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Effect of ozone micro-nano bubbles on longan shelf life extension

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ABSTRACT

This study aimed to evaluate the effects of micro/nano ozone bubbles (O_3 MNBs) water on extending longan shelf life. The experiment was assigned using a completely random design (CRD), which consisted of 4 treatments, including soaking with 1) distilled water for 5 minutes (control), 2) 10 minutes of ozone micro nanobubbles (O_3 MNBs) water, 3) 20 minutes of O_3 MNBs water, and 4) 30 minutes of O_3 MNBs water. All treatments contained four replications. Then, they were stored at 5°C with 80% relative humidity for fifteen days for further examination. The results revealed that the control had the highest weight loss than other treatments, especially 6 to 9 days after being treated with O_3 MNBs water. Moreover, longans with the O_3 MNBs water treatments were more significant in the L* and b* color indexes than the untreated fruit on longans outside the pericarp. In addition, the control appeared to have pericarp browning after storage for nine days and found fungal on the fruit twelve days after treatment.

Keywords: ozone, micro/nanobubbles, longan

INTRODUCTION

Longan (*Dimocarpus longan* L.) is a commercial fruit commonly grown in Thailand, the largest exporter of longan in the world. Longan can be induced flowering in the off-season and has a year-round supply, contributing roughly 141.42 million USD in 2022. The production area in the year 2022 was 279,845.92 hectares, which yielded approximately 1,567,087.36 metric tons (Sritontip et al., 2005; OAE, 2022). Although production export levels have increased, longan has a concise shelf life, deteriorating easily and quickly within 3 to 4 days at room temperature (Jiang et al., 2002). The short shelf life of fresh longan fruit is a common issue. In deteriorated longan, fruit shells turn from yellow-brown to dark brown into black color caused by dehydration, cooling damage, and microbes. For microbes damaged, it may result from microbial invasion before or after the harvesting process (Pan, 1994). It is not easy to store longan in good condition even for short periods. The main crucial postharvest losses are microbes, fungus spoilage, and pericarp browning of longan. The pericarp browning reaction can be associated with dehydration, heat stress, senescence, chilling injury, or disease of the longan fruit (Apai, 2010). At present, sulfur dioxide (SO_2) fumigation is a commercially accepted method to solve the problems of postharvest losses in longan, but it risks consumer health and safety concerns. (Drinnan, 2004; Sevilai et al., 2020). For this reason, there is a need to develop effective methods to replace the protocol using SO_2 fumigation to be less harmful to humans and the environment, such as using ozone and Micro-nanobubbles (MNBs) technology as an alternative method.

In Thailand, MNBs are a novel technology. The MNBs produce little gas bubbles between 50 micrometers and 200 nanometers in diameter, raising the gas or oxygen concentration in water. Moreover, MNBs can help as a filter for wastewater to eliminate pesticide and microbe residues from crops and promote seed germination and vegetative growth of plants (Oshita and Liu, 2013; Sritontip et al., 2019). Due to their extended stay in aqueous solutions and huge specific surface area, MNBs are used extensively in environmental engineering, environmental remediation, agriculture, agronomy, horticulture, aquaculture, and hydroponics. Reactive oxygen species (ROS) produced by MNB can also purify wastewater, eliminate persistent organic contaminants from food, and inactivate infections in water (Marcelino et al., 2022; Seridou and Kalogerakis, 2021; Zhang et al., 2022).

Ozone (O_3) is triatomic oxygen and is widely used to control microorganism growth (bacteria, fungi, viruses, and protozoa) and chemical

residue in various food industries as well as in the exportation of vegetable and fruit produce (Kim et al., 1999). Aslam et al. (2021) report that aqueous ozone treatment was effective in reducing the microbial population, maintaining quality parameters, and extending the shelf life of fresh-cut onion slices. This agrees with Chamnan et al. (2021), who reported that the longan exposed to ozone gas at 8,500 ppm for 5 min was considered a suitable treatment to extend shelf-life up to 35 days, which is 57% longer than the shelf-life of non-ozonated longan. Yang and Chen (2022) report that the preservation technology of ozone micro-nano bubble treatment has improved the preservation of fruits and vegetables by 12%. Therefore, this study aimed to investigate the effect of ozone micro-nanobubble applications on reducing pericarp browning and disease incidence to extend the shelf-life of longan and sulfur residue reduction.

MATERIALS AND METHODS

In this study, non-infected, mature fresh longan fruit of the Daw variety from the off-season was employed. The fruit poles of longans were chopped to a maximum length of 0.5 cm after they were chosen for their identical size and skin tone. The skins of the fruits were cleaned with 200 mg/L of sodium hypochlorite (NaClO) solution. Then, longan fruits were soaked in the NaClO solution for two minutes and dried at room temperature before further processes. The experiment was assigned using a completely randomized design. It included four treatments with four replications, i.e., 1) soaking longan fruits in distilled water for 5 minutes or control, 2) soaking longan fruits in O_3 MNBs water for 10 minutes, 3) soaking longan fruits in O_3 MNBs water for 20 minutes, and 4) soaking longan fruits in O_3 MNBs water for 30 minutes. There were 50 mature logan fruits in each replication.

The MNBs generator model KVM-25 used in this study was modified by the Faculty of Engineering, Rajamangala University of Technology Lanna, Thailand, to produce O_3 MNBs water for the experiment. The O_3 MNBs water had an oxidation-reduce potential (ORP) level at 600 mV. The total bubble distribution was 2.7388 x 10^{11}/mL, with a median size of 38.67 nm, mode size of 62 nm, and average size of 66.43 nm, measured by Horiba-960A laser scattering particle size distribution analyzer®. This could support a water flow rate of 25 L/minute, an airflow rate of 2 L/minute, an operating pressure of 0.25–0.4 MPa, and a 0.75 KW pump (Figure 1).

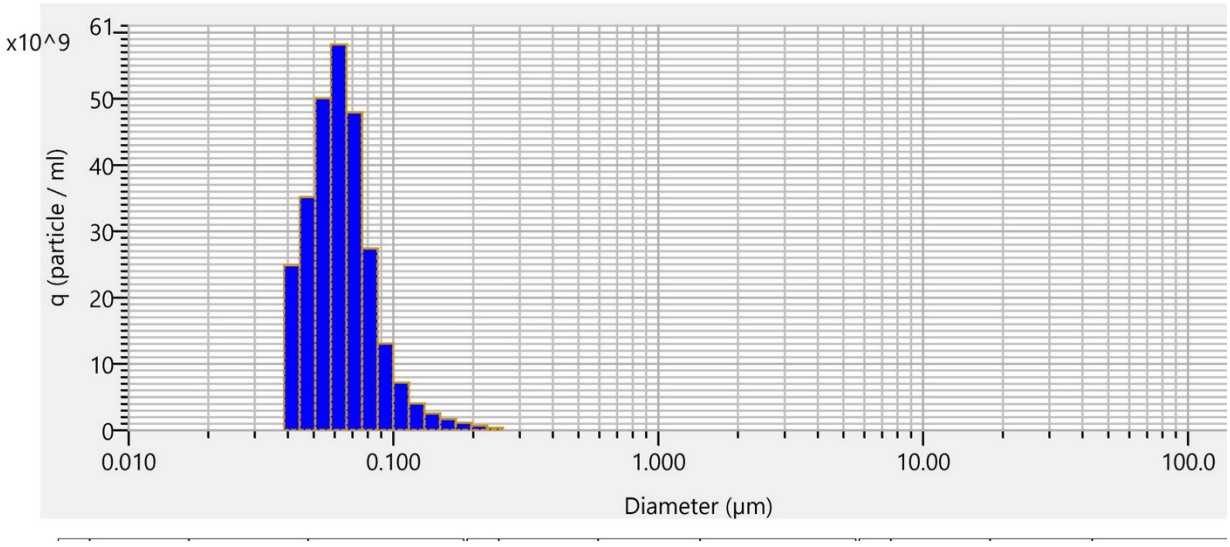

Figure 1. The analysis of the diameter of micro-nano bubbles produced by KVM-25 detected by Horiba-960A.

Longan fruit samples were soaked in according to treatment protocols simultaneously, then packed into a transparent box of perforated fruits and stored in the refrigerator at 5°C temperature and 80% relative humidity. Data were recorded every three days at a time for 15 days, or the longan fruits were counted until a fungal growth covering more than 25% of the longan fruit's surface was discovered or the level of change in pericarp browning was more than 50 %. Weight loss in each treatment was recorded every three days from the first day until the end of the experiment using an electrical balance (Ohaus, model PX3202, Maximum 3,200 g, USA). The formula for determining the percentage of water loss was:

Percentage of weight loss = (A – B)/A x 100

Where: A = sample weight on the first day of experimentation (g)
B = Sample weight, date of measurement (g).

The firmness of the fruit pulp was measured using the force gauge (RS232 Output Model 840060). The longan peel was removed, and the firmness of the fruit pulp was measured by pressing down at the center of the longan's fruit pulp. Skin color changes of the outer shell and fruit pulp were measured using the 3NH brand machine model NR200. The values are expressed as color brightness (L*), green (a*), and yellow (b*). The percentage of disease and total soluble solids using a hand refractometer were also recorded. The data were analyzed for variance, and the average was compared with Duncan's New Multiple Range Test (DMRT) at a significant level of 0.05.

RESULTS AND DISCUSSION

Weight loss percentage

Longan samples lost weight throughout the retention period, which lasted 15 days after the treatment. The highest weight loss percentage was found in the distilled water (control) treatment 6–9 days after treatment (Figure 2).

Fruit firmness

The changes in the fruit firmness tester showed a non-statistical difference among treatments after being treated with distilled water and O_3MNBs (Figure 3).

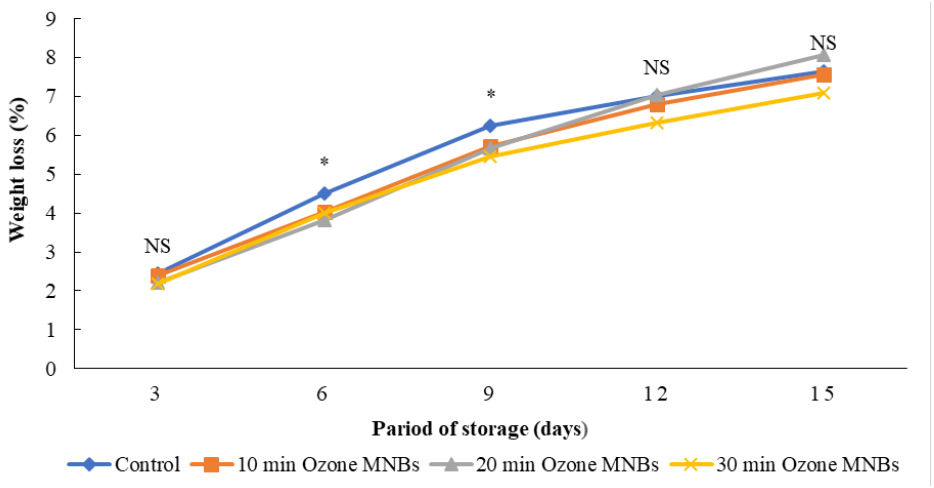

Figure 2. Weight loss of longan fruit after being treated and storage
NS = non-significant and * = significant differences at P < 0.05.

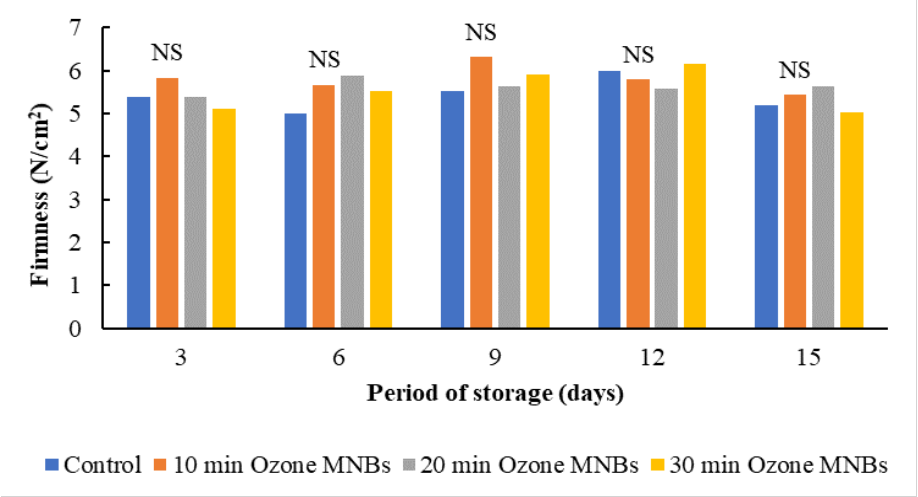

Figure 3. Firmness of longan fruit after being treated and storage
NS = non-significant.

Changes in the color of the outside of the pericarp and fruit pulp

The brightness value (L*) of the control had been reduced on days 3, 6, and 15 following the storage period, according to alterations in the color values of the outside of the pericarp. Moreover, the a* value in the control treatment was greater than in the O_3 MNBs, although the b* value suggested a lower value in the control treatment (Table 1).

After storage at a cool temperature, changes in the color value of the pulp revealed that the brightness value (L*) and b* values did not significantly differ. In contrast, the a* value changed unforeseen (Table 2).

Table 1. Effects of O_3 MNBs on the L*, a*, and b* values of the outer shell in longan fruits

	Treatments	Period of storage (days)					
		0	3	6	9	12	15
L*	Control	55.98	48.10b	48.01	46.10^{b}	44.25	39.93^{c}
	10 min O_3 MNBs	55.94	51.37a	50.71	48.05^{ab}	44.55	45.27b
	20 min O_3 MNBs	55.54	51.59a	49.53	47.96^{ab}	45.92	49.76^{a}
	30 min O_3 MNBs	55.98	49.96a	49.14	49.37^{a}	46.76	47.88^{ab}
	F-test	NS	**	NS	*	NS	**
a*	Control	11.07	11.64	11.48	11.55	11.86	13.03^{a}
	10 min O_3 MNBs	11.30	11.18	11.44	11.76	11.56	12.10^{ab}
	20 min O_3 MNBs	10.99	10.61	10.35	11.68	11.46	11.29^{b}
	30 min O_3 MNBs	11.07	10.74	11.34	11.31	11.55	11.85^{b}
	F-test	NS	NS	NS	NS	NS	**
b*	Control	24.93	26.59	28.70	24.37^{c}	22.98^{b}	21.43^{b}
	10 min O_3 MNBs	24.93	26.49	29.42	27.30^{b}	25.61^{a}	25.71^{a}
	20 min O_3 MNBs	24.96	26.20	28.74	28.64^{ab}	26.13^{a}	28.43^{a}
	30 min O_3 MNBs	24.93	25.45	28.46	29.94^{a}	24.11^{ab}	27.12^{a}
	F-test	NS	NS	NS	**	*	**

*Means within the column followed by the same superscript were not significantly different at P > 0.05 by DMRT,
NS = non-significant, * = significant differences at P < 0.05, ** = significant differences at P < 0.01 according to the DMRT test.

Table 2. Effects of O_3 MNBs on the L*, a*, and b* values of the pulp in longan fruits

	Treatments	Period of storage (days)					
		0	3	6	9	12	15
L*	Control	31.36	31.22	30.77	36.98	33.04	32.82
	10 min O_3 MNBs	30.54	32.54	31.89	37.42	33.63	34.11
	20 min O_3 MNBs	30.86	32.23	31.26	35.84	33.05	33.39
	30 min O_3 MNBs	31.36	32.40	31.80	37.78	32.99	33.37
	f-test	NS	NS	NS	NS	NS	NS
a*	Control	0.04	0.03	–0.01	0.02	0.16^{b}	0.02^{b}
	10 min O_3 MNBs	0.01	–0.09	–0.04	–0.01	0.18^{b}	0.06^{b}
	20 min O_3 MNBs	0.08	–0.17	–0.08	0.06	0.00^{b}	0.34^{a}
	30 min O_3 MNBs	0.04	–0.10	–0.03	0.15	0.62^{a}	0.07^{b}
	f-test	NS	NS	NS	NS	*	**
b*	Control	–0.95	–1.03	–1.37	–0.82	–0.55	–0.65
	10 min O_3 MNBs	–1.11	–0.97	0.03	–0.44	–0.62	–0.37
	20 min O_3 MNBs	–1.03	–1.32	–0.84	–0.32	–0.63	–0.44
	30 min O_3 MNBs	–0.95	–1.59	–1.12	–0.26	–0.09	–0.52
	F-test	NS	NS	NS	NS	NS	NS

*Means within the column followed by the same superscript were not significantly different at P > 0.05 by DMRT,
NS = non-significant, * = significant differences at P < 0.05, ** = significant differences at P < 0.01 according to the DMRT test.

Pathogenesis

After low-temperature storage, disease incidence in longan fruit was risen. The fruit's surface fungus started to develop during the period of storage. Only with the control approach did symptoms begin to appear on the 12th day of the experiment. The occurrence of the two treatments was then discovered on the experiment's fifteenth day, with the control therapy having the highest incidence and using 10-minute O_3MNBs. In contrast, 20 and 30 minutes of O3MNBs were not found in the disease (Figure 4).

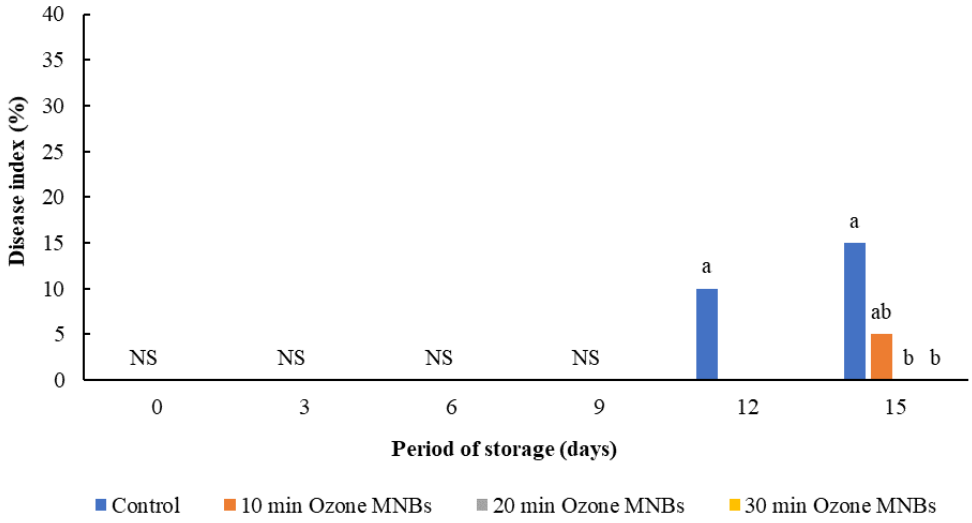

Figure 4. The effect of treatments on pathogenesis in longan fruit after storage.
NS = non-significant, * = significant differences at $P < 0.05$, according to the DMRT test.

Total soluble solids

It was found that the total amount of soluble solids in longan fruit tends to decrease gradually throughout the storage period compared to the default. However, the differences were found during the 1 5 days of the treatment. The control treatment was the greatest in total soluble solid contents (Figure 5).

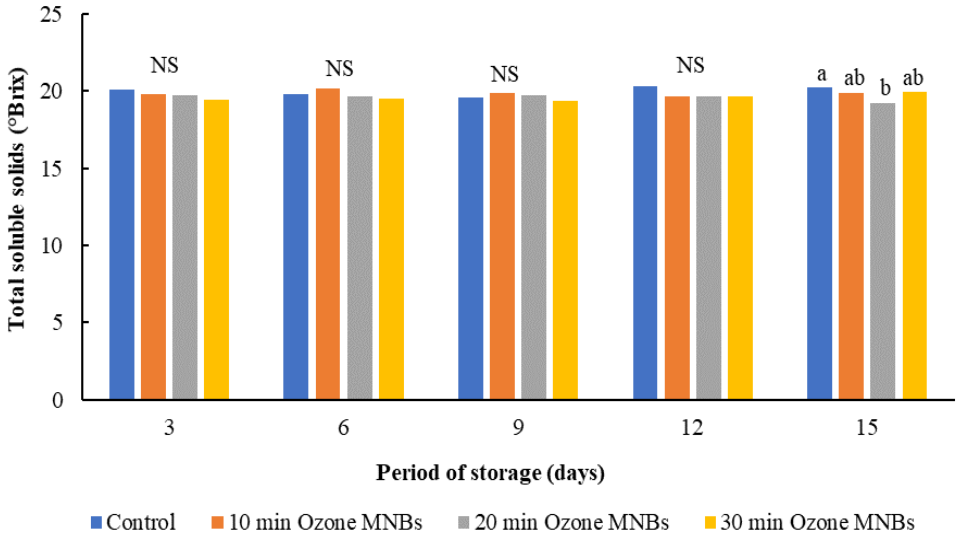

Figure 5. Effects of treatments on total soluble solid in longan fruits
NS = non-significant, * = significant differences at $P < 0.05$, ** = significant differences at $P < 0.01$ according to the DMRT test.

To increase shelf life and decrease disease, this study examined the impact of washing fresh longan fruits with O_3MNBs water on their quality. The outcome showed that longans were stored at a cool temperature for longer than the control. O_3 MNBs water has an impact on the fruit quality. The O_3 MNBs could lower the proportion of weight loss by reducing microbial infection regarding the spread of diseases. The L* and b* color indices of the longan fruit skin showed that the O_3 MNBs water treatments outperformed the untreated fruit.

A study of the effects of using ozone microbubbles (O_3 MBs) to reduce chlorpyrifos and anthracnose in sweet pepper has been reported. The sweet peppers were washed with O_3 MBs at each temperature and during each exposure period to O_3 MBs. Furthermore, O_3 MNBs affected the control of anthracnose caused by *Colletotrichum capsica* by about 96%, while the control unit was reduced by only 14.3% (Tamjapo et al., 2017). Lee et al. (2016) reported that chestnuts were washed with MNBs in combination with ozone for 10 minutes and found to reduce rot disease. This method could also extend the shelf life of chestnuts after harvesting. Moreover, the residual activity and ozone disinfection capacity could be significantly increased by O_3 MNB. When ozone ultra-fine bubbles were combined with high mechanical action in acidic electrolyzed water to wash fresh vegetables, the lowest viable bacterial count was detected, compared to other treatments, such as sodium hypochlorite. The effectiveness of ozone microbubbles in disinfecting *F. oxysporum f.* sp. melonis spores was investigated, and the results showed that they were more effective than macrobubbles. (Seridou and Kalogerakis, 2021; Ushida et al., 2017). In this study, the O_3 MNBs could extend the shelf life of logan while disinfecting microorganisms because O_3 can damage the enzymes of the DNA and RNA of microbial cells because it has two methods for suppressing bacteria. It is created by ozone molecules that oxidize compounds found in microbial cells or by unrelated substances that direct them to kill membrane cells and other internal cells in the cell. Microbial cells' cytoplasm, proteins, and lipid layers produce intracellular leakage, leading to cell disintegration. (Leowchavalit et al., 2003; Restainno et al., 1995; Victorin, K. 1992).

CONCLUSIONS

The soaking of O_3 MNBs for about 10 to 30 minutes could prolong the life of treatment and slow down the occurrence of diseases that may cause adverse effects on longan. The optimum O_3 MNBs soaking time in this experiment was 20 minutes.

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