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Pesticide Residue Reduction on Curly Chili (*Capsicum annum* L.) Using Ozone Fine Bubble Technology

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Abstract

Pesticide residues in curly chilies may cause health problems in consumers. Washing curly chilies using ozone fine-bubble water is a promising method for reducing pesticide residues. The aim of this study was to determine the optimal dose and duration to degrade pesticide residues, especially for profenofos, and to determine their effect on the shelf life and physical quality of curly chilies. After washing, the curly chilies were stored at room temperature and observed every two days. The results showed that washing with 1 ppm ozone fine bubble water for 10 min was effective in reducing profenofos residue on curly chili by up to 89.8% without reducing its quality. The shelf life of curly chilies was observed, and they started losing their commercial value after 6-8 days.

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1. Introduction

Chili are a commodity that are susceptible to pests and diseases. This is certainly the main reason chili farmers use pesticides to control pests and diseases in chili cultivation. Pesticide residues that remain on the surface of chili fruit have a negative impact on human health, causing various disease symptoms.

On the post-harvest line, pesticide residue degradation is performed in various ways; however, these methods have their own weaknesses. The use of ozone has attracted considerable interest because of its high effectiveness in the degradation of pesticide residues and sanitizing contaminants. Asgar et al., 2017, reported that soaking red chili peppers using ozone water can reduce profenofos pesticide residues between 32-52%.

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Despite its high oxidizing ability, ozone is an unstable gas. Ozone can only last for five minutes in distilled water at room temperature and is then released into air (Andoyo et al., 2018). This is dangerous, because ozone is a toxic gas that can be inhaled by the operator on duty. Various methods have been used to overcome this problem, including diffusion of ozone gas into water with fine bubbles.

Fine bubbles are bubbles with sizes less than 100 μm, which are divided into bubbles with sizes greater than 1 μm (microbubbles) and bubbles with a size below 1 μm (ultrafine bubbles) (ISO 20480-1, 2017). Fine bubbles have several characteristics, including increasing gas solubility, reducing friction, and forming free radicals (Batagoda et al., 2019).

Fine bubbles have been applied in various fields, such as wastewater treatment, improving water quality, drinking water treatment, increasing vigor and viability of seeds, and accelerating the breaking of seed dormancy (Batagoda et al., 2019; Purwanto et al., 2019). Some of the most interesting characteristics of fine bubbles are their low buoyancy and ability to form reactive oxygen species (ROS), which have high oxidative capabilities (Liu et al. 2016). Therefore, ozone gas, which has a short retention time, can stay longer in water when combined with fine bubbles and can also improve the overall oxidative ability.

Ozone microbubbles were reported to degrade 59-86% of pesticide residues on pokchoi and 57%-94% in celery (Li et al., 2023). However, Liu et al., 2020, reported an increase in the respiration rate and shrinkage in minimally treated cabbage after washing with ozone water which was influenced by the concentration of ozone used. Therefore, this study was conducted to determine the optimal concentration and duration of ozone fine bubble treatment to achieve an equilibrium between the effect of reducing pesticide residues and the quality of curly chili.

2. Materials and Methods

2.1 Time and Place of Observation

This study was conducted between June and July 2023. The washing of curly chili with ozone fine bubbles was conducted at the Techno Park Laboratory, IPB. The shelf life and quality tests were conducted at the Environmental and Biosystem Engineering Laboratory, Department of Mechanical and Biosystems Engineering, Faculty of Mechanical Engineering and Technology, IPB University. The quantitative testing of pesticide residue levels was performed at the Balai Besar Pascapanen Laboratory in Bogor, West Java.

2.2 Tools and Materials

The material used in this study was red curly chili with a harvest age of ± 90 days after planting (HST) obtained from farmers in Lembang, West Java, with a full red color. The chili washing equipment used included an oxygen generator, an ozone fine bubble generator, and a washing chamber/container. The tools used were a dissolved ozone meter, smartphone camera, colorimeter, rheometer, refractometer, and a digital scale.

2.3 Research Procedures

2.3.1 Material Preparation

Curly chilies were harvested by picking the chili stalks. The selected chilies were chilies with a complete red color at 90 days after planting with uniform size, no disease attack, and no mechanical damage due to transportation, such as broken fruit or loose stalks.

2.3.2 Curly Chili Washing

Curly chilies (1.5 kg) were placed in a chamber containing 10 liters of water. The oxygen generator and ozone fine bubble generator were then turned on (Figure 1). Ozone concentration was measured periodically using a dissolved ozone meter. When the ozone level reached the desired concentration, the ozone fine bubble generator machine was turned on and regulated so that the concentration remained fixed. The chili peppers were shaken occasionally during washing. After washing, the chilies were placed in a perforated container and aerated until completely dry.

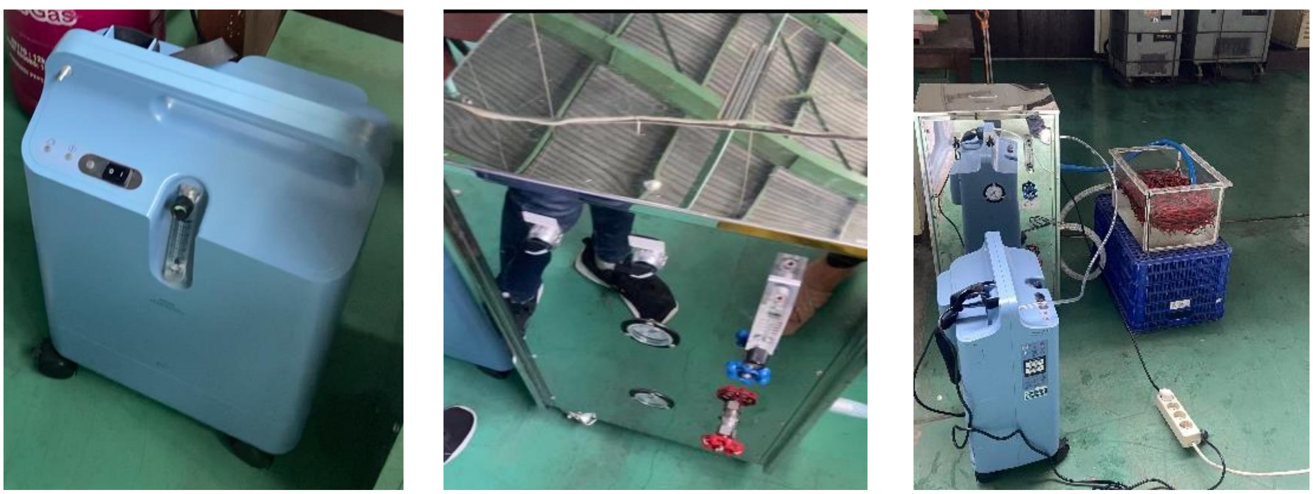

Figure 1. a) oxygen generator machine, b) ozone fine bubble generator machine, and c) chili washing process.

2.3.3 Pesticide Residue Testing

Pesticide residue testing was conducted both qualitatively and quantitatively using a test kit and gas chromatography (GC-MS). The test kit used is "K-nai instrument pesticide test kits." After testing, the sample with the best performance was selected. The selected sample was tested using GC-MS to determine the level of profenofos in chili.

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2.3.4 Storability Test

Curly chili peppers were stored at room temperature in a shaded place and were not exposed to sunlight. The storability test was conducted through visual observation, and photographs were taken every day. During storage, the chili peppers were rated on a damage scale from to 1-5, with the following description: 1, no wrinkles or damage; 2, slightly damaged, 1-15% of the chili surface wrinkled/damaged; 3, moderately damaged, 16-30%; 4, moderately damaged, 31-50%; 5, severely damaged, >50% (Lacap et al., 2020). Chili is considered commercially unfit when its condition exceeds scale 3.

2.3.5 Quality Test

Quality tests were divided into non-destructive (shelf life, weight loss, color) and destructive (hardness, total soluble solids) quality tests. The shelf life was measured using damage scale scoring (Lacap et al., 2020).

The samples used for the non-destructive quality tests were three replicates for each treatment. For the destructive quality test, three replicates were used for each treatment multiplied by the number of observation days (12 days). Each sample contained 100 g curly chili. Chili peppers were tested every other day for all tests.

2.4 Data Processing

Data processing was performed using a completely randomized design (CRD). The first factor is the ozone fine bubble concentration (C), which consists of three levels: 1, 2, and 4 ppm. The second factor was the duration of washing (T), which consisted of three levels: 3, 5, and 10 min. Furthermore, the data were analyzed using Analysis of Variance (ANOVA) using MiniTab 16.0 software to determine the effect of each treatment group. If there is a significant effect on the treatment, it will be tested further with the Duncan Multiple Range Test (DMRT) further test at a 5% significance level.

3 Results and Discussions

3.1 Shelf Life

Based on the observation results, the critical point of chili occurred on the 4th and 6th days of observation On these point, the chili has started to lose its commercial value. Figure 2 shows a photograph of one of the chili samples during the observation period, namely treatment C1T3. The best performing chili samples were those treated with C1T2, C1T3, C2T2, and the control. In these four samples, the chili peppers were given a score of 4 (moderately damaged) on day 8, while the other treatments had reached a score of 5 on the same day (Figure 2). All chili peppers treated with 4 ppm ozone experienced severe damage on day 6. Damage that occurs includes the surface of the chili skin

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and stalk wrinkling; the tip of the fruit begins to dry, the color of the skin and stalk is dull, the skin surface is dull, and there is a loose stalk.

Figure 2. Deterioration of chili sample quality at observation C1T3 a) day 0, b) day 2, c) day 4, d) day 6, e) day 8, f) day 10, g) day 12

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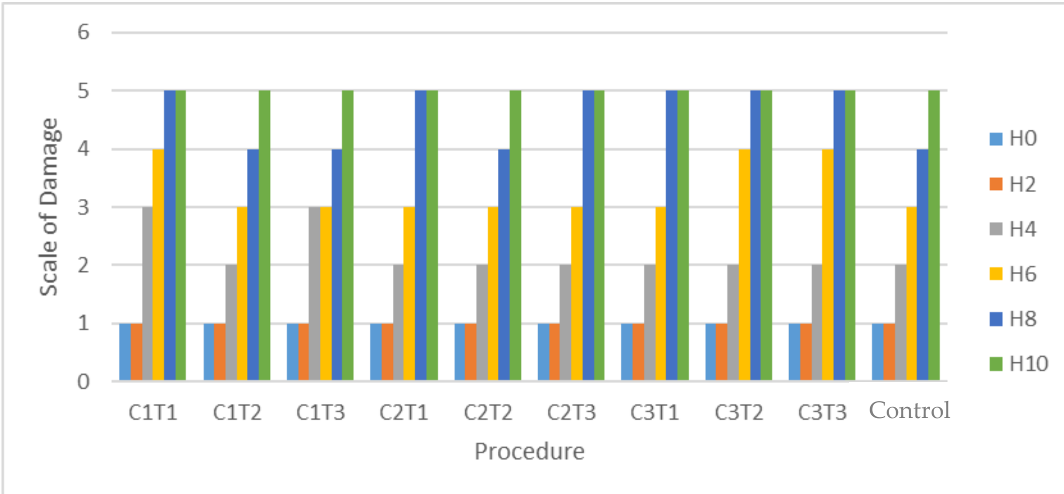

Figure 3. Damage value of chili peppers

The shorter shelf life of curly chili peppers washed with high concentrations of fine-bubble ozone water can be attributed to the fact that immersion of ingredients in ozonated water can trigger stress and cause several types of damage at the cellular level, which results in an increase in the respiration rate (Silveira et al., 2018).

3.2 Pesticide Residues

Pesticide testing began with qualitative testing using a pesticide residue testing kit. The results of the qualitative tests are shown in Figure 4.

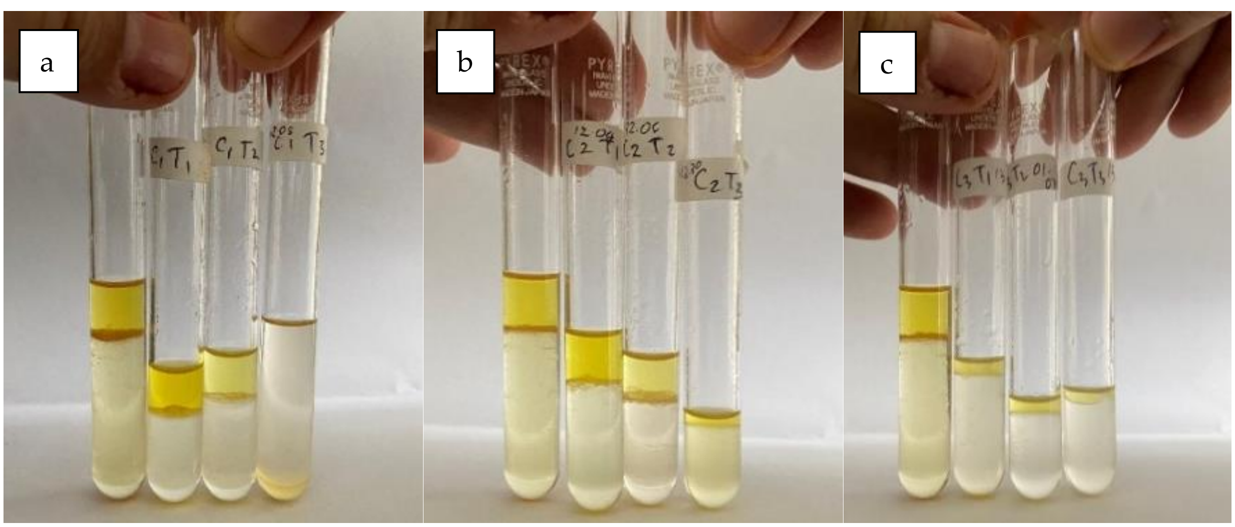

Figure 4. The results of the qualitative test comparison of pesticide residues with the control (far left), a) treatment C1T1, C1T2, C1T3; b) treatment C2T1, C2T2, C2T3; and c) treatment C3T1, C3T2, C3T3.

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Through testing with this test kit, the effectiveness of the reduction in organophosphate pesticide residues can be observed. The lower the amount of yellow liquid or the more transparent it becomes, it indicates that the pesticide residue levels were reduced. Based on the test results, it can be seen that samples C1T3, C3T2, and C3T3 produced less yellow liquid and were more transparent, especially sample C1T3. This shows that these treatments had the strongest residue reduction effect compared with the other treatments.

The test results of C3T2 and C3T3 looked similar based on screening; therefore, quantitative testing was only carried out for the C2T3 treatment, along with the control and C1T3 samples. The quantitative test results obtained using the GC-MS method are listed in Table 1.

Table 1. Pesticide residue test results on chili peppers

Treatment	Profenofos (mg/kg)
Control	0.305
C1T3	0.031

The test results showed that ozone fine-bubble treatment can reduce profenofos residues in curly chili. Treatment with 1 ppm ozone fine bubbles for 10 min succeeded in reducing profenofos residues by 89.8%, whereas ozone fine bubble treatment at 4 ppm for 5 min reduced profenofos residues by 66.9%. Asgar et al., (2017) previously reported that washing chili peppers using 1 ppm ozone solution can reduce profenofos residues by 52.49%. Compared to this study, fine bubble ozone was proven to be superior to regular ozone solutions for degrading pesticide content.

The increased effectiveness of reducing pesticide residues in fine-bubble ozone can be caused by the increased ozone retention in water, and the ROS content of fine bubbles can also increase the oxidation capacity (Li et al., 2023).

Based on SNI 4480:2016, the maximum residue level (MRL) was 5 mg/kg. In this study, all chili peppers tested contained profenofos, but were still below the threshold limit. Even so, chilies treated with 1 ppm fine-bubble ozone for 10 min produced chilies with the lowest profenofos residue of 0.031 mg/kg.

3.3 Weight Loss

Weight loss is one of the parameters that is considered crucial by businessess because of its negative influence on the distribution, storage, and sales processes in retail stores. All samples showed a uniform trend, where the weight loss value of the chili increased every day of observation (Figure 5).

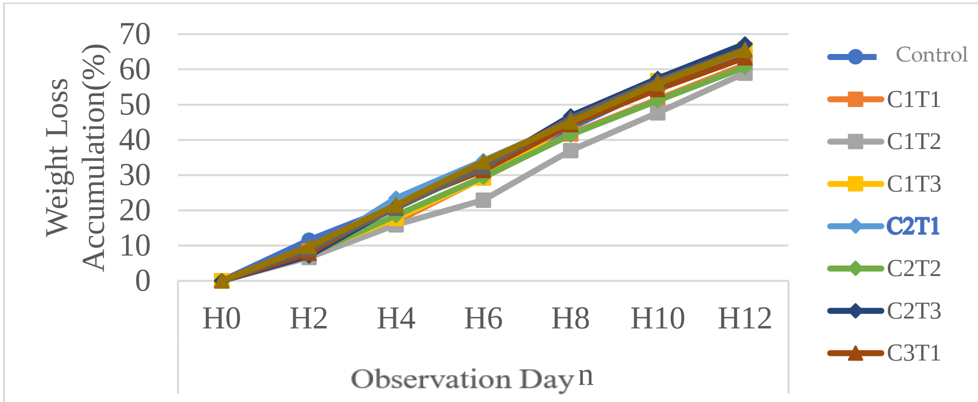

Figure 5. Accumulated weight loss of curly chili

Based on statistical analysis, an ozone fine-bubble concentration of 1 ppm significantly produced chili with the lowest weight loss compared with the control and other concentrations. On day 6, the average weight loss for chili with an ozone fine bubble concentration of 1 ppm was 27.09%, whereas of that the control was 31.9%.

In addition, a treatment duration of 5 min also significantly produced chili with the lowest weight loss compared to other durations, with an average of 28.23% (day 6); however, this difference was not significant. Treatment for 10 min resulted in the highest weight loss (45.56 %) on day 8. Based on the results of the analysis, it can be seen that the effectiveness of ozone based on the weight loss value of the sample is optimal at a level of 1 ppm with a duration of 5 min. However, the combination of 1 ppm concentration and 10 min duration was not significantly different from the 1 ppm treatment for 5 min.

Ozone treatment limits water loss by maintaining the stoma structure of the chili skin and suppressing the respiration rate of the fruit (Özen et al., 2020; Han et al., 2017). Glowacz (2016) also reported that ROS affect lipoxygenase (LOX) activity, which can reduce membrane damage and damage the surface of the outer skin structure.

3.4 Total Dissolved Solids

Total soluble solids are the total elements or mineral elements dissolved in a solution. According to Khandpur and Gogate (2015), total soluble solids are the nutrients accumulated in vegetables. Testing of the total soluble solids for samples treated with C3T2 stopped on day 6, whereas treatments C1T2, C2T1, and C3T3 stopped on day 8 because the material was very dry.

The results showed that the majority of chili peppers treated showed a trend where the average value of total soluble solids increased at the end of storage (Figure 6). The increase in the total soluble solid value is due to the accumulation of glucose formed by carbohydrate hydrolysis, which occurs faster than the process of converting glucose into energy and H2O (Aguilar-Veloz et al., 2022).

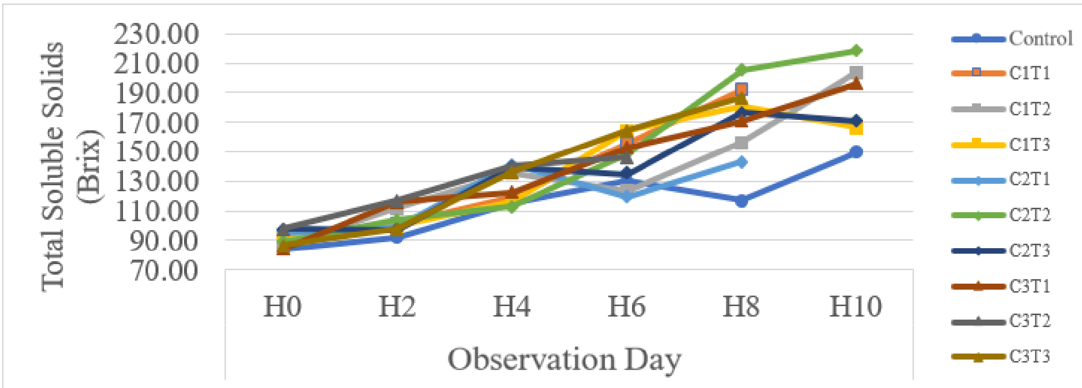

Figure 6. Change in total soluble solids of curly chili peppers

This is in accordance with the weight loss observations, where chilli experienced greater weight loss every day. The reduced weight indicates reduced water content in the chili, so that the mass of solids in the chili will be higher, which is followed by an increasing value of total soluble solids. ANOVA showed no significant difference in the total soluble solids between the control and treatment groups.

3.5 Texture

The texture value in this study refers to the hardness of chilies; hence, it is stated as the hardness value onwards. Almost all treatments in this study experienced the same phenomenon, where the hardness value increased at the beginning of storage. However, over time, the hardness value fluctuated and ended with a decrease in the hardness value at the end of the shelf life (Figure 7).

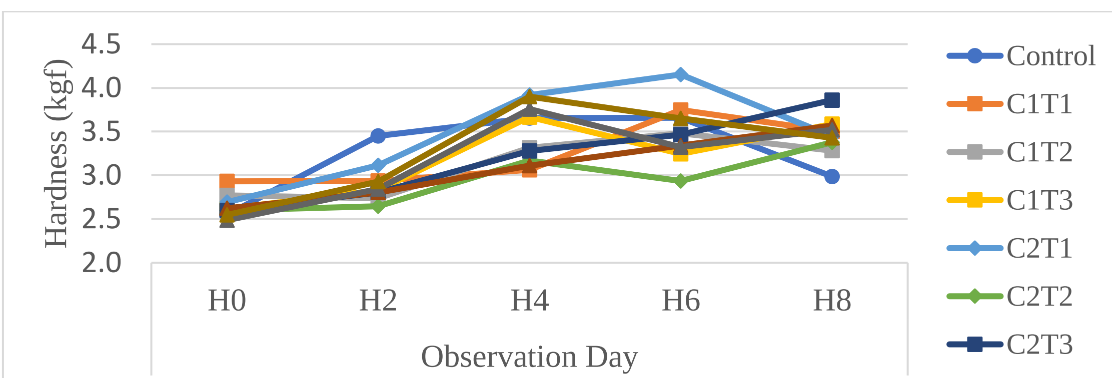

Figure 7. Change in hardness of curly chili

The fluctuation in the chili hardness value illustrates how the chili samples were damaged. Under fresh conditions, the hardness of chili tends to be low because the probe easily penetrates the surface, which occurs during the first two days. On the 4th day, the majority of chili peppers began to wither,

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where the surface of the chili peppers became more clay-like and it was increasingly difficult for the probe to penetrate the material, which means that the hardness value increased. On days 6th and 8th day the chili began to dry out and the hardness value decreased again because the probe penetrated the surface more easily.

Based on this, it can be seen that the critical point for assessing the hardness of chili peppers is when the chili peppers are at the lowest, of which the majority is on day 6. This is in accordance with the results of visual observations, where the critical point of chili began to occur on day 6.

Based on statistical tests, there was no significant difference in hardness between the control and treatment groups. In line with previous findings, ozone treatment up to 2 ppm did not significantly affect the hardness of green chili after washing with the ozone solution. (Glowacz et al., 2016; Ozen et al., 2020).

4. Conclusion

Ozone fine bubble technology has been proven to reduce profenofos pesticide residue levels in curly chili. Washing curly chili with ozone fine bubble solution at a concentration of 1 ppm for 10 min is recommended to degrade profenofos residues, with pesticide degradation levels reaching 89.8%. Ozone fine bubble treatment at 1 ppm for 10 min did not reduce the quality of curly chili.

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