CHAPTER 4

Results

Experiment 1 Effects of gaseous ClO₂ fumigation on mitochondrial energy levels and redox status of harvested 'Daw' longan pericarp during storage

1.1 Effects of exogenous ATP on antioxidant capacity and fruit quality of harvested 'Daw' longan during storage

Changes in antioxidant capacity and storage fruit quality and the effects of exogenous ATP on these parameters were observed in longan cv. Daw during storage.

1. Antioxidant capacity

In order to determine the antioxidant capacity of harvested longan pericarp during storage, three assays, ABTS, DPPH and FRAP, were utilized. The results from all assays generally exhibited similar and comparable changes. Antioxidant capacity of the untreated group immediately decreased until the end of the experiment (Figures 4.1-4.3 and Appendix: Table 1-3). ATP treatment altered antioxidant capacity of the samples.

It was found that alteration of antioxidant capacity by ATP was both concentration and time dependent (Figures 4.1-4.3). The higher the ATP concentration used the more pronounced the effects could be seen. The effects of 1 and 2 mM treatment were indiscernible. However, antioxidant capacity increased to maximum (between 18 to 35% depending on the assay) within 1 day of treatment and increased significantly to a higher value than that of the control. Interestingly, the pattern of decrease in antioxidant capacity and the time at which it started were similar regardless of the treatment (Figures 4.1-4.3 and Appendix: Tables 1-3).



Figure 4.1 Effects of exogenous ATP on antioxidant capacity assayed by ABTS method of 'Daw' longan pericarp during storage at 25±1°C (BD, before dipping; AD, after dipping; each value is presented as mean ± standard deviation; n = 3)



Figure 4.2 Effects of exogenous ATP on antioxidant capacity assayed by DPPH method of 'Daw' longan pericarp during storage at 25±1°C (BD, before dipping; AD, after dipping; each value is presented as mean ± standard deviation; n = 3)



Figure 4.3 Effects of exogenous ATP on antioxidant capacity assayed by FRAP method of 'Daw' longan pericarp during storage at 25±1°C (BD, before dipping; AD, after dipping; each value is presented as mean ± standard deviation; n = 3)

2. Storage fruit quality

Pericarp browning

Pericarp browning of longan fruit in both control and 0.5 mM ATP treated groups increased rapidly with increasing storage time and reached the unacceptable threshold, browning index > 3, on Day 2. However, treatment with 1 or 2 mM ATP hindered browning of the pericarp. Browning index increased slowly and reached the unacceptable level on Day 5 (Figure 4.4 and Appendix: Table 4).

Color of pericarp

L* value, representing the color of pericarp, of the control fruit, decreased rapidly with increasing storage time. Dipping with 1 and 2 mM ATP delayed the decrease in L* value compared to the control (Figure 4.5 and Appendix: Table 5).

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Figure 4.4 Effects of exogenous ATP on browning index of 'Daw' longan pericarp during storage at 25±1°C (BD, before dipping; AD, after dipping; each value is



Figure 4.5 Effects of exogenous ATP on L* value of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BD, before dipping; AD, after dipping; each value is presented as mean \pm standard deviation; n = 3)

Fruit disease

No disease symptoms were observed during the first 4 days of storage in any of the treatments. Subsequently, the symptom could be increasingly apparent, reaching the maximum level of 2.6 on Day 7. The effects of delaying disease development were more pronounced with prior treatment with 1 and 2 mM ATP (Figure 4.6 and Appendix: Table 6).



Figure 4.6 Effects of exogenous ATP on disease index of 'Daw' longan during storage at 25±1°C (BD, before dipping; AD, after dipping; each value is presented as mean ±

standard deviation; n =

Overall consumer acceptance by Chiang Mai University

The overall consumer acceptance of untreated control fruit decreased rapidly and became unacceptable on Day 3 of storage with an acceptance score below 5. ATP treatment delayed the decrease in the consumer acceptance score comparing to the control fruit. Treatments with 1 and 2 mM ATP had the highest acceptance scores above the acceptable limit for 4 days, whereas it was only 2 days for 0.5 mM ATP (Figure 4.7 and Appendix: Table 7).



Figure 4.7 Effects of exogenous ATP on overall quality acceptance of 'Daw' longan during storage at $25\pm1^{\circ}$ C (BD, before dipping; AD, after dipping; each value is presented as mean \pm standard deviation; n = 3)

3. Pearson correlation analysis

Correlation analysis during storage of longan fruit cv. Daw at $25\pm1^{\circ}$ C showed that ATP concentration used, antioxidant capacity and overall quality acceptance (QA) were positively and significantly (p < 0.01) correlated as shown in Table 4.1.

Table 4.1 Pearson correlation coefficients of ATP concentration used, antioxidant capacity and fruit quality of 'Daw' longan during storage at 25±1°C

Trait	<i>r</i> value					
	[ATP]	ABTS	DPPH	FRAP	QA	BI
[ATP]	AII	rig	hts	res	erv	e d
ABTS	0.719*	1				
DPPH	0.823**	0.949**	1			
FRAP	0.793**	0.916**	0.914**	1		
QA	0.816**	0.978**	0.982**	0.916**	1	
BI	-0.634*	-0.828**	-0.876**	-0.926**	-0.900**	1

*Significant at p < 0.05; **Significant at p < 0.01

[ATP], ATP concentration used; QA, overall quality acceptance; BI, browning index

1.2 Effects of ClO₂ on mitochondrial energy levels and redox status of 'Daw' longan pericarp during storage

Changes in energy status, respiratory enzyme activities, redox status in energy production and storage fruit quality of longan cv. Daw during storage at $25\pm1^{\circ}$ C and the effects of ClO₂ fumigation on these parameters were observed.

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1. Energy status

ATP content

Changes in ATP content of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C are shown in Figure 4.8 (Appendix: Table 8). The ATP content of control fruit decreased rapidly within the first day and continued so with time. The ATP level decreased by 66% during storage. Upon exposure to the ClO₂, ATP content increased, however, the rate and the extent of which varied with the concentration used in the fumigation. At the lowest concentration (5 mg L⁻¹), ATP content was moderately higher than that of the control and started to decrease after 1 day of storage. On the other hand, ATP content of the fruits surged immediately after fumigation with higher concentrations (10 and 25 mg L⁻¹) and continued to rise at a lesser rate to the highest level on Day 1. No significant difference was seen between the 10 and 25 mg L⁻¹ ClO₂ treatments, showing 10-88% and 10-87%, respectively, higher than that of the control (Figure 4.8 and Appendix: Table 8).

ADP content

Throughout the storage, ADP content of all groups decreased slowly to 71%

(from 18.56 to 5.11 μ g g⁻¹ FW). However, no significant differences in ADP content were found between the experimental groups (Figure 4.9 and Appendix: Table 9).



Figure 4.8 Effects of ClO₂ fumigation on ATP content of 'Daw' longan pericarp during storage at 25±1°C (BF, before fumigation; AF, after fumigation; each value is presented



Figure 4.9 Effects of ClO₂ fumigation on ADP content of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented as mean \pm standard deviation; n = 3)

AMP content

Changes in AMP content of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C are shown in Figure 4.10 (Appendix: Table 10). The AMP content of the control fruit increased gradually throughout storage and reached 252% on Day 7. The AMP content in ClO₂- fumigated fruit increased gradually throughout the storage time. However, the onset of the change depended upon whether the fruits were fumigated. A significantly lower level of AMP in comparison with that of the control was found immediately after ClO₂ treatment. The AMP content continued to be lower until Day 1 after which it rose steadily until the end of the experiment. No significant difference in the AMP content was found between each concentration (Figure 4.10 and Appendix: Table 10).



Figure 4.10 Effects of ClO₂ fumigation on AMP content of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented as mean ± standard deviation; n = 3)

Energy charge

EC of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C is shown in Figure 4.11 (Appendix: Table 11). Expectedly, EC changes during the storage followed the same trend as that of ATP content, decreased gradually with the storage time. ClO₂ treatment

at any concentrations increased EC immediately after fumigation and reached their respective highest EC value on Day 1 and decreased gradually thereafter. No significant difference in EC between the 10 and 25 mg L^{-1} treatments, showing 18-65% and 15-63%, respectively, higher than the control during storage (Figure 4.11 and Appendix: Table 11).



Figure 4.11 Effects of ClO₂ fumigation on energy charge of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented as mean ± standard deviation; n = 3)

2. Respiratory enzyme activities

Figure 4.12 and Appendix: Table 12 represents changes in SDH activity of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C. SDH activity of the control fruit declined rapidly within the first day of storage and continued gradually thereafter. SDH activity in ClO₂-treated fruits increased during the same period as the control. In addition, the activity was significantly higher until Day 3 (5 mg L⁻¹) and Day 4 (10 and 25 mg L⁻¹). Treatment with 10 mg L⁻¹ ClO₂ yielded maximum induction of SDH activity approximately 53-247% higher than the control during storage.

CCO activity

The changes in CCO activity differed from that of SDH. The activity of the control increased until reaching the highest value on Day 1, then decreased gradually thereafter. In contrast, CCO activity surged immediately to its respective highest level after fumigation, at all ClO₂ concentrations. Treatments with 10 and 25 mg L⁻¹ ClO₂ resulted in the highest increase in CCO activity (123 and 117%, respectively) during the storage. There was no significant difference in CCO activity between 10 and 25 mg L⁻¹ ClO₂, showing 40-123% and 44-117%, respectively higher than the control during storage. Interestingly, the rate of reduction in CCO activity is comparable regardless of whether or not the fruits were fumigated (Figure 4.13 and Appendix: Table 13).



Figure 4.12 Effects of ClO₂ fumigation on SDH activity of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented as mean ± standard deviation; n = 3)



Figure 4.13 Effects of ClO₂ fumigation on CCO activity of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented as mean ± standard deviation; n = 3)

3. Redox status in energy production

3.1 Pyridine nucleotide content and its redox state

NAD⁺ content

Changes in NAD⁺ content of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C are shown in Figure 4.14 (Appendix: Table 14). The NAD⁺ content of control fruits gradually raised and reached the level of 33% higher at the end of the storage. In all ClO₂ treatments, the NAD⁺ content climbed sharply until the fourth day, then, fell significantly reaching the level of 66% higher on the last day of the experiment. At its peak, the NAD⁺ content was 250% higher when fruits were fumigated with 10 and 25 mg L⁻¹ ClO₂ (Figure 4.14 and Appendix: Table 14).

NADH content

In the control, the NADH content increased gradually until it reached the highest level on Day 4, and dropped slightly. The changes were more pronounced in those fumigated with ClO₂. Moreover, higher concentrations of ClO₂ yielded higher NADH contents during storage (Figure 4.15 and Appendix: Table 15).

NAD⁺/NADH ratio

Change in NAD redox level as shown by NAD⁺/NADH ratio of 'Daw' longan pericarp during storage is shown in Figure 4.16 (Appendix: Table 16). The NAD⁺/NADH ratio of the control decreased only slightly and remained in a more reduced state throughout the time. ClO_2 treatments caused the ratio to increase immediately after fumigation. It reached its highest level on Day 4 (10 and 25 mg L⁻¹) and Day 5 (5 mg L⁻¹) before falling down to about 1.0 at the end.



Figure 4.14 Effects of ClO₂ fumigation on NAD⁺ content of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented as mean ± standard deviation; n = 3)



Figure 4.15 Effects of ClO_2 fumigation on NADH content of 'Daw' longan pericarp during storage at 25±1°C (BF, before fumigation; AF, after fumigation; each value is

presented as mean \pm standard deviation; n = 3)



Figure 4.16 Effects of ClO₂ fumigation on NAD⁺/NADH ratio of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented as mean ± standard deviation; n = 3)

3.2 Ubiquinone content and its redox state

Q content

The Q content of the untreated control showed little fluctuation throughout the experiment. In contrast, ClO_2 treatments resulted in continual increase in Q content. Treatments with 10 and 25 mg L⁻¹ were more effective. The final Q concentration was almost 3 times higher than the untreated control (Figure 4.17 and Appendix: Table 17).

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QH₂ content

The QH_2 content of the control group increased about 1 fold within 1 day of storage and decreased gradually until Day 3. The level dropped sharply within 1 day before reaching plateau. CIO_2 treatments caused a rapid increase in the QH_2 content. The level stayed significantly higher than that of the control during storage. It was interesting to note that immediately upon reaching the highest respective level, the QH_2 content fell suddenly to the starting level (Figure 4.18 and Appendix: Table 18).

Q/QH₂ ratio

In all experiments, the Q/QH_2 ratio was comparable during the first day. The control fruit exhibited slight decrease before dramatically rose on Day 3 to the level comparable with those treated with 5 mg L⁻¹ ClO₂ at the end of the storage. In the treatments with 10 and 25 mg L⁻¹ ClO₂, significantly higher Q/QH_2 ratios were found especially after the third day of storage (Figure 4.19 and Appendix: Table 19).

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Figure 4.17 Effects of ClO₂ fumigation on Q content of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented



Figure 4.18 Effects of ClO₂ fumigation on QH₂ content of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented as mean \pm standard deviation; n = 3)



Figure 4.19 Effects of ClO₂ fumigation on Q/QH₂ ratio of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented as mean ± standard deviation; n = 3)

3. Storage fruit quality

Pericarp browning

Change in pericarp browning as shown by browning index of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C is shown in Figure 4.20 (Appendix: Table 20). Browning occurred with increasing storage time and became unacceptable on Day 2 of storage in the control group. ClO₂ treatments delayed this further 2 (5 mg L⁻¹) or 4 days (10 and 25 mg L⁻¹). Changes in the browning index were not dissimilar when higher ClO₂ concentrations were used (Figure 4.20 and Appendix: Table 20).

Color of pericarp

L* value of the control gradually decreased during storage. ClO₂ fumigation initially and increased L* value significantly. The effects were prolonged until Day 3. Pericarp color darkened continuously thereafter (Figure 4.21 and Appendix: Table 21).

Fruit disease

No diseases developed during the first 4 days of storage for the control fruit. Subsequently, the disease symptom increased with increasing storage time. CIO_2 fumigation delayed the onset of disease development one day longer and caused the symptom to be less observable (Figure 4.22 and Appendix: Table 22).

Overall consumer acceptance

The overall consumer acceptance of untreated control fruit rapidly decreased and became unacceptable on Day 3 of storage with the acceptance score below 5. ClO_2 fumigation at the concentrations of 5 and 10 mg L⁻¹ delayed the decrease in the consumer acceptance score with the latter being the most effective. The acceptance score of fruits fumigated with 25 mg L⁻¹ ClO₂ was surprisingly lowest reaching the unacceptable level immediately after treatment (Figure 4.23 and Appendix: Table 23).



Figure 4.20 Effects of ClO₂ fumigation on browning index of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented as mean ± standard deviation; n = 3)



Figure 4.21 Effects of ClO₂ fumigation on L* value of 'Daw' longan pericarp during storage at 25±1°C (BF, before fumigation; AF, after fumigation; each value is presented



Figure 4.22 Effects of ClO₂ fumigation on disease index of 'Daw' longan during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented as mean \pm standard deviation; n = 3)



Figure 4.23 Effects of ClO₂ fumigation on overall quality acceptance of 'Daw' longan during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented as mean \pm standard deviation; n = 3)

Experiment 2 Effects of gaseous ClO₂ fumigation on redox status in free radical scavenging of harvested 'Daw' longan pericarp during storage

The effects of ClO₂ fumigation on changes in redox status, ASA-GSH cycle enzyme activities, NADPH regenerating enzyme activities and H₂O₂ content of longan cv. Daw during storage were studied.

Redox status in free radical scavenging
ASA and DHA contents and its redox couple

ASA content

ASA content of the control fruit decreased about 60% during storage. However, in fruit exposed to ClO_2 , the content immediately increased to maximum before declining gradually. The changes were more pronounced in those fumigated with higher ClO_2 concentrations. No significant difference was seen between treatments with 10 and 25 mg L⁻¹ ClO₂, showing 13-37% and 15-47%, respectively, higher than the control

during storage. The rates of which ASA decreased were comparable regardless of whether the fruit was exposed to ClO_2 (Figure 4.24 and Appendix: Table 24).

DHA content

As shown in Figure 4.25 (Appendix: Table 25), the changes in the DHA content were a mirror image of the ASA content. In the control experiment, the DHA content immediately but gradually increased to about 10% higher at the end of the experiment while that of the CIO_2 treated groups decreased abruptly before increasing gradually. No significant difference was seen between the 10 and 25 mg L⁻¹ CIO₂ treatments, showing 3-7% and 5-9%, respectively, lower than the control during storage (Figure 4.25 and Appendix: Table 25).

ASA/DHA ratio

As expected, ASA/DHA ratio of the control fruit decreased during storage. CIO_2 treatments only affected the ASA/DHA ratio right after fumigation. The ratio increased significantly (about 20 to 30%) compared to that of the control and this effect was concentration dependent. Statistical analysis of the data revealed no differences in its redox couple ratio between the 10 and 25 mg L⁻¹ ClO₂ treatments. After initial increase, the ratio declined gradually to about 50 to 70% of the original level (Figure 4.26 and Appendix: Table 26).

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Figure 4.24 Effects of ClO_2 fumigation on ASA content of 'Daw' longan pericarp during storage at 25±1°C (BF, before fumigation; AF, after fumigation; each value is



Figure 4.25 Effects of ClO₂ fumigation on DHA content of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented as mean ± standard deviation; n = 3)



Figure 4.26 Effects of ClO₂ fumigation on ASA/DHA ratio of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented as mean ± standard deviation; n = 3)

1.2 GSH and GSSG contents and its redox couple

GSH content

In the control, GSH content was found to decrease rapidly within 1 day before decreasing gradually thereafter. ClO_2 fumigation briefly but significantly increased GSH content and the effects were concentration dependent. Fumigation with either 10 or 25 mg L⁻¹ ClO₂ resulted in higher levels of GSH content (43-314% and 60-327%, respectively) during 7 days of storage. The decline of GSH started immediately afterward, however, the rate of decline was not unlike that of the control (Figure 4.27 and Appendix: Table 27).

GSSG content

The GSSG content of both control and treated fruit decreased continuously for 3 to 4 days before rising thereafter. In the control group, the content returned to the original level while those of the treated fruit only increased to about 90% of original.

The fruit treated with 10 and 25 mg L^{-1} ClO₂ had the lowest GSSG content on Day 4 before increasing sharply thereafter (Figure 4.28 and Appendix: Table 28).

GSH/GSSG ratio

Alteration of the GSH/GSSG ratio during storage followed the same trend as that of GSH content. ClO₂ treatment at any concentrations significantly increased GSH/GSSG ratio immediately after fumigation and gradually decreased thereafter. At 25 mg L⁻¹of ClO₂, the highest GSH/GSSG ratio was observed on Day 1 after fumigation while the lower concentrations (5 and 10 mg L⁻¹) reached the highest ratio instantly after fumigation (Figure 4.29 and Appendix: Table 29).



Figure 4.27 Effects of ClO₂ fumigation on GSH content of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented as mean ± standard deviation; n = 3)



Figure 4.28 Effects of ClO₂ fumigation on GSSG content of 'Daw' longan pericarp during storage at 25±1°C (BF, before fumigation; AF, after fumigation; each value is

presented as mean \pm standard deviation; n = 3)



Figure 4.29 Effects of ClO₂ fumigation on GSH/GSSG ratio of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented as mean ± standard deviation; n = 3)

1.3 NADPH and NADP contents and its redox couple

NADPH content

NADPH content of both control and ClO₂ treated fruit were found to decrease slightly over time. Moreover, no significant different was seen between all treatments (Figure 4.30 and Appendix: Table 30).

NADP⁺ content

While NADPH content of both control and ClO₂ treated fruit decreased only slightly and no significant difference was observed between the groups. The effects of ClO₂ treatment on NADP⁺ content differed statistically from the control. In the control, NADP⁺ content rose more rapidly and reached almost 2 times of the original level at the end of the experiment. ClO₂ treatment at any concentrations caused the content to rise slowly. The fruit treated with 10 and 25 mg L⁻¹ ClO₂ had the lowest NADP⁺ content of 5-23% and 6-24%, respectively, lower than the control during storage (Figure 4.31 and Appendix: Table 31).

NADPH/NADP⁺ ratio

Since only changes were observed in NADP⁺ content, the NADPH/NADP⁺ ratio were under the influence of these changes. The NADPH/NADP⁺ ratio of the control fruit markedly dropped within 1 day after fumigation and decreased continuously during storage. ClO_2 caused slight increase in the ratio immediately after treatment but dropped steadily at the same rate of the control fruit afterward. The fruit treated with 10 and 25 mg L⁻¹ ClO₂ were most effective and had the highest NADPH/NADP⁺ ratio during storage, showing 10-31% and 14-35%, respectively, higher than the control. At the end of the experiment, the ratio was 50 to 60% lower than the initial level (Figure 4.32 and Appendix: Table 32).



Figure 4.30 Effects of ClO₂ fumigation on NADPH content of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is



Figure 4.31 Effects of ClO₂ fumigation on NADP⁺ content of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented as mean ± standard deviation; n = 3)



Figure 4.32 Effects of ClO₂ fumigation on NADPH/NADP⁺ ratio of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented as mean ± standard deviation; n = 3)

2. ASA-GSH cycle enzyme activities

APX activity

APX activity of the control fruit only increased slightly during the first 2 days of the experiment before decreased sharply thereafter. Change in APX activity in the fruits fumigated with ClO₂ exhibited a similar pattern to the control but at higher levels. ClO₂ fumigation significantly increased APX activity and reached the highest activity on Day 2, before dropping sharply. The 10 and 25 mg L⁻¹ treatments with ClO₂ was the most effective in promoting APX activity approximately 4-62% and 7-79%, respectively, higher than the control during storage (Figure 4.33 and Appendix: Table 33).

DHAR activity

DHAR activity of the control fruit declined during the storage but the rate of which varied with time. The decrease was gradual during the first 2 days and the last 4 days while at Day 2, the activity changed abruptly. This abrupt change was also seen in ClO₂-treated samples. ClO₂ fumigation caused significant increase in DHAR activity.

The higher concentrations were the most effective. This enhancement effects could be seen until Day 2. The 10 and 25 mg L⁻¹ treatments with ClO_2 was most effective in promoting DHAR activity approximately 9-47% and 14-75%, respectively, higher than the control during storage (Figure 4.34 and Appendix: Table 34).

MDHAR activity

As shown in Figure 4.35 and Appendix: Table 35, MDHAR activity of all experiments increased rapidly on Day 1, and gradually decreased thereafter. All CIO_2 treated fruit significantly increased MDHAR activity and maintained significantly higher activity throughout storage time. The 10 and 25 mg L⁻¹ treatments with CIO_2 was most effective in promoting MDHAR activity approximately 11-66% and 21-85%, respectively, higher than the control during storage (Figure 4.35 and Appendix: Table 35).

GR activity

Change in GR activity of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C is shown in Figure 4.36 (Appendix: Table 36). In the control fruit, GR activity gradually decreased during 7 days of storage. ClO₂ fumigation increased the activity immediately after fumigation. The effects were most pronounced (up to 3 folds) in 10 and 25 mg L⁻¹ ClO₂ treatment and lasted only 1 day after which the activity declined gradually. However, the 10 and 25 mg L⁻¹ treatments with ClO₂ was most effective in promoting GR activity approximately 109-239% and 105-264%, respectively, higher than the control during storage (Figure 4.36 and Appendix: Table 36).

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Figure 4.33 Effects of ClO₂ fumigation on APX activity of 'Daw' longan pericarp during storage at 25±1°C (BF, before fumigation; AF, after fumigation; each value is



Figure 4.34 Effects of ClO₂ fumigation on DHAR activity of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented as mean ± standard deviation; n = 3)



Figure 4.35 Effects of ClO₂ fumigation on MDHAR activity of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented as mean ± standard deviation; n = 3)



Figure 4.36 Effects of ClO₂ fumigation on GR activity of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented as mean \pm standard deviation; n = 3)

3. NADPH regenerating enzyme activities

G6PDH activity

G6PDH activity of the control fruit decreased gradually during the storage. Fumigation with 5-25 mg L⁻¹ ClO₂, however, increased and maintained higher G6PDH activity for 2 days before gradually declined for the rest of the storage time. Treatment using either 10 or 25 mg L⁻¹ ClO₂ was the most effective yielding about 11-27% and 15-29%, respectively, higher than the control during storage (Figure 4.37 and Appendix: Table 37).

6PGDH activity

6PGDH activity declined slowly throughout the experiment. The changes in the activity of 6PGDH of both the control fruit and 5 mg L⁻¹ ClO₂ treatment were comparable. In contrast, the higher concentrations (10 and 25 mg L⁻¹) of ClO₂ increased 6PGDH to the maximum in Day 1 and maintained higher G6PDH activity during the gradual decline afterward, showing 15-43% and 18-48%, respectively, higher than the control during storage (Figure 4.38 and Appendix: Table 38).



Figure 4.37 Effects of ClO₂ fumigation on G6PDH activity of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented as mean ± standard deviation; n = 3)



Figure 4.38 Effects of ClO₂ fumigation on 6PGDH activity of 'Daw' longan pericarp during storage at $25\pm1^{\circ}$ C (BF, before fumigation; AF, after fumigation; each value is presented as mean ± standard deviation; n = 3)

4. H₂O₂ content

 H_2O_2 content of the control fruit increased rapidly and continuously before reaching 4-fold increase on Day 7. ClO₂ fumigation at the concentration of 5 mg L⁻¹ could not significantly reduce or delay the increase in H_2O_2 content. In contrast, a delay of 2 days could be seen if the fruit was fumigated with higher concentrations of ClO₂. The content reduced approximately 19-68% and 21-74% in the treatment with 10 and 25 mg L⁻¹, respectively, lower than control during storage. Interestingly, the rate at which H_2O_2 content increased was comparable in all experimental scenarios (Figure 4.39 and Appendix: Table 39).



Figure 4.39 Effects of ClO₂ fumigation on H_2O_2 content of 'Daw' longan pericarp during storage at 25±1°C (BF, before fumigation; AF, after fumigation; each value is presented as mean ± standard deviation; n = 3)

