

Original research

## Evaluation of the effectiveness of ozone gas and Gamma radiation on the *Oryzaephilus Surinamensis* (L.) Coleoptera: Cucujidae for protection stored dates.

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### Abstract:

The present work conducted to determine the effect of ozone gas concentrations and gamma radiation doses that would serve as phytosanitary treatment against the saw – toothed grain beetle *Oryzaephilus surinamensis* (L.) (Coleoptera: Cucujidae) adults on two varieties of stored dates. The results clearly that the LC<sub>50</sub> to ozone gas against *O. surinamensis* adults to 500, 1000, 2000 and 3000 ppm were 2.79, 1.06, 1.05 and 0.46 and 5.18, 1.37, 1.39 and 0.64 days for dates (Bermuda and Kandella varieties), respectively. Also, the results showed that the treatment *O. surinamensis* adults on mentioned variety with nine doses of gamma radiation (10, 20, 40, 50, 60, 100, 200, 400 and 800 Gy) compared with untreated insects. The results indicated that mortality of *O. surinamensis* increased with increasing of doses and exposure time. While, at 800 Gy exposed treatment the mortality % was 91.11 % and 98.89 % for *O. surinamensis* adults at Bermuda and Kandella varieties, respectively.

**Keywords:** stored dates, ozone gas, Gamma radiation, *O. Surinamensis*.

### 1- Introduction

The date palm (*Phoenix dactylifera* L.) plays an important economic role for foreign trade in many countries that are located in arid and semi-arid regions of the world. Dates are rich in certain nutrients and provide a good source of energy, due to their high carbohydrate content (70 - 80%). Moreover, it contains protein (2.30 - 5.60%), fat (0.20-0.50%), dietary fiber (6.40-11.50%), minerals about (0.10- 916 mg/100 g dry weight) and vitamins (C, B1, B2, B3 and A) (AlShahib and Marshal, 2003). In the developing countries, dry dates were attacked by great stored insects during storage causing the loss of their quality and quantity (Talukder *et al.*, 2004). Stored products of agriculture are attacked by more than 1200 species of pests (Rajendran, 2002).

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The saw-toothed grain beetle, *O. surinamensis*, is one of the pests known to attack stored foods and it is cosmopolitan in distribution and is likely to be found in almost any stored food of vegetable origin (Panagiotakopulu and Buckland, 2017). Recently, many researchers are working on developing less risky materials against insect pests in warehouses as non-chemical alternative avenues. Currently, ozone gas is used as ecofriendly new alternative avenue to control insect pests (Mahroof *et al.*, 2018). Some non-chemical and non-thermal methods have been encountered in stored insects notably, microwave heating, infrared heating, ozone treatment, ultraviolet and cold plasma (Paul *et al.*, 2020 & Srivastava and Mishra, 2021). Gamma irradiation is considered one of the successful control methods against various insect pest and also it was recommended as an effective technique against stored product insects (Ayvaz and Tuncbilek, 2006). It is widely used as disinfestation treatment in many regions. The use of irradiation treatments as disinfestation treatment depends on kill or inhibit the development of immature stages in irradiated products (Ayvaz and Yilmaz, 2015 & Hassan *et al.*, 2019). This study aim to evaluate the effectiveness of ozone gas and gamma irradiation on the adults of *O. surinamensis* at dates varieties in Aswan Governorate.

## 2- Materials and Methods

### 2.1.- Insect rearing:

The original cultures of saw-toothed grain, *Oryzaephilus surinamensis* adults was obtained from Stored Grain Research Department, Plant Protection Research Institute, Agricultural Research Center, Egypt. The adults were introduced to Bermuda date variety in glass jars (2 kg. capacity each). These jars were incubated at  $27 \pm 1$  °C and  $65 \pm 5$  % RH for two weeks, then the adults were removed and the dates were kept in the jars under the previous conditions till adult emergence (Darwish, *et al.*, 2019).

### 2.2.-Controlled experiments:

Ozone gas was produced from air using an ozone generator Model OZO 6 VTTLOZO Max Ltd, Shefford, Quebec Canada (OZO Max Ltd, Shefford, Quebec, Canada) from purified extra dry oxygen feed gas at the laboratory of Food Toxicology & Contaminants, National Research Center, Egypt. The amount of ozone output was controlled by a monitor controller having a plug-in sensor onboard which is changed for different ranges of ozone concentration and a belt pan in the monitor-controller allows controlling the concentration in a selected range according to (Darwish, *et al.*, 2019).

#### 2.2.1.- Application Ozone gas on the adult of *O. surinamensis*:

Fifteen pairs of *O. surinamensis* and 50 grams of dates for the types Bermuda and Kandella were placed in cloth bags and closed well with rubber band. These bags were placed in glass jars covered with a rubber stopper. The gas is connected with the ozone unit and the jar by a thin tube. The concentrations used in this experiment were 500, 1000, 2000 and 3000 ppm for 1 hour, 2 hours and 3 hours. Mortality of adults was assessed after 1, 3, 5, 7, 10, 14 days from treatment and after 45 days calculated the number of individuals produced.

### **2.2.2. Irradiation of adults of *O. surinamensis*:**

#### **Irradiation application:**

This work was carried out at the Radiation Unit of the Egyptian atomic Energy authority, Nasr City, Cairo.

The adult instar of *O. surinamensis* was exposed to nine concentrations of radiation (10, 20, 40, 50, 60, 100, 200, 400 and 800 Gy). In a cloth bag, 50 grams from dates of two varieties (Bermouda & Kandella) were placed, in addition to 15 pairs of *O. surinamensis* and they were tied tightly, three replications were made for each type of dates and for each concentration. Mortality of adults was assessed after 1, 3, 5, 7, 10, 14 days from treatment and after 45 days calculated the number of individuals produced.

### **2.2.3. Calculation of mortality & reduction:**

For the evaluation of the effect of ozone gas, and radiation, mortality of all treated stages was corrected according to **Abbott (1925)** as following:

$$\text{Corrected mortality} = \frac{\% \text{ mortality in treatment} - \% \text{ mortality in control}}{100 - \% \text{ mortality in control}}$$

### **2.3.- Statistical analysis of the obtained data:**

The average percent mortality of the tested insects was calculated and corrected using Abbott's formula **Abbott (1925)**. The corrected percentages of mortalities were statistically computed according to the method of **Finney (1971)**. Computed percentage of mortality was plotted versus the corresponding concentrations using Ldp line software program to obtain the toxicity regression lines.

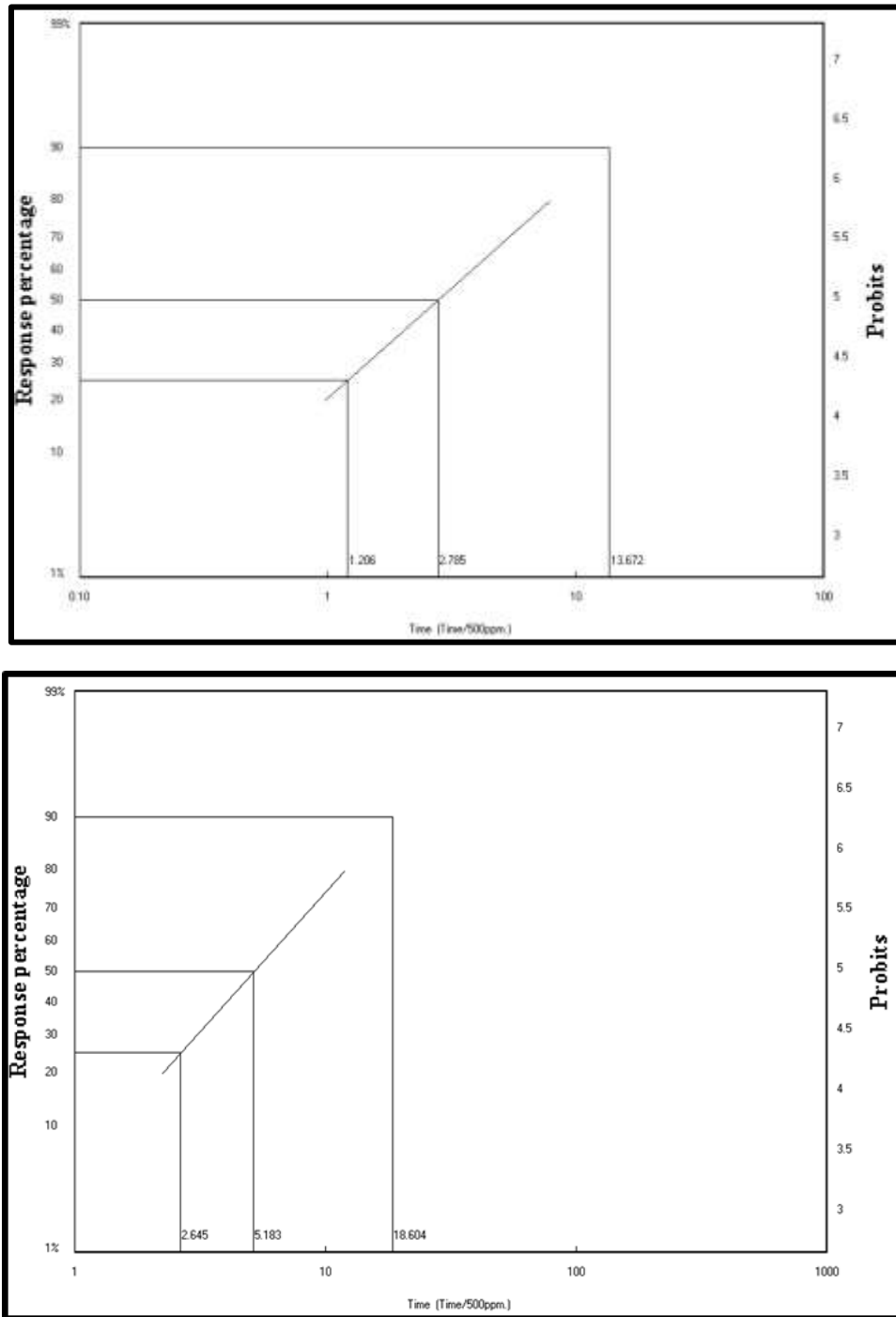
## **3- Results and Discussion**

### **The effect of ozone gas on *O. surinamensis* (L.) adults at 30± 1 °C and 65±5 %RH. with Bermuda and Kandella varieties:**

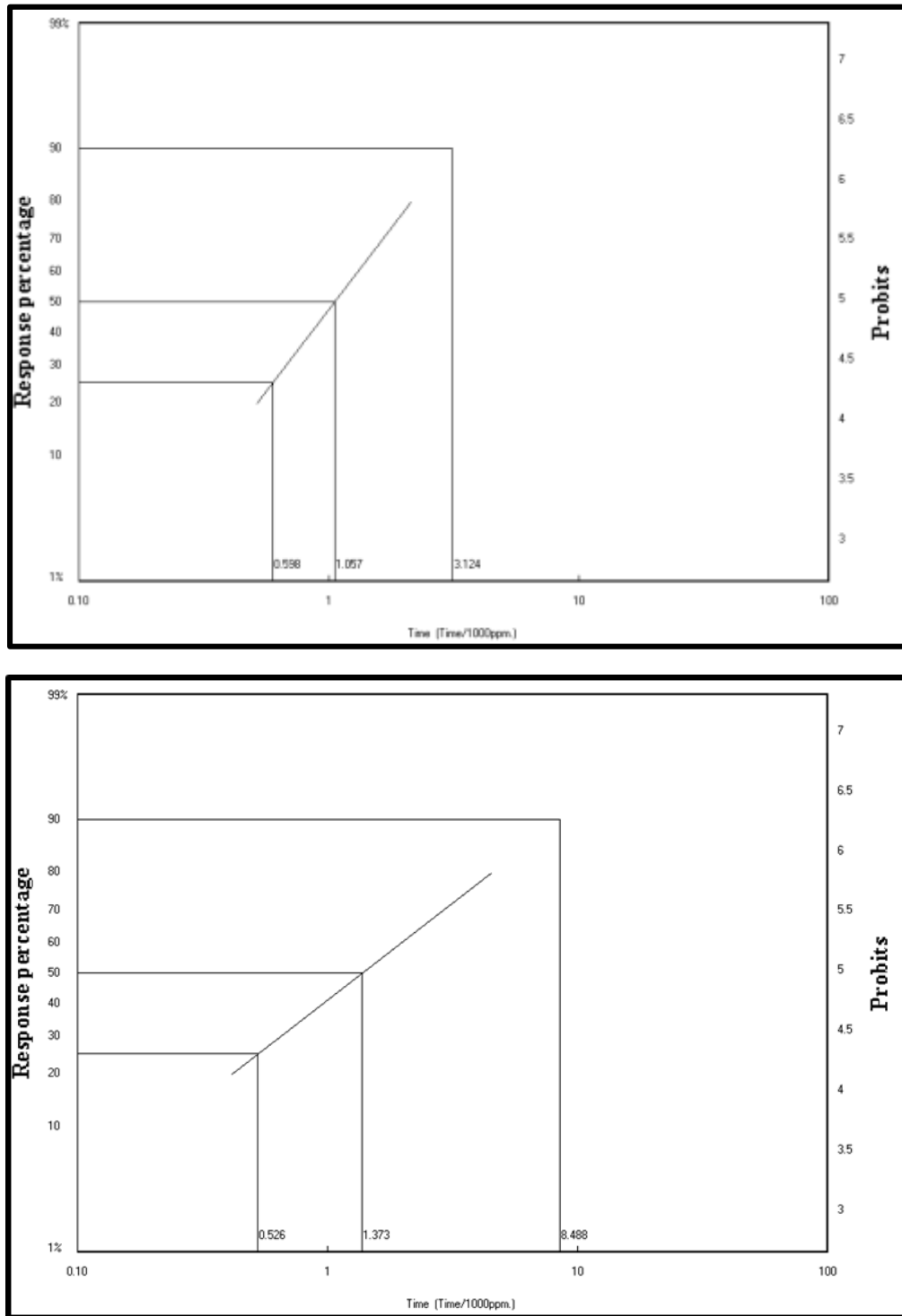
The effect of ozone gas at 500 ppm on *O. surinamensis* (L.) at Bermuda and Kandella varieties date were shown in Table (1) and Fig. (1) the data revealed that the corrected mortality % increased gradually by increasing the exposure time after treatment. The results showed that the corrected mortality at 1 hr. after 1 day were 16.67% and 0.0 % post treatment and the mortality increased after 14 days to 23.33% and 3.33% on Bermuda and Kandella varieties, respectively. While, these mortality values were 40.00 and 46.67 % & 18.89 and 24.44% at 3 hrs. to ozone gas release after one and 14 days on Bermuda and Kandella varieties respectively.

The results obtained from experiments dealing with the effect of ozone gas of *O. surinamensis* at 1000 ppm on Bermuda and Kandella varieties are summarized in Table (2) and Fig. (2) the results showed the corrected mortality % of *O. surinamensis* adults increased with increasing exposure times to ozone gas as well as the days post treatment. The results cleared that mortality of adults were 17.78 and 22.22% & 34.44 and 46.67 % at 1 hr. exposure period after 1 and 14 days post treatment, while at 3 hrs. of ozone gas release. These values of mortality were (90.00

and 94.45 %) & (71.11 and 80.00%) after 1 and 14 days post treatment at 3 hrs. on Bermuda and Kandella varieties, respectively.



**Fig. (1):** Lethal time values and confidence limits of ozone at 500 ppm adults of *O. surinamensis* after three hours for 14 day post treatment.



**Fig (2):** Lethal time values and confidence limits of ozone at 1000 ppm adults of *O. surinamensis* after three hours for 14 day post treatment.

**Table (1):** Mortality of adults of *O. surinamensis* ascending periods (1, 2, 5, 7, 10 and 14 day) after treatment when exposed to 500 ppm of ozone.

Date variety	Exposure time (hours)	Mortality % of <i>O. surinamensis</i> after days					
		1	3	5	7	10	14
Bermuda	One hour	16.67±0.88	17.78±0.88	18.89±1.20	21.11±0.88	21.11±0.88	23.33±1.53
	Two hours	24.44±2.08	30.00±2.08	31.11±1.45	31.11±2.33	31.11±2.33	31.11±2.33
	Three hours	40.00±2.96	45.56±2.96	45.56±3.93	45.56±2.96	45.56±2.96	46.67±3.21
Kandella	One hour	0.00±0.00	1.11 ±0.00	2.22±0.33	2.22±0.33	2.22±0.33	3.33±0.00
	Two hours	16.67±0.58	20.00±0.33	20.00±0.58	22.22±0.67	22.22±0.67	22.22±0.67
	Three hours	18.89±0.88	24.45±0.58	23.33±1.15	23.33±1.15	24.44±1.45	24.44±0.88

**Table (2):** Mortality of adults of *O. surinamensis* ascending periods (1, 2, 5, 7, 10 and 14 day) when exposed to 1000 ppm of ozone.

Date variety	Exposure time (hours)	Mortality % of <i>O. surinamensis</i> after days					
		1	3	5	7	10	14
Bermuda	One hour	17.78±1.20	21.11±1.67	21.11±1.67	21.11±1.67	21.11±1.67	22.22±1.33
	Two hours	67.78±0.88	70.00±1.20	70.00±1.20	70.00±1.20	70.00±1.20	71.11±1.20
	Three hours	90.00±0.58	90.00±1.20	90.00±1.20	90.00±1.20	90.00±1.20	94.45±1.20
Kandella	One hour	34.44±1.33	40.00±1.73	42.22±0.23	44.44±2.60	45.56±2.33	46.67±2.30
	Two hours	48.89±1.20	53.33±1.15	55.56±1.45	56.67±1.73	56.67±1.73	56.67±1.73
	Three hours	71.11±1.67	76.67±0.58	76.67±0.58	77.78±0.67	78.89±0.88	80±0.58

Table (3) and Fig. (3) showed the outcomes of the 2000 ppm ozone gas treatment against adults of *O. surinamense* at 30°C and 65 RH.% the adults mortality were 21.11% and 22.22% at 1 hr. after 1 day post treatment and the mortality increased to 51.11% and 42.22% post 14 days with Bermuda and Kandella varieties, respectively. While, the adults mortality were 84.45% and 57.78% at 3 hrs. after one day these values increased to 94.44% and 72.22% after 14 days post treatment on Bermuda and Kandella varieties, respectively.

**Table (3):** Mortality of adults of *O. surinamensis* ascending periods (1, 2, 5, 7, 10 and 14 day) when exposed to 2000 ppm of ozone.

Date variety	Exposure time(hours)	Mortality % of <i>O. surinamensis</i> after days					
		1	3	5	7	10	14
Bermuda	On hour	67.78±0.67	71.11±0.67	72.22±0.58	73.33±1.00	73.33±1.00	73.33±1.00
	Two hours	67.78±0.88	75.56±0.88	75.56±0.88	75.56±1.53	75.56±1.53	75.56±1.53
	Three hours	88.89±1.86	93.33±1.53	94.44±1.67	94.44±2.89	94.44±2.89	94.44±2.89
Kandella	On hour	67.78±2.19	68.89±2.33	68.89±2.33	68.89±2.33	68.89±2.33	68.89±2.33
	Two hours	71.11±1.20	77.78±0.33	81.11±0.33	81.11±0.33	82.22±0.67	82.22±0.67
	Three hours	92.22±0.88	96.67±0.58	97.78±0.33	97.78±0.33	100±0.000	100±0.000

Effect of ozone gas against adult of *O. surinamensis* (L.) at 3000 ppm on Bermuda and Kandella varieties are presented in Table (4) and Fig. (4) the results showed that highly effect of ozone gas on adult *O. surinamensis* at high concentration 3000 ppm at 1 hr. release gas after one day post treatment (67.78 and 67.78%) on Bermuda and Kandella varieties, respectively. These values increased to 73.33% and 68.89% after 14 days post treatment on the same conditions. On the other hand, these values increased to 88.89 & 94.44% and 92.22 & 100% after one and 14 days post treatment at the same concentration and varieties.

**Table (4):** Mortality of adults of *O. surinamensis* ascending periods (1, 2, 5, 7, 10 and 14 day) when exposed to 3000 ppm of ozone.

Date variety	Exposure time (hours)	Mortality % of <i>O. surinamensis</i> after days					
		1	3	5	7	10	14
Bermuda	On hour	21.11±1.20	38.89±1.86	47.78±2.73	50.00±3.00	51.11±3.18	51.11±3.18
	Two hours	60.00±1.73	67.78±1.67	68.89±1.33	68.89±1.33	68.89±1.33	68.89±1.33
	Three hours	84.45±1.86	92.22±1.20	94.44±0.88	94.44±0.88	94.44±0.88	94.44±0.88
Kandella	On hour	22.22±2.19	35.56±1.45	40.00±1.53	41.11±1.86	41.11±1.86	42.22±1.67
	Two hours	45.00±0.88	54.44±1.45	56.67±1.15	56.67±1.15	56.67±1.15	56.67±1.15
	Three hours	57.78±2.19	67.78±1.67	70.00±1.53	70.00±1.53	70.00±1.53	72.22±1.86

Lethal concentration values and parameters of mortality regression lines to ozone at 30± 1°C at the different concentrations on *O. surinamensis* were presented in Table (5) the LC<sub>50</sub> for 500, 1000, 2000 and 3000 ppm were 2.79, 1.06, 1.05, and 0.46, & 5.18, 1.37, 1.39 and 0.64 days for Bermuda and Kandella varieties, respectively. While, the LC<sub>90</sub> were 13.67, 3.12, 3.09 and 3.11 & 18.60, 8.49, 8.92 and 2.37 days for the same varieties, respectively. The results showed clearly that the Bermuda variety had tolerant infested with *O. surinamensis* than Kandella variety at all concentrations of ozone gas.

**Table (5):** Lethal time values and confidence limits for the adults, *O. surinamensis* at four exposure period of ozone.

Date variety	concentrations	LC <sub>25</sub>	LC <sub>50</sub>	LC <sub>90</sub>	slope
Bermuda	500ppm	1.21	2.79	13.67	+/- 0.40
	1000ppm	0.60	1.06	3.12	+/- 0.42
	2000ppm	0.60	1.05	3.09	+/- 0.43
	3000 ppm	0.17	0.46	3.11	+/- 0.43
Kandella	500ppm	2.65	5.18	18.60	+/- 0.52
	1000ppm	0.53	1.37	8.49	+/- 0.38
	2000ppm	0.52	1.39	8.92	+/- 0.38
	3000 ppm	0.32	0.64	2.37	+/- 0.46

Similar results were obtained by (Kells, *et al.*, 2001) indicated that high mortality was achieved for adults, of *Sitophilus zeamais* and *Tribolium castaneum* exposed to 50 ppm ozone for 3 days. (Zakladony, *et al.*, 2003) found that ozone application in concentration of 1.35g/m<sup>3</sup>



caused 100% mortality after 1 and 3 days post treatment for adults of *Sitophilus oryzae* and *Sitophilus granarius*, respectively. (Sausa, et al., 2008) tested ozone on phosphine – resistant insects in the laboratory, and found them to be susceptible to ozone at concentration of 0.321 g/m<sup>3</sup>. (Bonjour, et al., 2011) studied ozone fumigation of hard red winters wheat and found that increasing the exposure time may decrease the survival ratios of egg, larvae and pupa of *plodia interpunctella* at 5 ppm ozone concentration at exposure of 30, 60, 90, and 120 min were reported to result in mortality rates of 71.66% , 68.33%, 78. 33% and 85 % respectively. (Sadeghi, et al., 2017) reported that the effect of ozone on the mortality of *O. surinamensis* and *E. kuehniella* at dried figs was statistically proven that ozone concentration and exposure time had significant effects on mortality. Complete mortality was observed for 90 min at 5ppm ozone concentration for both insects. Our results from the present study demonstrate that increasing in exposure time or in gas concentration of ozone resulted increasing in adult mortality percent of *O. surinamensis* reaching high mortality 94.44% and 100% of 3000 ppm 3 hrs. at Bermuda and Kandella varieties, respectively.

**Effect of gamma irradiation against *O. surinamensis* adults at 30± 1°C and 65±5% RH with Bermuda and Kandella varieties:**

**Table (6):** Mortality of adults of *O. surinamensis* after ascending periods (1, 2, 5, 7, 10 and 14 day) when exposed to different doses of gamma irradiation on Bermuda variety.

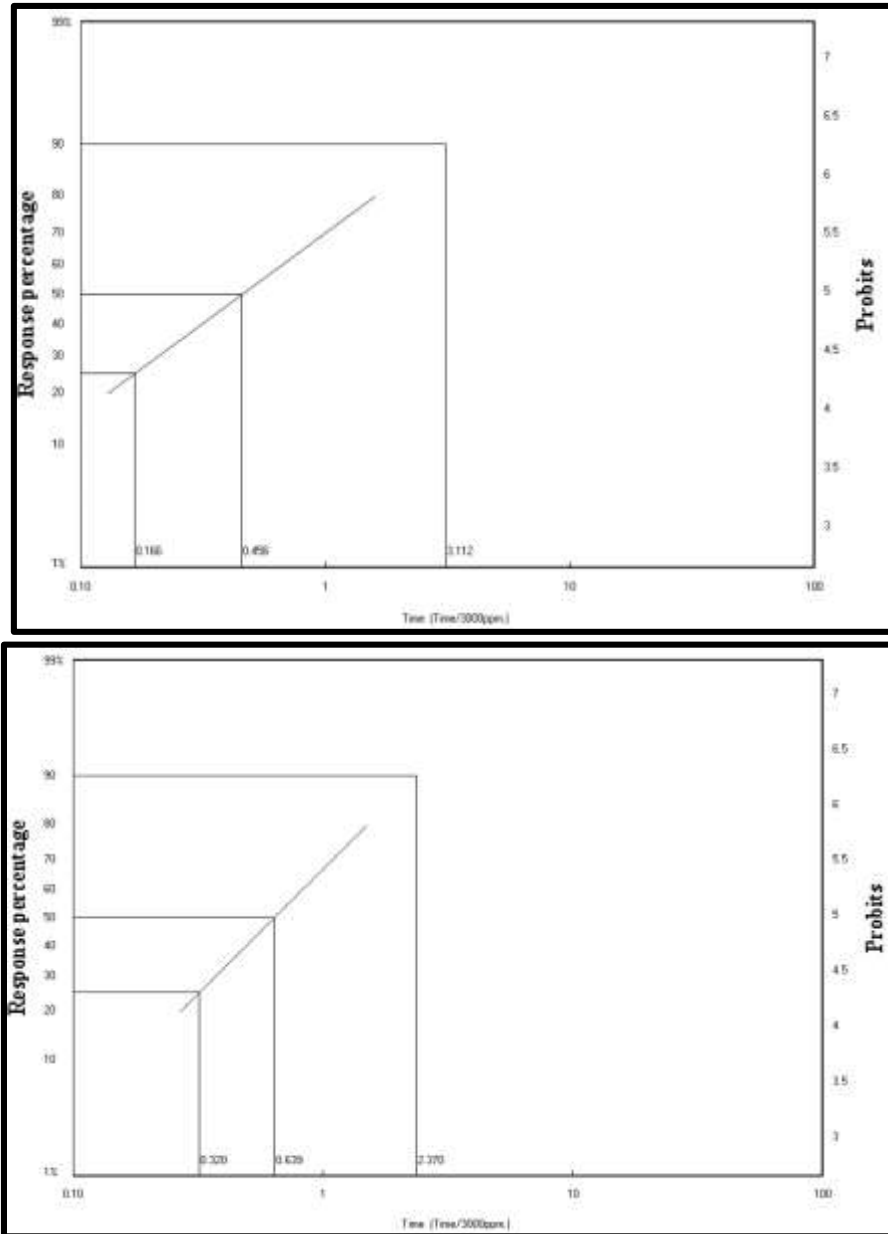
Concentration Gy	Mortality % of <i>O. surinamensis</i> after days					
	1	3	5	7	10	14
10	2.22±0.67	3.33±0.58	6.67±0.58	6.67±0.58	10.00±1.00	10.00±1.00
20	2.22±0.33	2.22±0.33	5.56±0.67	7.78±0.33	10.00±0.58	14.44±0.88
40	0.00±0.0	0.00±0.00	1.11±0.33	3.33±0.00	11.11±0.33	15.56±0.88
50	2.22±0.33	5.56±0.33	11.11±0.88	15.56±0.33	17.78±0.33	21.11±0.33
60	4.44±0.88	5.56±0.67	8.89±1.20	19.00±2.91	22.22±2.96	25.00±1.67
100	7.78±1.33	17.78±0.88	43.33±1.53	45.56±1.20	46.67±1.53	50.00±1.00
200	13.22±1.0	22.22±1.67	73.33±0.58	76.67±0.0	77.78±0.33	77.78±0.33
400	12.22±1.20	15.56±0.88	73.33±1.73	90.00±1.0	93.33±0.58	93.33±0.58
800	12.22±0.33	23.33±0.58	86.67±0.0	98.89±0.33	98.89±0.33	98.89±0.33

**Table (7):** Mortality of adults of *O. surinamensis* after as reading periods (1, 2, 5, 7, 10 and 14 day) when exposed to different concentration of gamma irradiation on Kandella variety.

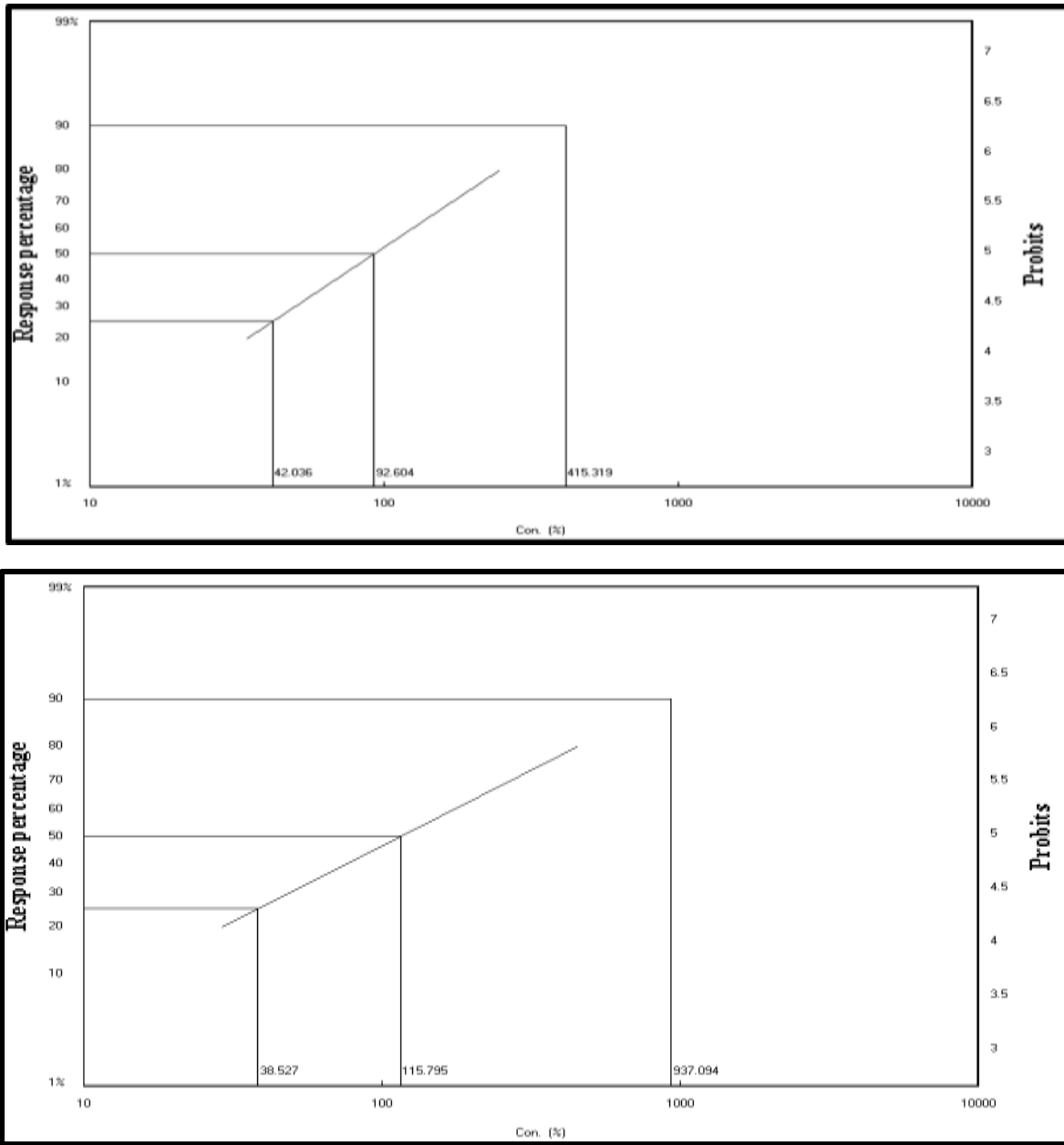
Concentration Gy	Mortality % of <i>O. surinamensis</i> after days					
	1	3	5	7	10	14
10	1.11±0.33	1.11±0.33	2.22±0.33	4.45±.67	10.0±1.53	12.20±1.20
20	3.33±0.58	6.67±0.00	8.89±0.33	8.89±0.33	18.89±0.33	20.0±0.58
40	7.78±0.33	10.00±0.00	10.00±0.0	10.00±0.0	14.45±0.67	21.11±0.33
50	3.33±1.00	11.11±0.67	14.45±0.67	18.89±1.45	18.89±1.45	20.0±1.55
60	12.22±0.33	14.45±0.67	14.45±0.67	16.67±0.58	22.22±0.67	23.33±0.58
100	14.45±0.67	23.33±1.15	51.11±0.88	55.56±1.20	53.33±1.00	56.67±1.00
200	7.78±0.67	13.33±1.73	48.89±1.33	60.00±1.53	64.44±0.88	66.67±1.00
400	4.44±0.33	11.11±0.33	58.89±3.38	70.00±2.52	75.56±2.40	76.67±2.65
800	4.44±0.33	22.22±1.86	66.67±1.73	68.18±1.53	87.78±0.88	91.11±0.88



The data obtained from experiments dealing with the irradiation of *O. surinamensis* on Bermuda and Kandella varieties were summarized in Table (6,7 and 8) and Fig. (5) data showed that the mortality increased markedly with increase of radiation dose exposure and time. As irradiation to 10Gy caused 1.11% and 2.22% mortality after one day post treatment for Bermuda and Kandella varieties, respectively. These values reached to 12.20% and 10.00% after 14 days. After a day of exposure the mortality at 800 Gy were 4.44% and 12.22 after one day. While, the values mortality after 14 day 91.11% and 98.89% for Bermuda and Kandella varieties, respectively.



**Fig. (4):** Lethal time values and confidence limits of ozone at 3000 ppm adults of *O. surinamensis* after three hours for 14 day after treatment.



**Fig. (5):** Lethal time values and confidence limits for the adult's *O. surinamensis* at different concentration from gamma irradiation

**Table (8):** Lethal time values and confidence limits for the adults, *O. surinamensis* at different concentration four different of gamma irradiation

Date variety	LC <sub>25</sub>	LC <sub>50</sub>	LC <sub>90</sub>	slope
Bermuda	38.53	115.80	937.09	+/- 0.09
Kandella	42.04	92.61	415.32	+/- 0.12

Lethal dose values and parameters of mortality regression lines to irradiation at  $30 \pm 1$  °C at the different concentrations on *O. surinamensis* are presented in Table (8) and Fig. (5) the LC<sub>50</sub>

of adult mortality *O. surinamensis* adult were 115.80 and 92.61 days for **Bermuda and Kandella varieties, respectively**. Similar results were obtained by (Abd El- Aziz *et al.*, 2017) reported that the complete reduction in adult emergence of *Sitotroga cerealella* (100%) when the treat all stages by 1500Gy to gamma radiation. (Mansour, 2016) showed that the sensitivity of 1-2 day old *Trigoderma granarium* adults to irradiation increased with increasing dose. Females were more susceptible to irradiation than males. The results of this study showed the mortality of *O. surinamensis* increased with increasing of doses and exposure time. While, at 800 Gy exposed treatment the mortality% was 91.11 % and 98.89 % for *O. surinamensis* adults at Bermuda and Kandella varieties, **respectively**.

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