### **Appendix** A

### Pictures of lime juice



Figure 1a Fresh lime juice and after HPP- treated lime juices at 400, 500 and 600 MPa







Figure 5a HPP-treated lime juices at 400 (A), 500 (B) and 600 MPa (C) for 15

minutes after 4 weeks storage at ambient temperature

# **Appendix B**

# Reagents and Instruments

Table 1b The instruments used in this study

	Brand
1. High Pressure equipment	Stansted, England
2. Colorimeter	Minolta, Japan
3. Spectrophotometer	Thermo-Spectronic, England
4. Centrifuge	Hettich-Zentrifugen, German
5. Kjeldahl, Protein Distillation unit	Foss, Germany
6. Digestion unit for Kjeldahl distillation	Foss, Germany
7. pH meter	Mettler-Toledo, Switzerland
8. Hand Refractometer	Belling Ham and Stanley, UK
9. Juice Stainless Extractor	Crocodile, Thailand
10. Incubator 35±2°C	Memmert, Germany
11. Incubator 25±2°C	Sanyo, Japan
12. Mini Freezer -80±2°C	Heto-Holten, Denmark
13. Water bath	Memmert, Germany
14. Furnaces	Eurotherm, Germany
15. Hot air oven	Sanyo, Japan
16. Refrigerator	Sanyo, Japan
	Sanyo, Japan



Figure 1B The picture of High Pressure Processing

prototype of Stansted "Food Lab" model 900

Dehydrated Culture media	Brand
1. Orange Serum Agar	Merck, Germany
2. Potato Dextrose agar (PDA)	Merck, Germany
3. Lauryl tryptose broth	Merck, Germany
4. Brilliant Green Lactose Bile broth (BGLB)	Merck, Germany
5. Peptone	Merck, Germany
6. <i>Escherichia coli</i> broth (EC broth)	Merck, Germany
7. Phosphate buffer	Merck, Germany

Table 2b The dehydrated culture media used for the experimental study

Reagents	Brand	Grade
1. Chloroform	BDH, England	AR
2. Sulfuric acid	Merck, Germany	AR
3. Hydrochloric acid	Merck, Germany	AR
4. Petroleum ether (b.p. 60-80°C)	BDH, England	AR
5. 4-dimethyl amino benzaldehyde	Fisher, England	AR
6. 70% Perchloric acid	BDH, England	AR
7. Glacial acetic acid	BDH, England	AR
8. Sodium hydroxide	BDH, England	AR
10. d-limonin Standard	Sigma, Germany	HPLC- grade
11. Phenolphthalein	Fisher, England	AR
12. Oxalic acid	Merck, Germany	AR
13. Boric acid	BDH, England	AR
14. Methylene blue	Fisher, England	AR
15. Ammonia	BDH, England	AR
16. Ethyl alcohol	BDH, England	AR
17. Diethyl ether	BDH, England	AR
18. Indophenol	BDH, England	AR
19. Gum acasia	OV, Thailand	Food grade
20. Carboxymethylcellulose (CMC)	OV, Thailand	Food grade
21. Pectin	OV, Thailand	Food grade
22. κ-carrageenan	OV, Thailand	Food grade
23. Pectin	OV, Thailand	Food grade

Table 3b The reagents used for the experimental study

# Appendix C

# Estimation of d-limonin

#### 1. Method of estimation of limonin

The d-limonin was estimated from the chloroform extract of sample by colorimetric method (Vaks and Lifshitz, 1981). Five ml of centrifuged juice, made to 25 ml with distilled water was extracted with petroleum ether (b.p. 60-80°C) in a separatory funnel (250 ml) to extract the coloring matter. The petroleum ether extract was discarded and aqueous solution was extracted with chloroform (3x25 ml). The chloroform extract was washed with distilled water (4x50 ml). The volume was made to 50 ml with chloroform. A known quantity of this solution was used for determination of d-limonin by developing color with Burham's reagent as mentioned below for preparation of standard solution of limonin.

**Burham's reagent** : 0.1 g of 4-dimethyl amino benzaldehyde

2.4 ml of 70 percent perchloric acid

3 ml of glacial acetic acid

0.1 g of 4-dimethyl amino benzaldehyde dissolved in 3 ml of glacial acetic acid and mixed with 2.4 ml of 70% perchloric acid, AR followed by stirring vigorously on an electric stirrer. The Burham's solution was made fresh prior in every estimations.



Figure 1c Estimation of d-limonin by a colorimetric method

(Vaks and Lifshitz, 1981)

#### 2. Preparation of d-limonin standard

Standard solution of d-limonin was prepared by dissolving 1.0 mg of dlimonin (Sigma, Germany) in chloroform and volume was made to 100 ml.

Different volumes i.e. 1, 2, 3, 4 and 5 ml of chloroform solution containing limonin concentrations 10, 20, 30, 40 and 50 micrograms respectively were taken in separate test tubes along with blank.

The tubes containing chloroform with different concentrations of standard were heated in a water bath for 10 minutes and cooled. To the residue of each test tube, added 3 ml of after 30 minutes, the intensity of red color so developed was measured with Spectrophotometer at 503 nm. The standard curve was plotted between different concentrations of d-limonin and the corresponding optical densities (abs). Limonin content was calculated from the standard curve (Fig.1b)



# Appendix D

# Physical, chemical, microbiological and

# nutritional analyses

#### 1. Physical method

#### 1.1 Color analysis

The L\*, a\* and b\* values were measured with a colorimeter (Colorimeter, Minolta CR-300, Japan). Samples were prepared by pouring about 30 ml of lime juice into a white plastic cup.



Figure D1 A graphic of the Lab color model

Source : <u>http://www.colorspan.com/support/tutorials/cmpl/lab.asp</u>

Source . In	tp://www.colorspan.coll/support/tatorials/empi/tao.asp
L*-value	= A lightness factor with a value range of 0-100%
a*-value	$\stackrel{\bigcirc}{=}$ Describes how red/green a color is, with a value range from
	(-) 60 to (+) 60, + values are more red; - values are more green)
b*- value	= Describes how yellow/blue a color is, a value range from (-)
	60 to (+) 60; + values are more yellow; - values are more blue)

#### 1.2 Yield analysis

The yield of lime juice was measured based on a formula :

yield = weight of lime juice x 100 weight of lime fruit

#### 2. Chemical method

#### 2.1 Total titratable acidity analysis (AOAC, 2000)

Pipette 10 ml lime juice sample into a flask and dilute the sample with distilled water. Then pipette 10 ml diluted solution sample into another flask. Drop phenolphthalein 2-3 drops and titrate the sample with 0.1 M NaOH until the samples reach the end point (sample solution became pink that was persisted for 30 s.)

% Citric acid = ml of 0.1 M NAOH x 0.1 x 0.007 x 100Vol. of sample

#### 2.1 Moisture and Total solid analysis (AOAC, 2000)

Heated an empty moisture dish in a hot air oven about 20-30 min. Cool in a desicator and weigh the dish. Into the cooled and weighed dish (provided with cover), previously heated to  $100 \pm 30^{\circ}$ C, accurately weigh 2 g of lime juice sample. Uncover the dish and dry the dish with its cover and contents for 3 h in an oven provided with opening for ventilation and maintained at  $100 \pm 3^{\circ}$ C. Cover the dish while it is still in the oven, transfer to a desicator, and weigh the dish soon after it reached a room temperature. Dry the sample again for several times until the sample has a constant weight.

% total solid = 
$$100 - \%$$
 moisture content

% moisture content = Loss in the sample weight during drying x 100

Initial weight of the sample

#### 2.2 Sugar analysis (AOAC, 2000)

#### 2.2.1 Reducing sugar

Transfer 15 ml of lime juice sample into a 100 volumetric flask and adjust to 100 ml with deionized water. Add 5 ml of Carrez I and II solutions. Shake and adjust to 200 ml with distilled water. Put aside the mixed solution for precipitation about 20 min and filter the solution with a Whatman filter paper no.4. Pour the solution filtrate into a 50 ml burette and pipette and pipette 5 ml of Fehling solution no. 1 and 2 into a flask.

Heat the Fehling solution on hot plate and add one drop of methylene blue indicator. Titrate the Fehling solution with the filtrate solution in the burette until the Fehling solution has a color of orange-red.

#### 2.2.2 Inversion sugar

Pipette 50 ml of filtrate solution from the previous determination into a volumetric flask. Add 10 ml of 6.34 N hydrochloric acid. Place the volumetric flask in a water bath at  $70^{\circ}$ C for 10 min and cool immediately. Make the mixture solution neutral with 5 N NaOH and adjust to 100 ml with water. Do titration following the procedure of reducing sugar.

% sucrose (S) = % different between  $D_1$  and  $D_2 \ge 0.95$ 

% total sugar =  $D_1 + S$ 

When  $D_1 = \%$  Reducing sugar

 $D_2 = \%$  Inversion sugar

#### 2.3 Protein analysis (AOAC, 2000)

Place sample (15 ml) in a digestion flask. Add 8 g catalyst mixture and 20 ml  $H_2SO_4$ . Place the flask in an inclined position in a digestion machine and heat the machine gently until frothing ceases. Continue boil briskly until the solution clears (~2 h).

Cool, add distilled water to dilute the mixture solution and pour into a distilling flask. Add 400 ml H<sub>2</sub>O (ammonia-free water) and a few Zn granules to prevent bumping. Immediately immerse a condenser tip into a receiver that contains 50 ml of 2% boric acid solution in a 500 ml flask and 5-7 drops indicator. Add 75 ml of 50% sodium hydroxide using a funnel into the distilling equipment0 Rotate the distilling flask to mix the contents thoroughly; then heat until all NH<sub>3</sub> has been distilled ( $\geq$ 150 ml distillate). Remove the receiver, wash the tip of the condenser and titrate excess standard acid in distillate with 0.05 M H<sub>2</sub>SO<sub>4</sub>. Do blank determination to correct any nitrogen content in reagents.

$$% N = (V_a - V_b) \times N. H_2 SO_4 \times 1.4007$$

When  $V_a = ml$  of standard acid for sample titration  $V_b = ml$  of standard acid for blank titration  $H_2SO_4 = normality$  acid VS = vol. of sample (g)

% Protein = % N x factor (factor value = 6.25)

#### 2.4 Fat analysis (AOAC, 2000)

Pippetted sample (20 ml) and place into a separated funnel. Add 10 ml water and shake. Add 1.25 ml ammonia solution, 10 ml ethyl alcohol and 25 ml diethyl ether, close with a stopper and shake vigorously for 1 min. Careful to release the pressure of the funnel. Add 25 ml petroleum ether, close the stopper and shake vigorously for 1 min. Careful to release the pressure. Let stand until an upper liquid is practically clear (~30 min). Pour the upper clear solution into a previously weighed beaker. Take the beaker to stand in a hood until diethyl ether and petroleum are evaporated and place the beaker in a hot air oven (T =  $102 \pm 2^{\circ}$ C) for 2 h. Cool in a desicator and weigh the sample.

% Fat content =  $(W_2 - W_3) \times 100$ 

 $V_1 =$ Vol. of sample

 $W_2$  = Weight of beaker and fat

 $W_3$  = Weight of beaker

#### 2.5 Ash analysis (AOAC, 2000)

Weigh 3-5 g sample into an ashing dish that has been heated, cooled in a desicator, and weighed soon after reaching room temperature. Before ashing the sample, heat the sample on a bunchen lamp until no more black smoke appeared. Then ash the sample in a muffle furnace at 550<sup>o</sup>C until light gray ash results or until it reaches a constant weight. Cool in a desicator and weight soon after reaching room temperature.

% Ash = Weight of ash x 100 Weight of sample

#### 2.6 Fiber analysis (AOAC, 2000)

Weigh 5 g sample into a 500 ml beaker. Transfer 1.25 M sulfuric acid (200 ml) into the beaker. Boil the sample solution on a hot plate for 30 min. Filter the sample solution using a Whatman paper no. 4 until it dries by applying a vacuum pump and wash the residue with boiling water until the sample does not have acid (do a test using a litmus paper). Place 200 ml of 1.25% NaOH into a beaker and boil the beaker on a hot plate. Wash the residue on the filter paper with distilled water. Boil the sample again on the hot plate for 30 min. Filter the sample using a Whatman paper no. 4 and wash the residue with boiling water. Transfer the filter paper with the sample residue into a crucible and dry at  $102 \pm 2^{\circ}$ C for 3 h. Cool in a desicator and weigh. Then ash the residue for 2 h at  $550 \pm 10^{\circ}$ C, cool in the desicator, and weigh.

% Crude fiber = 
$$(W_4 - W_3 + W_2) + (W_5 - W_3) (100 - \%H_2O - \%fat)$$

 $W_1$ 

 $W_1$  = Weight of sample

 $W_2$  = Weight of filter paper

 $W_3 =$  Weight of crucible

 $W_4$  = Weight of crucible + filter paper + sample after drying

- $W_5 =$  Weight of crucible + ash
- %H<sub>2</sub>O = Moisture content of sample

%fat = Fat of sample

#### 2.7 Carbohydrate content (AOAC, 2000)

Carbohydrate content was determined by measuring the difference of the original sample minus the moisture, protein, crude fat and mineral contents calculated at the same moisture level.

#### 2.9 pH value

The pH value of lime juices were measured with a pH meter. Samples were prepared by pouring about 40 ml of lime juice into a beaker. Before this analysts the pH meter was calibrated by pH buffer at pH 4.0 and 7.0.

#### 3 Microbiological analysis

#### 3.1 Total Plate Counts (FDA, 2001)

The number of total microorganisms were enumerated using a pour plate method on Orange Serum Agar. Incubation was performed at  $35\pm2^{\circ}$ C for 48 h.

#### 3.2 Total yeast and mounds (FDA, 2001)

were enumerated on Potato Dextrose Agar acidified to pH 3.5 with 10% tartaric acid by a pour plating technique. The incubation for total yeast and mold counts was done at  $22^{\circ}$ C for 5 days.

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#### 4 Nutritional analysis

#### 4.1 Vitamin C content (AOAC, 2000)

Pipette 50 ml sample into 100 ml volumetric flask. Add 25 ml of 0.4% oxalic acid and dilute with distilled water. Then pipette 10 ml of the diluted sample into a 125 ml flask. Titrate this sample with an indophenol standard solution. At the end point, an excess unreduced dye will produce a rose pink color in solution.

A similar procedure as above is done for 0.05 g of vitamin C standard.

1 ml of vitamin C standard 1 ml had a vitamin C content of 0.2 mg

10 ml of vitamin C standard 10 ml had a vitamin C content of  $0.2 \times 10 = 2 \text{ mg}$ 

If 2 mg of vitamin C used an indophenol standard solution of a ml, then the sample that used a b ml indophenol standard solution would have a vitamin C content of :

$$= 2 \times b \text{ mg}$$

Since the initial sample volume was 50 ml then for 100 ml of lime juice sample, the sample would contained vitamin C of:  $2 \times b \times 100 \text{ mg/100 ml} = \frac{2 \times b \times 100 \text{ mg/100 ml}}{a \times 50}$ 

# **Appendix E**

### Statistical analysis

A AI UN

#### limonin

Duncan <sup>a,b</sup>	
Duncan	

			Sub	oset	
hydro	Ν	1	2	3	4
CMC	40	6.1064			
carrageenan	40		7.2688		
pectin	40			7.5618	
gum	40				8.9835
Sig.		1.000	1.000	1.000	1.000

Means for groups in homogeneous subsets are displayed.

Based on Type III Sum of Squares

The error term is Mean Square(Error) = .061.

- a. Uses Harmonic Mean Sample Size = 40.000.
- b. Alpha = .05.
- Figure 1 E The statistical analysis of different types and levels of hydrocolloids on the d-limonin content of lime juices during one month storage at ambient temperature

vitc

Duncan <sup>a,b</sup>					9
			Sub	oset	
condition	Ν	1	2	3	4
control	36	13.7853			
500 MPa	36		22.2625		
600 MPa	36		22.4683	22.4683	
400 MPa	36	TIN	22.5061	22.5061	
400+cmc	36	UT		22.7697	
500+cmc	36			22.7928	
600+cmc	36				24.0867
Sig.		1.000	.287	.171	1.000

Means for groups in homogeneous subsets are displayed. Based on Type III Sum of Squares

The error term is Mean Square(Error) = .823.

- a. Uses Harmonic Mean Sample Size = 36.000.
  - b. Alpha = .05.

Figure 2 E The statistical analysis of ascorbic acid of HPP treated-pressured-lime juice during one month storage

#### limoinreft

Duncan <sup>a,b</sup>					
			Sub	oset	
condition	Ν	1	2	3	4
400+cmc	18	2.1522			
600+cmc	18	2.2069	2.2069		
500+cmc	9 18	2.2395	2.2395		
600 MPa	18		2.2911		
500 MPa	18	~ ~		2.6320	
400 MPa	18			2.6999	
control	18				19.5708
Sig.		.185	.201	.273	1.000

Means for groups in homogeneous subsets are displayed. Based on Type III Sum of Squares

The error term is Mean Square(Error) = .034.

- a. Uses Harmonic Mean Sample Size = 18.000.
- b. Alpha = .05.

### Figure 3 E The statistical analysis of HPP treated-lime juice and HPP CMC-added lime juice during one month storage at 4-6°C

limoinAT

Duncan <sup>a,b</sup>					
			Sub	set	
condition	Ν	1	2	3	4
400+cmc	18	2.0143			
600+cmc	18	2.0472	2.0472		
500+cmc	18		2.1576	$\mathbf{k}$	
600 MPa	18	TIN		2.3593	
500 MPa	18	UI		2.3870	
400 MPa	18			2.4031	
control	18				12.7058
Sig.		.630	.109	.550	1.000

Means for groups in homogeneous subsets are displayed. Based on Type III Sum of Squares The error term is Mean Square(Error) = .042.

a. Uses Harmonic Mean Sample Size = 18.000.

b. Alpha = .05.

Figure 4 E The statistical analysis of HPP treated-lime juice and HPP CMC-added lime juice during one month storage at ambient temperature.

4-6°C	$6.85 \pm 0.06^{a}$	$34.89 \pm 0.16^{b}$	$28.42 \pm 0.11^{\circ}$	$21.68 \pm 0.09^{d}$	$18.74 \pm 0.10$	e
Mean $\pm$ SD and values within a row followed	by different letters we	re significantly different (p	j<0.05)		0	
Table 2 E The effect of different §	gum acasia levels o	on the d-limonin conte	ent (ppm) of lime juic	e during one month sto	orage at ambient tempe	rature
h			St	orage time (weeks)		
Gum acasia (g/l)(w/v)		0	1	2	300	4
0 (control)		$6.85 \pm 0.06^{\circ}$	$23.34 \pm 0.53^{d}$	$19.88 \pm 0.15^{d}$	$16.58 \pm 0.13^{\circ}$	$2.72 \pm 0.07^d$
0.5		$5.48 \pm 0.42^{b}$	$18.82 \pm 0.10^{\circ}$	$14.31 \pm 0.04^{\circ}$	$8.51 \pm 0.20^{b}$	$2.60\pm0.08^{\mathrm{b}}$
ar		$5.22 \pm 0.34^{b}$	$17.23 \pm 0.17^{b}$	$9.86 \pm 0.06^{b}$	$8.71 \pm 0.02^{b}$	$2.38 \pm 0.06^{\circ}$
150		$3.29 \pm 0.28^{a}$	$7.52 \pm 0.02^{a}$	$2.33 \pm 0.01^{a}$	$2.18 \pm 0.02^{a}$	$1.84\pm0.20^{\mathrm{ab}}$
Mean $\pm$ SD and values within a column follow	wed by different letters	were significantly differer	nt (p<0.05)		57	

The d-limonin content (ppm) of fresh lime juice during one month storage at different storage temperature Table 1 E

 $\pm 0.07^{e}$ 

2.72

 $16.58 \pm 0.13^{d}$ 

 $19.88 \pm 0.15^{\circ}$ 

 $23.345 \pm 0.53^{b}$ 

 $6.85 \pm 0.06^{a}$ 

Ambient temperature

C

Storage Conditions

3

Storage time (weeks)

2

Table 3 E The effect of different pectin levels on the d-limonin content (ppm) of lime juice during one month storage at ambient temperature

			Storage time (weeks)	1/2.	
Pectin (g/l)(w/v)	0	I	2	3	4
0 (control)	$6.85 \pm 0.06^{a}$	$23.34 \pm 0.53^{d}$	$19.88 \pm 0.15^{\circ}$	$16.58 \pm 0.13^{d}$	$2.72 \pm 0.07^{d}$
	$6.44 \pm 0.57^{a}$	13.56±0.08°	$8.92\pm0.02^{b}$	$4.82\pm0.08^{\mathrm{b}}$	$2.05\pm0.10^\circ$
	$6.30\pm0.36^{\circ}$	<sup>4</sup> 90.0 ± 00.6	$8.22\pm0.04^{b}$	$7.52\pm0.02^\circ$	$1.14\pm0.01^{\mathrm{b}}$
	$6.01 \pm 0.32^{\circ}$	$3.40\pm0.04^{\circ}$	$2.74 \pm 0.02^{*}$	$1.42\pm0.07^{\mathrm{a}}$	$0.32\pm0.02^{a}$
Mean ± SD and values within a column followed by differer	nt letters were significantly dif	ferent (p<0.05)			

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		mn ann ann ann ta fuidd			
			<b>Storage time (weeks)</b>		
CMC (g/l)(w/v)	0		2	3	4
0 (control)	$6.85\pm0.06^{\rm bc}$	$23.34\pm0.53^{\rm d}$	$19.88 \pm 0.15^{b}$	$16.58\pm0.13^{\circ}$	$2.72\pm0.07^{b}$
	$5.75 \pm 0.04^{ab}$	$10.16\pm0.22^{\circ}$	$1.40\pm0.47^{\rm a}$	$2.07\pm0.23^{\mathrm{ab}}$	$2.28\pm0.08^{ab}$
	$5.35 \pm 0.62$ <sup>a</sup>	$6.08\pm0.23^{\rm b}$	$1.19 \pm 0.26^{a}$	$2.30\pm0.03^{\rm b}$	$2.22\pm0.01^{\rm a}$
	$6.34\pm0.30^{\rm b}$	$3.01\pm0.14^{\mathrm{a}}$	$1.31\pm0.29^{a}$	$1.77\pm0.25^{\mathrm{a}}$	$2.02\pm0.30^{\rm a}$
Mean $\pm$ SD and values within a column followed by difference.	nt letters were significantly diff	èrent (p<0.05)		9	

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Table 4 E. The effect of different CMC levels on the d-limonin content (nom) of lime inice during one month storage at ambient temnerature

Table 5 E The effect of different CMC levels on the d-limonin content (ppm) of lime juice during one month storage at ambient temperature

				a man a dama array array	
K-Carra geenan		St	orage time (weeks)	112	
(g/l) (w/v)	0 week	1 week	2 week	3 week	4 week
0 (control)	$6.85\pm0.06^{\rm a}$	$23.34\pm0.53^\circ$	$19.88\pm0.15^{\rm c}$	$16.58\pm0.13^{\rm d}$	$2.72\pm0.07^{ m c}$
0.5	$7.23 \pm 0.88^{ m ab}$	$12.58 \pm 0.42^{b}$	$2.74 \pm 0.02^{b}$	$1.42\pm0.04^{\mathrm{a}}$	$0.32\pm0.02^{\mathrm{a}}$
r	$7.44\pm0.43^{\rm ab}$	$12.25 \pm 0.38^{b}$	$1.68\pm0.01^{\rm a}$	$1.98\pm0.04^{\mathrm{b}}$	$1.26\pm0.09^{\mathrm{b}}$
	$7.97\pm0.07^{b}$	$10.54 \pm 0.28^{a}$	$1.58 \pm 0.22^{a}$	$2.26\pm0.04^{\circ}$	$1.02\pm0.14^{\mathrm{b}}$
Mean $\pm$ SD and values within a column followed by difference of the second structure of the second s	ent letters were significantly diff	ferent (p<0.05)		2	

 $18.74 \pm 0.10^{e}$  $1.35\pm0.52^{bc}$  $1.63\pm0.32^{d}$  $1.35\pm0.14^{\rm b}$  $2.18\pm0.22^{\rm e}$  $1.10 \pm 0.22^{\circ}$  $2.12\pm0.12^{\rm e}$ 4  $21.68 \pm 0.09^{d}$  $1.61 \pm 0.43^{d}$  $1.42 \pm 0.17^d$  $3.56 \pm 0.46^{\circ}$  $1.18\pm0.17^{\rm c}$  $2.55 \pm 0.25^{d}$  $3.11 \pm 0.06^{d}$ ~  $28.42 \pm 0.11^{\circ}$  $1.26\pm0.09^{cd}$  $2.18 \pm 0.22^{d}$  $1.28\pm0.06^{\rm c}$  $1.88 \pm 0.14^{\circ}$  $1.33 \pm 0.11^{\circ}$  $1.14 \pm 0.14^{\circ}$ 2 Storage period (weeks)  $34.89 \pm 0.16^{b}$  $0.98\pm0.03^{\rm bc}$  $1.17\pm0.06^{c}$  $1.34\pm0.32^{\rm b}$  $1.15 \pm 0.25^{\circ}$  $0.74 \pm 0.05^{b}$  $1.46\pm0.22^{\rm c}$  $0.80 \pm 0.01^{\rm b}$  $0.80 \pm 0.08^{\text{b}}$  $0.78 \pm 0.07^{\rm b}$  $6.85 \pm 0.06^{a}$  $1.19\pm0.02^{\rm b}$  $0.74\pm0.27^{b}$  $0.77 \pm 0.04^{b}$ 9  $Mean \pm SD \text{ and } NS= not significant different \\ values within a column followed by different letters were significantly different (p<0.05)$ Before processing<sup>NS</sup>  $6.85 \pm 0.06$  $6.85 \pm 0.06$  $6.85\pm0.06$  $6.85 \pm 0.06$  $6.85 \pm 0.06$  $6.85 \pm 0.06$  $6.85\pm0.06$ **Process Condition** + 400 MPa CMC 1.0 g/L + 500 MPa CMC 1.0 g/L + 600 MPaCMC 1.0 g/L 500 MPa 600 MPa 400 MPa Control

Table 6 E The effect of HPP on the d-limonin (ppm) of CMC-treated and non-treated lime juice during storage at 4-6°C

Table 7 E The effect of HPP on the d-limonin (ppm) of CMC-treated and non-treated lime juice during storage at ambient temperature

<b>8</b> ;	V	Bry W	Storage pe	riod (weeks)	6	
Process Condition	Before processing <sup>NS</sup>	0	1	2	3	4
Control	$6.85 \pm 0.06$	$6.85 \pm 0.06^{a}$	$23.34 \pm 0.53^{b}$	$19.88 \pm 0.15^{\circ}$	$16.58 \pm 0.13^{d}$	$2.72 \pm 0.07^{\rm e}$
400 MPa	$6.85 \pm 0.06$	$0.80 \pm 0.01^{\rm b}$	$1.27 \pm 0.05^{\circ}$	$1.84 \pm 0.07^{d}$	$2.33 \pm 0.10^{e}$	$1.34 \pm 0.06^{\circ}$
CMC 1.0 g/L + $400 \text{ MPa}$	$6.85 \pm 0.06$	$0.80\pm0.08^{ m b}$	$1.17\pm0.16^{b}$	$1.23 \pm 0.20^{b}$	$1.11 \pm 0.11^{b}$	$1.05 \pm 0.14^{\rm b}$
500 MPa	6.85 ± 0.06	$1.19 \pm 0.02^{b}$	$1.34 \pm 0.30^{b}$	$1.66 \pm 0.55^{bc}$	$2.00 \pm 0.11^{\circ}$	$1.13 \pm 0.14^{b}$
CMC 1.0  g/L + 500  MPa	$6.85 \pm 0.06$	$0.74 \pm 0.27^{b}$	$1.25 \pm 0.22^{b}$	$1.31 \pm 0.10^{b}$	$1.36\pm\ 0.08^{\rm b}$	$1.43 \pm 0.11^{b}$
600 MPa	$6.85 \pm 0.06$	$0.77 \pm 0.04^{ m b}$	$1.22 \pm 0.22^{b}$	$1.92 \pm 0.79^{cd}$	$2.08 \pm 0.07^{d}$	$1.32 \pm 0.08^{\rm bc}$
CMC 1.0 g/L + 600 MPa	$6.85 \pm 0.06$	$0.78\pm0.07^{ m b}$	$0.99 \pm 0.33^{\mathrm{bc}}$	$1.18\pm0.16^{\rm c}$	$1.26 \pm 0.19^{\circ}$	$1.23 \pm 0.15^{\circ}$
Moon I CD and MC- not significant different	and the second se					

Mean ± SD and NS= not significant different values within a column followed by different letters were significantly different (p<0.05)

able 8 E The effect of HPP on t	otal acidity (%Citric acid)	of CMC-treated and non	n-treated lime juice du	tring storage at 4-6°C		
			Storage pe	eriod (weeks)		
Process Condition	Before processing <sup>NS</sup>	0 <sup>NS</sup>	CES	2	3	4
Control	$7.03 \pm 0.12$	$7.03 \pm 0.12$	$6.87 \pm 0.11^{ab}$	$6.82 \pm 0.08^{b}$	$6.76 \pm 0.06^{b}$	$6.72 \pm 0.05^{b}$
400 MPa	$7.03 \pm 0.12$	$7.02 \pm 0.06$	$7.02 \pm 0.07^{a}$	$6.90 \pm 0.04^{ab}$	$6.84 \pm 0.05^{b}$	$6.80 \pm 0.08^{b}$
CMC 1.0 g/L + 400 MPa	$7.03 \pm 0.12$	$7.02 \pm 0.06$	$7.01 \pm 0.04^{a}$	$6.94 \pm 0.06^{ab}$	$6.88 \pm 0.02^{bc}$	$6.80 \pm 0.08^{\circ}$
500 MPa	$7.03 \pm 0.12$	$7.05 \pm 0.13$	$7.04 \pm 0.03^{a}$	$6.97 \pm 0.05^{a}$	$6.79 \pm 0.02^{b}$	$6.80 \pm 0.05^{b}$
CMC 1.0  g/L + 500  MPa	$7.03 \pm 0.12$	$7.02 \pm 0.11$	$7.00 \pm 0.01^{ab}$	$6.98 \pm 0.03^{ab}$	$6.90 \pm 0.07^{ab}$	$6.85 \pm 0.12^{b}$
600 MPa	$7.03 \pm 0.12$	$7.05 \pm 0.13$	$7.08 \pm 0.13^{a}$	$6.88 \pm 0.09^{ab}$	$6.77 \pm 0.04^{b}$	$6.73 \pm 0.09^{b}$

÷ 8 2 ξ Ē F ---

 $Mean \pm SD \text{ and } NS= not significant different$ values within a column followed by different letters were significantly different (p<0.05)

 $6.96 \pm 0.24^{a}$ 

 $6.97 + 0.06^{a}$ 

 $6.98 \pm 0.20^{a}$ 

 $6.94 \pm 0.08^{a}$ 

 $7.03 \pm 0.12$ 

7.03 + 0.12

CMC 1.0 g/L + 600 MPa

Table 9 E The effect of HPP on t	total acidity (%Citric acid)	of CMC-treated and n	on-treated lime juice d	uring storage at ambier	it temp	
			Storage perio	d (weeks)		
Process Condition	Before processing <sup>NS</sup>	SN0		2	3	4
Control	$7.03 \pm 0.12$	$7.03 \pm 0.12$	$6.86 \pm 0.06^{b}$	$6.77 \pm 0.02^{bc}$	$6.66 \pm 0.06^{cd}$	$6.58 \pm 0.07^{d}$
400 MPa	$7.03 \pm 0.12$	$7.02 \pm 0.06$	$6.87 \pm 0.11^{b}$	$6.82 \pm 0.08^{b}$	$6.76 \pm 0.05^{\rm b}$	$6.72 \pm 0.05^{b}$
CMC 1.0  g/L + 400  MPa	$7.03 \pm 0.12$	7.02 <u>+</u> 0.06	$6.86 \pm 0.09^{bc}$	$6.79 \pm 0.08^{cd}$	$6.65 \pm 0.12^{d}$	$6.67 \pm 0.05^{d}$
500 MPa	$7.03 \pm 0.12$	$7.05 \pm 0.13$	$6.86 \pm 0.05^{\rm b}$	$6.83 \pm 0.06^{b}$	$6.74 \pm 0.07^{\text{b}}$	$6.72 \pm 0.08^{b}$
CMC 1.0  g/L + 500  MPa	$7.03 \pm 0.12$	$7.02 \pm 0.11$	$6.82 \pm 0.05^{b}$	$6.71 \pm 0.05^{bc}$	$6.62 \pm 0.06^{\circ}$	$6.60 \pm 0.09^{\circ}$
600 MPa	$7.03 \pm 0.12$	$7.05 \pm 0.13$	$6.94 \pm 0.08^{ab}$	$6.81 \pm 0.06^{bc}$	$6.75 \pm 0.11^{\circ}$	$6.70 \pm 0.07^{c}$
CMC 1.0  g/L + 600  MPa	$7.03 \pm 0.12$	$7.03 \pm 0.12$	$6.73 \pm 0.06^{b}$	$6.73 \pm 0.20^{b}$	$6.71 \pm 0.15^{b}$	$6.72 \pm 0.11^{b}$
Mean + SD and NS= not significant differe	ent					

Mean  $\pm$  SU and NS= not significant different values within a column followed by different letters were significantly different (p<0.05)

Process Condition			Storage perio	d (weeks)		
	Before processing <sup>NS</sup>	0 <sup>NS</sup>		2	3	4
Control	$2.34 \pm 0.08$	$2.34 \pm 0.08$	$2.34 \pm 0.05^{ab}$	$2.37 \pm 0.03^{b}$	$2.37 \pm 0.05^{\text{b}}$	$2.37 \pm 0.03^{b}$
400 MPa	$2.34 \pm 0.08$	$2.28 \pm 0.03$	$2.34 \pm 0.00^{a}$	$2.36 \pm 0.02^{a}$	$2.34 \pm 0.02^{a}$	$2.36 \pm 0.03^{a}$
CMC 1.0 g/L + 400 MPa	$2.34 \pm 0.08$	$2.29 \pm 0.05$	$2.34 \pm 0.04^{a}$	$2.32 \pm 0.06^{a}$	$2.34 \pm 0.08^{a}$	$2.36 \pm 0.06^{a}$
500 MPa	$2.34 \pm 0.08$	$2.28 \pm 0.03$	$2.29 \pm 0.06^{a}$	$2.30 \pm 0.09^{a}$	$2.31 \pm 0.01^{a}$	$2.30 \pm 0.01^{a}$
CMC 1.0 g/L + 500 MPa	$2.34 \pm 0.08$	$2.30 \pm 0.02$	$2.32 \pm 0.04^{a}$	$2.32 \pm 0.06^{a}$	$2.31 \pm 0.07^{a}$	$2.33 \pm 0.06^{a}$
600 MPa	$2.34 \pm 0.08$	$2.28 \pm 0.02$	$2.29 \pm 0.06^{a}$	$2.29 \pm 0.02^{a}$	$2.34 \pm 0.01^{a}$	$2.33 \pm 0.03^{a}$
CMC 1.0  g/L + 600  MPa	$2.34 \pm 0.08$	$2.30 \pm 0.04$	$2.33 \pm 0.02^{a}$	$2.31 \pm 0.03^{a}$	$2.31 \pm 0.08^{a}$	$2.37 \pm 0.05^{a}$
Aean ± SD and NS= not significant differe alues within a column followed by differe Table 11 F. The effect of HPP on r	ent ent letters were significantly differe oH value of CMC-treated ar	ant (p<0.05) nd non-treated lime iui	ce during storage at am	hient temberature		
<b>b</b> : <b>r</b> :	V	R'Y	Storage peri	od (weeks)	100	
Process Condition	Before processing <sup>NS</sup>	0 week <sup>NS</sup>		2	3	4
Control	$2.34 \pm 0.08$	$2.34 \pm 0.08$	$2.39 \pm 0.04^{\text{b}}$	$2.41 \pm 0.03^{b}$	$2.43 \pm 0.04^{\text{b}}$	$2.43 \pm 0.05^{b}$
400 MPa	$2.34 \pm 0.08$	$2.28 \pm 0.03$	$2.33 \pm 0.06^{a}$	$2.28 \pm 0.01^{a}$	$2.33 \pm 0.02^{a}$	$2.31 \pm 0.06^{a}$
CMC 1.0 g/L + 400 MPa	$2.34 \pm 0.08$	$2.29 \pm 0.05$	$2.30 \pm 0.02^{a}$	$2.29 \pm 0.04^{a}$	$2.28 \pm 0.06^{a}$	$2.29 \pm 0.02^{a}$
500 MPa	$2.34 \pm 0.08$	$2.28 \pm 0.03$	$2.33 \pm 0.06^{a}$	$2.29 \pm 0.03^{a}$	$2.34 \pm 0.01^{a}$	$2.32 \pm 0.04^{a}$
CMC 1.0  g/L + 500  MPa	$2.34 \pm 0.08$	$2.30 \pm 0.02$	$2.28 \pm 0.02^{a}$	$2.31 \pm 0.04^{a}$	$2.29 \pm 0.02^{a}$	$2.29 \pm 0.02^{a}$
600 MPa	$2.34 \pm 0.08$	$2.28 \pm 0.02$	$2.33 \pm 0.06^{a}$	$2.29 \pm 0.02^{a}$	$2.31 \pm 0.03^{a}$	$2.30 \pm 0.02^{a}$

CMC 1.0 g/L600 MPa $2.34 \pm 0.08$  $2.30 \pm 0.30$ Mean  $\pm$  SD and NS= not significant different $2.34 \pm 0.08$  $2.30 \pm 0.30$ values within a column followed by different letters were significantly different (p<0.05)</td>

 $2.30 \pm 0.02^{a}$  $2.28 \pm 0.03^{a}$ 

 $2.31 \pm 0.03^{a}$  $2.30 \pm 0.03^{a}$ 

 $2.30 \pm 0.05^{a}$ 

 $2.31 \pm 0.05^{a}$ 

 $2.30 \pm 0.04$ 

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