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RESEARCH ARTICLE

Phytosanitary ozone treatment for guava infested with *Bactrocera zonata* and mandarin infested with *Ceratitis capitata* (Diptera: Tephritidae)

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Abstract

Both the peach fruit fly, *Bactrocera zonata* (Saunders), and the Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann), attack a wide variety of tree fruits worldwide. Exporting fruit hosts of these pests from countries where they are endemic to those where they are absent but may spread requires phytosanitary treatments. This research is to determine if *C. capitata* and *B. zonata* could be phytosanitary controlled by ozone treatment schedules. Moreover, elucidates the efficiency of ozone application in controlling microbial growth during the storage of fruits under cooling. Existing results proved that exposing infested fruits (guava and mandarin) with tephritids (*C. capitata* and *B. zonata*) to ozone gas for a time reaching 8 hours is sufficient for getting rid of immature stages (eggs, 1st, 2nd, and 3rd larval instars) inside fruits. On the other hand, the results indicated that the ozonation process at 1000 ppm for 6 and 8 hours reduced the total fungal count (TFC) compared to the control samples. Ozone application may be an effective strategy for guava and mandarin fruits to retain post-harvest quality and have a longer period of storage, even though ozone-treated fruits have increased ascorbic acid levels.

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Keywords: Peach fruit fly; Mediterranean fruit fly; Phytosanitary; Ozonation.

Introduction

There are many species in the family Tephritidae, many of which are significant agricultural pests. In specifically, *Ceratitis capitata* and the 46 identified species of the genus *Bactrocera* (EFSA, 2022). A polyphagous species known as *Bactrocera zonata* has been seen to attack more than 50 kinds of both wild and cultivated plants, primarily those bearing fresh fruits (EPPO, 2023). Tropical Asia is the original home of *B. zonata* (White and Elson Harris, 1992). Various countries in southern and Southeast Asia have this species. *B. zonata* has expanded its geographic range to include the Middle East and northern Africa, which have drier climates. The species was discovered for the first time in Egypt in 1924 but reports of its establishment and significant impact on the yield of fruit crops start to the year 1993 (CABI, 2020). The Mediterranean fly, *C. capitata* (Medfly) (Diptera: Tephritidae), is one of the most destructive fruit pests in the world because it can attack commercially important fruit, resulting in significant economic losses that are estimated to be more than $2 billion annually. The damage caused when larvae eat directly on the pulp, causing premature fruit drop, is the main cause of the

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decreased yields. Additionally, oviposition holes optimize subsequent bacterial and fungal infections, which further reduce yields (Cappelli et al., 2022; EPPO, 2011). A list of Union quarantine pests, including *B. zonata* and *C. capitata*, can be found in Annex II of Commission Implementing Regulation (EU) 2019/2072. Additionally, the insect is designated as a priority pest under Commission Delegated Regulation (EU) 2019/1702, resulting in the necessity of pest surveys every year. Currently, the import of fruits and plants used as hosts for a variety of non-EU Tephritidae is either forbidden or subject to particular regulations (Commission Implementing Regulation (EU) 2019/2072, Commission Implementing Regulation (EU) 2018/2019). Additionally, certain import regulations are in place for the import of growth media (Commission Implementing Regulation (EU) 2019/2072; EFSA, 2022). As a quarantine disinfestation technique, ozone treatment is frequently employed to stop pests from establishing themselves in new locations during export (Botondi et al., 2021). Ozone is safe for use in food applications since it spontaneously decomposes without leaving behind dangerous residues in the treatment media. Ozone can have negative consequences on products when employed improperly. To use ozone effectively and safely, treatment conditions should be established individually for all types of materials. As a quarantine disinfestation technique, ozone treatment is frequently employed to stop pests from establishing themselves in new locations during export (Botondi et al., 2021). Ozone is safe for use in food applications since it spontaneously decomposes without leaving behind dangerous residues in the treatment media. Ozone can have negative consequences for products when employed improperly. To use ozone effectively and safely, treatment conditions should be established individually for all types of materials (Karaca and Sedat, 2007). Therefore, the present study was conducted in order to evaluate the effectiveness of the ozonation process on *B. zonata* and *C. capitata* pupation and the adult emergence of infected mandarin and guava fruits, respectively. As well as study the effect of ozone gas on microbiological quality and the content of ascorbic acid in the treated fruits.

Materials and methods

Fruit samples

Guava (*Psidium guajava*) and Mandarin (*Citrus reticulate*), also known as the Clementine variety, were obtained from a private farm located on the Cairo-Alexandria Desert Road. The mandarin and guava fruits utilized in these tests for fake infestation were fresh, ripe, healthy, and uninfested by microbes or insects.

Laboratory rearing technique for Tephritids

The peach fruit fly, *B. zonata,* and the Mediterranean fruit fly, *C. capitata*, were continually cultivated in the same conditions at the Plant Protection Research Institute's Entomology Laboratory in Dokki, Giza, Egypt, at a temperature of 25± 2 ℃ and relative humidity of 60–70%.

Infected with guava and mandarin by eggs stage

Guava fruits were infested by *B. zonata* while mandarin fruits were infested by *C. capitata*. Every 24 hours, the laid eggs were removed from the oviposition containers. Under a stereo microscope, the gathered eggs were strung on the card (black paper, 2 x 2 cm) by using a channel-hair brush. The fruits were artificially infested with eggs. A small cut was made for fruits to see the card inside. Each mandarin or guava fruit was infested with a card-carrying 30 eggs (1-day-old). Fruits infested with eggs were exposed to ozone gas at 1000 ppm for 2, 4, 6, and 8 hours. Five mandarin or guava fruits replicated with 30 eggs for each were used. After treatment, the fruits were put into plastic vials and kept in a cabinet with a controlled environment (25 ± 2 °C and 60–70%RH). All fruits infested with eggs were incubated in plastic boxes with 1cm sand under lab conditions (25 ± 2 °C and 60–70% RH) to determine the percentage of pupations, and adult emergences.

Treating of larvae stage with ozone gas inside host fruit

The first, second, and third larval instars of *B. zonata* or *C. capitata* were artificially injected into each guava or mandarin fruit. The larvae-infested fruits were exposed to ozone gas at 1000 ppm for 2, 4, 6, and 8 hours. For each treatment, 30 larvae were replicated on five infected fruits. Following treatment, the fruits were put into plastic vials and kept in a cabinet with a controlled atmosphere (25 ± 2 °C and 60–70% RH) to check the percentage of pupation and the adult emergency.

Large-scale confirmatory tests

Large-scale confirmatory tests were conducted by exposing the third larval instar of *B. zonata* or *C. capitata* to ozone in each guava or mandarin fruit at 1000 ppm for 8 hours (the ozone dose estimated to prevent adult emergency). This was repeated 50 times. For control, 50 late third instars of *B. zonata* or *C. capitata* were placed in guava or mandarin fruit, and this was repeated five times. All treated and untreated infested guava or mandarin fruits were incubated in a controlled environment cabinet at 25 ± 2 °C and 70% ±5RH. All infested fruits were observed to check for adult emergencies.

Exposure of fruits to ozone gas

Ozone gas was applied to guava and mandarin fruit using an ozone generator (Model OZO6 VTTL OZO Max Ltd., Shefford, Quebec, Canada). Samples are put in a square container with a tight-fitting lid and are joined to an inlet valve on one side and an output valve on the other. Except for the control fruit, which was left untreated with ozone, the fruit was exposed to ozone gas at 1000 ppm for 2, 4, 6, and 8 hours.

Estimation of total fungal count (TFC)

According to Feroz et al. (2016), in 90 ml of sterile physiological saline, 10 grams of the samples (ozonated for 6 and 8 hours) were homogenized for 1 minute. Serial dilutions were surface plated on Sabaroud Dextrose Agar (SDA) using this homogenate, and plates were incubated at 25 o C for 5 days. TFC per gram was gradually taken into consideration from SDA plates after incubation.

Determination of Ascorbic acid

Ascorbic acid was quantified and provided as mg/100 g fresh weight using 2, 6-dichlorophenol indophenol for the titration of the juice of samples (Ranganna, 1979).

Statistical analysis

One-way analysis of variance (ANOVA) (Tukey's test) was used to analyses experimental data with SPSS (statistical package for social sciences, version 20.0), and sample significance was determined at P≤0.05. The mean ± standard deviation of the data (n = 5) is displayed.

Results and Discussion

Effect of ozone gas on Bactrocera zonata pupation and adult emergence in infected guava

Results concerning the evaluation of the efficacy of exposure to ozone gas on different stages of *B. zonata* in infested guava are shown in Table (1). Data indicated that the percentages of pupation and adult emergence of *B. zonata* from untreated guava infested with eggs, 1st, 2nd, and 3rd larval instars were (96 and 93.67%), (98.67 and 94.67%), (95.33 and 94.67%), and (96.67 and 91.33%), respectively. The percentages of pupation and adult emergence at the different time of exposure for ozone (2, 4 and 6 h) for fruits infested with eggs in the 1st, 2nd and 3rd larval instars were (20.67, 15.33%), (34%, 18%), (38.67 , 26%), (49.33, 38%) & (12.67, 8.67), (22, 12.67), (31.33, 19.33), (41.33, 27.33) & (6.67, 5.33), (8, 6.67), (10.67,1.33), (32.67, 17.33), respectively. Exposure of infested guava with eggs in the 1st, 2nd and 3rd larval instars to ozone gas (1000 ppm) at 8 hours completely inhibited pupation and adult emergence.

Table 1. Effect of exposure time and ozone concentration on *Bactrocera zonata* pupation and adult emergence in infected guava.

Life stage	Exposure time (h)	No. Pupae Mean ±SE	Pupation %	No. Emerging Adult Mean ±SE	Adult emergence %
Egg	Control	28.8 ± 0.58[a]	96	28.1 ± 1.1 [a]	93.67
	2	6.2 ± 0.20 [b]	20.67	4.6 ± 0.24 [b]	15.33
	4	3.8 ± 0.20 [c]	12.67	2.6 ± 0.75 [c]	8.67
	6	2.0 ± 0.32[d]	6.67	1.6 ± 0.40 [d]	5.33
	8	0.0 ± 0.0 [e]	-------	0.0 ± 0.0 [e]	---------
1st larval instar	Control	29.6 ± 0.55[a]	98.67	28.4 ± 0.4[a]	94.67
	2	10.2 ± 0.37[b]	34	5.4 ± 0.51 [b]	18
	4	6.6 ± 0.24 [c]	22	3.8 ± 0.37[c]	12.67
	6	2.4 ± 0.60[d]	8	2.0 ± 0.45[d]	6.67
	8	0.0 ± 0.0 e	---------	0.0 ± 0.0 e	----------
2nd larval instar	0	28.6 ± 0.86[a]	95.33	28.4 ± 0.4[a]	94.67
	2	11.6 ± 0.24[b]	38.67	7.8 ± 0.49[b]	26
	4	9.4 ± 0.24 [c]	31.33	5.8 ± 0.45 [c]	19.33
	6	3.2 ± 0.37[d]	10.67	0.4 ± 0.24 [d]	1.33
	8	0.0 ± 0.0[e]	---------	0.0 ± 0.0 [e]	----------
3rd larval instar	Control	29 ± 0.0[a]	96.67	27.4 ±0.51[a]	91.33
	2	14.8 ± 0.89[b]	49.33	11.4 ±0.51[b]	38
	4	12.4 ± 0.24[c]	41.33	8.2 ± 0.2[c]	27.33
	6	9.8 ± 0.24[d]	32.67	5.2 ± 0.84[d]	17.33
	8	0.0 ± 0.0[e]	--------	0.0 ± 0.0[e]	----------

Effect of ozone gas on Ceratitis Capitata pupation and adult emergence in infected mandarin

Data in Table (2) indicated that exposing mandarin fruits infested with eggs of *C. capitata* to ozone gas for 2 hours resulted in reducing the percentage of pupation and adult emergence to (11.33 & 1.33) against (96 & 70.67%) for the untreated group. Pupation and adult emergence were inhibited in mandarin infested with eggs at 4 hours of exposure to ozone. For percentage pupation and adult emergence from mandarin infested with 1st and 2nd larval instars reached to (33.33 and 15.33%), (15.33 and 0.0 %) & (35.33, 20), (24, 12%), respectively, at exposing times 2 and 4 hours. Complete prevention for pupation and adult emergence from mandarin infested with 1st and 2nd larval instar after exposing mandarin to ozone gas for a period of 6 and 8 hours. For mandarin infested with the 3rd larval instar the pupation and adult emergence percentages for 2,4 and 6 hours of exposure to ozone gas were (40.67,28.67%), (30.67, 16%), (16, 3.33%), respectively. Pupation and adult emergence from infested mandarin with 3rd larval instar was prevented at the exposing period reached 8 hours.

Large-scale confirmatory tests

No F1 adults were developed in the large-scale confirmatory experiment after estimated third larval instars of *C. capitata* or *B. zonata* were exposed to 1000 ppm ozone for 8 hours in infested mandarin or guava as shown in Table (3). In agreement with Husain et al. (2015), who tested the effect of exposing date fruits infested with 4th instar larvae of the almond moth, *Ephestia cautella* (Walker) (Lepidoptera: Pyralidae), to ozone gas at 22 ppm for 24, 48 and 72 hours.

Table 2. Effect of ozone gas on *Ceratitis capitata* pupation and adult emergence in infected mandarin

Life stage	Exposure time (h)	No. Pupae Mean ±SE	Pupation %	No. Emerging Adult Mean ±SE	Adult emergence %
Egg	Control	25.0 ± 0.45[a]	96	21.2 ± 0.58[a]	70.67
	2	3.4 ± 0.24[b]	11.33	0.4 ± 0.24 [b]	1.33
	4	0.0 ± 0.0 [c]	--------	0.0 ± 0.0 [c]	---------
	6	0.0 ± 0.0 [c]	---------	0.0 ± 0.0 [c]	---------
	8	0.0 ± 0.0 [c]	-------	0.0 ± 0.0 [c]	---------
1st larval instar	Control	24.2 ± 0.37[a]	80.67	21.6 ± 0.51[a]	72
	2	10 ± 0.32[b]	33.33	4.6 ± 0.24[b]	15.33
	4	4.6 ± 0.24[c]	15.33	0.0 ± 0.0[c]	--------
	6	0.0 ± 0.0 [d]	---------	0.0 ± 0.0 [c]	---------
	8	0.0 ± 0.0 [d]	---------	0.0 ± 0.0 [c]	----------
2nd larval instar	Control	24.2 ± 0.37[a]	80.67	20.8 ± 0.66[a]	69.33
	2	10.6 ± 0.24[b]	35.33	6 ± 0.32[b]	20
	4	7.2 ± 0.37[c]	24	3.6 ± 0.24[c]	12
	6	0.0± 0.0 [d]	---------	0.0± 0.0 [d]	---------
	8	0.0 ± 0.0 [d]	---------	0.0 ± 0.0 [d]	----------
3rd larval instar	Control	22 ± 0.71[a]	73.33	20 ± 0.55[a]	66.67
	2	12.2 ± 0.37[b]	40.67	8.6 ± 0.4[b]	28.67
	4	9.2 ± 0.37[c]	30.67	4.8 ± 0.37[c]	16
	6	4.8 ± 0.37[d]	16	1 ± 0.32[d]	3.33
	8	0.0 ± 0.0 [e]	--------	0.0 ± 0.0 [e]	----------

The most damaging fumigant was ozone, which caused 100% mortality after various exposure times. Also, the present data is similar to the finding of Ghazawy et al. (2021) when exposed date Palm was infested with larvae of *Plodia interpunctella* and *E. cautella* to 200 and 400 ppm of ozone gas at 0.5, 1, 2, 4, 6 and 8 hours. The mortality percentage increased gradually at two doses and at different exposure times until reaching 100% mortality for both pests after exposure to 200 and 400 ppm for 8 hours. The following pests are damaged by ozone due to synthase activity and oxidative stress caused by enzymes like superoxide dismutase (SOD) and nitric oxide (NO) activity to damage the brain tissues and neurosecretory cells: *Trogoderma granarium*, *Tribolium castaneum*, *Ephestia cautella*, and *Plodia interpunctella* (Ghazawy et al., 2021). In the post-embryonic life of insects, neurosecretory cells in the central nervous system regulate several important physiological processes. During post-embryonic development, neuroblasts were seen, but they stopped dividing before adult emergence (Ganeshina et al., 2000). Some insects responded well to fumigation with ozone (Jian et al., 2013). For caterpillars and grubs exposed to ozone, ultrastructural analysis revealed that the muscles, fat body, and neurosecretory cells in the brain were all impacted (Ghazawy et al., 2021). Lipids, proteins, and nucleic acids are oxidized and changed at the molecular and cellular level as a result of ozone action (Iriti and Faoro, 2008). According to Khadre et al. (2001), oxygen generated from the breakdown of ozone may cause "oxidative stress," which eventually results in cellular damage and death. Ghazawy et al. (2021) observed that ozone motivates oxidative stress and death without leaving pollutants. It can harm an insect pest's neurosecretory cells and brain tissues. This could result in safe pest management methods.

Table 3. Large-scale confirmatory tests of 3rd larval instars after exposing to 1000 ppm ozone gas for 8 h in the host fruit of *C. capitata* or *B. zonata*

Exposing time for ozone	No. of replicates	No. of treated	No. of F1 adults
8 h	50	1500	-
0 (control) for mandarin	5	150	116
0 (control) for guava	5	150	144

Effect of ozone gas on total fungal count

In the present work, samples of guava and mandarin were ozonated by 1000 ppm ozone gas for 6 and 8 h and subjected to mycological examination. The results presented in Table (4) showed the total fungal count in guava samples after being ozonated for 6 and 8 h at 1000 ppm. The results showed that the total fungal count (TFC) isolated decreased in ozonated samples compared with control samples. The TFC in the control samples recorded 3.54±0.66, 3.57±0.61, 3.576±0.52 and 3.58±0.15 (log10CFU/g) after storage for one, two, three and four weeks, respectively. Guava samples treated with ozone gas for 6 and 8 h did not show any TFC with storage at 4°C for one week. Concerning, mandarin samples stored for two weeks after being treated with ozone gas for 6 and 8 h did not show any TFC, while the control sample was 3.61±0.24 (log10CFU/g). The infection percent (TFC) decreased after ozonation for 6 and 8 h to 2.24±0.22 and 1.47±0.16 (log10CFU/g) after storage for three weeks compared with control samples, which was 3.66±0.33 (log10CFU/g). Ozone in gaseous or aqueous form has been reported to reduce levels of natural microflora as well as bacterial and fungal contamination in fruits. Fungal inactivation depends on several factors, including ozone concentration and exposure time (Naito and Takahara, 2006). Researchers have looked into the antibacterial effectiveness of ozone for a variety of microorganisms, including viruses, bacterial spores, and vegetative bacteria. Ozone exposure for preventing fungal deterioration in thorn less blackberries was assessed by Barth et al. (1995). The fruit was collected, kept for 12 days at 2 ºC, and exposed to ozone concentrations of 0.0, 0.1, and 0.3 ppm before being tested for fungal decay (*Botrytis cinerea*), anthocyanins, color, and peroxidase activity. According to Khadre (2001), ozone affects a wide range of biological elements, including proteins, unsaturated lipids, and respiratory enzymes in cell membranes; peptidoglycans in cell envelopes; enzymes and nucleic acids in the cytoplasm; and proteins and peptidoglycans in spore coatings and viral capsids. As a result, the mechanism of ozone's microbicidal action is a complex process. Additionally, according to Skog and Chu (2001), ozone gas increases membrane permeability and decreases cellular compartmentation.

Table 4. Total fungal count in guava treated by ozone gas stored at 4°C

Treatments of samples	Total fungal count (log_{10}CFU/g)				
	Zero time	First week	Second week	Third week	Fourth week
Un treated	3.38±0.33	3.54±0.66	3.57±0.61	3.57±.52	3.58±0.15
Ozonated for 6 h	0.0	0.0	2.69±0.27	2.87±0.25	2.9±0.34
Ozonated for 8h	0.0	0.0	1.95±0.26	2.07±0.15	2.77±0.27

Table 5. Total fungal count in mandarin orange treated by ozone gas that stored at 4°C

Treatments of samples	Total fungal count ($(log_{10}CFU/g)$				
	Zero time	First week	Second week	Third week	Fourth week
Un treated	3.4±0.35	3.5±0.22	3.61±0.24	3.66±0.33	3.69±0.62
Ozonated for 6 h	0	0	0	2.24±0.22	2.36±0.21
Ozonated for 8 h	0	0	0	1.47±0.16	1.84±0.25

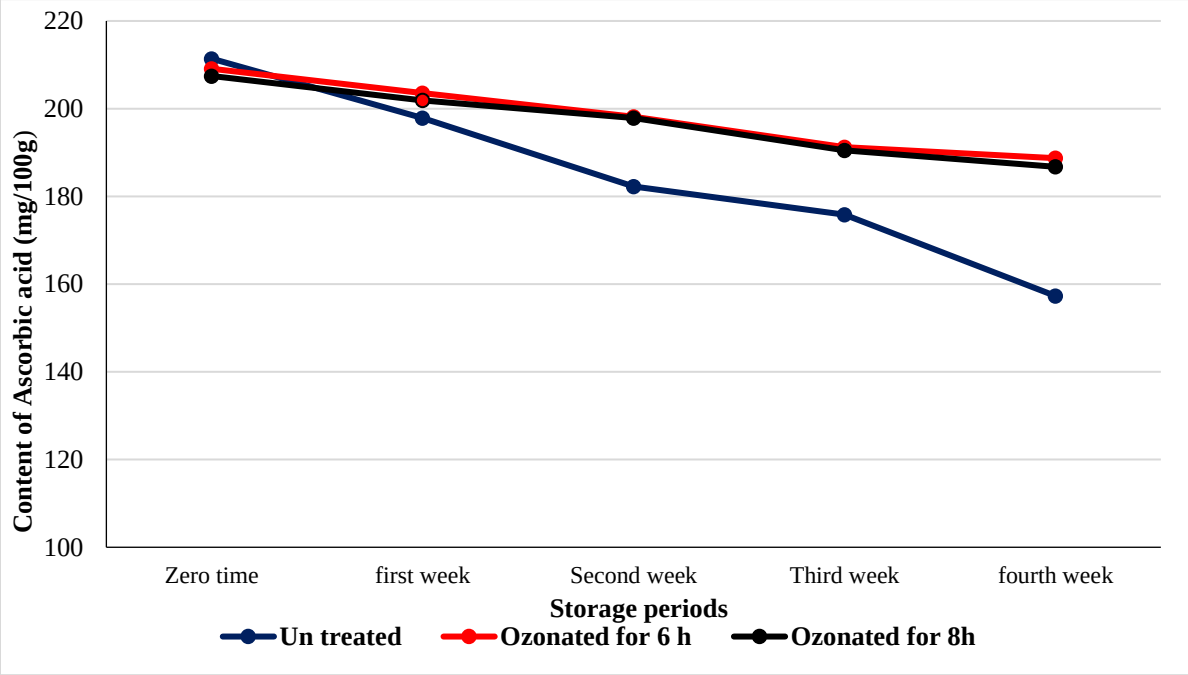

Figure 1. Content of Ascorbic acid in guava infected by *Bactrocera zonata* after ozonation

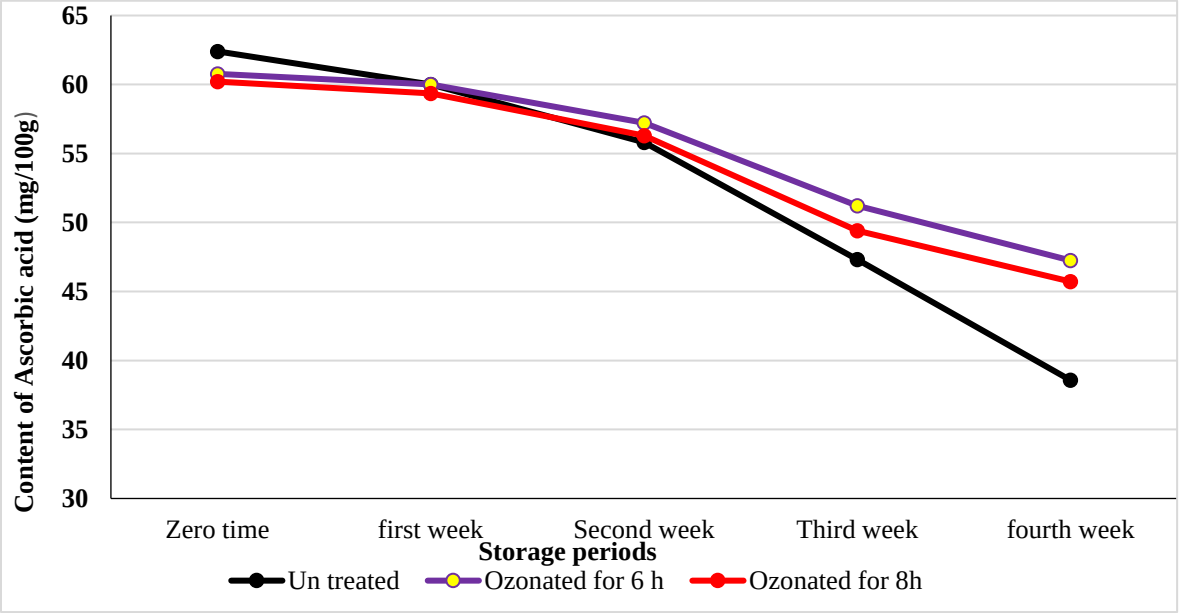

Figure 2. Content of Ascorbic acid in mandarin fruits infected by *Ceratitis capitata* after ozonation

Impact of ozonation on ascorbic acid content

The ascorbic acid concentration of guava and mandarin stored at 4°C and 85–90% RH showed a clear tendency to decline with all treatments, including the control sample, during the duration of the storage periods. After four weeks of storage, guava exposed to ozone gas for 6 and 8 h recorded the highest values of ascorbic acid, 188.72 and 186.75 mg/100 ml, respectively, compared with the control sample (untreated by ozone gas), which recorded 157.31 mg/100 ml as shown (Fig. 1). On the other hand, the concentrations of ascorbic acid in guava at zero time were 211.4 mg/100 ml in control sample, while ozonated samples for 6 and 8 h were 209.1 and 207.45 mg/100 ml, respectively.

The ascorbic acid content in the control sample of mandarin fruit is 62.4 mg/100 g, and in the samples treated with ozone gas for 6 and 8 hours, the ascorbic acid content at zero time is 60.77 and 60.21 mg/100 g, respectively. After four weeks of storage, the concentrations of the ascorbic acid content were 38.57, 47.25, and 45.72 mg/100 in the control sample and the ozonated sample for 6 and 8 hours, respectively as shown (Figure 2). Ascorbic acid's value rapidly degrades as a result of fungal infection and storage time. So, keeping any fungi from growing on the fruits will keep the fruits' value as sources of ascorbic acid. According to Ayón-Reyna et al. (2019), as the fruits were exposed to air, the ascorbic acid content was likely first transformed into dehydroascorbic acid through a reversible reaction and then perhaps into 2-3-dicetogulonic acid. The ascorbic acid content of avocado slices was better stabilized during storage with ozone treatments. These outcomes are comparable to those attained by Pérez et al. (1999) on strawberries, Ladaniya (2004) on lime, Barboni et al., (2010) on kiwi fruit, and Khalil (2016) on Zaghloul date palm, as well as EL-Hadidy (2017) on Washington Navel orange. This is because ozone gas treatments delayed ascorbic acid content degradation and increased its concentration in the fruit under cold storage conditions. The high oxidative capacity of ozone creates toxic molecular species, acting as a potent phytotoxic agent and activating the fruit defense system (anti-oxidative system), which promotes the biosynthesis of ascorbic acid from the carbohydrate pool to avoid the oxidative activity caused by ozone, according to Klopotek et al. (2005), Allende et al. (2007), Alwiand Ali (2015).

Conclusions

Exposing the immature stages (eggs, 1st, 2nd & 3rd) of fruit flies (*C. capitata* and *B. zonata*) to 1000 ppm ozone gas for 8 h is sufficient for preventing F1 adults. This dose was efficient and satisfactory in providing quarantine security for the export or import of mandarin or guava fruits. TFCs were not found during the two-week storage period. As a result, it is suggested that fresh guava and mandarin be exposed to ozone at a concentration of 1000 ppm for eight hours to preserve their freshness for 15 days when kept in a refrigerator at 4 ±1 °C and 85–90% relative humidity.

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Author's Contributions

All authors have reviewed and edited the manuscript.

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Declarations

Conflicts of Interest

The authors disclosed no conflict of interest starting from the conduct of the study, data analysis, and writing until the publication of this research work.

Ethics Approval

Not applicable

Consent for publication

The content of the submitted article has been carefully examined and approved by all authors who are all aware of its submission to this journal.

Availability of data and materials

All data from this study are included in this published article.

Ethics Approval and Consent to Participate

Not applicable

References

Allende, A., Marín, A., Buendía, B., Tomás-Barberán, F., Gil, M.I. (2007) 'Impact of combined postharvest treatments (UV-C light, gaseous O3, super atmospheric O2 and high CO2) on health promoting compounds and shelf-life of strawberries.' *Postharvest Biology and Technology* 46(3), pp.201-211. DOI:10.1016/j.postharvbio.2007.05.007

Alwi, N.A., Ali, A. (2015) 'Dose-dependent effect of ozone fumigation on physiological characteristics, ascorbic acid content and disease development on Bell pepper (*Capsicum annuum* L.) during storage.' *Food and Bioprocess Technology*, 8, pp. 558–566. 10.1007/s11947-014-1419-2

Ayón-Reyna, L.E., Ayón-Reyna, L.G., López-López, M.E., López-Angulo, G., Pineda- Hidalgo, K.V., Zazueta-Niebla, J.A., Vega- García, M.O. (2019) 'Changes in ascorbic acid and total phenolics contents associated with browning inhibition of pineapple slices.' *Food Science and Technology*, Campinas, 39(3), pp. 531-537.https://doi.org/10.1590/fst.21117

Barboni, T., Cannac, M., Chiaramonte, N. (2010) 'Effect of cold storage and ozone treatment on physic chemical parameters, soluble sugars, and organic acids in Actinidia deliciosa.' *Food Chemistry*,121(4), pp.946–951. DOI:10.1016/j.foodchem.2010.01.024

Barth, M.M., Zhou, C., Mercier, J., Payne, F.A. (1995) 'Ozone storage effects on anthocyanin content and fungal growth in blackberries.' *Journal of Food Science*, 60(6), pp. 1286-1288. DOI:10.1111/j.1365-2621.1995.tb04575.x

Botondi, R., Barone, M., Grasso, C. (2021) 'A review into the effectiveness of ozone technology for improving the safety and preserving the quality of fresh-cut fruits and vegetables, Foods.' 10(4), pp. 748-775. https://doi.org/10.3390/foods10040748

CABI (Centre for Agriculture and Bioscience International), (2020) 'Invasive Species Compendium. Data sheet *Bactrocera zonata (peach fruit fly)'*. 10/02/2020. Wallingford, UK: *CAB International. Available online*: https://www.cabi.org/isc/datasheet/17694

Cappelli, A., Petrelli, D., Gasperi, G., Serrao, A.G.M., Ricci, I., Damiani, C., Favia, G. (2022) 'Bacterial Symbionts in *Ceratitis capitata*. Insects.' 13(5), pp. 474. https://doi.org/10.3390/insects13050474

Commission Implementing Regulation (EU) 2019/2072. *Bactrocera zonata* is a Union quarantine pest listed in Annex II of Commission Implementing Regulation (EU) 2019/2072. The pest is also listed as a priority pest under Commission Delegated Regulation (EU) 2019/1702 implying the obligation for annual surveys of the pest. https://www.legislation.gov.uk/eur/2019/2072

EFSA (European Food Safety Authority), (2022) 'Pest survey card on *Bactrocera dorsalis*.'EFSA supporting publication 2022: EN-7206. Available online: https://efsa.europa.eu/plants/planthealth/monitoring/surveillance/bactrocera-dorsalis . Last updated: 1 March 2022.

EL-Hadidy, G.A.M. (2017) 'Effect of postharvest treatments with Oxilite on Washington "Navel Orange" fruits under cold storage conditions.' *International Journal of Chem. Tech. Research, 10(2)*, pp. 523-533.

EPPO (2023) 'EPPO Global Database (available online). https://gd.eppo.int

EPPO (2011) 'PQR-EPPO data base on quarantine pests. http://www.eppo.int (accessed on 01 September 2011).

Feroz, F., Mori, M., Sakagami, Y. (2016) 'Bacterial and fungal loads in raw sea foods, fruits and vegetables collected from Dhaka Bangladesh and the effect of heat on its growth.' *Bangladesh Journal of Microbiology*, 33 (1-2), pp. 23-28. DOI:10.3329/bjm.v33i1.39599

Ganeshina, O., Schäfer, S., Malun, D. (2000) 'Proliferation and programmed cell death of neuronal precursors in the mushroom bodies of the honeybee. *Journal of Comparative Neurology*, 417(3),pp. 349–365.DOI:10.1002/(SICI)1096-9861(20000214)417:33.0.CO;2-E

Ghazawy, N.A., Zinhoum, R.A., Ali, M.M., Afify, A., Hussain, H.B. (2021) 'Efficacy of Ozone on Mortality, Super Oxide Dismutases, Nitric Oxide Enzymes and Ultrastructural Alterations on Some Stored Product Insect Larvae in Egypt.' *African Entomology*, 29(1), pp. 17-31. https://doi.org/10.4001/003.029.0017

Husain, M., Rasool, K.G., Tufail, M., Alhamdan, A.M., Mehmood, K., Aldawood, A.S. (2015) 'Comparative

efficacy of CO2 and ozone gases against *Ephestia cautella* (Lepidoptera: Pyralidae) larvae under different temperature regimes.' *J. Insect Sci*., 15 (1), pp. 126. DOI: 10.1093/jisesa/iev108

Iriti, M., Faoro, F. (2008) 'Oxidative stress, the paradigm of ozone toxicity in plants and animals.*' Water, Air and Soil Pollution*, 187, pp.285–301. https://doi.org/10.1007/s11270-007-9517-7

Jian, F., Jayas, D.S., White, N.D.G. (2013) 'Can ozone be a new control strategy for pests of stored grain?' *Agriculture Research*, 2(1),pp. 1–8.DOI:10.1007/s40003-012-0046-2

Khadre, M. A., Yousef, A. E., Kim, J. G. (2001) 'Microbiological aspects of ozone applications in food: a review.' *Journal of Food Science*, 66(9), pp.1242–1252. https://doi.org/10.1111/j.1365-2621.2001.tb15196.x

Khalil,H.A .(2016) 'Effect of ozone application on postharvest quality and microbiological state of "Zaghloul" date palm fruits.' *J. Plant Production, Mansoura Univ*., 7 (1), pp. 43-51. DOI:10.21608/jpp.2016.43456

Klopotek, Y., Otto, K., Böhm, V. (2005) 'Processing strawberries to different products alters contents of vitamin C, total phenolics, total anthocyanins, and antioxidant capacity.' *J. Agric. Food Chem*., 53(14), pp.5640–5646. https://doi.org/10.1021/jf047947v

Karaca, H., Sedat Velioglu, Y. (2007) 'Ozone Applications in Fruit and Vegetable Processing.' *Food Reviews International*, 23(1), pp.91-106. https://doi.org/10.1080/87559120600998221

Ladaniya,M.S.(2004) 'Response of 'Kagzi' acid lime to low temperature regimes during storage. *Journal of Food Science and Technology Mysore*, 41(3), pp.284-288.

Naito, S., Takahara, H. (2006) 'Ozone contribution in food industry in Japan.' *Ozone: Science and Engineering*, 28(6), 425-429. https://doi.org/10.1080/01919510600987347

Pérez, A.G., Sanz, C., Ríos, J.J., Olías, R., Olías, J.M. (1999) 'Effect of ozone treatment on postharvest strawberry quality.' *J. Agric. Food Chem*., 47(4), pp. 1652–1656. DOI: 10.1021/jf980829l

Ranganna, S. (1979) 'Manual of Analysis of Fruit and Vegetable Products, second ed.' *Tata McGraw Hill, New Delhi*, pp.634.

Skog, L.J., Chu, C.L. (2001) 'Effect of ozone on qualities of fruits and vegetables in cold storage.' *Canadian Journal of Plant Science*, 81, pp.773–778. DOI:10.4141/P00-110

White, I.M., Elson Harris, M.M. (1992) 'Fruit Flies of Economic Significance: Their Identification and Bionomics.' *Environmental Entomology*, 22, pp. 1408-1408. https://doi.org/10.1093/ee/22.6.1408a