ASIAN JOURNAL OF PHARMACEUTICAL AND CLINICAL RESEARCH

NNOVARE
ACADEMIC SCIENCES
Knowledge to Innovation

Vol 9, Issue 6, 2016

Online - 2455-3891 Print - 0974-2441

Research Article

ANTHOCYANIN PROFILE AND ITS ANTIOXIDANT ACTIVITY OF WIDELY USED FRUITS, VEGETABLES, AND FLOWERS IN THAILAND

CHAIYAVAT CHAIYASUT¹, BHAGAVATHI SUNDARAM SIVAMARUTHI¹, NOPPAWAT PENGKUMSRI¹, SASITHORN SIRILUN¹, SARTJIN PEERAJAN², KHONTAROS CHAIYASUT², PERIYANAINA KESIKA¹*

¹Department of Pharmaceutical Sciences, Faculty of Pharmacy, Chiang Mai University, Chiang Mai, Thailand. ²Health Innovation Institute, Chiang Mai, Thailand. Email: p.kesika@gmail.com

Received: 22 July 2016, Revised and Accepted: 27 July 2016

ABSTRACT

Objective: Anthocyanins are the water-soluble pigments most commonly present in flowers, fruits, and vegetables. Anthocyanins have been reported for is antioxidant, anti-inflammatory, antibacterial properties, and other health benefits in human. This study was performed to extract and determine the anthocyanin and to access the antioxidant properties of the most commonly used fruits, vegetables, and flowers in Thailand.

Methods: Extraction of anthocyanin and its aglycones from the selected samples and determination of anthocyanin and anthocyanidins using reversed-phase high-performance liquid chromatography analysis. Determination of total antioxidant ability using 2,2'-azino-bis-3-ethylbenzothiazoline-6-sulfonic acid, 1, 1-diphenyl-2-picryl-hydrazil, and Ferric reducing antioxidant power assay.

Results: Maximum distribution of tested anthocyanin and anthocyanidin contents was observed in the extracts of Jambolan plum, Ceylon spinach, purple sweet potato, purple lettuce (PLe) followed by common plum (CP) and red dragon fruit. The extracts of ma-kiang, purple eggplant (PE), red grape, and PLE showed the maximum amount of cyanidin 3-glucoside, delphinidin 3-glucoside, peonidin 3-glucoside, and malvidin 3-glucoside, respectively. Likewise, the maximum concentration of cyanidin, delphinidin, peonidin, and malvidin was found in common plum, red khae, red cabbage (RC), and PLe, respectively. Ma-kiang extract exhibited the maximum antioxidant activity followed by RC, CP, and PE extract compared to the other extracts.

Conclusion: The present study primarily profiled the anthocyanin content of selected fruits, vegetables, and flowers. Among the tested samples, making extract showed the high cyanidin 3-glucoside content and antioxidant activity. Further, detailed study on the anthocyanin content at different climate and geographical conditions and other factors are necessary to develop nutraceutical or cosmetic product with a functional ingredient.

Keywords: Anthocyanin, Anthocyanidin, Antioxidant activity.

© 2016 The Authors. Published by Innovare Academic Sciences Pvt Ltd. This is an open access article under the CC BY license (http://creativecommons.org/licenses/by/4. 0/) DOI: http://dx.doi.org/10.22159/ajpcr.2016.v9i6.14245

INTRODUCTION

Anthocyanins are the most important and largest subgroups of flavonoids and are responsible for the red, blue, and purple color of the flower, fruits, and vegetables. These water soluble pigments occur primarily as glycosylated anthocyanidin, i.e., one or more sugar moiety (mono-or di- or trisaccharide forms of sugar such as rhamnose, galactose, glucose, arabinose, and xylose) bonded with the aglycone [1]. Anthocyanins are stable due to the glycosylation, whereas anthocyanidins are less stable and are rarely found in nature [2]. Almost 635 anthocyanins were identified [3] and among these pigments, glycoside forms of cyanidin, delphinidin, pelargonidin, peonidin, petunidin, and malvidin are the most commonly distributed anthocyanins in nature [4,5]. Anthocyanins are most commonly present in the flower and fruits. Apart from these plant materials, they are present in other parts of plants such as stem, leaves, and storage organs [6]. The edible parts of the plants that are rich in color pigments are widely used in the beverage and food industries [7].

Many studies have been reported for the anthocyanin content in vegetables and fruits. The present study focused on the fruits (Jambolan plum (JP), roselle (Ro), red dragon fruit (RDF), mamao (Ma), red grape (RG), common plum (CP), pomegranate (Po), ma-kiang (MK), and cherry (Ch)), vegetables (ceylon spinach (CS), purple sweet potato (PSP), red cabbage (RC), purple eggplant (PE), purple lentils (PL), purple lettuce (PLe), and purple taro (PT)), and flowers (red khae (RK), butterfly pea (BP)) that are widely used in Thailand. *Syzygium cumini* (L.) Skeels is a tree native to India, and these trees are also cultivated in

most of the tropical and subtropical countries [8,9]. The tree parts such as leaves, fruits, seeds, and barks have been reported for its medicinal properties and also known for its use in traditional herbal medicines [10,11]. The fruit of S. cumini is a berry and commonly known as JP. Hibiscus sabdariffa L. is an edible herb commonly known as roselle, which is native to Africa and also grown in tropical and subtropical countries [12-16]. Roselle (Ro) flower petals are also widely used as food colorants, an ingredient in traditional medicine and reported for its health benefits in human [13,17]. McKay et al. (2010) revealed that the daily intake of roselle tea reduces the blood pressure in both pre and mild hypertensive adults [18]. Hylocereus costaricensis is a tropical plant native to Mexico and South America and commonly known as super RDF or red pitaya [19,20]. The red berries of Vitis vinifera L. are commonly known as RG, and these berries are rich in anthocyanins and flavonols [21]. The grapevine is mainly cultivated for the wine industries [22]. Prunus domestica is also known as European or CP, which is rich in phenolic phytochemicals [23] and CP are reported as a good source of natural antioxidants [23-25]. Punica granatum L. is commonly known as pomegranate (Po) native to the Middle East [26] and also cultivated in tropical and subtropical regions [27]. Po is reported as a functional food that is rich in antioxidants [26]. Cleistocalyx nervosum var. paniala is known as ma-kiang (MK), which is found in Northern Thailand and other parts of Southeast Asia [28,29]. Prunus avium is known as cherry (Ch), and these fruits are rich in phenolic compounds including anthocyanins [30]. Basella rubra L. is commonly known as CS, which are rich in phenolic compounds and exhibits higher antioxidant

property [31]. *Ipomoea batatas* (L.) Lam. is known as PSP native to South America and grown in tropical and subtropical regions [32]. Teow *et al.*, 2007, reported that the anthocyanins in PSPs showed high antioxidant activity compared to the other colored sweet potatoes [33]. The PSP are used as natural food colorants in Asia [34]. *Brassica oleracea* Linn is commonly known as RC and used as a food colorant due to its stable anthocyanins [35]. The eggplant (*Solanum melongena* L.) has different color patterns such as purple, white, and green [36]. *Lactuca sativa* L. is commonly known as lettuce, which is a leafy vegetable ranging in colors from green to purple. *L. sativa* L. is native to the Mediterranean region [37]. *Sesbania grandiflora* (L.) is commonly known as corkwood tree, vegetable-hummingbird (English), khae (Thai) [38], and its flower are pink, red, or white in color. *Clitoria ternatea* L. is commonly known as BP plant. The anthocyanin isolated from its blue colored flower is used as natural food colorants in Thailand [39].

The current study aimed to extract and to determine the anthocyanin present in the selected fruits, vegetables, and flowers and also to determine the free radical scavenging ability of the anthocyanin extracts using 2,2'-azino-bis-3-ethylbenzothiazoline-6-sulfonic acid (ABTS), 1, 1-diphenyl-2-picryl-hydrazil (DPPH), and Ferric reducing antioxidant power (FRAP)assay. Few studies have reported that several external factors such as climate, nature of the soil in the place of cultivation and other factors such as different varieties, and its stage influence the composition of compounds responsible for the antioxidant property [40,41]. The purpose of the present study was to catalog the anthocyanin content and antioxidant properties of fruits, vegetables, and flowers, which were commonly used for dietary consumption and/or medicinal purpose in Thailand. Moreover, this is the first study explaining about the anthocyanin content of mamao, red khae, and purple taro.

METHODS

Sample collection and extraction

The fruits such as JP (*S. cumini* (L.) Skeels), Ro (*H. sabdariffa* L.), RDF (*H. costaricensis*), Ma (*A. puncticulatum* Miq.), RG (*V. vinifera* L.), CP (*P. domestica*), Po (*P. granatum* L.), MK (*C. nervosum* var. paniala), ch (*P. avium*), and vegetables such as CS (*B. rubra* L.), PSP (*I. batatas* (L.) Lam.), RC (*B. oleracea* Linn), PE (*S. melongena* L.), PL (*V. unguiculata* subsp. Sesquipedalis), PLe (*L. sativa* L.), PT (*C. esculenta* var. Esculenta), and flowers such as RK (*S. grandiflora* (L.) Desv.), BP (*C. ternatea* L.) were collected from the Chiang Mai local market, Thailand. All these samples were compared with the herbarium specimen of Faculty of

Pharmacy, Chiang Mai University, Thailand.

The fresh samples were washed thoroughly using the distilled water. The washed samples were dried at 50°C for 48 h and then chopped or grinded before extraction. Anthocyanin and anthocyanidins content of all the samples (each 10 g) were solvent extracted (methanol: 0.1 N HCl; $85\text{:}\ 15$), filtered, evaporated, and the percentage of yield was calculated as described in the previous publication [42].

Determination of anthocyanin and anthocyanidins

Anthocyanins and anthocyanidins present in the extracted samples were determined using reversed-phase high-performance liquid chromatography (HPLC) as described in the previous publication [42]. Cyanidin 3-glucoside, delphinidin 3-glucoside, malvidin 3-glucoside, and peonidin 3-glucoside (Tokiwa Phytochemical Co., Ltd, Japan) were used as standards for the determination of anthocyanin. Cyanidin chloride, delphinidin chloride, malvidin chloride, and peonidin chloride, (Extrasynthese, France) were used as standards for the determination of anthocyanidin.

Determination of antioxidant capacity

Free radical scavenging ability was determined by performing the ABTS, DPPH, and FRAP assay as described in the previous publication [42,43]. The antioxidant capacity was represented as mg trolox equivalent antioxidant activity (TEAC) per gram of extract for ABTS and DPPH assay. The reducing power was represented as mg ${\rm FeSO}_4$ equivalent per gram of extract.

Statistical analysis

The experiments were carried out in triplicates. The values in the results were represented as mean \pm Standard deviation. Statistical analysis was performed using one way - analysis of variance (SPSS version 17 statistical software [Chicago, SPSS Inc., U.S.A]). The significant differences in anthocyanin content and antioxidant activities among the fruits and vegetables used in this study were determined using the *post-hoc* test (Least Significant Difference). Differences at a p<0.05 were considered to be statistically significant.

RESULTS AND DISCUSSION

The samples were collected, and the contents were extracted by standard extraction (methanol: 0.1 N HCl) method. The yield of extracts obtained from the samples (fruits, vegetables, and flowers used in this study) was

Table 1: Percentage of the yield of extracted fruits, flowers and vegetables

Common name	Scientific name	Sample code	% of yield
Jambolan plum	Syzygium cumini (L.) Skeels.	JP	63.90±6.71
Ceylon spinach	Basella rubra L.	CS	27.65±2.90
Roselle	Hibiscus sabdariffa L.	Ro	56.45±5.93
Red dragon fruit	Hylocereus costaricensis	RDF	63.75±6.69
Mamao	Antidesma puncticulatum Miq.	Ma	79.90±8.39
Red grape	Vitis vinifera L.	RG	90.00±9.45
Purple sweet potato	Ipomoea batatas (L.) Lam.	PSP	82.15±8.63
Red khae	Sesbania grandiflora (L.) Desv.	RK	62.75±6.59
Red cabbage	Brassica oleracea Linn	RC	29.35±3.08
Common plum	Prunus domestica	CP	89.20±9.37
Pomegranate	Punica granatum L.	Po	56.25±5.91
Purple eggplant	Solanum melongena L.	PE	29.90±3.14
Butterfly pea	Clitoria ternatea L.	BP	33.20±3.49
Ma kiang	Cleistocalyx nervosum var. paniala	MK	39.55±4.15
Purple lentils	Vigna unguiculata subsp. Sesquipedalis	PL	18.50±1.94
Purple lettuce	Lactuca sativa L.	PLe	17.20±1.81
Cherry	Prunus avium	Ch	89.65±9.41
Purple taro	Colocasia esculenta var. Esculenta	PT	5.80±0.61

S. cumini: Syzygium cumini, B. rubra: Basella rubra, H. sabdariffa: Hibiscus sabdariffa, H. costaricensis: Hylocereus costaricensis, A. puncticulatum: Antidesma puncticulatum, V. vinifera: Vitis vinifera, I. batatas: Ipomoea batatas, S. grandiflora: Sesbania grandiflora, B. oleracea: Brassica oleracea, P. domestica: Prunus domestica, P. granatum: Punica granatum, S. melongena: Solanum melongena, C. ternatea: Clitoria ternatea, C. nervosum: Cleistocalyx nervosum, V. unguiculata: Vigna unguiculata, L. sativa: Lactuca sativa, C. esculenta: Colocasia esculenta, JP: Jambolan plum, CS: Ceylon spinach, Ro: Roselle, RDF: Red dragon fruit, Ma: Mamao, RG: Red grape, PSP: Purple sweet potato, RK: Red khae, RC: Red cabbage, CP: Common plum, Po: Pomegranate, PE: Purple eggplant, BP: Butterfly pea, MK: Ma-kiang, PL: Purple lentils, PLe: Purple lettuce, Ch: Cherry, PT: Purple taro

tabulated (Table 1). Each extract was subjected to anthocyanins, and anthocyanidins estimation by HPLC analysis. The major anthocyanidins such as cyanidin, delphinidin, peonidin, malvidin, and its aglycone forms such as cyanidin 3-glucoside, delphinidin 3-glucoside, peonidin 3-glucoside, and malvidin 3-glucoside have been examined.

All samples (each 10 g) were extracted using methanol: 0.1 N HCl (85: 15). The values are represented as mean \pm SD (n=3).

The distribution of anthocyanin and anthocyanidin contents studied in this study was observed to be varied in each extract. Among the extracted samples, maximum distribution of tested anthocyanin and anthocyanidin contents was observed in the extracts of IP, CS, PSP and PLe followed by CP and RDF. Many studies have revealed the presence of anthocyanin in fruits and vegetables. The anthocyanins such as glycosides of delphinidin-diglucoside, malvidin-diglucoside and petudinindiglucoside [44] have been reported to be present in the JP. In the present study, the presence of anthocyanins in IP include cyanidin 3-glucoside, delphinidin 3-glucoside, malvidin 3-glucoside, and anthocyanidins include cyanidin were identified by HPLC analysis (Figs. 1a-c and 2a). Malvidin 3-glucoside, cyanidin, peonidin, and malvidin was found in the CS extract (Figs. 1d and 2a, c, d). Usenik et al. (2009) reported the content of anthocyanin in *P. domestica* L. during the ripening of fruit using HPLC. The study stated that the anthocyanins namely, cyanidin 3-rutinoside followed by peonidin 3-rutinoside, cyanidin 3-glucoside, cyanidin 3-xyloside, and peonidin 3-glucoside were present in the ripe plum. This study concluded that the content of anthocyanin is increased during the ripening process [45]. Similarly, Miletić et al. (2012) reported that the anthocyanin content increased during the ripening process [23]. The mono- or di- acylated derivatives of cyanidin and peonidin are the major anthocyanins of PSP [46]. Cyanidin 3-glucoside, delphinidin 3-glucoside, delphinidin, and peonidin were found to be present in the PSP extract (Fig. 1a, b and 2b, c). The PLe extract contains malvidin (Fig. 2d) and peonidin 3-glucoside, malvidin 3-glucoside, and less amount of cyanidin 3-glucoside (Fig. 1a, c, d). CP extract contains cyanidin 3-glucoside, delphinidin 3-glucoside, and cyanidin (Fig. 1a, b, and 2a). RDF extract contains delphinidin 3-glucoside, peonidin 3-glucoside, and malvidin 3-glucoside (Fig. 1b-d).

Among the extracted samples, only two of the tested anthocyanin and anthocyanidin contents were observed in the extracts of RG, BP, and PL. Kallithraka et al. (2009) reported the anthocyanin content in the red V. vinifera spp. grapes of forty-six varieties cultivated in Greece and revealed that Syrah variety was rich in anthocyanin content compared to the other varieties. The study also reported that malvidin 3-0-glucoside was prevalent compared to other anthocyanins (peonidin 3-0-glucoside, petunidin 3-0-glucoside, cyanidin 3-0-glucoside, and delphinidin 3-0-glucoside) present in the grapes [47]. Figueiredo-González et al. (2012) reported the profiles of anthocyanin and flavonol content in red V. vinifera L. berries of three varieties namely Brancellao, Mouratón, and Gran Negro. The skin of red berries from V. vinifera L. contains glycosides of cyanidin, delphinidin, malvidin, peonidin, and petunidin. The study reported that the content of anthocyanin in the skin of RG varied in each variety. Brancellao variety contains high cyanidin-3-0-glucoside content, whereas Gran Negro variety contains high malvidin- and peonidin-3-0-glucoside content. Mouratón variety contains a high content of delphinidin- and petunidin-3-0-glucoside [48]. In the present study, the RG extract contains 3-glycosides of cyanidin and peonidin (Figs. 1a and c). The anthocyanins present in the blue colored BP flowers are malvidin-3-\u03b3glucoside, and delphinidin-3-β-glucoside [49]. The BP extract contains malvidin 3-glucoside (Fig. 1d) and its aglycone (Fig. 2d). The PL extract contains cyanidin-3-glucoside, delphinidin 3-glucoside (Figs. 1a and b).

The extract of Ro, Ma, RK, PT, RC, Po, PE, MK, and Ch showed only one of the tested anthocyanin and anthocyanidin contents (Figs. 1 and 2). Kouakou et al. (2015) reported that the calyx of roselle contains predominantly delphinidin-3-0-sambubioside, cyanidin-3-0-sambubioside and also contains cyanidin-3-0-glucoside, delphinidin-3-0-glucoside. The callus of roselle contains predominantly cyanidin-3-0-sambubioside, delphinidin-3-0-glucoside, and also contains cyanidin-3-0-glucoside, delphinidin-3-0-sambubioside, malvidin-3-0-glucoside, and petunidin-3-0-glucoside [50]. In the present study, cyanidin-3-glucoside was observed in the Ro and Ma extract (Fig. 1a). There are no previous reports on the anthocyanin present in Ma, RK, and PT. Delphinidin was observed in RK and PT extracts in the present study (Fig. 2b). The major anthocyanins of RC are cyanidin-3-diglucoside-5-glucoside derivatives

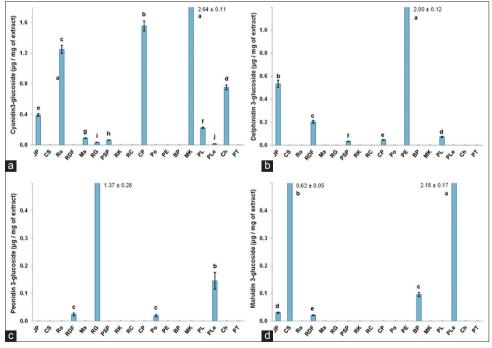


Fig. 1: Content of anthocyanin in the extracts of fruits, vegetables and flowers used in this study. (a) Cyanidin 3-glucoside, (b) delphinidin 3-glucoside, (c) peonidin 3-glucoside, (d) malvidin 3-glucoside. The values are represented as mean ± standard deviation (n=3). Alphabets (a-j) indicates the order of significant differences in the respective anthocyanin content between the extracts (p<0.05)

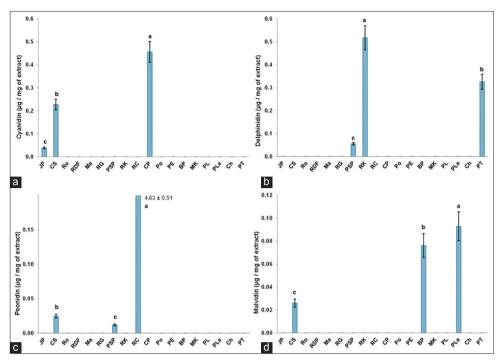


Fig. 2: Content of anthocyanidins in the extracts of fruits, vegetables and flowers used in this study. (a) Cyanidin, (b) delphinidin, (c) peonidin, (d) malvidin. The values are represented as mean ± Standard deviation (n=3). Alphabets (a-c) indicates the order of significant differences in the respective anthocyanidin content between the extracts (p<0.05)

acylated (mono- or di-) with p-coumaric, caffeic, ferulic, sinapic acids, and also contain pelargonidin-3-glucoside and di-acylated forms of cyanidin-3-0-triglucoside-5-0-glucoside with hydroxycinnamic acids [51]. In the current study, the presence of peonidin was observed in the RC extract (Fig. 2c). The major anthocyanin present in P. granatum L. was cyanidin-3,5-diglucoside, which was reported to be stable during the pasteurization of pomegranate juice (PJ) of Hicaznar variety, and PJ also contains 3-glucoside of cyanidin and delphinidin [52]. Similarly, Turkyılmaz (2013) reported cyanidin-3,5-diglucoside as major anthocyanins present in the PJs of Turkey varieties and PJ also contains cyanidin-3-glucoside, delphinidin-3-glucoside, pelargonidin-3-glucoside, delphinidin-3,5-diglucoside, and pelargonidin-3,5-diglucoside [53]. The content of anthocyanin and antioxidant property of methanolic extract of Po has already been reported [54]. Borochov-Neori et al. (2011) reported the effect of climate on the composition of anthocyanin and its content Po fruit arils [55]. In the present study, the presence of peonidin 3-glucoside was observed in the Po extract (Fig. 1c). The major anthocyanin of PE was delphinidin-3-rutinoside [56]. Nasunin (delphinidin 3-(4-(p -coumaroyl) -L- ramnosil -(1,6) glucopyranoside)5- gluco-pyranoside) is an anthocyanin and showed high radical scavenging activity, which can be used as natural antioxidant source and colorant for food, cosmetic, and pharmaceutical products [57]. The PE extract contains 3-glucoside of delphinidin (Fig. 1b). The major anthocyanin found in *C. nervosum* var. paniala was cyanidin-3-glucoside [58]. The major anthocyanin present in the P. avium is cyanidin-3-glucoside and cyanidin-3-rutinoside [59]. Similar to the previous reports, cyanidin-3-glucoside was found in the MK and Ch extract in the present study (Fig. 1a). The samples namely, MK, PE, RG, and PLe were recorded with maximum amount of cvanidin 3-glucoside (2.64±0.11 μg/mg of extract), delphinidin 3-glucoside (2.00±0.12 μg/ mg of extract), peonidin 3-glucoside (1.37±0.28 μg/mg of extract), and malvidin 3-glucoside (2.18±0.17 μg/mg of extract), respectively. Likewise, maximum concentration of cyanidin (0.46 \pm 0.05 $\mu g/mg$ of extract), delphinidin (0.52 \pm 0.05 µg/mg of extract), peonidin (4.63 \pm 0.51 µg/mg of extract), and malvidin (0.09±0.01 µg/mg of extract) were noticed in CP, RK, RC, and PLe, respectively.

The total antioxidant ability of each extract was determined by ABTS, DPPH, and FRAP assays. The inhibitory concentration (IC_{E0}) values of

Table 2: IC₅₀ values of extracted fruits, flowers and vegetables

Sample code	ABTS assay	DPPH assay
JP	51.48±1.40	120.26±3.72
CS	64.37±1.60	123.87±3.64
Ro	32.45±0.86	118.74±3.52
RDF	100.88±2.70	126.35±3.86
Ма	119.62±3.50	132.53±3.65
RG	54.15±1.50	123.64±3.80
PSP	105.58±3.10	127.06±3.88
RK	84.27±2.80	124.78±3.76
RC	20.68±0.62	110.88±3.50
CP	24.00±0.72	115.06±3.50
Po	614.83±12.20	143.06±3.87
PE	25.95±0.82	117.75±3.50
BP	130.62±3.50	134.97±4.00
MK	14.70±0.71	109.84±3.60
PL	100.20±2.71	125.73±3.50
PLe	48.00±1.50	119.69±3.71
Ch	53.27±1.44	121.66±3.72
PT	88.61±2.31	125.46±3.75

IC_{so}: Inhibitory concentration, JP: Jambolan plum, CS: Ceylon spinach, Ro: Roselle, RDF: Red dragon fruit, Ma: mamao, RG: Red grape, PSP: Purple sweet potato, RK: Red khae, RC: Red cabbage, CP: Common plum, Po: Pomegranate, PE: Purple eggplant, BP: Butterfly pea, MK: Ma-kiang, PL: Purple lentils, PLe: Purple lettuce, Ch: Cherry, PT: Purple taro

each extract were calculated and were represented in Table 2. The MK extract exhibited the maximum antioxidant activity followed by RC, CP, and PE extract compared to the other extracts in all the tested *in vitro* models of antioxidant assays used in this study (p<0.05). MK extract showed the maximum free radical scavenging ability (11.62±0.16 and 10.96±0.01 mg TEAC/g of MK extract (Figs. 3a and b) with an IC $_{50}$ 14.70±0.71 and 109.84±3.60 µg of MK extract (Table 2) in ABTS and DPPH assay, respectively) and reducing power (5.26±0.14 mg FeSO $_{4}$ equivalent/g of MK extract in FRAP assay) (Fig. 4). The anthocyanin content and the antioxidant ability of the RC were higher compared to the white cabbage [60]. The total phenolic content of the fresh plum and prunes (the dried plum) influences its antioxidant capacity [61].

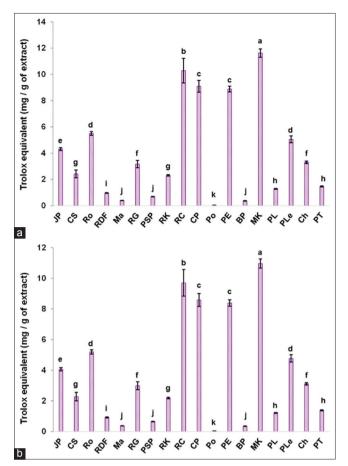


Fig. 3: Antioxidant capacity of the extracted fruits, vegetables, and flowers determined by (a) ABTS and (b) DPPH assay. The antioxidant capacity of the extracted samples was represented as mg trolox equivalent antioxidant activity (TEAC) per gram of extract for ABTS and DPPH assay. The values are represented as mean ± Standard deviation (n=3). Alphabets (a-k) indicates the significant differences in the respective antioxidant activity between the extracts (p<0.05)

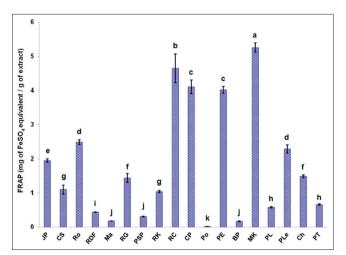


Fig. 4: The reducing power of the extracted fruits, vegetables and flowers were determined by FRAP assay. The reducing power was represented as mg FeSO4 equivalent per gram of extract. The values are represented as mean ± Standard deviation (n=3). Alphabets (a-k) indicates the significant differences in the respective antioxidant activity between the extracts (p<0.05)

The purple color small sized eggplant showed a high total phenolic, anthocyanin content, and possess high antioxidant activity [62]. In the present study, RC, CP, and PE extract showed ABTS** radical scavenging activity of about 10.28±0.93, 9.09±0.45, and 8.88±0.22 mg TEAC/g of extract, respectively (Fig. 3a), DPPH radical scavenging ability of about 9.70±0.87, 8.58±0.42, and 8.38±0.21 mg TEAC/g of extract, respectively (Fig. 3b), and reducing power of about 4.65±0.42, 4.11±0.20, and 4.02±0.10 mg FeSO_A equivalent/g of extract, respectively (Fig. 4).

Mohd-Esa et al. (2010) reported that roselle seeds showed the strong and highest antioxidant activity compared to the extracts of roselle calyces, leaves, and stems and stated that roselle seeds as a potent functional food [63]. Yang et al. (2012) have reported that the extract of roselle calyx is a good natural source of antioxidant [15]. Veigas et al. (2007) have reported high antioxidant activity and stability of the anthocyanins present in the JP extract using the DPPH radical scavenging, reducing power, lipid peroxidation assay [44]. The darker cultivars of Ch exhibited higher antioxidant activity than the lighter cultivars grown in Turkey [64]. The skin of V. vinifera grape berries of blue-black varieties showed higher total polyphenol content, antioxidant activity than white grape varieties and stated that skin of blue-black varieties are a good natural source of antioxidants [65]. Loganayaki et al. (2012) reported the high antioxidant and anti-inflammatory properties of the methanol extract of RK flower and stated that the edible flower can be used as a natural source of nutraceutical supplement [66]. The PSP have been reported to contain high anthocyanin content, total phenol content, and possess high antioxidant activity compared to the red, yellow, and white sweet potatoes [67]. The leaves and flowers of BP showed high antioxidant activity and considered as a natural source of antioxidants [68]. The Ro extract also showed antioxidant activity followed by PLe, JP, Ch, RG, CS, RK in the present study (Figs. 3 and 4).

Moreover, many of the natural pharma products were derived from colored fruits, vegetables, and flowers for the natural bioactive properties. The ability of the bioactivities is attributed to the content of the phenolic compounds, especially anthocyanins. During the processing of such compounds for pharmaceutical preparations, the activity may lose during the physical treatments [69]. Thus, it is necessary to cataloging the anthocyanin content and free radical scavenging property of every commonly used natural source such as fruits, flowers and vegetables.

CONCLUSION

Overall, the current study results conclude that ma-kiang extract contained a high amount of cyanidin 3-glucoside and showed maximum antioxidant activity compared to the other tested extracts. This is the primary study about the anthocyanin content in the extract (methanol: 0.1 N HCl) of mamao, red khae, and purple taro. Anthocyanins and anthocyanidins are prone to degradation. And the content of both anthocyanins and anthocyanidins also influenced by the geographical and climate conditions, and cultivation. Hence, further detailed study at different climate and geographical conditions and other factors may provide a detailed knowledge of the content and distribution of anthocyanins and anthocyanidins in these samples that can be used to develop nutraceutical or cosmetic product with a functional ingredient.

ACKNOWLEDGMENT

PK gratefully acknowledges the CMU Post-Doctoral Fellowship. Authors gratefully acknowledge the Faculty of Pharmacy, and Chiang Mai University, Thailand, for the support and instrumentation facilities.

REFERENCES

- Prior RL, Wu X. Anthocyanins: Structural characteristics that result in unique metabolic patterns and biological activities. Free Radic Res 2006;40(10):1014-28.
- 2. Borkowski T, Szymusiak H, Gliszczynska-Swiglo A, Tyrakowska B.

- The effect of 3-O-beta-glycosylation on structural transformations of anthocyanins. Food Res Int 2005;38:1031-7.
- Andersen ØM, Jordheim M. Anthocyanins-food applications. Presented at Proceeding 5th International Congress. Pigments Foods: For Quality and Health, 14-16 August, Helsinki, Finland; 2008.
- Eder R. Pigments. In: Nollet LML, editor. Food Analysis by HPLC. Monticello, NY: Marcel Dekker; 2000. p. 845-80.
- Kong JM, Chia LS, Goh NK, Chia TF, Brouillard R. Analysis and biological activities of anthocyanins. Phytochemistry 2003;64(5):923-33.
- Delgado-Vargas F, Paredes-Lopez O. Anthocyanins and betalains. In: Delgado-Vargas F, Paredes-Lopez O, editors. Natural Colorants for Food and Nutraceutical Uses. Boca Raton, FL: CRC Press; 2003. p. 167-219.
- He J, Giusti MM. Anthocyanins: Natural colorants with healthpromoting properties. Annu Rev Food Sci Technol 2010;1:163-87.
- 8. Srivastava S, Chandra D. Pharmacological potentials of *Syzygium cumini*: A review. J Sci Food Agric 2013;93:2084-93.
- Sehwag S, Das M. Nutritive, therapeutic and processing aspects of Jamun, Syzygium cuminii(L.) Skeels- An overview. Indian J Nat Prod Resour 2014;5(4):295-307.
- Chagas VT, França LM, Malik S, Paes AM. Syzygium cumini (L.) skeels: A prominent source of bioactive molecules against cardiometabolic diseases. Front Pharmacol 2015;6:259.
- 11. Rupeshkumar M, Kavitha K, Haldar PK. Role of herbal plants in the diabetes mellitus therapy: An overview. Int J Appl Pharm 2014;6(3):1-3.
- 12. Chang YC, Huang HP, Hsu JD, Yang SF, Wang CJ. Hibiscus anthocyanins rich extract-induced apoptotic cell death in human promyelocytic leukemia cells. Toxicol Appl Pharmacol 2005;205(3):201-12.
- 13. Amor BB, Allaf K. Impact of texturing using instant pressure drop treatment prior to solvent extraction of anthocyanins from Malaysia roselle (*Hibiscus sabdariffa*). Food Chem 2009;115(3):820-5.
- Chew YL, Goh JK, Lim YY. Assessment of *in vitro* antioxidant capacity and polyphenolic composition of selected medicinal herbs from Leguminosae family in Peninsular Malaysia. Food Chem 2009;116:13-8.
- Yang L, Gou Y, Zhao T, Zhao J, Li F, Zhang B, et al. Antioxidant capacity of extracts from calyx fruits of roselle (*Hibiscus sabdariffa* L.). Afr J Biotechnol 2012;11(17):4063-8.
- 16. Camelo-Méndez GA, Ragazzo-Sánchez JA, Jiménez-Aparicio AR, Vanegas-Espinoza PE, Paredes-López O, Del Villar-Martínez AA. Comparative study of anthocyanin and volatile compounds content of four varieties of Mexican roselle (*Hibiscus sabdariffa* L.) by multivariable analysis. Plant Foods Hum Nutr 2013;68:229-34.
- Wang CJ, Wang JM, Lin WL, Chu CY, Chou FP, Tseng TH. Protective effect of *Hibiscus* anthocyanins against tert-butyl hydroperoxideinduced hepatic toxicity in rats. Food Chem Toxicol 2000;38:411-6.
- McKay DL, Chen CY, Saltzman E, Blumberg JB. Hibiscus sabdariffa L. tea (tisane) lowers blood pressure in prehypertensive and mildly hypertensive adults. J Nutr 2010;140:298-303.
- Mizrahi Y, Nerd A, Nobel PS. Cacti as crops. Hortic Rev 1997;18:291-320.
- Aziz FA, Noor MM. Ethanol extract of dragon fruit and its effects on sperm quality and histology of the testes in mice. Biomed Res 2010;21(2):126-30.
- Montealegre RR, Peces RR, Vozmediano JL, Gascuena JM, Romero EG. Phenolic compounds in skins and seeds of ten grape *Vitis vinifera* varieties grown in a warm climate. J Food Comp Anal 2006;19:687-93.
- Orak HH. Total antioxidant activities, phenolics, anthocyanins, polyphenoloxidase activities of selected red grape cultivars and their correlations. Sci Hortic 2007;111:235-41.
- Miletić N, Popović B, Mitrović O, Kandić M. Phenolic content and antioxidant capacity of fruits of plum cv. 'stanley' (*Prunus domestica L.*) As influenced by maturity stage and on-tree ripening. Aust J Crop Sci 2012;6(4):681-7.
- 24. Gil MI, Tomás-Barberán FA, Hess-Pierce B, Kader AA. Antioxidant capacities, phenolic compounds, carotenoids, and Vitamin C contents of nectarine, peach, and plum cultivars from California. J Agric Food Chem 2002;50:4976-82.
- Kristl J, Slekovec M, Tojnko S, Unuk T. Extractable antioxidants and non-extractable phenolics in the total antioxidant activity of selected plum cultivars (*Prunus domestica* L.): Evolution during on-tree ripening. Food Chem 2011;125:29-34.
- Johanningsmeier SD, Harris GK. Pomegranate as a functional food and nutraceutical source. Annu Rev Food Sci Technol 2011;2:181-201.
- Stover E, Mercure EW. The pomegranate: A new look at the fruit of paradise. Hortic Sci 2007;42:1088-92.

- Taya S, Punvittayagul C, Inboot W, Fukushima S, Wongpoomchai R. Cleistocalyx nervosum extract ameliorates chemical-induced oxidative stress in early stages of rat hepatocarcinogenesis. Asian Pac J Cancer Prev 2014;15(6):2825-30.
- Natakankitkul S, Poontawee W, Wongmekiat O, Suttajit M. Potential Antioxidant Activities of Extracts from Proanthocyanidin rich Fruits for Food Supplement and Cosmetic. The 6th International Conference on Natural Products for Health and Beauty (NATPRO6); 2016.
- Kelebek H, Selli S. Evaluation of chemical constituents and antioxidant activity of sweet cherry (*Prunus avium* L.) cultivars. Int J Food Sci Technol 2011;46:2530-7.
- Oloyede FM, Oloyede FA, Obuotor EM. Comparative studies of chemical compositions of two species of Basella. App Sci Report 2013;3(2):121-4.
- 32. Milind P, Monika. Sweet potato as a super-food. Int J Res Ayurveda Pharm 2015;6(4):557-62.
- Teow C, Truong VD, McFeeters R, Thompson R, Pacota K, Yencho G. Antioxidant activities, phenolic and β-carotene contents of sweet potato genotypes with varying flesh colours. Food Chem 2007;103:829-38.
- Steed LE, Truong VD. Anthocyanin content, antioxidant activity, and selected physical properties of flowable purple-fleshed sweetpotato purees. J Food Sci 2008;73:S215-21.
- Markakis P, editor. Stability of anthocyanins in foods. In: Anthocyanins as Food Colors. New York, USA: Academic Press; 1982. p. 163-80.
- Hanson PM, Yang RY, Tsou SC, Ledesma D, Engle L, Lee TC. Diversity in eggplant (*Solanum melongena*) for superoxide scavenging activity, total phenolics, and ascorbic acid. J Food Compost Anal 2006;19(6-7):594-600.
- Rubatzky EV, Yamaguchi M. Lettuce and other composite vegetables.
 In: World Vegetables: Principles, Production and Nutritive Values.
 2nd ed. Boston, MA: Springer US, 1997. p. 333-70.
- 38. Jiraungkoorskul K, Jiraungkoorskul W. Sesbania grandiflora: New nutraceutical use as antidiabetic. Int J Pharm Pharm Sci 2015;7(7):26-9.
- Kungsuwan K, Singh K, Phetkao S, Utama-Ang N. Effects of pH and anthocyanin concentration on color and antioxidant activity of *Clitoria* ternatea extract. Food App Biosci J 2014;2(1):31-46.
- Belancic A, Agosin E, Ibacache A, Bordeu E, Baumes R, Razungles A, et al. Influence of sun exposure on the aromatic composition of Chilean Muscat grape cultivars Moscatel de Alejandra, and Moscatel rosada. Am J Enol Vitic 1997;48:181-6.
- 41. Reynolds AG, Wardle DA, Hall JW, Dever M. Fruit maturation of four *Vitis vinifera* cultivars in response to vineyard location and basal leaf removal. Am J Enol Vitic 1995;46:542-58.
- Pengkumsri N, Chaiyasut C, Saenjum C, Sirilun S, Peerajan S, Suwannalert P, et al. Physicochemical and antioxidative properties of black, brown and red rice varieties of northern Thailand. Food Sci Technol (Campinas) 2015a;35(2):331-8.
- 43. Pengkumsri N, Chaiyasut C, Sivamaruthi BS, Saenjum C, Sirilun S, Peerajan S, *et al.* The influence of extraction methods on composition and antioxidant properties of rice bran oil. Food Sci Technol (Campinas) 2015b;35(3):493-501.
- Veigas JM, Narayan MS, Laxman PM, Neelwarne B. Chemical nature, stability and bioefficacies of anthocyanins from fruit peel of Syzygium cumini skeels. Food Chem 2007;105:619-27.
- Usenik V, Stampar F, Kastelec D. Phytochemicals in fruits of two *Prunus domestica* L. plum cultivars during ripening. J Sci Food Agric 2013;93(3):681-92.
- Montilla EC, Hillebrand S, Winterhalter P. Anthocyanins in purple sweet potato (*Ipomoea batatas* L.) varieties. Fruit, Veg Cereal Sci Biotechnol 2011;5(2):19-24.
- Kallithraka S, Aliaj L, Makris DP, Kefalas P. Anthocyanin profiles of major red grape (*Vitis vinifera* L.) varieties cultivated in Greece and their relationship with *in vitro* antioxidant characteristics. Int J Food Sci Technol 2009;44:2385-93.
- 48. Figueiredo-González M, Simal-Gándara J, Boso S, Martínez MC, Santiago JL, Cancho-Grande B. Anthocyanins and flavonols berries from *Vitis vinifera* L. cv. Brancellao separately collected from two different positions within the cluster. Food Chem 2012;135:47-56.
- Srivastava BK, Pande CS. Anthocyanins from the flowers of Clitoria ternata. Planta Med 1977;32:138-40.
- Kouakou TH, Konkon NG, Ayolié K, Obouayeba AP, Abeda ZH, Koné M. Anthocyanin production in calyx and callus of Roselle (*Hibiscus sabdariffa* L.) and its impact on antioxidant activity. J Pharm Phytochem 2015;4(3):9-15.
- McDougall GJ, Fyffe S, Dobson P, Stewart D. Anthocyanins from red cabbage - stability to simulated gastrointestinal digestion. Phytochemistry 2007;68(9):1285-94.

- Turfan O, Türkyılmaz M, Yemiş O, Özkan M. Anthocyanin and colour changes during processing of pomegranate (*Punica granatum* L., cv. Hicaznar) juice from sacs and whole fruit. Food Chem 2011;129(4):1644-51
- Türkyılmaz M. Anthocyanin and organic acid profiles of pomegranate (*Punica granatum* L.) juices from registered varieties in Turkey. Int J Food Sci Technol 2013;48:2086-95.
- 54. Rengarajan S, Melanathuru V, Munuswamy D. Comparative studies of antioxidant activity from the petals of selected Indian medicinal plants. Int J Pharm Pharm Sci 2015;7(6):29-34.
- Borochov-Neori H, Judeinstein S, Harari M, Bar-Ya'akov I, Patil BS, Lurie S, et al. Climate effects on anthocyanin accumulation and composition in the pomegranate (*Punica granatum* L.) fruit arils. J Agric Food Chem 2011;59(10):5325-34.
- Sadilova E, Stintzing FC, Carle R. Anthocyanins, colour and antioxidant properties of eggplant (Solanum melongena L.) and violet pepper (Capsicum annuum L.) peel extracts. Z Naturforsch C 2006:61(7-8):527-35.
- Gallo M, Naviglio D, Ferrara L. Nasunin, an antioxidant anthocyanin from eggplant peels, as natural dye to avoid food allergies and intolerances. ESJ 2014;10:9.
- Jansom C, Bhamarapravati S, Itharat A. Major anthocyanin from ripe berries of *Cleistocalyx nervosum* var. *paniala*. Thammasat Med J 2008;8(3):364-70.
- Mozeti B, Trebse P, Hribar J. Determination and quantitation of anthocyanins and hydroxycinnamic acids in different cultivars of sweet cherries (*Prunus avium* L.) from Nova Gorica Region (Slovenia). Food Technol Biotechnol 2002;40(3):207-12.
- Leja M, Kamińska I, Kołton A. Phenolic compounds as the major antioxidants in red cabbage. Folia Hortic Ann 2010;22(1):19-24.
- 61. Miletić N, Mitrović O, Popović B, Nedović V, Zlatković B, Kandić M.

- Polyphenolic content and antioxidant capacity in fruits of plum (*Prunus domestica* L.) cultivars "Valjevka" and "Mildora" as influenced by air drying. J Food Qual 2013;36:229-37.
- Nisha P, Abdul Nazar P, Jayamurthy P. A comparative study on antioxidant activities of different varieties of *Solanum melongena*. Food Chem Toxicol 2009;47(10):2640-4.
- Mohd-Esa N, Hern FS, Ismail A, Yee CL. Antioxidant activity in different parts of roselle (*Hibiscus sabdariffa* L.) extracts and potential exploitation of the seeds. Food Chem 2010;122:1055-60.
- Hayaloglu AA, Demir N. Physicochemical characteristics, antioxidant activity, organic acid and sugar contents of 12 sweet cherry (*Prunus avium* L.) cultivars grown in Turkey. J Food Sci 2015;80(3):C564-70.
- 65. Bunea CI, Pop N, Babes AC, Matea C, Dulf FV, Bunea A. Carotenoids, total polyphenols and antioxidant activity of grapes (*Vitis vinifera*) cultivated in organic and conventional systems. Chem Cent J 2012;6(1):66
- Loganayaki N, Suganya N, Manian S. Evaluation of edible flowers of agathi (Sesbania grandiflora L. Fabaceae) for in vivo anti-inflammatory and analgesic, and in vitro antioxidant potential. Food Sci Biotechnol 2012;21(2):509-17.
- 67. Ji H, Zhang HX, Li HT, Li YC. Analysis on the nutrition composition and antioxidant activity of different types of sweet potato cultivars. Food Nutr Sci 2015;6:161-7.
- Rabeta MS, An Nabil Z. Total phenolic compounds and scavenging activity in *Clitoria ternatea* and *Vitex negundo* linn. Int Food Res J 2013;20(1):495-500.
- Sivamaruthi BS, Pengkumsri N, Saelee M, Kesika P, Sirilun S, Peerajan S, et al. Impact of physical treatments on stability and radical scavenging capacity of anthocyanidins. Int J Pharm Pharm Sci 2016;8(1):162-7.