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## Effect of high concentration-ozone fumigation on chemical and physical changes in fresh chilli

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### Abstract

Live insect such as fruit fly inside chilli fruit is one of major problems for Thai chilli export industry. This research dealt with an application of high concentration gaseous ozone affecting chemical and physical changes in chilli fruit. Fresh chilli cv. “Super Hot” was fumigated with ozone in 25 liter-polycarbonate container under pressure 12 kPa with flow rate 7.5 l/min through ozone generator. Chilli was treated with high concentration ozone in a range of 444 – 1000 ppm and holding time of 5 – 15 min. Changes in chemical properties were measured using hydrogen peroxide content in exocarp and placenta of chilli fruit as an indicator of ozone penetration. Color and firmness were measured as indices of physical properties. Results indicated that after ozone fumigation, the hydrogen peroxide content in chilli significantly increased ( $p < 0.05$ ) during first week and then significantly declined ( $p < 0.05$ ) during the second week in storage of 5°C. Color difference index  $\Delta E$  of chilli fruit did not significantly change ( $p \geq 0.05$ ) but that of chilli stem was significantly different ( $p < 0.05$ ) as it became darker than that of the control. Firmness of treated chilli fruit was not significantly different ( $p \geq 0.05$ ) compared to that of the control. This work provides a preliminary guideline for further study for selection of high concentration ozone treatments as an alternative to disinfest the fruit fly in chilli.

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*Keywords:* Chilli; Ozone Fumigation; Fruit fly disinfestation; Chemical and physical properties

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### 1. Introduction

A major persistent problem affecting export of Thai fresh chilli is an existence of live insect pest that cannot disinfest completely. The EU regulation requires residue of pesticide such as organophosphate in fresh chilli to be as low as 3 ppm (Pornsiripratharn, 2010), causing less capacity to control of live insect pest, especially the chilli fruit fly (*Bactrocera latifrons*). Fruit fly normally lays eggs 1 cm below surface of chilli fruit. As egg hatches, the 1<sup>st</sup> instars grows within 2-3 days (Wingsanoil and Siri1, 2011). Currently, there is no suitable treatment to substitute the pesticide control to disinfest the fruit fly from farm to fork. Hot water and vapor heat treatments are among methods to disinfest

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fruit fly for export commodities such as mango (Varith et al., 2007). However, these methods have not been intensive studied to suit for fruit fly disinfestation in chilli because of heat sensitivity to the chilli fruit.

Ozone gas is a strong oxidize and potent disinfecting agent that can react directly with double bonds of polyunsaturated fatty acids to organic substance (Muthukumarappan et al., 2009). During degradation of ozone, it can be transformed into peroxide or hydrogen peroxide which potent to destroy vital molecules. Thus such effects can result in cellular injuries and death of the insect which can be used for pest control (Holmstrup et al., 2011) or disease control in fruit and vegetable (Pengphol et al., 2006). Ozone is also rapidly and spontaneously degraded to O<sub>2</sub>, and therefore is not associated with toxic residues remaining in the products or environment. The aim of this work was to preliminary study the effect of ozone to overall quality of chilli which can lead to the process to disinfest the fruit fly eggs and larvae.

## 2. Materials and methods

### 2.1. Ozone fumigation system

The ozone fumigation system consisted of a corona discharge-ozone generator. The generator connected with the control system using Labview™ program with wireless network. The system connected to fumigation chamber of 0.4×0.4×1.2 m<sup>3</sup>, able to hold two 25 l-polycarbonate containers. The system conveyed ozone gas via silicone tube. The optimum flow rate of ozone gas was 7.5 l/min with system pressure in a range of 11-25 kPa to generate ozone gas at high concentration of 5.5 g/h. (Varith et al., 2010)

### 2.2. Sample preparation

Fresh chilli cv. “Super Hot” harvested less than 1 day was obtained locally from wholesale market in Chaing Mai, Thailand. Chilli sample of 4 kg was filled in polycarbonate container to fit a half tank. For fumigation process, ozone gas with concentration of 444 – 1000 ppm was fed into the container and held under pressure for 5 – 15 min, resulting in equivalent concentration-time (CT) of 4,445 – 13,335 ppm-min (Ozkan et al., 2011) as shown in Table 1. After fumigation, the samples was stored at 5 °C in polystyrene trays and wrapped with clear polypropylene plastic bag with holes for ventilation following the commercial practice.

Table 1 Test conditions of ozone fumigation on chilli.

Condition	Concentration Ozone (ppm)	Holding time (minute)	Dose (ppm×minute)
0 (Control)	-	-	-
1	889	5	4,445
2	1,000	5	5,000
3	444	15	6,660
4	778	10	7,780
5	889	10	8,890
6	1,000	10	10,000
7	778	15	11,670
8	889	15	13,335

### 2.3. Measurement of hydrogen peroxide

The hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) assay followed the modified method of Abnova (2013) was applied to indicate the ozone gas penetration. Treated chilli was separated and blended between exocarp and placenta. The 0.1 g exocarp and

placenta slurries were diluted with a mixture of 2 ml of 50 mmol K<sub>3</sub>PO<sub>4</sub> (pH 6.8), 0.1 mmol EDTA and 100%PVPP, and incubated for 30 min at 20°C. The slurry was centrifuged at 2,500 rpm for 15 min. The clear-top solution of mixture was separated and added with reagent at concentration of 1:10 (by volume) for hydrogen peroxide analysis. Absorbance test was immediately performed at 550 nm using photometer (SLT Spectra, Germany) with commercial H<sub>2</sub>O<sub>2</sub> was used as standard curves. All measurement were carried out at room temperature room (28 °C).

#### 2.4. Color measurement

A spectrophotometer (Miniscan XE, Hunter Lab, USA) was used to assess the color of chilli and stem. Ten readings per pack were taken to ensure that the data obtained represented the all variation color within fruit and stem for chilli. The means of L\*, a\* and b\* from 10 readings were reported and calculated for color difference ΔE (Eq. 1) and hue angle (Eq. 2 to Eq. 4), which were then used compared to those of the control (Hansuebsia, 2011).

$$\Delta E = \sqrt{(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2} \quad (1)$$

$$\text{Hue angle} = \arctangent (b^*/a^*) ; a^* > 0, b^* \geq 0 \quad (2)$$

$$\text{Hue angle} = \arctangent (b^*/a^*) + 180^\circ ; a^* < 0 \quad (3)$$

$$\text{Hue angle} = \arctangent (b^*/a^*) + 360^\circ ; a^* > 0, b^* < 0 \quad (4)$$

Hue angle was measured to scale the color of sample between 0-360 degree. The 0-45 degree represented purple-red to orange-red; the 45-90 degree represented to orange-red to yellow; the 90-135 degree represented yellow to yellow-green; the 135-180 degree represented yellow to green; the 180-225 degree represented green to blue color; the 225-315 degree represented blue to purple; and the 315-360 degree represented purple to purple-red.

#### 2.5. Texture measurement

The firmness of chilli was measured using a Texture Analyzer TA.XT2i Plus (Texture Technologies Corp, MA, USA) attached with stainless steel cylinder probe 2 mm. Five chilli fruits were selected randomly and penetrated at 1 cm distance from the stem. Test was conducted using cross-head speed of 2 mm/s penetrated through chilli. The maximum force was recorded using the Texture Expert software to monitor firmness of treated samples compared to that of the control.

#### 2.6. Statistical analysis

Samples from each treatment were analyzed during days in storage using the Statistical Package for the Social Sciences (SPSS, IBM Corp., USA) by one way analysis of variance (ANOVA). The Sheffe's method test was used to determine significant differences at p<0.05.

### 3. Results and discussion

#### 3.1. Ozone penetration

The ozone penetration of chilli exocarp and placenta of is shown in Fig. 1. The H<sub>2</sub>O<sub>2</sub> values exhibited the same trend between exocarp and placenta of chilli where H<sub>2</sub>O<sub>2</sub> concentration tended to increase during first week of storage,

then reaching the peak in second week and finally decrease to the lowest level in the third week. Comparing the magnitude, the H<sub>2</sub>O<sub>2</sub> concentration in exocarp was always higher than in the placenta. It can be explained that as ozone fumigation finished, the ozone had already penetrate into the placenta (core) of chilli. This effect is importance to ensure that ozone gas always goes thorough entire chilli, especially through the core of chilli where the live fruit fly larvae is hardly spotted. The increase of H<sub>2</sub>O<sub>2</sub> after fumigation may cause from lipid peroxidation of cell membranes to generate various such as peroxide radical, H<sub>2</sub>O<sub>2</sub> or singlet oxygen (Purvis et al., 1993). However, the H<sub>2</sub>O<sub>2</sub> finally degraded as it declined from observation during the last week.

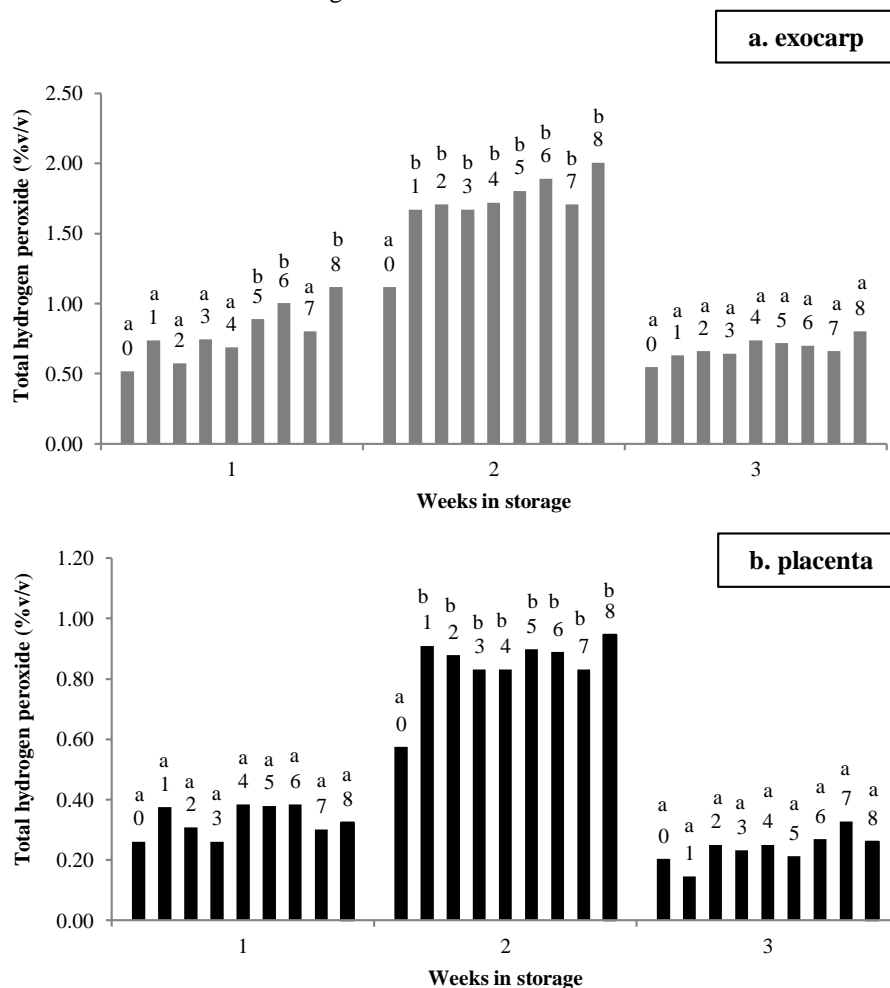


Fig. 1. Total hydrogen peroxide in (a) exocarp and (b) placenta of chilli stored at 5 °C for 3 weeks. Number on each column represents treatment according to Table 1. Letter on each column indicates significant difference at p<0.05.

### 3.2. Color and texture

The color differences of chilli fruit and stem expressed by  $\Delta E$  and hue angle values are shown in Table 2. Values of  $\Delta E$  and hue angle of the fruit was not significantly different ( $p \geq 0.05$ ) from that of the control during storage for 14 days. Color of chilli fruit was not affected by the high concentration ozone. In contrast,  $\Delta E$  in stem shows significant difference ( $p < 0.05$ ) from that of the control while hue angle values were statistically insignificant ( $p \geq 0.05$ ). It means

that the high concentration ozone fumigation had effect on the stem, as the chilli stem became darker than that of the control.

Table 2 Color measurements of chilli fruit and stem during storage at 5 °C

Condition	Day 0		Day 4		Day 8		Day 11		Day 14	
	$\Delta E$	Hue	$\Delta E$	Hue	$\Delta E$	Hue	$\Delta E$	Hue	$\Delta E$	Hue
<b>Fruit</b>										
Control	0	0.60	0	0.60	0	0.59	0	0.62	0	0.65
1	0.99	0.59	1.76	0.58	1.91	0.51	1.54	0.60	1.91	0.69
2	1.75	0.59	1.87	0.58	1.30	0.58	1.04	0.60	1.36	0.70
3	0.65	0.60	1.25	0.58	1.72	0.54	1.92	0.60	1.01	0.67
4	1.37	0.59	1.02	0.59	2.08	0.56	1.62	0.58	1.55	0.67
5	1.83	0.60	1.12	0.58	1.64	0.58	1.89	0.58	1.67	0.59
6	1.56	0.62	1.76	0.58	1.22	0.55	1.10	0.62	1.93	0.58
7	1.25	0.60	1.76	0.60	1.90	0.58	1.00	0.61	1.11	0.57
8	1.82	0.60	1.5	0.58	1.35	0.54	1.56	0.62	1.86	0.61
<b>Stem</b>										
Control	0	178.72	0	178.72	0	178.70	0	178.68	0	178.70
1	5.67	178.73	4.18	178.71	5.08	178.68	3.75	178.71	4.76	178.67
2	4.89	178.72	4.90	178.72	4.16	178.70	4.14	178.68	2.18	178.68
3	4.05	178.70	1.93	178.68	2.47	178.66	3.39	178.65	3.34	178.64
4	2.56	178.72	4.03	178.71	3.38	178.71	4.77	178.73	2.83	178.69
5	4.70	178.72	4.45	178.72	3.58	178.70	4.54	178.68	7.13	178.70
6	2.07	178.74	2.76	178.74	1.62	178.71	3.27	178.71	7.9	178.71
7	3.73	178.71	2.50	178.73	1.25	178.70	3.34	178.69	5.52	178.69
8	5.56	178.79	4.79	178.77	2.62	178.72	3.18	178.70	9.05	178.74

Fig. 2. shows that firmness of chilli slightly decreased on treated samples compared to that of the control. There were no significant differences ( $p \geq 0.05$ ) for all treatments. It means that ozone fumigation did not affect the firmness of chilli as it gradually decreased during storage for 14 days due to the natural decay. Wang *et al.* (2004) stated that oxidizing agents can cause oxidation of phenolic cross-linkages among cell wall pectin, structural proteins or other polymers causing changes in firmness of the fruit. However, in our study, it is possible that the concentration of ozone as high potent-oxidizing agent may not strong enough to cause such changes of firmness of chilli, or the cell wall of chilli has enough resistance to endure the high concentration as such.

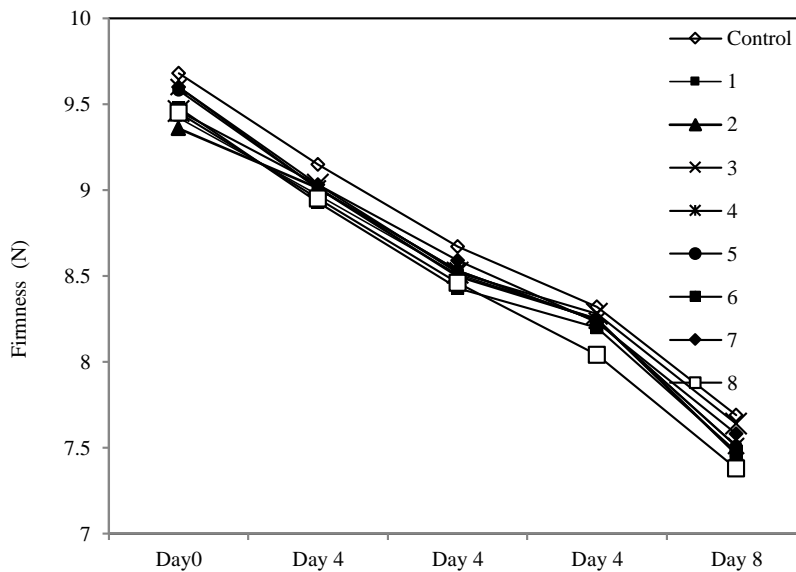


Fig. 2. Effect of ozone fumigation on firmness of chilli during storage 5 °C.

#### 4. Conclusions

The high concentration-ozone fumigation on fresh chilli resulted to increase of  $H_2O_2$  in exocarp and placenta. Ozone affected the stem as it appeared darker than that of the control but did not affect color of the fruit. Ozone did not affect the firmness of chilli during storage for 14 days as well. All treatments exhibited of great potential for disinfestation fumigation of the chilli fruit fly since the ozone had penetrated throughout the fruit, from skin to core. This work exhibited great potential for further study of ozone to be used as disinfestation agent for pest fumigation such as fruit fly.

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