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联合国粮食及农业组织 Food and Agriculture Organization of the United Nations Organisation des Nations Unies pour l'alimentation et l'agriculture Продовольственная и сельскохозяйственная организация Объединенных Наций Organización de las Naciones Unidas para la Alimentación y la Agricultura منظمة الأغذية والزراعة للأمم المتحدة

# COMMISSION ON PHYTOSANITARY MEASURES

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| **Fifteenth Session** |
| **Rome, 30 March - 3 April 2020** |
| **Adoption of International Standards for Phytosanitary Measures - Ink amendments to adopted international standards for phytosanitary measures (ISPMs) - Annexes to ISPM 28 (*Phytosanitary treatments for regulated pests*): irradiation treatments for tephritid fruit flies - Modified atmosphere usage in irradiation treatments** |
| **Agenda item 10.2** |
| **Prepared by the IPPC Secretariat** |

## I. Introduction

1. The IPPC Technical Panel on Phytosanitary Treatments (TPPT) discussed the effects of low oxygen on irradiation efficacy at their meeting in July 2019[1], considering that almost all currently adopted phytosanitary treatments (PTs) for irradiation treatments[2] contain the following disclaimer: "This irradiation treatment should not be applied to fruits and vegetables stored in modified atmospheres.". The only exception is PT 11 (*Irradiation treatment* for Grapholita molesta *under hypoxia*) as the supporting study has tested the treatment in low oxygen environment.

1 2019-07 TPPT Meeting Report (Vienna, Austria): https://www.ippc.int/en/publications/87681/

2 Adopted ISPMs: https://www.ippc.int/en/core-activities/standards-setting/ispms/

## II. Background

2. When drafting the first irradiation treatments, more than 10 years ago, the TPPT decided to include a limitation as the studies available at the time (Hallman 2001, 2004a, b)[3] indicated that irradiation under low-oxygen conditions might reduce the efficacy of the treatment.

3. Multiple studies have shown a loss of irradiation treatment efficacy at very *low* oxygen levels (near 0%), and it is agreed that very low oxygen during irradiation should not be allowed. However it was proposed that as fruit flies have been well studied at *moderate* oxygen levels and oxygen levels of 5-7% or higher did not cause a loss of irradiation treatment efficacy in the studied fruit flies this caveat may be removed (Hallman, 2004a, b; Follett *et al*., 2013; Srimartpirom *et al*., 2018; Follett *et al*., 2018)[4].

4. The TPPT reviewed the preliminary results of a FAO/IAEA/USDA Project on Phytosanitary Treatments in which research was carried out regarding the effect of low oxygen storage on efficacy of phytosanitary irradiation against Tephritid fruit flies. In laboratory trials, no difference in survival of four Tephritid fruit fly species was found when stored in low oxygen before and during irradiation. The research is summarized in Attachment 1 (to English version only) to provide technical justification. The result of this study is also expected to be published in a peer-reviewed journal.

5. The TPPT recommended the removal of the restriction for Tephritid fruit fly species and noted that there is information available of trials that resulted in 5% survival of *Grapholita molesta* treated under hypoxia and thus the restriction would need to be further considered for other insect group, such as the Lepidoptera.

6. The SC agreed based on the TPPTs recommendation, to present to the CPM-15 (2020) as ink amendments the removal of the disclaimer “This irradiation treatment should not be applied to fruits and vegetables stored in modified atmospheres.” from irradiation treatments for Tephritid fruit flies concerning the adopted Annexes to ISPM 28 listed in decision point (1) below.

## III. Decision

7. The CPM is invited to:

1) *note* the ink amendments to the following adopted Annexes to ISPM 28 (Attachment 02 to English version only):

- PT 1: Irradiation treatment for *Anastrepha ludens* (2009)
- PT 2: Irradiation treatment for *Anastrepha obliqua* (2009)
- PT 3: Irradiation treatment for *Anastrepha serpentina* (2009)
- PT 4: Irradiation treatment for *Bactrocera jarvisi* (2009)
- PT 5: Irradiation treatment for *Bactrocera tryoni* (2009)
- PT 7: Irradiation treatment for fruit flies of the family Tephritidae generic (2009)
- PT 14: Irradiation treatment for *Ceratitis capitata* (2011)

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3 Hallman, G J. 2001b. Irradiation as a Quarantine Treatment. In: R. Molins (ed) Food Irradiation: Principles and Applications. Wiley Interscience, New York, pp. 113-130.
Hallman, G J. 2004a. Irradiation Disinfestation of Apple Maggot (Diptera: Tephritidae) in Hypoxic and Low-Temperature Storage. Journal of Economic Entomology, 97(4), 1245-8.
Hallman, G.J. 2004b. Ionizing irradiation quarantine treatment against Oriental fruit moth (Lepidoptera: Tortricidae) in ambient and hypoxic atmospheres. Journal of Economic Entomology, 97: 824−827.

4 Follett, P A, Wall M, and Bailey W, 2013. Influence of modified atmosphere packaging on radiation tolerance in the phytosanitary pest melon fly (Diptera: Tephritidae). J. Econ. Entomol., 106 (5): 2020–2026.
Srimartpirom M, Burikam I, Limohpasmanee W, Kongratarporn T, Thannarin T, Bunsiri A, and Follett PA. 2018. Low-Dose Irradiation With Modified Atmosphere Packaging for Mango Against the Oriental Fruit Fly (Diptera: Tephritidae). Journal of Economic Entomology 111(1): 135 – 140.
Follett P A., Swedman A, and Mackey B. 2018. Effect of Low-Oxygen Conditions Created by Modified Atmosphere Packaging on Radiation Tolerance in Drosophila suzukii (Diptera: Drosophilidae) in Sweet Cherries. Journal of Economic Entomology 111(1): 141 – 145.

2) *note* that the ink amendments will be implemented into the language versions of the concerned standards as resources permit.
3) *agree* that, once the Secretariat has applied the ink amendments, the previous versions of the standards are replaced by the newly noted versions.

**Title: Ink amendments Attachment 1** *(English only)*

## Effects of low oxygen on irradiation efficacy

*(Prepared by Ms Vanessa S. Dias[1] and Mr Guy Hallman as requested by the TPPT in July 2019)*

### 1. Background

Modified atmosphere storage consisting of low levels of oxygen is often used to enable fresh and minimally processed foods to maintain visual, textural and nutritional appeal and prolong shelf life.

Currently most adopted phytosanitary irradiation (PI) treatments are restricted to commodities that have not been stored in modified atmosphere and contain the disclaimer: "This irradiation treatment should not be applied to fruit and vegetables stored in modified atmospheres" because research has indicated that irradiation in the presence of low oxygen might reduce efficacy.

It was proposed to review the effect of modified atmosphere on the efficacy of PI treatments as the United States Department of Agriculture, Animal and Plant Health Inspection Service (USDA-APHIS) has recently relaxed their restrictions on the use of PI on fruits and vegetables in low oxygen storage.

In this endeavour the literature was reviewed and the FAO/IAEA/USDA Project on Phytosanitary Treatment carried out research on the effect of low oxygen storage on efficacy of PI against tephritid fruit flies. This document reports the results of that research.

### 2. Objective

To evaluate if low-oxygen conditioning can increase the radiotolerance of third-instar larvae of *Anastrepha fraterculus, A. ludens, Bactrocera dorsalis,* and *Ceratitis capitata* reared in mangoes or mandarins.

### 3. Methodology

**Tephritids:** *Anastrepha fraterculus* (Argentina), *A. ludens* (Mexico), *B. dorsalis* (Kenya), and *C. capitata* (Argentina) colonies were used in our study. All colonies were maintained at the Insect Pest Control Laboratory (IPCL) of the Joint FAO/ IAEA Division of Nuclear Techniques in Food and Agriculture in Seibersdorf, Austria.

**Fruit Infestation:** Only naturally infested mangoes and mandarins were used in our experiments. To prevent fruit contamination and decrease larval mortality due to fungi infection, multiple sanitization measures were applied before and after infestation. Before infestation, fruits were washed, rinsed, soaked for 15 min in antifungal solution (4% sodium benzoate and 1% sodium hypochlorite), and re-rinsed. Natural infestation consisted of placing pre-sanitized fruits into a screen-mesh cage (45 × 45 × 45 cm) containing sexually mature insects. Mangoes were infested by either *A. fraterculus* or *A. ludens* for up to 6h and mandarins were infested by either *B. dorsalis* or *C. capitata* for up to 2h. After infestation, a second sanitization round was applied to all infested fruits to reduce fungi development. Following the re-sanitization procedures after infestation, each fruit was individually numbered with a black permanent marker, weighted, and its perimeter measured. Subsequently, infested fruit were incubated for developing larvae reach the 3rd instar stage. Third instars were used in all experiments because they were the most radiotolerant stage for tephritid fruit fly eggs and larvae (Hallman *et al*., 2010).

**Low-oxygen Treatments:** Prior to irradiation, infested mangoes were placed individually in a plastic chamber (13.0 cm diameter × 18.5 cm high) built with a Lock & Lock lid containing four sides interlocking, one plastic luer-lock valve attached to the left side of the lid, and a hole (0.6 cm diameter)

[1] Joint FAO /IAEA Division of Nuclear Techniques in Food and Agriculture

placed in the right side of the lid to allow the inside air to be expelled during gas flushing. After gas flushing, the top hole placed in the mangoes' chamber was sealed with an adhesive septum (0.6 cm diameter). Similarly, two infested mandarins were placed inside a plastic chamber (12.5 cm diameter × 19.5 cm high) constructed with a screw lid on the top sealed with vacuum grease, two plastic luer-lock valves attached to the bottom and up sides for gas flushing, and a hole (0.6 cm) covered by an adhesive septum (1.5 cm diameter) placed on the top side to allow for further determination of the gas concentration inside the chamber. Before gas flushing, three pieces (1 cm x 1 cm) of Gafchromic® films were positioned under and above each mandarin or mango that was later treated with ionizing radiation, allowing the absorbed irradiation dose to be measured at different positions. Hypoxia (5-7% $O_2$, ~16% $CO_2$) and severe-hypoxia (< 1% $O_2$, ~20% $CO_2$) treatments were achieved by flushing the plastic chambers with gas mixtures containing argon, carbon dioxide, nitrogen and oxygen at different concentrations for 2 to 3 minutes. These gas mixtures simulated the possible mild to extreme atmospheric conditions during phytosanitary irradiation of controlled atmospheres, in which oxygen can be partially or completely replaced by carbon dioxide due to fruit respiration. Oxygen and carbon dioxide concentrations were monitored hourly using a CheckMate 3 gas analyzer (Dansensor, Denmark). Infested mandarins and mangoes were either conditioned under low-oxygen atmospheres (hypoxia or severe-hypoxia) for six hours or kept under ambient air (normoxia) before irradiation.

**Irradiation Treatments:** Mandarins and mangoes infested with third-instar larvae treated with a given atmospheric regime were irradiated in a Gammacell 220 (MDS Nordion, Ottawa, Canada) (dose rates ranging from 75 to 95 $Gy.min^{-1}$) located at the IPCL in Seibersdorf, Austria. Irradiation of infested fruits with gamma rays covered a range of doses with sub-lethal and lethal effects for each fruit fly species. Briefly, mandarins infested with *B. dorsalis* larvae were irradiated at 30, 40, 80, 116, and 150 Gy. Mandarins infested with *C. capitata* larvae were irradiated at 20, 30, 50, 70, and 100 Gy. Mangoes infested with either *A. fraterculus* or *A. ludens* larvae were irradiated at 25, 35, 50, and 70 Gy. Non-irradiated fruits (normoxia, hypoxia, and severe-hypoxia controls) were handled similarly to irradiated fruits. Absorbed dose was verified using HD-V2 Gafchromic® film placed in three levels for mandarins (bottom, middle, and top) or two levels for mangoes (bottom and top). HD-V2 films were read through an optical density meter (DoseReader 4, RadGen®) 24 h after exposure.

**Post-treatment Evaluations:** After treatment, each fruit was individually labelled and placed in a plastic container. Fruit were dissected within seven days after treatment, and the number of puparia and dead and live third-instars were recorded. Treatment efficacy was determined by prevention of adult emergence.

## 4. Results

Our results suggest that hypoxic and severe-hypoxic conditioning before and during irradiation can increase the emergence of *A. fraterculus* (Table 1), *A. ludens* (Table 2), B. *dorsalis* (Table 3), and *C. capitata* (Table 4) only at low doses of gamma radiation. At high doses of irradiation, low-oxygen conditioning treatments did not increase the emergence rates of any fruit fly species evaluated. Dosimetry, oxygen and carbon dioxide measures were systematically obtained for all mangos and mandarins treated with radiation and modified atmosphere (Table S1 and Table S2).

**Table 1:** Numbers of replicates, larvae per fruit, treated larvae, dead insects, and adult emergence (mean ± SE) of *Anastrepha fraterculus* third instars irradiated at different doses in normoxia, hypoxia, and severe-hypoxia atmospheres

| Atmospheric conditions | Nominal irradiation dose (Gy) | Replicates | No. larvae per fruit (mean ± SE) | Total no. larvae treated | Total no. insects dead | Adult emergence (%) |
|---|---|---|---|---|---|---|
| Normoxia (21% $O_2$, 0% $CO_2$) | 0 (control) | 35 | 178 ± 33 | 9119 | 2194 | 74.27 ± 2.81 |
| | 25 | 31 | 210 ± 28 | 6521 | 6354 | 1.96 ± 0.43 |
| | 35 | 12 | 206 ± 46 | 2483 | 2476 | 0.14 ± 0.09 |
| | 50 | 17 | 130 ± 24 | 2224 | 2224 | 0.00 ± 0.00 |

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|---|---|---|---|---|---|---|
| | 70 | 29 | 201 ± 31 | 5835 | 5835 | 0.00 ± 0.00 |
| Hypoxia (5.51 ± 0.06% $O_2$, 15.73 ± 0.22% $CO_2$)) | 0 (control) | 34 | 218 ± 35 | 7440 | 1139 | 79.50 ± 2.61 |
| | 25 | 33 | 264 ± 36 | 8724 | 8327 | 4.86 ± 1.00 |
| | 35 | 10 | 327 ± 94 | 3275 | 3274 | 0.25 ± 0.25 |
| | 50 | 11 | 223 ± 65 | 2462 | 2462 | 0.00 ± 0.00 |
| | 70 | 32 | 195 ± 29 | 6251 | 6251 | 0.00 ± 0.00 |
| Severe-hypoxia (0.35 ± 0.02% $O_2$, 22.28 ± 0.18% $CO_2$) | 0 (control) | 41 | 160 ± 25 | 6581 | 1293 | 70.14 ± 3.62 |
| | 25 | 28 | 260 ± 48 | 7290 | 4871 | 29.28 ± 3.20 |
| | 35 | 12 | 225 ± 78 | 2701 | 2625 | 4.42 ± 2.78 |
| | 50 | 17 | 136 ± 27 | 2318 | 2318 | 0.00 ± 0.00 |
| | 70 | 33 | 220 ± 39 | 7278 | 7278 | 0.00 ± 0.00 |

**Table 2:** Numbers of replicates, larvae per fruit, treated larvae, dead insects, and adult emergence (mean ± SE) of *Anastrepha ludens* third instars irradiated at different doses in normoxia, hypoxia, and severe-hypoxia atmospheres

| Atmospheric conditions | Nominal irradiation dose (Gy) | Replicates | No. larvae per fruit (mean ± SE) | Total no. larvae treated | Total no. insects dead | Adult emergence (%) |
|---|---|---|---|---|---|---|
| Normoxia (21% $O_2$, 0% $CO_2$) | 0 (control) | 30 | 98 ± 14 | 6194 | 1799 | 68.00 ± 3.49 |
| | 25 | 38 | 231 ± 30 | 8797 | 8602 | 1.65 ± 0.48 |
| | 35 | 25 | 163 ± 27 | 4088 | 4082 | 0.07 ± 0.05 |
| | 50 | 24 | 110 ± 29 | 2751 | 2751 | 0.00 ± 0.00 |
| | 70 | 20 | 100 ± 26 | 1990 | 1990 | 0.00 ± 0.00 |
| Hypoxia (5.20 ± 0.07% $O_2$, 15.81 ± 0.19% $CO_2$) | 0 (control) | 18 | 208 ± 44 | 3757 | 648 | 81.29 ± 2.60 |
| | 25 | 25 | 221 ± 38 | 5539 | 5261 | 5.55 ± 2.11 |
| | 35 | 12 | 238 ± 65 | 2864 | 2863 | 0.12 ± 0.12 |
| | 50 | 13 | 249 ± 62 | 2996 | 2996 | 0.00 ± 0.00 |
| | 70 | 14 | 176 ± 49 | 2468 | 2468 | 0.00 ± 0.00 |
| Severe-hypoxia (0.32 ± 0.03% $O_2$, 21.40 ± 0.13% $CO_2$) | 0 (control) | 24 | 161 ± 30 | 3863 | 1247 | 64.72 ± 4.47 |
| | 25 | 27 | 98 ± 22 | 2640 | 1891 | 31.21 ± 5.41 |
| | 35 | 16 | 144 ± 35 | 2315 | 2293 | 0.97 ± 0.59 |
| | 50 | 19 | 170 ± 36 | 3237 | 3233 | 0.08 ± 0.08 |
| | 70 | 19 | 128 ± 38 | 2435 | 2435 | 0.00 ± 0.00 |

**Table 3:** Numbers of replicates, larvae per fruit, treated larvae, dead insects, and dult emergence (mean ± SE) of *Bactrocera dorsalis* third instars irradiated at different doses in normoxia, hypoxia, and severe-hypoxia atmospheres

| Atmospheric conditions | Nominal irradiation dose (Gy) | Replicates | No. larvae per fruit (mean ± SE) | Total no. larvae treated | Total no. insects dead | Adult emergence (%) [mean ± SE] |
|---|---|---|---|---|---|---|
| Normoxia (21% $O_2$, 0% $CO_2$) | 0 (control) | 35 | 123 ± 10 | 18397 | 3057 | 83.73 ± 1.47 |
| | 30 | 8 | 78 ± 17 | 1172 | 1105 | 4.79 ± 1.56 |
| | 40 | 16 | 119 ± 17 | 4899 | 4787 | 3.30 ± 1.00 |
| | 80 | 8 | 143 ± 33 | 2289 | 2288 | 0.01 ± 0.01 |
| | 116 | 52 | 70 ± 8 | 6405 | 6405 | 0.00 ± 0.00 |

| | | | | | | |
|---|---|---|---|---|---|---|
| | 150 | 15 | 187 ± 31 | 6175 | 6175 | 0.00 ± 0.00 |
| Hypoxia (5.27 ± 0.04% $O_2$, 14.75 ± 0.08% $CO_2$) | 0 (control) | 23 | 86 ± 11 | 4050 | 844 | 82.40 ± 2.75 |
| | 30 | 16 | 141 ± 25 | 4240 | 3872 | 12.81 ± 3.28 |
| | 40 | 19 | 133 ± 21 | 4523 | 4264 | 9.65 ± 2.64 |
| | 80 | 16 | 119 ± 22 | 3699 | 3699 | 0.00 ± 0.00 |
| | 116 | 36 | 80 ± 12 | 4967 | 4967 | 0.00 ± 0.00 |
| | 150 | 14 | 66 ± 10 | 1852 | 1852 | 0.00 ± 0.00 |
| Severe-hypoxia (0.35 ± 0.02% $O_2$, 21.65 ± 0.09% $CO_2$) | 0 (control) | 45 | 108 ± 10 | 11168 | 2269 | 81.00 ± 2.22 |
| | 30 | 8 | 94 ± 20 | 1509 | 816 | 40.61 ± 6.14 |
| | 40 | 16 | 89 ± 14 | 3820 | 3143 | 18.12 ± 3.09 |
| | 80 | 7 | 76 ± 23 | 1069 | 1069 | 0.00 ± 0.00 |
| | 116 | 36 | 63 ± 9 | 4511 | 4511 | 0.00 ± 0.00 |
| | 150 | 15 | 141 ± 20 | 3938 | 3938 | 0.00 ± 0.00 |

**Table 4:** Numbers of replicates, larvae per fruit, treated larvae, dead insects, and adult emergence (mean ± SE) of *C. capitata* third instars irradiated at different doses in normoxia, hypoxia, and severe-hypoxia atmospheres

| Atmospheric conditions | Nominal irradiation dose (Gy) | Replicates | No. larvae per fruit (mean ± SE) | Total no. larvae treated | Total no. insects dead | Adult emergence (%) [mean ± SE] |
|---|---|---|---|---|---|---|
| Normoxia (21% $O_2$, 0% $CO_2$) | 0 (control) | 21 | 67 ± 7 | 5901 | 1293 | 78.31 ± 2.51 |
| | 20 | 4 | 76 ± 21 | 529 | 514 | 8.94 ± 4.50 |
| | 30 | 12 | 78 ± 15 | 2590 | 2470 | 2.76 ± 1.14 |
| | 50 | 5 | 73 ± 36 | 654 | 653 | 0.13 ± 0.13 |
| | 70 | 6 | 86 ± 21 | 1031 | 1031 | 0.00 ± 0.00 |
| | 100 | 39 | 39 ± 5 | 2669 | 2669 | 0.00 ± 0.00 |
| Hypoxia (5.17 ± 0.05% $O_2$, 15.60 ± 0.08% $CO_2$) | 0 (control) | 9 | 63 ± 19 | 1004 | 267 | 74.77 ± 3.10 |
| | 20 | 10 | 49 ± 14 | 977 | 796 | 22.23 ± 4.56 |
| | 30 | 9 | 32 ± 9 | 543 | 489 | 8.72 ± 3.78 |
| | 50 | 10 | 62 ± 16 | 1248 | 1247 | 0.13 ± 0.13 |
| | 70 | 11 | 124 ± 27 | 2727 | 2727 | 0.00 ± 0.00 |
| | 100 | 19 | 59 ± 9 | 2138 | 2138 | 0.00 ± 0.00 |
| Severe-hypoxia (0.39 ± 0.03% $O_2$, 21.47 ± 0.08% $CO_2$) | 0 (control) | 29 | 46 ± 6 | 2462 | 424 | 77.61 ± 3.59 |
| | 20 | 10 | 66 ± 18 | 1320 | 591 | 54.20 ± 4.85 |
| | 30 | 12 | 44 ± 9 | 1053 | 846 | 14.81 ± 3.88 |
| | 50 | 10 | 44 ± 11 | 880 | 880 | 0.00 ± 0.00 |
| | 70 | 10 | 70 ± 18 | 1334 | 1334 | 0.00 ± 0.00 |
| | 100 | 33 | 27 ± 4 | 1550 | 1550 | 0.00 ± 0.00 |

## 5. Discussion

After discussion of these findings the TPPT recommended that the disclaimer "*This irradiation treatment should not be applied to fruit and vegetables stored in modified atmospheres*" be removed from all PI treatments against tephritids. It was not considered to remove the disclaimer for other insects because insufficient research has been conducted with all other insects.

## 6. References cited

**Hallman, G.J., Levang-Brilz, N.M., Zettler, J.L., Winborne, I.C**., 2010. Factors affecting ionizing radiation phytosanitary treatments, and implications for research and generic treatments. J. Econ. Entomol. 103: 1950–1963.

## Supplementary Material

**Table S1:** Dosimetry, oxygen ($O_2$) and carbon dioxide ($CO_2$) levels for infested mangoes irradiated at different doses in normoxia, hypoxia, and severe-hypoxia atmospheres

| Tephritid species | Atmospheric conditions | $O_2$ (%) [mean ± SE] | $CO_2$ (%) [mean ± SE] | Nominal dose | Absorbed dose (mean ± SE) [min, max] | |
|---|---|---|---|---|---|---|
| | | | | | Bottom (Gy) | Up (Gy) |
| *Anastrepha fraterculus* | Normoxia | 21 | 0 | 0 Gy | - | |
| | Hypoxia | 5.64 ± 0.11 | 15.55 ± 0.40 | | | |
| | Severe-hypoxia | 0.32 ± 0.04 | 22.47 ± 0.35 | | | |
| | Normoxia | 21 | 0 | 25 Gy | 24.47 ± 0.56 [21, 34] | 27.42 ± 0.59 [21, 29] |
| | Hypoxia | 5.52 ± 0.11 | 15.91 ± 0.48 | | 24.47 ± 0.56 [21, 30] | 26.19 ± 0.33 [20, 30] |
| | Severe-hypoxia | 0.34 ± 0.06 | 22.41 ± 0.36 | | 25.52 ± 0.82 [22, 39] | 27.42 ± 0.59 [21, 34] |
| | Normoxia | 21 | 0 | 35 Gy | 37.74 ± 1.15 [32, 44] | 34.20 ± 0.68 [30, 38] |
| | Hypoxia | 4.98 ± 0.17 | 17.38 ± 0.72 | | 39.68 ± 0.90 [36, 44] | 37.85 ± 1.28 [32, 45] |
| | Severe-hypoxia | 0.40 ± 0.03 | 21.46 ± 0.31 | | 39.89 ± 1.37 [31, 49] | 38.84 ± 1.64 [31, 48] |
| | Normoxia | 21 | 0 | 50 Gy | 55.51 ± 1.23 [45, 69] | 52.57 ± 1.10 [43, 59] |
| | Hypoxia | 5.01 ± 0.12 | 16.38 ± 0.65 | | 57.10 ± 0.63 [53, 60] | 54.22 ± 1.09 [48, 60] |
| | Severe-hypoxia | 0.41 ± 0.04 | 22.33 ± 0.44 | | 57.65 ± 0.89 [52, 66] | 52.46 ± 1.37 [40, 64] |
| | Normoxia | 21 | 0 | 70 Gy | 66.01 ± 1.12 [59, 88] | 73.51 ± 1.01 [59, 84] |
| | Hypoxia | 5.70 ± 0.12 | 15.00 ± 0.36 | | 67.18 ± 1.28 [57, 85] | 75.30 ± 0.89 [62, 85] |
| | Severe-hypoxia | 0.33 ± 0.05 | 22.19 ± 0.39 | | 69.66 ± 1.33 [59, 88] | 74.88 ± 0.88 [57, 85] |
| *Anastrepha ludens* | Normoxia | 21.0 | 0.0 | 0 Gy | - | |
| | Hypoxia | 5.32 ± 0.17 | 15.89 ± 0.25 | | | |
| | Severe-hypoxia | 0.29 ± 0.04 | 21.40 ± 0.29 | | | |
| | Normoxia | 21 | 0 | 25 Gy | 25.26 ± 0.33 [21, 30] | 26.41 ± 0.33 [24, 33] |
| | Hypoxia | 5.23 ± 0.12 | 15.68 ± 0.33 | | 26.78 ± 0.57 [21, 31] | 26.82 ± 0.44 [24, 32] |

| | | | | | |
|---|---|---|---|---|---|
| Severe-hypoxia | 0.33 ± 0.07 | 21.30 ± 0.17 | | 28.86 ± 0.62 [21, 35] | 26.76 ± 0.44 [22, 34] |
| Normoxia | 21 | 0 | | 34.43 ± 0.53 [30, 40] | 32.41 ± 0.71 [26, 42] |
| Hypoxia | 5.05 ± 0.10 | 16.77 ± 0.71 | 35 Gy | 40.76 ± 1.11 [37, 48] | 37.74 ± 1.15 [32, 44] |
| Severe-hypoxia | 0.28 ± 0.05 | 21.49 ± 0.37 | | 36.99 ± 1.27 [47, 67] | 34.59 ± 1.21 [27, 42] |
| Normoxia | 21 | 0 | | 55.34 ± 1.01 [47, 67] | 51.29 ± 1.24 [37, 67] |
| Hypoxia | 4.90 ± 0.08 | 15.96 ± 0.55 | 50 Gy | 56.97 ± 1.19 [49, 63] | 52.40 ± 0.96 [47, 58] |
| Severe-hypoxia | 0.27 ± 0.04 | 21.33 ± 0.33 | | 56.01 ± 1.37 [47, 67] | 51.93 ± 1.18 [42, 67] |
| Normoxia | 21 | 0 | | 68.80 ± 1.72 [56, 87] | 74.56 ± 1.25 [63, 85] |
| Hypoxia | 5.40 ± 0.16 | 15.00 ± 0.43 | 70 Gy | 72.87 ± 2.19 [63, 88] | 75.35 ± 0.94 [70, 84] |
| Severe-hypoxia | 0.40 ± 0.08 | 21.56 ± 0.38 | | 70.95 ± 1.70 [57, 84] | 73.95 ± 1.27 [60, 81] |

**Table S2**: Dosimetry, oxygen ($O_2$) and carbon dioxide ($CO_2$) levels for infested mandarins irradiated at different doses in normoxia, hypoxia, and severe-hypoxia atmospheres

| Tephritid species | Atmospheric conditions | $O_2$ (%) [mean ± SE] | $CO_2$ (%) [mean ± SE] | Nominal dose | Absorbed dose (mean ± SE) [min, max] | | |
|---|---|---|---|---|---|---|---|
| | | | | | Bottom (Gy) | Middle (Gy) | Up (Gy) |
| *Bactrocera dorsalis* | Normoxia | 21 | 0 | | | | |
| | Hypoxia | 5.23 ± 0.09 | 14.76 ± 0.21 | 0 Gy | - | | |
| | Severe-hypoxia | 0.28 ± 0.03 | 21.74 ± 0.13 | | | | |
| | Normoxia | 21 | 0 | | 34.48 ± 1.21 [28, 41] | 37.57 ± 1.10 [31, 43] | 38.03 ± 1.55 [33, 45] |
| | Hypoxia | 5.34 ± 0.15 | 15.03 ± 0.27 | 30 Gy | 35.06 ± 0.56 [31, 42] | 37.85 ± 0.68 [31, 44] | 35.86 ± 0.72 [26, 41] |
| | Severe-hypoxia | 0.56 ± 0.08 | 21.41 ± 0.33 | | 35.95 ± 0.87 [32, 40] | 37.99 ± 0.90 [32, 42] | 38.44 ± 1.07 [33, 45] |
| | Normoxia | 21 | 0 | | 46.47 ± 0.89 [39, 59] | 51.73 ± 1.46 [42, 62] | 46.90 ± 0.78 [39, 56] |
| | Hypoxia | 5.36 ± 0.09 | 14.54 ± 0.15 | 40 Gy | 45.72 ± 0.63 [39, 52] | 46.70 ± 0.67 [40, 52] | 45.42 ± 0.74 [35, 54] |
| | Severe-hypoxia | 0.38 ± 0.04 | 21.50 ± 0.14 | | 48.40 ± 0.99 [41, 59] | 55.43 ± 1.28 [41, 67] | 46.68 ± 0.85 [38, 56] |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| | Normoxia | 21 | 0 | | 72.58 ± 2.23 [65, 96] | 82.65 ± 1.46 [71, 90] | 77.33 ± 1.65 [69, 91] |
| | Hypoxia | 5.10 ± 0.09 | 14.54 ± 0.15 | 80 Gy | 79.73 ± 1.70 [62, 94] | 96.12 ± 0.83 [77, 94] | 79.78 ± 1.69 [66, 96] |
| | Severe-hypoxia | 0.68 ± 0.08 | 20.90 ± 0.17 | | 78.97 ± 2.75 [63, 90] | 82.09 ± 1.38 [74, 89] | 82.94 ± 2.60 [70, 98] |
| | Normoxia | 21 | 0 | | 114.54 ± 0.75 [101, 131] | 121.63 ± 0.72 [101, 136] | 110.06 ± 0.99 [94, 131] |
| | Hypoxia | 5.27 ± 0.08 | 14.52 ± 0.15 | 116 Gy | 117.50 ± 1.26 [99, 135] | 123.12 ± 1.45 [95, 149] | 121.33 ± 2.04 [85, 158] |
| | Severe-hypoxia | 0.37 ± 0.03 | 21.49 ± 0.15 | | 118.41 ± 0.84 [104, 132] | 121.63 ± 0.72 [114, 152] | 117.82 ± 1.79 [98, 152] |
| | Normoxia | 21 | 0 | | 148.99 ± 0.85 [142, 164] | 155.81 ± 0.49 [142, 173] | 136.68 ± 1.63 [120, 184] |
| | Hypoxia | 5.39 ± 0.15 | 14.84 ± 0.22 | 150 Gy | 152.01 ± 3.02 [127, 186] | 164.89 ± 3.11 [143, 194] | 159.50 ± 3.62 [130, 197] |
| | Severe-hypoxia | 0.21 ± 0.04 | 22.45 ± 0.50 | | 151.58 ± 1.27 [142, 161] | 156.10 ± 1.48 [142, 173] | 136.80 ± 3.18 [123, 157] |
| *Ceratitis capitata* | Normoxia | 21 | 0 | | | | |
| | Hypoxia | 5.11 ± 0.11 | 15.54 ± 0.29 | 0 Gy | - | | |
| | Severe-hypoxia | 0.30 ± 0.06 | 21.63 ± 0.14 | | | | |
| | Normoxia | 21 | 0 | | 20.63 ± 0.76 [18, 23] | 21.97 ± 0.85 [19, 25] | 20.14 ± 0.68 [19, 23] |
| | Hypoxia | 5.03 ± 0.11 | 15.98 ± 0.21 | 20 Gy | 21.63 ± 0.33 [19, 24] | 23.13 ± 0.57 [19, 28] | 20.06 ± 0.62 [16, 25] |
| | Severe-hypoxia | 0.56 ± 0.03 | 21.26 ± 0.12 | | 21.52 ± 0.42 [19, 25] | 21.59 ± 0.31 [20, 25] | 19.14 ± 0.48 [19, 23] |
| | Normoxia | 21 | 0 | | 32.41 ± 0.47 [26, 38] | 36.38 ± 1.03 [27, 44] | 31.61 ± 0.52 [25, 35] |
| | Hypoxia | 5.18 ± 0.12 | 15.39 ± 0.14 | 30 Gy | 33.14 ± 0.46 [30, 36] | 34.84 ± 0.58 [31, 39] | 33.51 ± 0.95 [25, 38] |
| | Severe-hypoxia | 0.43 ± 0.03 | 21.64 ± 0.31 | | 31.93 ± 0.59 [27, 35] | 34.69 ± 0.69 [27, 40] | 30.82 ± 0.65 [26, 38] |
| | Normoxia | 21 | 0 | | 49.57 ± 0.59 [48, 52] | 53.77 ± 1.06 [50, 58] | 49.57 ± 0.87 [46, 53] |
| | Hypoxia | 5.16 ± 0.15 | 15.92 ± 0.21 | 50 Gy | 53.48 ± 0.99 [47, 60] | 56.27 ± 0.73 [52, 62] | 53.20 ± 1.05 [49, 63] |
| | Severe-hypoxia | 0.59 ± 0.04 | 20.88 ± 0.06 | | 52.06 ± 0.50 [48, 56] | 53.76 ± 0.76 [47, 58] | 55.46 ± 1.07 [45, 65] |
| | Normoxia | 21 | 0 | | 70.62 ± 1.13 [64, 76] | 75.47 ± 0.90 [71, 79] | 71.60 ± 0.98 [67, 76] |
| | Hypoxia | 5.48 ± 0.10 | 15.44 ± 0.17 | 70 Gy | 70.84 ± 0.79 [61, 75] | 75.01 ± 1.19 [65, 88] | 72.95 ± 0.89 [69, 83] |
| | Severe-hypoxia | 0.59 ± 0.03 | 21.06 ± 0.09 | | 70.54 ± 0.86 [63, 75] | 77.09 ± 1.12 [71, 85] | 75.45 ± 0.88 [72, 84] |

| | | | | | | |
|---|---|---|---|---|---|---|
| Normoxia | 21 | 0 | | 101.74 ± 0.73 [85, 118] | 108.69 ± 0.95 [95, 128] | 97.08 ± 0.89 [81, 113] |
| Hypoxia | 5.08 ± 0.08 | 15.46 ± 0.16 | 100 Gy | 104.50 ± 1.39 [91, 123] | 108.91 ± 1.25 [94, 123] | 111.74 ± 1.08 [100, 127] |
| Severe-hypoxia | 0.26 ± 0.03 | 21.68 ± 0.16 | | 103.57 ± 0.78 [93, 120] | 108.16 ± 0.86 [91, 129] | 96.67 ± 1.24 [76, 115] |

## Attachment 02: Ink amendments to irradiation treatments of Tephritid fruit flies in adopted Phytosanitary Treatments (PTs) *(__English only__)*

Table 1: Ink amendments to remove the restriction of the use of the irradiation treatment to commodities that have been stored in modified atmosphere

| ISPM | CURRENT TEXT | PROPOSED INK AMENDMENT |
|---|---|---|
| ISPM 28 (*Phytosanitary treatments for regulated pests*) - PT 1 (*Irradiation treatment for* Anastrepha ludens) | “This irradiation treatment should not be applied to fruits and vegetables stored in modified atmospheres.” […] | ~~“This irradiation treatment should not be applied to fruits and vegetables stored in modified atmospheres.”~~ […] |
| ISPM 28 (*Phytosanitary treatments for regulated pests*) - PT 2 (*Irradiation treatment for* Anastrepha obliqua) | “This irradiation treatment should not be applied to fruits and vegetables stored in modified atmospheres.” […] | ~~“This irradiation treatment should not be applied to fruits and vegetables stored in modified atmospheres.”~~ […] |
| ISPM 28 (*Phytosanitary treatments for regulated pests*) - PT 3 (*Irradiation treatment for* Anastrepha serpentina) | . “This irradiation treatment should not be applied to fruits and vegetables stored in modified atmospheres.” […] | ~~“This irradiation treatment should not be applied to fruits and vegetables stored in modified atmospheres.”~~ […] |
| ISPM 28 (*Phytosanitary treatments for regulated pests*) - PT 4 (*Irradiation treatment for* Bactrocera jarvisi) | . “This irradiation treatment should not be applied to fruits and vegetables stored in modified atmospheres.” […] | ~~“This irradiation treatment should not be applied to fruits and vegetables stored in modified atmospheres.”~~ […] |
| ISPM 28 (*Phytosanitary treatments for regulated pests*) - PT 5 (*Irradiation treatment for* Bactrocera tryoni) | “This irradiation treatment should not be applied to fruits and vegetables stored in modified atmospheres.” […] | ~~“This irradiation treatment should not be applied to fruits and vegetables stored in modified atmospheres.”~~ […] |
| ISPM 28 (*Phytosanitary treatments for regulated pests*) - PT 7 (Irradiation treatment for fruit flies of the family Tephritidae (generic)) | “This irradiation treatment should not be applied to fruits and vegetables stored in modified atmospheres.” […] | ~~“This irradiation treatment should not be applied to fruits and vegetables stored in modified atmospheres.”~~ […] |
| ISPM 28 (*Phytosanitary treatments for regulated pests*) - PT 14 (*Irradiation treatment for* Ceratitis capitata) | . “This irradiation treatment should not be applied to fruits and vegetables stored in modified atmospheres.” […] | ~~“This irradiation treatment should not be applied to fruits and vegetables stored in modified atmospheres.”~~ […] |