP-HPLC system (Shimadzu) consisted of an auto sampler and column oven equipped with Inertsil ODS(4.6mm×250mm,5lm) with mobile phase of DCM: acetonitrile(6:4,v/v, containing 0.05:BHA as antioxidant) (eluentA) and MeOH(eluentB). The following gradient was used: initial condition was 70% (A) and 30%(B) for 5 min, followed by 80%(A) and 20%(B) for 5 min. The flow rate 1.5 ml/minute, Injection volume 20 µl and photodiode array detector at 472 nm for the analysis of lycopene and beta-carotene. Calibration curves were constructed with the external standards.

Determination of free radical scavenging using DPPH method

The antioxidant activities of all extracts was evaluated through free radical scavenging effect on 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical. The determination was based on the method proposed by Akowuah et al. (2005). Richer et al. (2004) Two ml of 0.1mM DPPH methanolic solution was added into 200 µl of sample extracts and 0.8 ml methanol. The mixture was thoroughly mixed and kept in the dark for 1 hr. The control was prepared by mixing 2 ml of DPPH and 1 ml methanol. The absorbance was measure at 517 nm using microplate reader spectrophotometers. Samples were measured in three replicates. Percentage of DPPH scavenging activity was calculated as % inhibition of DPPH = [Abs control –Abs sample / Abs control] x 100

Determination of ferric reducing/antioxidant power assay (FRAP)

FRAP assay was carried out using the method of Benzie and Strain (1996). Seddon et al. (1994) 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 prepared fresh, daily and was warmed to 37 ^{o}c in oven prior to use. A total of 50 µl samples extract were added to 1.5 ml of the FRAP reagent and mixed well. The absorbance was measured at 593 nm using microplate reader spectrophotometers after 4 mins. Samples were measured in three replicates. Standard curve of iron (II) sulfate solution was prepared using the similar procedure. The results were expressed as µmol Fe (II) /100 g extract sample.

Statistical Analysis

This study used the means of triplicate determination on three batches of each vegetable with completely randomized design. Data analyses of xanthophyll, lycopene, Beta-carotene and antioxidant activity were performed by SPSS software version 13. A significant difference was considered in values differing at the confidence level of $P<0.05$.

Result and Discussion

The selected 11 kind of Thai tropical fruits that people consumed in the market were studied for Xantophyll, Lycopene, Beta-carotene and Antioxidant activity. All selected fruits were sold in all of season in Thailand. So people can choose and buy for their meal around the year. In table 1 shown Thai common name, common name and scientific name of selected fruits.

Table 1 Thai common names, common names, scientific name and edible portion for analyze

Thai common name	Common name	Scientific name	Edible peroid
Mayom	Star gooseberry	Phyllanthus acidus (L.) Skeels	Mature
Chompu	Java apple, rose apple	Eugenia javanica Lam.	Mature
Keaw Mung-Kon	Pitaya, Dragon Fruit	Hylocercus undatus	Ripen
Farung	Guava	Psidium guajava L.	Mature
Malakor	papaya, or papaw	Carica papaya L.	Ripen
Cantaloupe	Cantaloupe	Cucumis melo	Ripen
Ma-Muang	Mango	Mangifera indica L.	Ripen
Sala	salak plum	Zalacca edulis	Mature
Tang Mo	Watermelon	Citrullus lanatus	Ripen
Klouy nam wha	Banana	Musa sapientum Linn.	Ripen
kra thon	Santol	Sandoricum koetjape Burm.f. Mer.	Mature

The edible period of fruits that Thai people usually consume is also shown in **Table 1**. Most of these fruits are consumed in its ripen period except Mayom, Chompu, Farung, Sala and Krathon which are usually consumed in mature period. The taste of 11 selected was divided in 2 groups; one is with sweet taste they are Keaw Mung-Kon, Malakor, Cantaloupe, Ma-Muang, Tang Mo, and Klouy nam wha; and another one is with spicy-salt with sour or some bitter taste they are Mayom, Chompu, Farung, Sala, and kra thon.

Table 2 illustrated the content of xanthophyll, lycopene and beta-carotene level of selected Thai fruit. The results shown that, there is xanthophyll in 11 fruits except Klouy nam wha. The highest of average xanthophyll found in cantaloupe (1.31±0.07 mg/100g edible portion), the second highest is Malarkor (1.07±0.02 mg/100g edible portion). The study could not detect xanthophyll in Klouy nam wha. Lycopene content in Tang Mo shows highest content (131.00±1.72 mg/100g edible portion), the second is cantaloupe (58.75±0.32 mg/100g edible portion). The study could not detect lycopene in Mayom, Chompu, Kaew mung-kon, and Farung. All kind of selected Thai tropical fruit contained Beta-carotene. Among the selected Thai tropical fruits, Beta-carotene content is shown highest in Ma-mung (127.12±2.12 mg/100g edible portion), the second highest is in cantaloupe (38.00±1.05 mg/100g edible portion).

Table 2 Amount of Xanthophyll, lycopene and Beta-carotene in selected Thai fruits determine by HPLC (mg/100g)

Thai common name	Common name	Xanthophyll, mg/100g $\bar{x} \pm SD$	Lycopene, mg/100g $\bar{x} \pm SD$	β-carotene, mg/100g $\bar{x} \pm SD$
Mayom	Star gooseberry	0.32±0.01	ND	4.82±0.15
Chompu	Java apple, rose apple	0.36±0.02	ND	3.38±0.09
Keaw Mung-Kon	Pitaya, Dragon Fruit	0.21±0.01	ND	3.03±0.13
Farung	Guava	0.60±0.05	ND	3.69±0.06
Malakor	Papaya or papaw	1.07±0.02	53.50±0.43	32.30±2.07
Cantaloupe	Cantaloupe	1.31±0.07	58.75±0.32	38.00±1.05
Ma-muang	Mango	0.33±0.01	3.17±0.10	127.12±2.12
Sala	salak plum	0.84±0.06	3.45±0.02	28.05±0.59
Tang Mo	Watermelon	0.52±0.01	131.00±1.72	24.76±0.50
Klouy nam wha	Banana	ND	4.35±0.07	5.00±0.15
kra thon	Santol	0.23±0.01	3.16±0.02	8.31±0.63

ND = not Detect, Values are shown in mean ± SD of triplicate measurement.

Table 3 Antioxidant activity (DPPH radical scavenging and FRAP) in selected Thai fruits.

Thai common name	Common name	DPPH %inhibition	FRAP Value mM FeSO4
Mayom	Star gooseberry	94.33±0.60	4.19±0.01
Chompu	Java apple, rose apple	67.36±3.45	1.07±0.01
Keaw Mung-Kon	Pitaya, Dragon Fruit	45.65±0.44	0.58±0.01
Farung	Guava	93.35±0.05	2.07±0.01
Malakor	papaya or papaw	93.64±0.12	0.17±0.02
Cantaloupe	Cantaloupe	91.09±0.12	2.91±0.01
Ma-Muang nam dok mai	Mango	22.03±1.33	0.34±0.01
Sala	salak plum	90.72±0.05	1.07±0.02
Tang Mo	Watermelon	87.31±0.68	0.46±0.01
Klouy nam wha	Banana	34.98±1.55	0.32±0.01
kra thon	Santol	16.73±1.14	1.85±0.01

Values are shown in mean ± SD of triplicate measurement.

The table-3 shows the antioxidant activity of the eleven selected Thai tropical fruits by DPPH radical scavenging and FRAP assay. The results show that all kind of selected Thai tropical fruits shown %inhibit by DPPH assay. Mayom displayed the highest antioxidant activity by DPPH assay, with 94.33±0.60 %inhibit. The lowest antioxidant activity by DPPH assay is Kra thon, with 16.73±1.14 %inhibit. All the fruits selected from Thai tropical fruits shown antioxidant power assay by ferric reducing. Mayom displayed the highest antioxidant activity by FRAP assay, with 4.19±0.01 mM $FeSO_4$. And Mar lar kor shows the lowest antioxidant activity by FRAP assay, with 0.17±0.01 mM $FeSO_4$.

Conclusions

The present study revealed two facts that are useful for health. One is result shown quantity of Xantophlly, Lycopene and Beta-carotene in selected Thai tropical fruits that are common fruits for Thai people. The result shown all kinds of selected Thai tropical fruits contained difference in Xantophlly, Lycopene and Beta-carotene content so people can choose the favorite one to eat. Two is antioxidant activity of Xantophlly, Lycopene and Beta-carotene in selected Thai tropical fruit shown high efficiency. From these results if people chose to consume any kind of selected Thai tropical fruits that are cheap and common in local market about 3-5 serving/day that recommend in Nutrition flag they can get appropriated carotenoid agent that have the effective antioxidant activity to regulate their health. So how to promote people eating native meal and side dish vegetables than choose food supplement that is very harmful and expensive is consider.

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