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Evaluation of quality changes of pressurized and pasteurized herbal-plant beverages during chilled storage

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Abstract

The physicochemical and microbiological qualities of pressurized herbal-plant beverage at 400 MPa and 25°C for 30 min and pasteurized beverage at 90°C for 2 min were investigated during storage at 4°C for 4 weeks. It was found that the brightness (L) of both processed beverages noticeably decreased (P < 0.05) with an increasing storage time, while other parameters such as a^* (reddish), b^* (yellowish), ΔE (total different colors) and BI (Browning Index) were trended to increase. In addition, the bioactive components, i.e. ascorbic acid, γ-oryzanol, asiatic acid, gallic acid, ellagic acid and total phenols, and antioxidant capacity (DPPH assay) in both processed beverages significantly declined (P < 0.05) throughout the entire storage. Overall the results displayed that pressurization could maintain the natural color and bioactive compounds of the product better than thermally process.

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Introduction

Consumption of herbal products or infusions can prevent certain diseases such as cancer and cardiovascular diseases, as these products are rich in various bioactive components and high antioxidant activity (Chunthanom *et al*., 2013). Herbal-plant beverage is a new product prepared from a mixture of dried longan (*Euphoria longana* Lam.), black glutinous aromatic rice (*Oryza sativa* L.) and fresh pennywort (*Centella asiatica* L.) extracts with a ratio of 1:1:1 by volume (Chaikham *et al*., 2014). Dried longan contains significant amounts of gallic acid, ellagic acid and corilagin (Rangkadilok *et al*., 2005) which have been established for their pharmacological properties such as anti-tyrosinase, anti-glycated, anti-fungal and anti-cancer (Yang *et al*., 2011). γ-Oryzanol, a predominant bioactive compound found in color rice, is also shows several health-promoting effects including antioxidant properties, anti-inflammatory activity and anti-cancer (Wilson *et al*., 2002; Lerma-Garcia *et al*., 2009). Moreover, the active principles of pennywort extract include asiatic acid, asiaticoside, madecassic acid and madecassoside exhibited to reduce chronic venous insufficiency and have wound-healing properties (Pointel *et al*., 1987; Apichartsrangkoon *et al*., 2012).

In general thermal processing such as pasteurization has been used to preserve several foods and beverages including medicine herbal extracts. However, this process could degrade their natural color and flavor as well as nutritional and antioxidant qualities, thus pressurization, a non-thermal technique, is favored to conserve the sensory attributes and nutritional values of the products (Apichartsrangkoon *et al*., 2009, 2012, 2013; Chaikham and Apichartsrangkoon, 2012a, b; Barba *et al*., 2013). Keenan *et al.* (2012) stated that thermal processing of smoothies apparently reduced total anthocyanins, total phenols, ascorbic acid and L attribute (lightness) compared to untreated and pressurized (450 MPa/20°C/5 min) samples. In addition, Chaikham and Apichartsrangkoon (2012a) found that pressurized longan juices at 300 and 500 MPa for 20 min were brighter and more transparent than pasteurized juice (90°C/2 min). They also elucidated that the amounts of ascorbic acid and total phenols as well as antioxidant capacity (DPPH assay) in pressurized samples were significantly higher than those in pasteurized batch. Similar observations were reported with strawberry and pomegranate juice (Cao *et al*., 2012; Varela-Santos *et al*., 2012).

The aim of this present work was to evaluate the alteration of color parameters and bioactive components including ascorbic acid, γ-oryzanol, asiatic acid, gallic acid ellagic acid and total phenols as well as antioxidant capacity of pressurized (400 MPa/25°C/30 min) and pasteurized (90°C/2 min)

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herbal-plant beverages on storage at 4°C for 4 weeks.

Materials and Methods

Herbal-plant beverage preparation

Dried longan, black glutinous aromatic rice and pennywort leaves were purchased from a local market in Chiang Mai, Thailand. Dried longan and blended black rice were boiled in water at a ratio of 1:4 (w/w) for 3 min and at a ratio of 1:15 (w/w) for 15 min, respectively; and then separately filtered by a sterile cotton cloth. Fresh pennywort leaves were extracted with drinking water at a ratio of 1:1 (w/w) then filtered by a cotton cloth. The formula of herbal-plant beverage was a mixture of dried longan, black rice and pennywort extracts at a ratio of 1:1:1 by volume with total soluble solids of 10°Brix (Chaikham *et al*., 2014).

A 150-ml of the mixture was then packed in a laminated bag (nylon plus polyethylene; Siampack, Thailand) and subjected to pressure 400 MPa at 25°C for 30 min. The high pressure vessel was a 'Food Lab' model 900 high pressure rig (Stansted Fluid Power; Essex, UK). The rate of pressure increase was about 330 MPa/min. During this high pressure treatment an adiabatic increase in temperature occurs. At ambient temperature (25°C), the monitored cell temperature increased by about 5°C to 400 MPa but decreased to the set equilibrium value in less than 1.5 min. The pressure transmitting medium used in this experiment was a mixture of castor oil (Chemical & Lab Supplies, Thailand) and 98% ethanol (Chemical & Lab Supplies) at a ratio of 20:80 (v/v) (Chaikham and Apichartsrangkoon, 2012a, b). For pasteurization, 250 ml of the mixture were packed in a retort pouch (Siampack, Tthailand), and then heated in boiling water until the inside core of the package reached 90±5°C for 2 min. Subsequently, both processed beverages were stored at 4°C and analyzed continuously once a week for 4 weeks.

Instrument color parameter measurements

A colorimeter, model Color Quest XE (HunterLab, Reston, VA) was used to measure the color of untreated and processed herbal-plant beverages. Color parameters, L, a^* and b^* were used to calculate total different colors [$\Delta E = ((\Delta L)^2 + (\Delta a^*)^2 + (\Delta b^*)^2)^{1/2}$] (Barba et al., 2013) and Browning Index [$BI = (100(x - 0.31))/0.172$, where $x = (a^* + 1.75L)/(5.645L + a^* - 3.012b^*)$] (Ferrari *et al*., 2010).

Determination of ascorbic acid

Ascorbic acid content was determined following the method described by Chaikham and Apichartsrangkoon (2012a, b) with some modifications. One gram of freeze dried sample was mixed with 9 ml diluted sulfuric acid (pH 2.2) (Merck, Germany), stirred for 30 min and then centrifuged at 2,500 rpm for 10 min. The supernatant was filtered through a 0.20-µm nylon membrane (Vertical, Thailand) and the filtrate used for HPLC analysis. The HPLC system (Shimadzu LC-10AD; Shimadzu, Japan) consisted of a low-pressure pump and a photodiode array detector (SPD-M20A; Shimadzu) adjusted to a lmax 250 nm. Chromatographic separation was performed with a C18 column (YMC-Pack ODS-AM, 5 µm, 4.6 mm ID x 250 mm; YMC, Japan). The isocratic eluent was 0.1 M acetic acid (Merck) in deionized water (RCI Lab-Scan, Thailand) with a flow rate of 1.5 ml/min. The temperature of the column was adjusted to 30°C and the injection volume was a 20-µl filtrate. The peak area was determined and converted to concentration of ascorbic acid.

Determination of gamma-oryzanol

γ-Oryzanol content was determined following the modified procedure as described by Iqbal *et al*. (2005). Two grams of freeze dried sample were extracted by dichloromethane (RCI Lab-Scan) in Soxhlet apparatus for 6 h. The crude oil was diluted with dichloromethane and filtered through a 0.20-µm nylon membrane. The filtrate was used for HPLC analysis. The isocratic eluent was a mixture of methanol (RCI Lab-Scan), acetonitrile (RCI Lab-Scan), dichloromethane and acetic acid at a ratio of 50:44:3:3 (v/v) with a flow rate of 1.4 ml/min. The UV detection was at a lmax 330 nm with an injection volume of 20 µl filtrate. The peak area of each component was determined and converted to concentration.

Determination of asiatic acid

The analysis of asiatic acid was carried out using a modified method as described by Inamdar *et al.* (1996). One gram of freeze dried sample was mixed with 9 ml methanol, stirred for 2 h and then centrifuged at 2,500 rpm at 25°C for 10 min. The supernatant was filtered through a 0.20-µm nylon membrane and the filtrate was used for HPLC analysis. The mobile phase was a mixture of acetonitrile (solvent A) and deionized water (solvent B) with a flow rate of 1.4 ml/min. The gradient system of the mobile phase commenced from 0 min (20% A/80% B) to 30 min (55% A/45% B), 35 min (55% A/45% B) and 45 min (80% A/20% B). A 20-µl filtrate was injected into the column. The peak area of asiatic acid was determined

and converted to concentration.

Determination of gallic and ellagic acids

Gallic and ellagic acids were determined using a modified HPLC method described by Rangkadilok *et al.* (2005). One gram of freeze dried sample was mixed with 9 ml of 100% methanol and stirred for 30 min, then centrifuged at 2,500 rpm for 15 min. The supernatant was filtered through a 0.20-µm nylon filter and the filtrate was used for HPLC assay. The mobile phase was a mixture of 0.4% formic acid (solvent A) (Merck) and 100% methanol (solvent B) with a flow rate of 1.0 ml/min. The gradient system of the mobile phase commenced from 0 min (100% A) to 4 min (95% A/5% B), 10 min (70% A/30% B), 16 min (66% A/34% B), 22 min (45% A/55% B), 28 min (55% A/45% B) and 34 min (100% A), and maintained at this state to 40 min. The temperature of the column was adjusted to 25°C and UV detection was performed at a lmax 270 nm with an injection volume of 20 µl. Peak areas were determined and converted to the content of each component.

Determination of total polyphenols

Total phenols were determined using the Folin-Ciocalteu reagent (Chaikham and Apichartsrangkoon, 2012a,b). Two milliliters of sample were stirred with 8 ml of 100% cooled ethanol (Chemical & Lab Supplies) for 15 min and centrifuged at 2,500 rpm for 15 min. A 0.5 ml of supernatant was mixed with 2.5 ml of 10% Folin-Ciocalteu reagent (Sigma, Germany) and allowed to react for 5 min. Subsequently, 2 ml of saturated sodium carbonate solution (Ajax, Australia) was added to the mixture and held for 2 h at room temperature. The apparent blue complex was determined at a lmax 765 nm (Spectophotometer; Perkin Elmer UV WINLAB, USA). Total phenols were expressed as mg gallic acid equivalent per 100 ml sample (mg GAE/100 ml).

Determination of antioxidant capacity

2,2-Diphenyl-1-picrylhydrazyl (DPPH) radical-scavenging activity was determined following the methods as described by Chaikham and Apichartsrangkoon (2012a, b). A 2-ml of herbal-plant beverage was mixed with 100% methanol for 10 min and centrifuged at 2,500 rpm for 10 min. Consequently, 1.6 ml of supernatant was mixed with 0.4 ml of 1.5 µM DPPH radical (Fluka, Switzerland) methanol solution. The mixture was shaken and allowed to stand for 30 min at room temperature. Absorbance of the solution was measured at a lmax 517 nm. A control was prepared using 1.6 ml methanol. Percentage inhibition of DPPH radicals was calculated:

$$\%\text{DPPH radical scavenging activity} = [1 - (\text{Abs}_{samples}/\text{Abs}_{control})] \times 100$$

Microbiological assessments

The assessments of standard plate counts, yeasts, moulds and fecal coliforms in fresh and processed beverages were followed the BAM method (U.S. Food and Drug Administration2, 001).

Data analysis

All data were the means of triplicate determinations with individual duplication (n=6). Analysis of variance (ANOVA) was carried out using SPSS Version 15.0 (SPSS Inc., USA), and determination of significant differences among treatment means was done by Duncan's multiple range tests (P < 0.05).

Results and Discussion

Changes of instrument color parameters during storage

Changing of color parameters of pressurized and pasteurized herbal-plant beverages during storage at 4°C are shown in Table 1. The lightness (L parameter) of pasteurized beverage (P < 0.05) exhibited significantly lower (P < 0.05) than that of untreated and pressurized samples in order. During storage, the L parameters of both processed beverages apparently decreased (P < 0.05) in accordance with the increase of storage time. Similarly, Barba *et al.* (2012) found that L parameters of pressurized blueberry juice at 600 MPa and 42°C for 5 min stored at 4°C for 56 days significantly decreased with the increase of storage time. Identical finding was reported by Keenan *et al.* (2010) with pressurized fruit smoothies at 450 MPa and ~ 20°C for 1-3 min stored at 2-4°C for 30 days. Besides pressurized beverages, Rivas *et al.* (2006) also described a decrease of the L parameter of pasteurized orange-carrot juice during refrigerated storage.

The a^* parameter (redness) of pressurized beverage was significant lower (P < 0.05) than that of untreated sample, which was shifted to the green zone, while the pasteurized sample showed the highest redness. During chilled storage, a_* parameters of both processed samples significantly increased (P < 0.05) according to the increasing storage time. The a^* parameter of pressurized sample was shifted from green zone ($-a^*$) to red zone ($+a^*$) after storage for 3 weeks. Close with the a^* parameter, b^* parameters (yellowness) of untreated and pressurized herbal-plant beverages were significantly lower (P < 0.05) than that of pasteurized sample. Upon storage, the b^* parameters of both processed samples were significantly rose (P < 0.05). The reduction of L

Table 1. Alteration of color parameters of untreated and processed herbal-plant beverages during storage at 4ºC for 4 weeks

Parameters	Storage time (weeks)	Untreated beverage	Pressurized beverage	Pasteurized beverage
L	Initial state	32.16 ± 0.05^{A}	31.98 ± 0.03^{Ba}	26.53 ± 0.04^{Ca}
	1		31.04 ± 0.04^{Ab}	26.42 ± 0.05^{Bb}
	2		30.11 ± 0.01^{Ac}	26.09 ± 0.04^{Bc}
	3^{ns}		26.05 ± 0.06^{d}	26.02 ± 0.03^{c}
	4		25.26 ± 0.02^{Be}	25.60 ± 0.02^{Ad}
*a**	Initial state	-0.06 ± 0.02^{B}	-0.12 ± 0.01^{Cd}	2.13 ± 0.04^{Ad}
	1		-0.09 ± 0.03^{Bd}	2.05 ± 0.02^{Ac}
	2		-0.03 ± 0.01^{Bc}	2.14 ± 0.02^{Ab}
	3		0.06 ± 0.01^{Bb}	2.29 ± 0.01^{Aa}
	4		0.13 ± 0.02^{Ba}	2.30 ± 0.01^{Aa}
*b**	Initial state	3.40 ± 0.02^{B}	3.43 ± 0.02^{Be}	4.16 ± 0.03^{Ac}
	1		3.58 ± 0.03^{Bd}	4.18 ± 0.05^{Ac}
	2		3.64 ± 0.01^{Bc}	4.19 ± 0.04^{Ac}
	3		3.70 ± 0.01^{Bb}	4.24 ± 0.01^{Ab}
	4		3.78 ± 0.03^{Ba}	4.30 ± 0.02^{Aa}

Means in the same column or row followed by the same lowercase or capital letters respectively are not significantly different (P > 0.05). Data are expressed as means ± standard deviation (n=6).

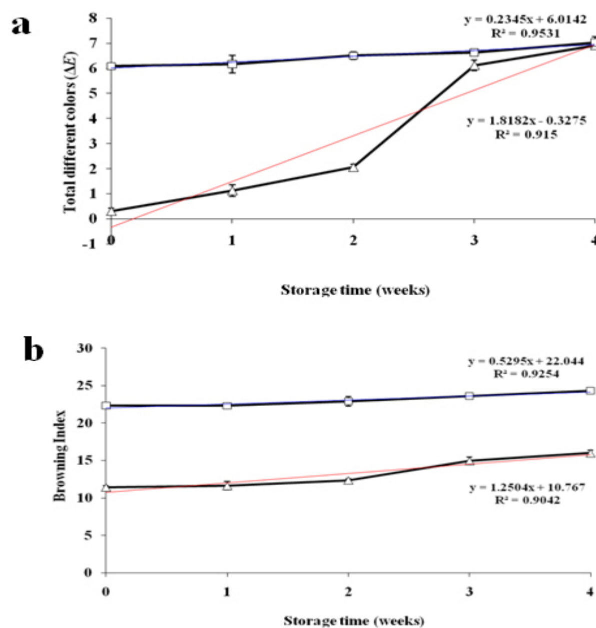

Figure 1. The intensities of (a) total different colors and (b) Browning Index of (Δ) pressurized and (□) pasteurized herbal-plant beverages during storage at 4ºC for 4 weeks. Data are expressed as means ± standard deviation (n=6).

parameter or the increase of a^* and b^* parameters in pressurized sample during processing and storage could be due to the effect of enzymatic browning reaction by residual oxidase enzymes including polyphenol-oxidase and peroxidase, while that in thermally treated sample might be due to the Maillard condensation (Keenan *et al*., 2010; Chaikham and Apichartsrangkoon, 2012a). Keenan *et al.* (2011a) illustrated that pressurized apple puree containing 8% (w/w) inulin at 500 MPa and 20ºC for 1.5 min had a^* and b^* parameters increasing on storage at 4°C for 30 days.

Total different colors (ΔE), which indicate the magnitude of the color difference between untreated and processed herbal-plant beverages during the storage period, are shown in Figure 1(a). The results showed that ΔE parameter of both processed herbal-plant beverages significantly increased (P < 0.05)

Table 2. Alteration of bioactive components and antioxidant capacity of untreated and processed herbal-plant beverages during storage at 4ºC for 4 weeks

Bioactive compounds	Storage time (weeks)	Untreated beverage	Pressurized beverage	Pasteurized beverage
Ascorbic acid (mg/100 ml)	Initial state	1.92 ± 0.04^{A}	1.64 ± 0.05^{Ba}	1.03 ± 0.04^{Ca}
	1		1.60 ± 0.03^{Aa}	0.95 ± 0.05^{Bab}
	2		1.48 ± 0.08^{Ab}	0.88 ± 0.03^{Bbc}
	3		1.32 ± 0.05^{Ac}	0.82 ± 0.05^{Bc}
	4		1.11 ± 0.10^{Ad}	0.73 ± 0.02^{Bd}
γ-oryzanol (mg/100 ml)	Initial state	1.25 ± 0.03^{A}	1.28 ± 0.06^{Aa}	0.96 ± 0.04^{Ba}
	1		1.22 ± 0.04^{Aa}	0.90 ± 0.05^{Ba}
	2		1.15 ± 0.02^{Ab}	0.73 ± 0.05^{Bb}
	3		1.11 ± 0.03^{Ab}	0.70 ± 0.07^{Bb}
	4		1.01 ± 0.08^{Ac}	0.54 ± 0.03^{Bc}
Asiatic acid (mg/100 ml)	Initial state	2.02 ± 0.04^{A}	1.85 ± 0.05^{Bab}	1.48 ± 0.04^{Ca}
	1		1.86 ± 0.02^{Aa}	1.40 ± 0.04^{Bab}
	2		1.80 ± 0.02^{Ab}	1.42 ± 0.02^{Bb}
	3		1.72 ± 0.03^{Ac}	1.40 ± 0.03^{Bb}
	4		1.64 ± 0.04^{Ad}	1.30 ± 0.02^{Bc}
Gallic acid (mg/100 ml)	Initial state	7.44 ± 0.05^{A}	7.10 ± 0.08^{Ba}	5.49 ± 0.06^{Ca}
	1		6.94 ± 0.04^{Ab}	5.40 ± 0.05^{Ba}
	2		6.52 ± 0.05^{Ac}	5.22 ± 0.07^{Bb}
	3		6.50 ± 0.06^{Ac}	5.01 ± 0.12^{Bc}
	4		6.08 ± 0.11^{Ad}	4.64 ± 0.09^{Bd}
Ellagic acid (mg/100 ml)	Initial state	22.85 ± 0.21^{A}	18.99 ± 0.20^{Ba}	15.26 ± 0.10^{Ca}
	1		18.14 ± 0.13^{Ab}	15.18 ± 0.05^{Ba}
	2		17.03 ± 0.09^{Ac}	14.03 ± 0.08^{Bb}
	3		15.46 ± 0.12^{Ad}	11.82 ± 0.10^{Bc}
	4		15.11 ± 0.15^{Ae}	11.53 ± 0.11^{Bd}
Total phenols (mg GAE/100 ml)	Initial state	98.70 ± 1.40^{A}	84.16 ± 1.20^{Ba}	70.84 ± 0.90^{Ca}
	1		85.02 ± 0.83^{Aa}	69.14 ± 0.25^{Bb}
	2		80.64 ± 0.52^{Ab}	63.55 ± 0.74^{Bc}
	3		78.40 ± 1.04^{Ac}	60.81 ± 0.66^{Bd}
	4		70.66 ± 0.87^{Ad}	54.10 ± 1.05^{Be}

Means in the same column or row followed by the same lowercase or capital letters respectively are not significantly different (P > 0.05). Data are expressed as means ± standard deviation (n=6).

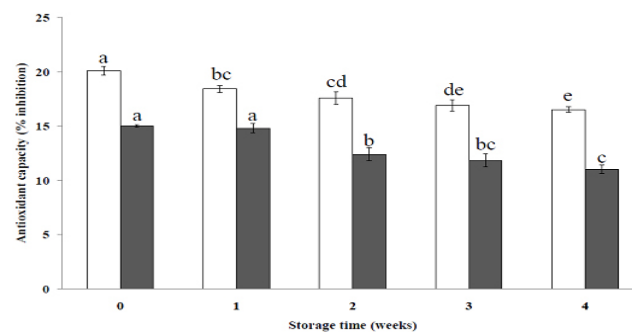

Figure 2. Alteration of antioxidant capacity (DPPH assay) of pressurized (non-filled bars) and pasteurized (solid bars) herbal-plant beverages during storage at 4ºC for 4 weeks, means followed by the same letters indicate non-significant difference (P > 0.05). Data are expressed as means±standard deviation (n=6).

with the increasing storage time. When considering the value of ΔE, the color difference between the stored and untreated samples can be estimated such as not noticeable (ΔE 0–0.5), slightly noticeable (ΔE 0.5–1.5), noticeable (ΔE 1.5–3.0), well visible (ΔE 3.0–6.0) and great visible (ΔE 6.0–12.0) (Cserhalmi et al., 2006; Barba et al., 2013). In this study great visible changes of brown color were observed (ΔE > 6.0) in pressurized and pasteurized samples at week 3 and at the initial state of storage, respectively. Overall the increasing rate of ΔE values in pressurized sample during storage (slope ~ 1.8) showed higher than that of the pasteurized sample (slope ~ 0.2). Similar results were observed by Keenan *et al.* (2011b) with high pressure (450 MPa/20°C/1, 3 or 5 min) and thermally (P_{70} > 10 min) treated fruit smoothies kept at 4°C for 30 days.

Browning reaction occurs in many fruit and vegetable products during processing and storage. The intensity of the total color change could also

Table 3. General microbiological qualities of untreated and processed herbal-plant beverages during storage at 4ºC for 4 weeks

Assessments	Storage time (weeks)	Untreated beverage	Pressurized beverage	Pasteurized beverage
Standard plate count (CFU/ml)	Initial state	$5.63 \pm 1.53^{A} \times 10^4$	nd^{Bb}	nd^{Bb}
	1^{ns}		nd^{b}	nd^{b}
	2^{ns}		nd^{b}	nd^{b}
	3^{ns}		nd^{b}	nd^{b}
	4		$<10^{Ba}$	$1.85 \pm 0.59^{Aa} \times 10^2$
Yeasts and moulds (CFU/ml)	Initial state	$8.20 \pm 2.40^{A} \times 10^2$	nd^{Ba}	nd^{Bb}
	1^{ns}		nd^{a}	nd^{b}
	2^{ns}		nd^{a}	nd^{b}
	3^{ns}		nd^{a}	nd^{b}
	4		nd^{Ba}	$<10^{Aa}$

Means in the same column or row followed by the same lowercase or capital letters respectively are not significantly different ($P > 0.05$). nd is not detected and ns is non significantly different.

be indicated by Browning Index (BI). BI values of untreated and processed herbal-plant beverages were determined as presented in Figure 1(b). The results showed BI value of the pressurized sample was significant lower ($P < 0.05$) than that of the pasteurized sample due to less Maillard condensation effect. The higher in BI of the pasteurized sample might be a reflection of high a^* and low L parameters. The BI values of both processed beverages significantly increased ($P < 0.05$) with the increase of storage time. The changing rate of BI of the pressurized sample (slope ~ 1.2) was higher than that of the pressurized batch (slope ~ 0.5), which could be due to the enzymatic degradation by residual oxidative enzymes. According to Polydera *et al.* (2005), who pasteurized (80°C/1 min) and pressurized (600 MPa/40°C/4 min) orange juices stored at 0-30°C for 60 days, they found that the rate of browning in both processed juices markedly rose when storage time increased.

Stability of bioactive components and antioxidant capacity during storage

As considering the storage period, ascorbic acid, γ-oryzanol, asiatic acid, gallic acid, ellagic acid ant total phenols in both processed beverages significantly diminished ($P < 0.05$) with an increase in storage time (Table 2). The antioxidant capacity (DPPH radical-scavenging activity) also apparently dropped ($P < 0.05$) with increasing storage time (Figure 2). However, most of these bioactive components in the high pressure treated beverage still remained in high amounts than those in the thermally treated beverage throughout the chilled storage period. Apichartsrangkoon *et al.* (2012) found that ascorbic acid in pressurized (400 MPa/30°C/20 min) and pasteurized (90°C/3 min) pennywort juices significantly decreased after storage for 4 months at 4°C. Similar result was discovered with pressurized (400 and 600 MPa/20°C/5 min) and pasteurized (75°C/5 min) acidified apple purées kept at 5°C for 3 weeks (Landl *et al.*, 2010).

A significantly decrease of γ-oryzanol content was observed in herbal-plant beverage after thermal processing compared to the raw sample. Similarly, Finocchiaro *et al.* (2007) found that γ-oryzanol in cooked brown rice was lost more than 8% when compared with raw brown rice. The diminution of this compound in cooked brown rice during storage closed to the work by de Simone Carlos Iglesias Pascual *et al.* (2013) with in an average γ-oryzanol loss of 20%. Besides ascorbic acid and γ-oryzanol, Komin (2005) suggested that thermal processing resulted in a several fold decreased of triterpene glycosides i.e. madecassoside, madecassic acid, asiaticoside and asiatic acid in Malaysian pennywort juice. While Apichartsrangkoon *et al.* (2012) illustrated that asiaticoside and madecassoside in processed pennywort juices were relatively stable during storage without the significant changes for a period of 4 months at 4°C. The health-promoting effects of triterpene glycosides found in pennywort extract have been reported in wound healing, UV-induced photoaging, glutamate- or β-amyloid-induced neurotoxicity and hepatofibrosis, strong neuroprotective and anti-cancer (Krishnamurthy *et al.*, 2009; Tang *et al.*, 2009).

The alteration of gallic acid, ellagic acid, total phenols and antioxidant capacity on storage were also discovered by several researchers. For instance, Chaikham and Apichartsrangkoon (2012b) found that gallic acid, ellagic acid, total polyphenols and antioxidant capacity (DPPH assay) in longan juice processed by high pressure (500 MPa/25°C/30 min) and thermal (90°C/2 min) processing significantly decreased when the storage time was increased. Additionally, Vega-Galvez *et al.* (2011, 2012) pressurized Aloe vera gel at 300-500 MPa and 20°C for 3 min and kept at 4°C for 35 days. They revealed that total phenols and antioxidant capacity (DPPH assay) of all both processed samples significantly declined with an increasing storage time. Zerdin *et al.* (2003) stated that oxygen can diffuse into the matrix of the products from the headspace present in the packaging material during storage. The formation of these free radical species will oxidize the antioxidant compounds, degrading various bioactive compounds and antioxidant activity over time.

Changes of standard microbiological qualities during storage

Standard microbiological assessments displayed that, at week 4, the amounts of total plate counts and yeasts-moulds in pressurized sample were less than 1 log CFU/ml and not detected respectively, while those in pasteurized sample were detected ~ 2 and

< 1 log CFU/ml, respectively (Table 3). In addition, fecal coliforms, *Escherichia coli* and *Staphylococus aureus* in both processed infusions were satisfactorily eliminated (data not shown). Similar findings with our study were presented by Varela-Santos et al. (2012) who pressurized pomegranate juice at 450-550 MPa for 30-150 s, and stored at 4°C for 35 days.

Conclusion

The results of this study showed that the brightness (L) of both processed beverages apparently decreased in accord with the increase of the storage time, while other parameters such as a^*, b^*, ΔE and BI trended to increase upon the storage. Additionally, all bioactive compounds and antioxidant capacity in both the pressurized and pasteurized beverages significantly declined throughout the storage period. To sum up the results displayed that pressurization could maintain the natural color and predominant bioactive compounds of this beverage better than pasteurization.

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