## RESEARCH REGARDING THE INFLUENCE OF ETHEPHON ON THE RIPE ACEROLA (MALPIGHIA GLABRA L.)

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**ABSTRACT.** These days, there are many types of chemicals which have ability to hasten the ripe fruit and affect the quality of fruits like calcium carbide, ethylene, acetylene, gibberellic acid. The objective of this research determines the influence of ethephon (2-chloroethyl phosphonic acid or ethrel) on the ripe acerolas (Malpighia glabra L.). Because the acerolas are the common fruit in Vietnam and another Asia countries, it had a lot of water and high nutrition, especially reducing sugars and acid ascorbic. The acerolas samples was solution soaked into ethephon at concentrations of 0, 1, 1.5, 2 and 2.5 % (v/v), then preserved in basket covered with cloth in conditions: temperature 30±1.4 °C, relative humidity 72±10 %. The use of specifications for evaluating were the percentage of ripe fruits, weight loss, the content of reducing sugar, total acidity and sensory evaluation of acerolas. The results showed that acerolas soaked into concentration of 2 % (v/v) ethephon have stimulated fruits that were the quick and uniform maturity; peel of acerolas was very smooth, red and characteristic flavour. Consumers are not unpleasant with the sensory evaluation quality of fruits soaked with ethephon. The ethephon residue in fruit after treatment did not harm in consumer's health and it was absolutely suitable for the food law in Vietnam.

**Key words:** Ethephon; 2-chloroethyl phosphonic acid; Ethrel; Acerolas; Maturity.

#### INTRODUCTION

Acerola (Malpighia glabra L.) is tropical fruit in the family а Malpighiaceae. Acerolas had a bright red drupe, is 1–3 cm in diameter with weight of 3–5 g. The fruit peel was glossy, thin, soft and easily damage. It was cultivated in the south Vietnam with the high yield. Acerola had a lot of water and the high nutrition so it material in beverage was raw processing and was the main fruit for with vietnamese dessert (vi.wikipedia.org, 2009). In Vietnam,

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it was usually preserved at normal condition in the local market store in .

Mature fruits produce a certain amount of ethylene which does not make uniform maturation in a batch. Therefore, a solution of chemical treatment is necessary. Ethephon (ethrel) was known as a plant growth regulator which decomposes into ethylene. It stimulates defoliation, ripe evenly fruit, decreasing preservation time and minimizing post-harvest losses (Pham, 2005).

Recently, there have been many mixed opinions on the toxicity of ethephon that confused the customers (Nguyen et al., 1999). It was classified organophosphate as an pesticide and was registered with EPA Environmental Protection (US Agency) since 1973 as a plant growth regulator used to promote fruit flower ripening and induction. Ethephon is irritant to the skin or the eves but is not a skin sensitizer, it was not a carcinogen and is classified by (International Agency IARC for Research on Cancer) as group D (not carcinogenic to humans) and FAO pointed out a maximum allowable daily intake for ethephone at 0.05 mg/kg body weight/day (Bui, 2007). In addition, using ethephon solution in ripe fruit was quite new and having no article for the influence of this chemical on the ripe acerolas. Nevertheless, ethephon could be hygroscopic and released ethylene during storage. Thus, ethephon could hasten the ripe acerolas and the ethephon residues did not affect the health of consumers with the tiny dose. Therefore, we conducted to research this subject.

## MATERIALS AND METHOD

## Materials

Acerolas are harvested in Go Cong, Ben Tre province, Vietnam. Fruits will be harvested after approximately 25 days from the date simultaneously bloom. Harvested fruits are  $20.5\pm1.87$  mm in diameter with weight 4-6 g/fruit.

Ethephon are in liquid form, pale yellow color, transparent and from China.

## Method

### Ripe acerolas processing

Acerolas  $\rightarrow$  Shorting  $\rightarrow$  Washing $\rightarrow$ Soaking  $\rightarrow$  Keeping dry  $\rightarrow$  Preserving

Acerolas were preserved into a basket covered with a cloth at temperature  $30\pm1.4$  °C and relative humidity  $72\pm10$  %.

## Analysis method

The percentage (%) of weight loss was used by electronic scales, the mass loss rate based on the volume of the preservation process results over the volumes initially.

The concentration of reducing sugar was determined by Lane – Eynon method. We used glucose solution 1%, the mixture of Fehling A and B. Sugars will discoloration of methylene blue indicator, which is the end of the reaction. Content of reducing sugars was expressed in percentage (%) of fruit mass.

The total acidity value was determined by (AOAC 942.15, 2007). Titration acidity was performed by NaOH 0.1 N with phenolphatalein 0.1% as an indicator and expressed in grams (g) of total acidity per 100 g of fruit.

Residue examination of ethephon on acerolas was determined by gas chromatography–mass spectrophotometer (GC – MS) (AOAC 2007.01, 2007). It was expressed in milligrams (mg) of ethephon per 1 kg of fruit.

Different testing on sensory evaluation: pair comparison test – the simple difference test (or the same/different test) (Lawless and Heymann, 2010).

#### Data analysis

Each sample of acerolas had weight of 250 g, corresponding to 55–60 units, repeated three times. Data would be analyzed by Statgraphics program (Centurion XV) with confidence interval p=0.05.

## **RESULTS AND DISCUSSION**

## Effect of ethephon concentration solution on acerola maturity and weight loss versus preservation time

In *Table 1*, the percentage of ripe acerolas at different concentrations increased corresponding to incubation time. In particular, the samples soaked with ethephon solution at higher concentration resulted higher in ripening rate and faster ripening time without ethephon than those treatment. At time 50 and 60 hours. the soaking sample in 2 and 2.5% of ethephon solution obtained the highest ripening grade. however, these samples decreased due to the spoilage. The samples at these concentrations matured quite equally, had specific aroma and dark red color. It is proved that ethephon was decomposed to release ethylene which promotes the ripening process of acerolas. The more ethylene was decomposed, the ripe fruit promoted fast (Le et al., 2008). The ripening rate of soaking samples in 2% of ethephon increased approximately 35% after 50 hours and 20% after 60 hours compared to those without ethephon treatment. However, at time 70 hours, the samples without ethephon treatment reached the highest ripening rate (75.85%)(Fig. 1).

During post harvest ripening, weight loss rate increased over the preservation time because the respiratory plant process released carbon dioxide, water and consumes glucose. In addition, acerolas have high water content 85-95%, the evaporation process may be one of the main reasons causing weight loss of fruits (Ton, 2008). For the first 40 hours. the weight loss was inconsiderable. From 40 to 80 hours. the weight loss increased rapidly (Table 2). At the time of 40 incubation hours, the weight loss of acerolas increased nearly two times when comparing with the time 30 hours. And at the time 50 hours, the weight loss raised approximate three times when comparing with the time 40 hours. The weight loss slowed from the time 50 to 60 hours. After 60 hours of incubation, the weight loss increase dramatically, particularly the soaked with 2.5% samples of ethephon. The weight reduction was slow in samples with 2% ethephon when comparing with treatment remaining samples (Fig. 2).

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Hour	Conc. (%,v/v)					
nour	0	1	1.5	2	2.5	
10	$0.00\pm0.00$	$0.00{\pm}0.00$	$0.00\pm0.00$	$0.00\pm0.00$	$0.00\pm0.00$	
10	Aa	Aa	Aa	Aa	Aa	
20	$\textbf{0.00} \pm \textbf{0.00}$	$0.00\pm0.00$	3.70± 3.21	$3.64 \pm 4.81$	$8.02{\pm}~4.08$	
20	Aa	Aa	Aab	Aab	Bb	
20	$6.35 \pm 3.70$	$15.93 \pm 2.61$	$38.68 \pm 2.61$	41.82± 1.82	44.09± 1.44	
	Ва	Bb	Bc	Bcd	Cd	
40	$30.44 \pm 2.05$	$50.31{\pm}0.54$	$65.69 \pm 4.00$	72.73± 1.82	$75.77 \pm 1.75$	
40	Ca	Cb	Cc	Cd	Dd	
50	48.86± 1.81	$71.78{\pm}0.40$	$82.82{\pm}0.92$	83.03± 1.05	$85.73 \pm 0.72$	
	Da	Db	Dc	Dc	Ed	
60	$65.53 \pm 0.99$	$81.59 \pm 0.51$	$84.67{\pm}0.93$	$86.67 \pm 1.05$	$89.51 \pm 4.09$	
00	Ea	Db	Dbc	Dcd	Ed	
70	$75.85 \pm 0.17$	$78.55 \pm 1.54$	$76.11 \pm 2.71$	$73.94 \pm 2.10$	$67.65 \pm 2.89$	
70	Fab	Eb	Eab	Ca	Fc	
80	$43.22 \pm 7.17$	$39.33{\pm}3.86$	41.74± 1.70	$48.48{\pm}3.78$	$43.47{\pm}2.46$	
00	Dab	Ba	Bab	Eb	Cab	
90	$5.09\pm5.00$	$0.00\pm0.00$	$0.00\pm0.00$	$0.00\pm0.00$	$0.00\pm0.00$	
30	ABa	Ab	Ab	Ab	Ab	

 Table 1 - Percentage of ripe acerolas (%) versus the preservation time



Figure 1 - The ripe acerolas rate (%) versus the preservation time

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Hour	Conc.(%,v/v)					
neu	0	1	1.5	2	2.5	
0	$0.00 \pm 0.00$					
	Aa	Aa	Aa	Aa	Aa	
10	$\textbf{0.67} \pm \textbf{0.23}$	$0.53 \pm 0.46$	$\textbf{0.67} \pm \textbf{0.23}$	$\textbf{0.67} \pm \textbf{0.23}$	$\textbf{0.27} \pm \textbf{0.23}$	
	ABa	Aba	Ba	ABa	ABa	
20	$\textbf{0.80} \pm \textbf{0.00}$	$\textbf{0.53} \pm \textbf{0.23}$	$\textbf{0.67} \pm \textbf{0.23}$	$\textbf{0.53} \pm \textbf{0.23}$	$\textbf{0.53} \pm \textbf{0.23}$	
20	ABa	Aba	Ва	ABa	ABCa	
30	$\textbf{1.20} \pm \textbf{0.01}$	$\textbf{1.20} \pm \textbf{0.01}$	$\textbf{1.33} \pm \textbf{0.23}$	$1.32\pm0.60$	$\textbf{0.93} \pm \textbf{0.23}$	
50	Ba	Ba	Ca	Ba	BCa	
40	$\textbf{2.14} \pm \textbf{0.24}$	$\textbf{2.40} \pm \textbf{0.68}$	$1.47\pm0.23$	$\textbf{2.65} \pm \textbf{0.22}$	$\textbf{1.33} \pm \textbf{0.23}$	
	Ca	Ca	Cb	Ca	Cb	
50	$\textbf{7.48} \pm \textbf{0.61}$	$\textbf{6.39} \pm \textbf{0.43}$	$6.39 \pm 0.69$	$5.96 \pm 0.77$	$\textbf{6.64} \pm \textbf{0.61}$	
50	Da	Dab	Dab	Db	Dab	
60	$\textbf{8.01} \pm \textbf{0.43}$	$\textbf{6.93} \pm \textbf{0.26}$	$\textbf{6.79} \pm \textbf{0.40}$	$\textbf{6.49} \pm \textbf{0.45}$	$\textbf{7.57} \pm \textbf{0.69}$	
00	Da	Dbc	Dbc	Dc	Eab	
70	$9.48\pm0.85$	$12.78\pm0.80$	$11.19\pm0.40$	$10.99 \pm 1.16$	$13.81\pm0.92$	
	Ea	Eb	Ec	Eac	Fb	
80	$12.28\pm0.90$	15.31 ± 1.00	$14.380\pm0.37$	14.04 ± 1.21	$16.60\pm0.82$	
00	Fa	Fbc	Fb	Fb	Gc	

Table 2 - The weight loss (%) versus the preservation time



Figure 2 - The weight loss (%) versus the preservation time

### Effect of ethephon concentration solution on reducing sugar and total acidity versus preservation time

The amount of reducing sugar in fruits tended to decrease slowly during preservation time. The fruits would produce more amount of reducing sugar than that of consumed sugar during ripening process (Le et al., 2008; Tran, 2004). In Table 3, the samples without ethephon treatment, the quantity of reducing sugar fluctuated slightly and increased gradually at greater ethephon concentrations. The samples treated with 2.5% of ethephon after 30 hours,

the amount of reducing sugar grew quite strongly (8.43%). Otherwise, this amount varied unstably and decreased rapidly, from 6.86% after 40 hours to 4.9% after 60 hours. Acerolas in this phase began to spoil which caused high respiration to consume much amount sugar (Ton, 2008). The treatment sample with 2% of ethephon, amount of reducing sugar did not increased rapidly when comparing with 2.5% of ethephon (reached 8.43% after 40 hours); in return, this amount had longer and slower stability reduction (retained 6.67% after 60 hours) (*Fig.* 3).

Hour	Conc.(%,v/v)					
nour	0	1	1.5	2	2.5	
0	4.71 ± 0.59	4.71 ± 0.00	4.51 ± 0.68	4.71 ± 0.59	4.71 ± 0.59	
	Aa	Aa	Aa	Aba	Aa	
10	4.90± 0.34	5.10 ± 0.34	4.90± 0.34	4.9± 0.34	5.49± 0.34	
10	ABa	ABa	Aa	ABa	BCa	
20	5.10 ± 0.34	5.49± 0.34	6.08± 0.34	5.88± 0.59	6.67± 0.34	
20	ABa	BCab	Bbc	CDb	Dc	
20	5.49± 0.34	5.69± 0.34	7.65± 0.59	8.24± 0.59	8.43± 0.34	
30	BCa	Bca	CDb	FGb	Eb	
40	6.08± 0.34	7.06± 0.59	8.24± 0.59	8.43± 0.34	6.86± 0.34	
	Ca	Eb	Dc	Gc	Dab	
50	6.86± 0.34	8.24± 0.59	7.84± 0.68	7.45± 0.34	5.88± 0.59	
	Da	Fb	CDb	EFab	Cc	
60	7.45± 0.34	6.67± 0.34	7.25± 0.34	6.67± 0.34	4.9± 0.34	
	Da	Deb	Cab	DEb	ABc	
70	7.06 ±0.59	6.08± 0.34	6.08± 0.34	5.49± 0.90	4.51± 0.34	
70	Da	Cab	Bab	BCbc	Ac	
90	6.86± 0.34	5.1±0.68	5.88± 0.59	4.51± 0.34	4.31± 0.34	
80	Da	Abc	Bb	Ac	Ac	

Table 3 - Amount of reducing sugar (%) versus preservation time

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Preservation time (hour)

Figure 3 - Amount of reducing sugar (%) of acerolas versus the preservation time

Hour	Conc.(%,v/v)					
nour	0	1	1.5	2	2.5	
0	3.164 ±0.06	3.127 ±0	3.127 ±0.11	3.201 ±0.06	3.238 ±0.11	
	Aa	Aa	Aa	Aa	Aa	
10	2.941 ±0.06	3.052 ±0.13	2.941 ±0.06	3.052 ±0.06	3.015 ±0.11	
10	BCa	Aba	ABb	ABc	Bc	
20	2.903 ±0.22	2.866 ±0.32	2.829 ±0.06	2.903 ±0.11	2.866 ±0.06	
20	Cb	Aba	BCa	BCb	BCa	
30	2.829 ±0.13	2.792 ±0.11	2.829 ±0.26	2.972 ±0.11	2.754±0.17	
	CDab	Bca	BCab	CDa	CDa	
40	2.792 ±0.11	2.754 ±0.13	2.754 ±0.13	2.717 ±0.06	2.606 ±0.13	
	CDab	Cb	BCb	Db	Dc	
50	2.717 ±0.28	2.717 ±0.06	2.68 ±0.11	2.606 ±0.06	2.308 ±0.13	
50	CDa	Cda	CDa	DEa	Eb	
60	2.643 ±0.06	2.457 ±0.22	2.531 ±0.06	2.494 ±0.23	2.122 ±0.11	
	DEa	Dea	Da	Ea	EFb	
70	2.419 ±0.06	2.196 ±0.06	2.271 ±0.06	2.084 ±0.06	2.01 ±0	
10	EFa	Efb	Eb	Fc	Fc	
80	2.271 ±0.06	2.084 ±0.13	2.047 ±0.06	2.084 ±0.06	2.047 ±0.06	
80	Fa	Fb	Fb	Fb	Fb	

Table 4 - Amount of total acidity value (g/l) versus the preservation time



Figure 4 - Amount of total acidity value (g/l) of acerolas versus the preservation time

In Table 4, the total acidity value in acerolas tended to decrease during the preservation time. It are the major respiratory substrates, utilized higher than others and could decrease to 50% during fruit formation. Reducing amount of total acidity in ripe fruits results as increasing of cell membrane penetration lead acid to diffuse into cells, this process released some derivatives of malic acid, which would convert into soluble sugar (Le et al., 2008; Tran, 2004). The amount of total acidity in 0% of ethephon sample decreased at the least and was significant different at level of p=5% when comparing with the remaining samples. Amount total acidity of all samples treated with ethephon changed inconsiderably (Fig. 4). Acerolas changed to ripening process, so damage process began to appear at the same time.

In this research, the samples treated with 2% of ethephon solution which had high ripening rate and reducing sugar amount, low weight loss, ripening evenly and having good sensory evaluation after incubation time from 50 to 60 hours.

## Testing the ethephon residue on the acerolas

After the preservation time from 50 to 60 hour, the ethephon residue on acerolas was covariant when comparing its with the ethephon concentration solution (Table 5). residue However, ethephon will reduce over preservation time cause ethephon could be hygroscopic and released ethylene (Tran, 2000) (Fig. 5). Nevertheless, residues on samples still remain in the permitted range (in accordance with the current regulation of Vietnam-Circular No. 68/2012/TT-BNNPTNT).

Table 5 -	The ethephon	residue on	the acerolas	(mg/kg)
				\ J' J/

Ethephon concentration in soaking solution (%,v/v)	1	1.5	2	2.5
Ethephon residue on the acerolas (mg/kg)	0.0715±0.011 <sup>a</sup>	0.0825±0.005 <sup>ab</sup>	0.11±0.013 <sup>bc</sup>	0.12±0.014 <sup>c</sup>

Various small lowercase letters are significant difference at the level of p=5%.

$$CICH_2-CH_2-P-OH \xrightarrow{H_2O} CH_2 = CH_2 + H_3PO_4 + HCI$$

$$/ OH$$

Figure 5 - Ethylene was formed from ethephon (Tran, 2000)

# Sensory evaluation of acerolas after soaking in ethephon solution

We used 24 participants who have evaluated the similarities and differences of the samples of acerola which are not soaked (*Figs. 6 and 7*), were bought in the local market and soaked into ethephon solutions with the preservation time 50 hour. Result was presented in *Table 6*.

 $\chi^2 < \chi^2$  <sub>cri</sub>, consumers were unable to differentiate the soaking and not soaking samples into ethephon solution. In other words, these two samples (A and B) are the similar at the level of p=5%.



Figure 6 - Acerolas before soaking in ethephon solution



Figure 7 - Acerolas after soaking in ethephon solution

Quantity of Response Sample order samples Similar Different 2 AA 6 4 AB 6 3 3 BΒ 6 5 1 ΒA 6 4 2

Table 6 - Responding summary of participants

A: samples soaked into ethephon solution

B: samples did not soaked into ethephon solution

 $\chi^2 = 0.75$ 

#### CONCLUSIONS

At an ethephon concentration of 2%, acerolas were preserved in about 50–60 hours, they had an uniform maturity, low weight loss, high reducing sugar, minor changes on total acidity.

The ethephon residues on acerolas was in the safe range for health of consumers. Consumers did not realize the differences of acerolas after soaking into the ethephon solution.

Acerolas which are hastened to ripe by using ethephon with small cost (2 USD per 500 ml of ethephon); while generate a considerable efficiency of commercial value (Ethephon can be dilute to 25 liters). This cost is likely acceptable in Vietnam.

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