Comparative Studies of Limonin and Naringin Distribution in Different Parts of Pummelo [Citrus grandis (L.) Osbeck] Cultivars Grown in Thailand

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ABSTRACT

Seven pummelo cultivars: Kao Numpueng (KNP), Thong Dee (TD), Kao Paen (KP), Kao Yai (KY), Tha Knoi (TK), Kao Tanggwa (KTG) and Pattavee (PV) grown under Thai pummelo fruit conditions were selected for comparative studies on the distribution of limonin and naringin in various parts of the fruit. In this study, the highest amount of limonin was detected in the seeds of all cultivars. Lesser amounts were detected in albedo followed by flavedo, segment membranes and juice. The limonin content in the seeds ranged from 1375.31- 2615.30 ppm, with the lowest amount in the juice being 10.07-29.62 ppm. The limonin in albedo in decreasing order of cultivars was: TK, TD, KTG, KNP, PV, KY and KP (352.72-133.58-ppm). The limonin in flavedo in decreasing order of cultivars was: PV, TD, KP, KNP, TK, KTG and KY (295.49-130.16 ppm), while in segment membranes the decreasing order of cultivars was: PV, KTG, KNP, TD, KY, KP and TK (293.14-85.81 ppm). Naringin was found in a greater amount than limonin in all fruit parts of the cultivars studied. The order of fruit parts that contained naringin in a decreasing amount was: albedo, flavedo, segment membranes, seeds and juice. The order of cultivars containing naringin in the albedo in a decreasing amount was: KTG, KNP, KP, KY, TK, PV, TD (28,508.01–10,065.06 ppm). In the flavedo in decreasing amounts, the cultivar order was: PV, KTG, KNP, KP, KY,TD and TK (8,964.24–2,483.96 ppm), while in the segment membranes the decreasing order was: KTG, KY, TD, TK, PV, KP and KNP (4,369.50-1,799.48 ppm). In the seeds, the decreasing order was: PV, KTG, KNP, TD, and KP (426.66-257.87 ppm). In the juice, the decreasing order was: PV, TK, KY, TD, KNP, KP and KTG (386.45-242.63 ppm). The chemical composition of the pummelo juice samples showed a high content of ascorbic acid in the range of 37.03-57.59 mg/100ml. The total soluble solids range was 7.14-9.45 OBrix, the titratable acidity range was 0.38-0.98(g/100ml) as citric acid, and the pH range was 3.69-4.05.

Key words: pummelo, cultivars, limonin, naringin

INTRODUCTION

Pummelo [Citrus grandis (L.) Osbeck] is widely grown in many countries in southeast Asia. It is considered the largest citrus fruit, with

a diameter that may reach more than 12" with a yellow or green skin, white or pinkish flesh, and a sweetish–acidic flavor. It is also a good source of vitamin C and antioxidants (Rahman *et al.*, 2003; Xu *et al.*, 2008). The pummelo juice industry is

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becoming increasingly important due to the excellent taste and nutrition of the pummelo. However the bitterness in the juice is a problem, like with other citrus juices.

The bitterness in citrus fruit is affected by limonin and naringin, which are generally recognized as the major two bitter compounds. Limonin is the bitter limonoid found in major citrus cultivars such as grapefruit, the Navel orange, and the Shamouti orange (Guadagni et al., 1973). Naringin is the bitter flavonoid which was first found in grapefruit. Limonin is synthesized as a non bitter form (limonoate A ring lactone) in leaves and transported to fruit and seeds. Thus each part of the fruit contains different amount of limonin (Maier et al., 1977) Naringin is not transported after being synthesized in the fruit or leaves. Limonin and naringin co-exist in most citrus cultivars, but in different amounts, and their threshold levels are different. The limonin in orange juice has been detected by taste panels at a minimum concentration of 6 ppm (Guadagni et al., 1973, Kimball and Norman, 1990), whereas naringin at a high concentration gives a bitter taste. Limoniod and flavonoid in orange or pummelo juice have been reported by many authors (Bennett and Hasegawa, 1982; Hasegawa et al., 1986; Hashinaga et al., 1990). The average amount of limonin content in the juice from 16 pummelo cultivars was found to be 18 ppm (Ohta and Hasegawa, 1995). The limonin and naringin content in the juice from eight pummelo cultivars namely: Kao Yai, Kao Numpueng, Kao Tanggwa, Kao Hom, Kao Paen, Tong Dee, Tha Khoi and one local variety were reported in the range of 6.82-32.40 ppm and 200.84-578.76 ppm, respectively (Wattanasiritham et al., 2005). The limonin and naringin content in each part of the pummelo fruit have not been reported. Orange peel is an essential ingredient of marmalade and the albedo of pummelo is used for candy pummelo. So there are many distinct cultivars available in the market, with each cultivar varying in its physical characteristics and juice composition. The objective of this study was to compare the presence of limonin and naringin in various parts of the fruit in seven pummelo cultivars which are commercially grown. The results of this study will be useful for the juice industry.

MATERIALS AND METHODS

Materials

Samples of the seven pummelo cultivars [Citrus grandis (L.) Osbeck] were collected from orchards in five provinces between November 2005 and February 2006. Fruit were harvested at the age of eight months. The cultivars were: Thong Dee (TD), Kao Paen (KP) and Kao Nampheung (KNP) (Nakhon Pathom Province); Kao Yai (KY) (Samut Songkhram Province); Tha Knoi (TK) (Phichit Province); Kao Tanggkya (KTG) (Chi Nat Province); and Pattavee (PV) (Nakhon Si Thammarat Province).

Preparation of samples

Five pummelo fruits were randomly chosen from different trees. Each whole fruit was weighed and divided into five different parts: flavedo, albedo, segment membranes, seeds and juice as shown in Figure 2. All tissue except for the juice was homogeneously blended in a blender (Moulinex Model A 327 R7) and dried at -40°C in a freeze dryer (Heto model LyoPro 3000) for 12-15 hrs. These components were then ground and vacuum-packed and kept in a freezer at -20 °C for further analysis. The juice was stored at -20°C. Before analysis, the frozen juice was thawed in a hot water bath at 80°C for 15 minutes which completely changed any limonin A-ring lactone to limonin (Couture and Rouseff, 1992).

Extraction of limonin from juices

To determine the limonin content in the juice, 10 ml of the juice sample was centrifuged at 2500×g for 10 min. Before use, the Millipore C18 Sep-pak cartridge was rinsed with 2 ml of methanol and then 5 ml of deionised water. Then 1 ml of juice supernatant was passed through the cartridge. The cartridge was rinsed with 5 ml of deionised water and limonin was slowly eluted from the cartridge with 1 ml of methanol. The methanol effluent was filtered through a 0.22 µm nylon filter prior to analysis using high performance liquid chromatography (HPLC) (Shaw and Wilson, 1984).

Extraction of naringin from fruit juice

The following process was used to determine the naringin content in the juice. A sample of 1-2 ml of the juice was extracted with 4 ml of methanol by shaking for 1 min using a Vortex Mixer and then followed by centrifuging at $2500 \times g$ for 10 min. The extract was passed through a 0.22 μ m nylon filter prior to the HPLC analysis (Rouseff, 1988).

Extraction of limonin and naringin from fruit parts

Samples of 5 g of fruit tissue were weighed and then extracted with 20 ml of methanol by shaking for 1 min with a Vortex Mixer, followed by centrifuging at 2500×g for 10 min. The extract was passed through a 0.22 μ m nylon filter prior to the HPLC analysis.

Standards

Limonin ($C_{26}H_{30}O_8$) and naringin ($C_{27}H_{32}O_{12}$) were obtained from Sigma-Aldrich Chemical Co. (St. Louis, USA). Standard solutions of limonin were prepared weekly by diluting the stock solution to 2.5, 5, 10, 15, 25 and 35 ppm with the mobile phase. Standard solutions of naringin were prepared weekly by diluting the stock solution to 20, 40, 60, 80, 100, 120, 140 and 160 ppm with the mobile phase.

Determination of limonin and naringin by HPLC

Limonin and naringin were determined by a reverse-phase HPLC method. The system consisted of a water HPLC (USA) system with two hydraulic pumps (model 515), an injection system (U6K), a Novapak C₁₈ Column (3.9 x 150 mm, pore size 4 µm), a C₁₈ guard column, a UV-VIS detector (model 2478) and a computerized recorder/integrator (model Millennium 32). For limonin determination, the mobile phase consisted of acetonitrile:deionised water (35:65) with a flow rate of 1 ml/min. The injection volume of the sample was 20 µl. The detection wavelength was 210 nm. For naringin, the mobile phase consisted of acetonitrile:deionised water (25:75) with a flow rate of 1 ml/min and the injection volume of the sample was 20 µl. The detection wavelength was 280 nm.

Ascorbic acid, pH, total titratable acidity and Brix

Ascorbic acid was determined by a titration method (AOAC, 1995). The pH was measured by a digital pH meter (CG 842 Schott, Germany). The total acidity of the juice samples was determined by titration with 0.1N NaOH using phenolphthalein as the indicator. The acidity of the juice samples was calculated as citric acid. A refractometer (Atago N1 Brix 0-32%, Japan) was used to measure the soluble solids as degrees Brix.

RESULTS AND DISCUSSION

In this study, the seven commercial pummelo cultivars sampled had a weight range from 897 to 2400 g and an average weight of 1651.36 g. KNP produced the heaviest fruit (2400 g) whereas the lightest one (897.5g) was KP (Table 1). Other research has reported a weight range from 700-1000 g (Cedeno-Maldonado *et al.*, 1993). The weight of pummelo fruit, even in the same cultivar, can vary due to differences in nutrition and the growing environment (Rahman

et al., 2003). The weight of the whole fruit was categorized into three groups: small, medium and large. Small fruit were obtained only from KP, while TD, PV, KTG and TK had medium weight whole fruit and large fruit were obtained from KNP and KY (Table 2).

No relationship was found between the weight of the whole fruit and fruit parts weight (flavedo, albedo, segment membranes, seeds and juice). However, juice weight was one of the important characteristics of edible quality (Vitamin C and mineral). The greatest weight of juice was in TD (44.38 g/100g whole fruit). The range in the weight of seeds was 1.37-0.01 g/100g whole fruit. There were two cultivars, TK and KY, which had no seeds. Rahman *et al.* (2003) reported that the seeds of 30 local accessions of pummelo varied in the range from 0.29 to 0.59 g. In this study, KNP had a greater seed weight than any previously reported.

Distribution of limonin and naringin in pummelo fruit parts

The linear regression equation for the curve for limonin content was y=15647x-5042.5 with a correlation coefficient (r^2) of 0.9976 (Figure 1). The limonin content from the seven pummelo cultivars was analyzed by an HPLC method. Figure 2 shows the chromatogram for limonin from a pummelo juice sample.

Each fruit part - flavedo, albedo, segment membranes, seeds and juice - was analyzed for limonin content. Figure 3 shows the cross-section of pummelo indicating the distribution of limonin in each part and the different concentrations of limonin (Table3). The limonin concentration was highest in seeds in all cultivars (1375.31-2615.30 ppm), while for the other parts, the ranking in decreasing order was albedo (133.58-352.72 ppm), flavedo (130.16-295.49 ppm), segment membranes (85.81-293.14 ppm), and juice (10.07-

Table 1 Fruit parts from seven pummelo cultivars (g/100g whole fruit).

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Cultivar	Whole	hole g/100g Whole fruit					
	fruit	Flavedo	Albedo	Segment	Seeds	Juice	
	(g)			membranes			
KNP	2400	8.27±0.43	26.47±3.9	25.18±2.17	1.37±0.34	38.71±3.21	
KP	897.5	14.42±1.02	41.34±2.44	8.27 ± 0.90	0.50 ± 0.16	35.43±2.11	
KTG	1563	10.34 ± 0.13	34.54±0.30	21.09±1.54	0.76 ± 0.10	33.28±1.59	
KY	2118	8.18±0.82	37.07±1.48	21.81±1.87	No-Seeds	32.95±0.43	
PV	1445	10.38±1.05	44.12±2.06	18.25±1.83	0.01 ± 0.00	27.24±1.81	
TD	1500	11.89±1.09	34.05±1.22	9.50 ± 0.55	0.18 ± 0.03	44.38±1.89	
TK	1636	10.25 ± 0.22	53.53±0.87	7.74 ± 0.88	No-Seeds	28.48±1.33	

KNP=Kao Nampheung, KP=Kao Paen, KTG=Kao Tanggkya, KY= Kao Yai, PV=Pattavee,

TD = Thong Dee, TK = Tha Knoi. Values are mean ±SD (Data are average of five fruit samples).

Table 2 Fruit parts from seven pummelo cultivars by weight class.

	1	1	-	0		
Class	Whole fruit	Flavedo	Albedo	Segment	Seeds	Juice
				membranes		
Small	KP	KNP,KY	KNP	KP,TD,TK	PV, TD	PV,TK
Medium	KTG, PV,	KTG,PV,	KP,KTG,KY,	PV	KP, KTG	KNP,
	TD, TK	TD,TK	PV,TD			KP,KTG,KY
Large	KNP, KY	KP	TK	KNP,KTG,KY	KNP	TD

KNP=Kao Nampheung, KP=Kao Paen, KTG=Kao Tanggkya, KY= Kao Yai, PV=Pattavee,

TD =Thong Dee, TK =Tha Knoi.

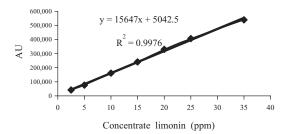


Figure 1 Calibration graph of limonin.

29.62 ppm), respectively. The decreasing order of limonin in fruit parts was similar to that reported by McIntoch *et al.* (1982) who stated that the highest limonin level within grapefruit tissue was found in the cotyledon, followed by the inner seed coat, outer seed coat, center pith, segment membranes, albedo, flavedo and juice vesicles.

The findings of Jungsakulruijirek (1997) showed similar results for the distribution of limonin in Thai tangerine, where the limonin concentration in decreasing order was seeds (1480 mg), albedo (205 ppm), flavedo (180 ppm), segment membranes (10 ppm) and juice sacs (1 ppm). The pattern of limonin distribution in pummelo fruit was similar to that in other citrus fruits, but differences in concentrations may exist,

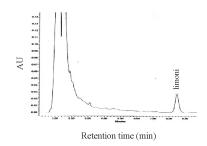


Figure 2 Chromatogram of limonin in pummelo juice.

which might be due to genetic characteristics. The outer tissues of citrus fruit (flavedo, albedo and segment membranes) showed a definite trend of increasing limonin concentration toward the distal end of the fruit (the seeds). Apparently, the juice contained small amounts of limonin.

In seeds, the greatest limonin content (2615.30-1375.31 ppm) was found in KNP, followed by KP, TD, PV and KTG, in decreasing amounts. In flavedo the greatest limonin content (295.49-130.16 ppm) was found in PV followed by TD, KP, KNP, TK, KTG and KY. In albedo, the limonin content (352.72-133.58ppm) was greatest in TK followed by TD, KTG, KNP, PV, KY and KP, while in segment membranes, the limonin content (293.14-85.81ppm) was the

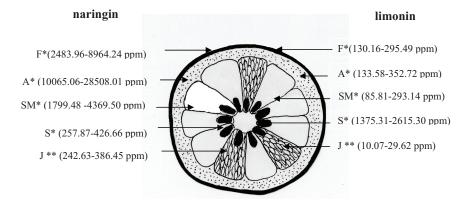


Figure 3 Cross section of pummelo showing distribution of limonin (ppm) and naringin (ppm) in each fruit part. F=Flavedo, A=Albedo, SM= Segment membranes, S=seeds, J=juice *Unit of limonin concentration in flavedo, albedo, segment membranes and seeds from dry weight (DW).

^{**}Unit of limonin concentration in juice from fresh weight (FW).

I WOIC C	Zimonin content	(ppin) in paining	o man parts.				
Cultivars	Tissues						
	Flavedo *	Albedo *	Segment membranes *	Seeds *	Juice **		
KNP	169.30±17.75	252.81±36.36	265.14±55.97	2615.30±453.37	22.69±3.94		
KP	215.06±32.93	135.20±14.89	87.64±8.79	2605.56±472.64	18.27±3.70		
KTG	150.52±29.12	259.31±48.25	267.34±42.04	1375.31±85.81	10.07±164		
KY	130.16±31.32	233.05±27.58	111.59±21.64	No-Seeds	21.43±3.57		
PV	295.49±33.78	239.17±47.24	293.14±48.85	2443.46±463.43	20.02±1.68		
TD	217.72±48.84	313.58±64.50	122.69±133.80	2594.60±524.63	29.62±5.42		
TK	159.88±31.01	352.72±72.59	85.81±18.54	No-Seeds	25.52±3.54		

Table 3 Limonin content (ppm) in pummelo fruit parts.

KNP=Kao Nampheung, KP=Kao Paen, KTG=Kao Tanggkya, KY=Kao Yai, PV=Pattavee,

greatest in PV followed by KTG, KNP, TD, KY, KP and TK. In the juice, the limonin content (29.62-10.07 ppm) was greatest in TD followed by TK, KNP, KY, PV, KP and KTG. The average limonin content in the juice samples was 21.09 ppm. In fact the juice is the most important component for industry. Some other reports on the limonin content in pummelo juice include Wattanasiritham et al. (2005) who found that the limonin concentrations in the juice of eight pummelo cultivars ranged from 6.82 to 32.40 ppm and the average was 21.07 ppm. Ohta and Hasegawa (1995) analyzed the limonin content in the pummelo juice of 16 cultivars and reported an average content of 18 ppm. The high concentration of limonin in pummelo juice is a problem for the pummelo juice industry and customers' acceptance, due to the threshold concentration of limonin being low at 6 ppm (Guadagni et al., 1973; Kimball and Norman, 1990). It is recommended to use the variety with the least amount of limonin for the juice industry. However the limonin concentration in each variety was not directly comparable, because of the effect of several conditions such as growing region, cultural practices and nutrition. It is suggested that the limonin content in the juice be determined before processing.

The linear regression equation for the

curve for the naringin content was y=20408x+55859 with the correlation coefficient (r^2) being 0.9991 (Figure 4). The naringin was analyzed by an HPLC method. Figure 5 shows the chromatogram of naringin in a pummelo juice sample.

The distribution pattern of naringin within the fruit tissues of pummelos was different from that of limonin. The major source of naringin was the albedo, while seeds and juice contained a minor amount. Similar findings were reported by Barthe et al. (1988). Fruit parts that contained naringin in a decreasing order were: albedo (10,065.06-28,508.01 ppm), flavedo (2,483.96-8,964.24 ppm), segment membranes (1,799.48-4,369.50 ppm), juice (242.63-386.45 ppm) and seeds (257.87-426.66 ppm). With respect to cultivar differences, PV had the highest naringin concentration in the juice (386.45 ppm), followed by TK, KY, TD, KNP and KP (381.24-315.71 ppm) while KTG had the lowest naringin concentration (242.63 ppm) (Table 4 and Figure 3). The average concentration of naringin in the juice was 337.46 ppm. Wattanasiritham et al. (2005) analyzed eight cultivars of pummelo and found the average naringin content in the juice was 366.42 ppm. Xu et al. (2008) analysed two pummelo cultivars -Miyou and Sijiyou - and found the level of naringin ranged from 108.52 to 125.79 ppm.

TD=Thong Dee, TK=Tha Knoi. Values are mean ±SD (Data are average from five fruit samples)

^{*}Unit of limonin concentration in flavedo, albedo, segment membranes, and seeds from dry weight (DW).

^{**}Unit of limonin concentration in the juice from fresh weight (FW).

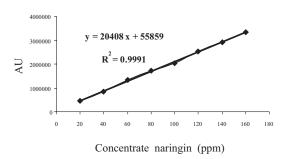


Figure 4 Calibration graph of naringin.

It is possible that naringin may not contribute to the bitterness of pummelo juice, because the threshold level was as high as 800 ppm (Rouseff *et al.*, 1988).

In terms of cultivar, KTG had the highest naringin concentration in the albedo (28,508.01 ppm) followed by KNP and KP (19,331.55 and 12,133.49 ppm), while TD had the lowest naringin concentration (10,065.06 ppm.) In the flavedo of PV, KTG and KNP, the naringin content was 8,964.24,7,716.76 and 5,739.44 ppm, respectively, while TK had the lowest naringin concentration (2,483.96 ppm).

In the segment membranes of KTG, KY and TD, the naringin content was 4,369.50,

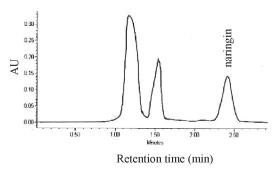


Figure 5 Chromatograms of naringin in the pummelo.

3,852.12 and 3391.61 ppm, respectively, while KNP had the lowest naringin concentration (1799.48 ppm).

In seeds of PV, KTG and KNP, the naringin content was 426.66, 388.88 and 297.48 ppm, respectively, while KP had the lowest naringin concentration (257.87 ppm).

Juice composition

It was obvious that the choice of cultivars affected the composition of the juice. The ascorbic acid content in all cultivars studied varied from 37.03 to 57.59 mg/100ml. With reference to the ADI for ascorbic acid (60 mg) (Rouseff and Nagy, 1994), and research showing the ascorbic acid

Table 4 The naringin content (ppm) in pummelo fruit parts.

Cultivars	Tissues						
	Flavedo*	Albedo*	Segment	Seeds*	Juice**		
	membranes*						
KNP	5739.44±430.40	19331.55±1129.35	1799.48±406.86	297.48±47.37	323.00±43.62		
KP	5344.56±924.32	12133.49±1602.34	2031.03±224.65	257.87±43.99	315.71±34.48		
KTG	7716.76±863.62	28508.01±2619.83	4369.50±493.35	388.88±28.58	242.63±33.64		
KY	5245.40±817.01	11683.58±1296.74	3852.12±506.42	No-Seeds	364.68±82.91		
PV	8964.24±870.79	11172.13±753.48	2067.92±523.08	426.66±80.66	386.45±80.22		
TD	4609.86±522.77	10065.06±248.34	3391.61±747.59	293.03±52.26	348.47±54.93		
TK	2483.96±875.84	11543.97±993.47	2095.47±349.26	No-Seeds	381.24±67.19		

KNP = Kao Nampheung, KP= Kao Paen, KTG= Kao Tanggkya, KY= Kao Yai, PV= Pattavee,

TD = Thong Dee, TK = Tha Knoi Values are mean ±SD. (Data are average from five fruit samples).

^{*} Unit of naringin concentration in flavedo, albedo, segment membranes and seeds from dry weight (DW).

^{**}Unit of naringin concentration in juice from fresh weight (FW).

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Cultivars	Chemical properties						
	Ascorbic acid	Total soluble	Titratable acidity	°Brix/acid ratio	pН		
	(mg/100 ml)	solid (°Brix)	(g/100 ml)				
TD	57.59±0.71	8.50±0.52	0.51±0.03	16.62±1.57	4.05±0.12		
KY	50.33±1.25	9.10±0.84	0.49 ± 0.05	18.67±1.02	3.96 ± 0.05		
KP	40.24±1.98	8.00 ± 0.00	0.38 ± 0.01	20.89±0.76	4.02±0.11		
KNP	43.75±0.92	9.45±0.28	0.98 ± 0.04	15.42±0.39	3.72 ± 0.10		
TK	37.03±2.04	8.45±0.43	0.61 ± 0.03	12.84±1.02	3.69 ± 0.08		
KTG	48.79±1.96	7.14±0.99	0.58 ± 0.03	12.43±1.91	3.85±0.17		
PV	44.62±1.65	8.00±1.05	0.58 ± 0.03	13.65±1.79	3.80±0.11		

Table 5 Juice composition of seven pummelo cultivars.

KNP = Kao Nampheung , KP= Kao Paen, KTG= Kao Tanggkya, KY= Kao Yai, PV= Pattavee,

TD =Thong Dee, TK =Tha Knoi Values are mean ±SD. (Data are average from five fruits)

content of citrus fruit varied from 20 to 79 mg/100ml (Nagy, 1980), pummelo fruit is a good source of vitamin C (Table 5).

TD had the highest ascorbic acid content 57.59 mg/100ml, followed by KY (50.33 mg/100ml) and KTG (48.79 mg/100ml), respectively, whereas the cultivar TK had the lowest ascorbic acid content, 37.03 mg/100ml. The Brix/acid ratio ranged from 12.43–20.89, whereas for the orange juice industry its range has been reported from 10-16 (Salumkhe and Desai, 1984).

The Brix/acid ratio was also identified as an important parameter related to the quality of citrus fruit (Xu *et al.*, 2008). The cultivar KP had the highest Brix/acid ratio (20.89), followed by KY and TD (18.67 and 16.62). The cultivar KTG had the lowest Brix/acid ratio of (12.43). Therefore pummelo juice was suitable for citrus juice processing. The pH of the pummelo juice was in the range 3.65–4.02. Ohta and Hasegawa (1995) reported that generally, the pH of pummelo juice was between 3 and 4.

Hasegawa *et al.* (1992) reported that the acidity of citrus plays a great role in terms of bitterness, because under low pH conditions, the A-ring lactone (LARL) can be converted to limonin.

CONCLUSIONS

Limonin and naringin were distributed in the flavedo, albedo, segment membranes, juice and seeds in different amounts. The cultivar type had an effect on the amount of limonin and naringin. The highest amount of limonin was detected in seeds from KNP, followed by KP, TD, PV, and KTG. The lowest amount of limonin was found in the juice of KTG followed by KP, PV, KY, KNP, TK and TD. Matthew *et al.* (1990) reported that limonin in citrus fruits was found primarily in the seeds, with smaller amounts in the central core and segment membrane.

The highest amount of naringin was detected in the albedo of KTG followed by KNP, KP, KY, TK, PV and TD. The lowest amount was found in the juice of KTG followed by KP, KNP, TD, KY, TK and PV.

LITERATURE CITED

AOAC. 1995. Official methods of analysis 16th ed., The Association of Official Analytical Chemists. Arlington, Virginia. USA. Chapter 45: P.16-17.

Barthe, G.A., P.S. Jourdan C.A. McIntosh and R.L. Mansell. 1988. Radioimmuno-assay for the quantitative determination of hesperidin and

- analysis of its distribution in *Citrus sinensis*. **Phytochem**. 27: 249-254.
- Bennett, R.D. and S. Hasegawa. 1982. 7a Oxygenated limonoids from the rutaceae. **Phytochem**. 21: 2349–2354.
- Cedeno-maldonado, A., W. Gonzalez and E. Fontanet.1993. Performance of pummelo clones in the central mountain region of Puerto Rico. **Bull. of Fruit Tree Res. Station**. 24: 14-22.
- Couture, R. and R. Rouseff. 1992. Debittering and deacidifying sour orange (*Citrus auarantium*) juices using neutral and anion exchanger resin. **J. Food Sci.** 57(2): 380-384.
- Guadagni, D.G., V.P. Maier, J.G. Turnbaugh. 1973. Effects of some citrus constituents on taste thresholds for limonin and naringin bitterness. **J. Sci. Food Agric**. 24: 1199-1205.
- Hashinaga, F., C. H. Fong and S. Hasegawa. 1990. Biosynthesis of limonoids in citrus sudachi. **Agri Bio. Chem.** 54: 3019–3020.
- Hasegawa, S., Z. Herman, E. Orme and P. Ou. 1986. Biosynthesis of limonoids in citrus: sites and translocation. **Phytochem.** 25: 2783-2785.
- Hasegawa, S., C.H. Fong., Z. Herman., M. Miya. 1992. Glucosides of limonoids. American Chemical Society 87-97.
- Jungsakulruijirek, S. 1997. Limonin in Thai Tangerine (*Citrus reticulata*, Blanco) in relation to reduction of juices bitterness. Ph. D. thesis, Asian Institute of Tecnology, Bangkok.
- Kimball, D.A. and S.I. Norman 1990. Change in California novel orange juices during commercial debittering. **J. Food Sci.** 55 (1): 273 -274.
- Maier, V.P., R.D. Bennett and S. Hasegwa. 1977.
 Limonin and other limonoids, pp.355-396. *In*S. Nagy, P.E. Shaw and M.K. Veldius (eds.).
 Citrus Science and Technology: Vol 1. The AVI Publishing Company, Inc.USA.

- McIntosh, C.A., R.L. Mansell and R.L. Rouseff. 1982.Distribution of limonin in the fruit tissue of nine grapefruit cultivar. **J. Agric Food Chem**. 30: 689-692.
- Nagy, S. 1980. Vitamin C contents of citrus fruit and their products: A review. **J. Agric. Food Chem**. 28: 8-18.
- Ohta, H. and S. Hasegawa. 1995. Limonoids in pummelos [*Citrus grandis* (L.) Osbeck] **J. Food Sci**. 60: 1284-1285.
- Rahman, M.M., M.G Rabbani, A.S.M.M.R. Khan, N. Ara and M.O. Rahman. 2003. Study on physio-morphological characteristics of different local pummelo accessions Pakistan. J. Bio. Sci. 6 (16): 1430-1434.
- Rouseff, R.L. 1988. Liquid chromatographic determination of naringin as a detector of Grapefruit juices in orange juices. **J. Assoc. Off. Anal. Chem.** 71: 798 802.
- Rouseff, R.L and S. Nagy. 1994. Potential health promoting properties of citrus fruit components. **Food Technol.**11: 125-132.
- Salumkhe, D.K. and B.B. Desai. 1984.

 Postharvest Biotechnology of Fruit Vol.1.

 Boca Raton: CRC press. Inc. Boca Raton.

 Florida.168 p
- Shaw, P.E. and C.W. Wilson. 1984. A rapid method for determination of limonin in citrus juices by high performance liquid chromatography.

 J. Food Sci. 49: 1216 -1218.
- Wattanasiritham, L., K.Taweesuk and B. Ratanachinakorn. 2005. Limonin and Naringin in Pummelos (Citrus grandis (L.) Osbeck). In 31st Congress on Science and Technology of Thailand at Suranaree University of Technology, 18–20 October 2005.
- Xu, G., D. Lin, J. Chen, X. Ye, Y. Me and J. Shi. 2008. Juices components and antioxidant capacity of citrus varieties cultivated in Chin. Food Chem. 106 (2): 545-551.