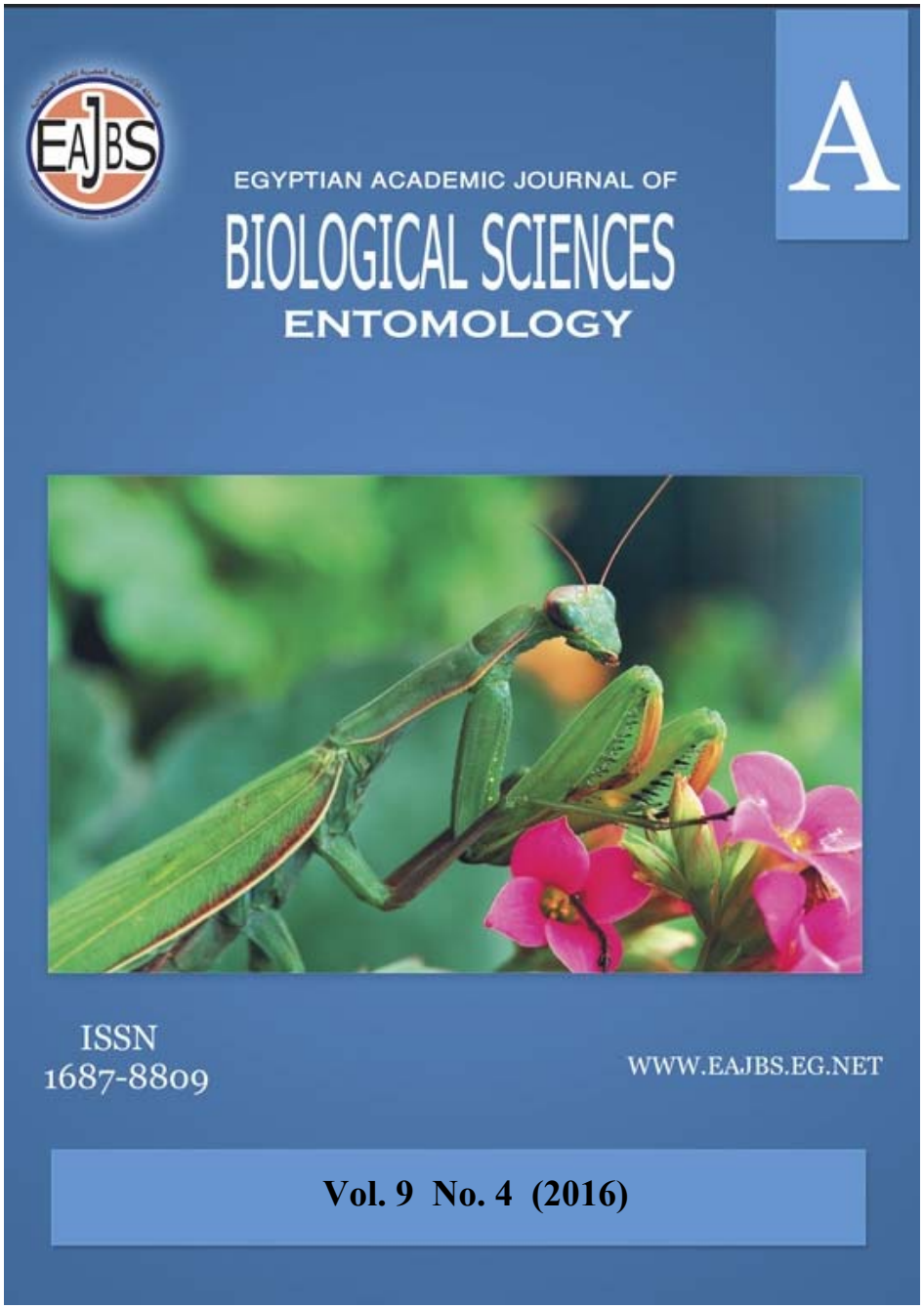


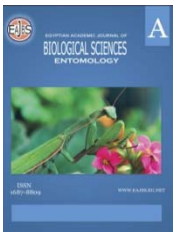
Egyptian Academic Journal of Biological Sciences is the official English language journal of the Egyptian Society for Biological Sciences, Department of Entomology, Faculty of Sciences Ain Shams University.
Entomology Journal publishes original research papers and reviews from any entomological discipline or from directly allied fields in ecology, behavioral biology, physiology, biochemistry, development, genetics, systematics, morphology, evolution, control of insects, arachnids, and general entomology.
www.eajbs.eg.net

Citation: *Egypt. Acad. J. Biolog. Sci. (A. Entomology) Vol.9 (4)pp. 1- 13(2016)*

Egypt. Acad. J. Biolog. Sci., 9(4): 1–13 (2016)

Egyptian Academic Journal of Biological Sciences
A. Entomology

ISSN 1687- 8809
www.eajbs.eg.net



Effect of Ozone on A serious Wheat Pest, *Sitotroga cerealella* (Olivier) and Its Progeny

Abd El-Ghaffar, M. M., Mohamed, H. A., Ibrahim, I.L. and Gad, H. A.
Plant Protection Department, Faculty of Agriculture. Al-Azhar Univ. Egypt
E-mail: hassan_jad_855@yahoo.com

ARTICLE INFO
Article History
Received:27/7/2016
Accepted: 29/8/2016

Keywords:
Ozone
Sitotroga cerealella
Tritium aestivum
F_1 generation.

ABSTRACT

Angoumois grain moth, *Sitotroga cerealella* (Olivier), is one of the most important pests of such stored products as maize and wheat. Different stages of this pest were treated with ozone as a gas at three concentrations (1, 3, and 5 g/m^3) for six different periods (0.5, 1, 2, 3, 4 and 5 h.) compared with untreated insects. The results indicated that increasing the concentration and exposure time led to increasing the rate of mortality for all tested stages at parental generation. The results showed that also, ozone affected biological aspects of the progeny resulted from treated stages (egg, larvae, pupae and adult) at F_1 generation.

INTRODUCTION

Wheat (*Triticum aestivum* L.) is the most widely grown food crop in the world, which ranks first in terms of area and production in the world (FAO 1988). Wheat is a major cereal crop in Egypt as most of the population utilizes it daily in one or more of the three meals. Interest in storing wheat is increasing to face the increasing demand for human consumption.

Angoumois grain moth *Sitotroga cerealella* (Olivier) is one of the main insect pests of stored grain in the tropics and subtropics. Sorghum, maize, wheat, barley, and millets are the main crops infested by this pest. Infestations start in the field when crops are carried to storage facilities, immature stages from the field infestations complete their life cycles to pupate and emerge as adult in storage. Population of *S. cerealella* multiplies 112.27 times between two successive generations (Teotia and Singh, 1976). Cogburn and Bollich (1980) reported that *S. cerealella* usually found in the upper 40 cm layer of the grain.

Alternative methods for effective post-harvest pest control have been required due to the restriction of chemical fumigants such as methyl bromide which has been a major stored-product and quarantine treatment but it has harmful effects on human health and the environment inducing significant ozone depleting substance (Ross, 1999). Currently, bulk commodities are often fumigated but the number of fumigants registered around the world is extremely limited and very few of the new treatments are acceptable for all applications.

Additionally, the growing demand for organic grains has generated a need for control strategies for this niche market.

Ozone, a powerful oxidant, has numerous beneficial applications and is very familiar to the food processing industry. It has been long been used in food processing as a water treatment to disinfect, eliminate odors, taste and color (Kim *et al.,* 1999; Legeron, 1984; Suffet *et al.,* 1986; EPA, 1999). Ozone (O_3) is an allotrope of oxygen, which can be generated by UV-light and electrical discharges in air (corona-discharge). Ozone generation by electrical discharge is most common and has several advantages, including greater sustainability of the unit and higher ozone production. Ozone has a half-life of 20-50 min, rapidly decomposing to diatomic oxygen, a natural component in the atmosphere. Because ozone can be easily generated at the treatment site using only electricity and air, it offers several safety advantages over conventional post-harvest pesticides. First, there are no stores of toxic chemicals, chemical mixing hazards, or disposal of left over insecticides or containers (Law and Kiss, 1991). Second, with a short half-life, it reverts back to naturally occurring oxygen leaving no residue on the stored products.

Researchers have been recently focused on the application of ozone as a fumigant to control stored-grain insects and microorganisms and to reduce mycotoxins on grain. for several reasons: It may be generated at the site of use; itself decomposes to molecular oxygen (McDonough *et al.,* 2011; Mylona *et al.,* 2014; Savi *et al.,* 2015), which prevents the need to store and dispose of hazardous chemicals; there is no need for aeration to remove the gas after application and it is classification as "GRAS" (Generally Recognized As Safe) by the United States Environmental Protection Agency (USEPA) (Isikber and Athanassiou 2015).The purpose of this study is to know the direct effect and latent effect of ozone gas on S. *cerealella.*

MATERIALS AND METHODS

A laboratory strain of the Angoumois grain moth, *Sitotroga cerealella* (Olivier) was obtained from the plant protection institute, Ministry of Agriculture, Egypt. Biology of *S. cerealella* was studied on wheat grains in laboratory conditions by maintaining them at room temperature.

Ozone generation techniques:

The schematic diagram of ozone generator is shown in Fig. (1). A cylindrical dielectric barrier discharge (DBD) cell has been used as an ozone generator (Located at Center of Plasma Technology, Faculty of Science, Al-Azhar University, Nasr City, Cairo, Egypt). The DBD cell consists of two cylindrical coaxial electrodes separated by a gap distance and dielectric barrier (glass). AC (50 Hz) high voltage (2-5 kV) was applied on the DBD cell to generate filamentary discharge. The DBD cell is fed by oxygen gas.

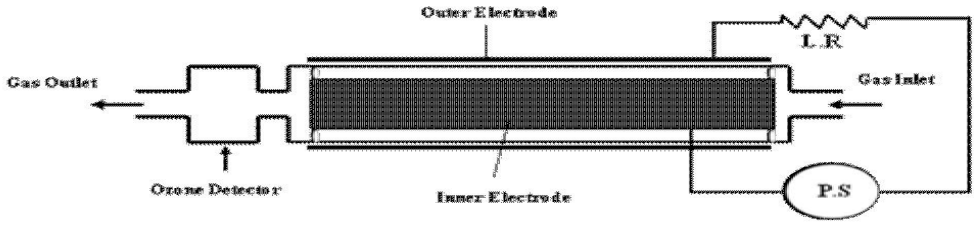


Fig. 1: Schematic diagram of the ozone generator

The basic mechanism of ozone generation simply consists of dissociation of oxygen molecules by the discharge electrons that are formed in the discharge filaments inside the discharge gap. The atomic oxygen, which is produced due to the dissociation, reacts with the oxygen molecules to form ozone. The concentration of the generated ozone was controlled by the discharge current and the gas flow rate was adjusted to 5 L/min.

The concentration of ozone formed inside the DBD system was measured using ozone detector (Model H1-AFX-Instrumentation, USA) (Garamoon *et al*., 2009). Ozone was applied directly into the tubes containing the various insect stages and wheat under investigation.

Exposure of different stages of S*. cerealella* to ozone:

Newly-laid eggs of *S. cerealella* (300 egg) and adults one day old (50 adult) were treated in glass tubes, while larvae (21-23 days) and pupae (25-27 days) were treated by exposing (10 g) wheat grains after artificial infestation to different concentrations of ozone gas viz. 1.0, 3.0, and 5.0 g/m^3 for 0.5, 1, 2, 3, 4 and 5 h. compared with untreated insects, the experiments were carried out in three replicates.

Mortality percentages and biological aspects of the different developmental stages:

By the end of the tested exposure periods, the treated eggs or adult or grains containing larvae or pupae were taken out and incubated under the optimum constant conditions of 25±2°C and 65 ± 5% r.h. Egg and adult remained inside the tubes and were examined daily to record hatchability, adult survival for treated eggs and mortality percentages for treated adults which were corrected according to Abbott's formula (Abbott, 1925). Treated grains infested with larvae or pupae were transferred to glass jars (5 cm in diameter and 15 cm in depth) covered with muslin cloth for incubation under the same optimum conditions and were examined daily until the emergence of adult moths (Due to the difficulty of detecting larvae or pupae inside the grains).

Latent effects on certain biological and reproductive potentials of F_1 generation:

To continue the F_1 generation, the newly adult (males were paired with females of the same age) emerged from treated egg or larvae or pupae or adult. The biological aspects fecundity, hatchability and mating percentages were studied. The data were statistically evaluated by analysis of variance (F) followed by Duncan's multiple range test 1955 to examine the significant differences between treatment to every generation separately. The 5% level of probability was used in all statistical tests. The statistical software program Costat was used for all analyses.

RESULTS AND DISCUSION

Effect of ozone treatment on eggs:

Parental generation:

The data in Table (1) clearly show that both exposure time to ozone and the gas concentrations have obvious influence on egg hatchability and adult survival of *S. cerealella*. The lowest egg hatchabilities (6.66, 5.7 and 0.0%) were recorded at 5h. exposure time when concentration of ozone was 1, 3 and 5g/m^3, respectively, comparing with control (88.13%). The same trend was obtained in respect to adult survival. The lowest percentages of adult survival (9.53 and 0.0 %) were recorded for insects treated for 5h. with gas concentration of 1 and 3g/m^3. However, at concentration of 5g/m^3, all insects perished with all exposure times comparing with control 85.5%. Obtained results indicate that increasing in exposure time or in gas

concentration resulted in decreasing in hatchability and adult survival.

Table 1: Effect of O_3 gas (g/m^3) on the biological aspects of *S. cerealella* treated as one day old egg

Exposure time(h)	Egg numbers	hatchability %	Adult survival %	Mating %
1g/m^3				
0	300	88.13 a	85.50 a	100.0 a
0.5	300	16.67 b	38.00 b	0.00 b
1	300	14.53 c	33.33 b	0.00 b
2	300	12.90 c	32.43 b	0.00 b
3	300	10.00 d	30.00 b	0.00 b
4	300	9.40 d	20.00 bc	0.00 b
5	300	6.66 e	9.53 c	0.00 b
L.S.D		2.1	16.38	7.95
3g/m^3				
0	300	88.13 a	85.50 a	100.0 a
0.5	300	16.00 b	35.00 b	0.00 b
1	300	7.90 c	23.33 c	0.00 b
2	300	7.50 c	22.23 c	0.00 b
3	300	6.70 c	20.00 cd	0.00 b
4	300	6.70 c	16.70 d	0.00 b
5	300	5.70 c	0.00 e	----------
L.S.D	300	2.08	4.86	7.95
5g/m^3				
0	300	88.13 a	85.50 a	100.0
0.5	300	11.60 b	0.00 b	----------
1	300	7.63 b	0.00 b	----------
2	300	6.40 b	0.00 b	----------
3	300	2.90 b	0.00 b	----------
4	300	2.73 b	0.00 b	----------
5	300	0.00 b	--------	----------
L.S.D		12.64	3.15	

Means followed by the same letter in each column are not significantly different at P>0.05.

The data given in Table (2) indicate that the developmental periods (egg incubation period, larval and pupal period from treated egg was prolonged compared to control.

Table 2: Delayed effect of O_3 gas (g/m^3) on the developmental periods of *S. cerealella* treated as one day old egg.

Exposure time (h)	Egg Incubation period (d)	larval period (d)	Pupal period (d)	Male longevity (d)	Female longevity (d)
1 g/m^3					
0	3.30 c	20.00 c	6.00 d	5.00 a	6.00 a
0.5	4.50 a	21.50 a	7.50 a	3.00 b	3.00 b
1	4.00 b	21.00 ab	7.50 a	3.00 b	3.00 b
2	4.50 a	20.40 bc	7.00 b	3.00 b	3.00 b
3	4.10 b	20.50 bc	6.60 bc	3.00 b	3.00 b
4	4.10 b	20.50 bc	6.60 bc	3.00 b	3.00 b
5	4.00 b	20.30 c	6.50 c	3.00 b	3.00 b
L.S.D	0.336	0.584	0.454	0.178	0.18
3 g/m^3					
0	3.30 a	20.00 b	6.00 c	5.00 a	6.00 a
0.5	3.60 a	20.30 b	6.30 bc	3.00 b	3.00 b
1	3.70 a	20.40 b	6.40 c	3.00 b	3.00 b
2	3.70 a	20.60 ab	6.60 ab	3.00 b	3.00 b
3	3.60 a	20.50 ab	6.60 ab	3.00 b	3.00 b
4	3.70 a	21.00 a	7.00 a	3.00 b	3.00 b
5	4.00 a	--------	--------	--------	--------
L.S.D	0.409	0.493	0.477	3.25	1.454
5 g/m^3					
0	3.30 c	20.00	6.00	5.00	6.00
0.5	4.00 b	--------	--------	--------	--------
1	4.20 ab	--------	--------	--------	--------
2	4.40 ab	--------	--------	--------	--------
3	4.50 a	--------	--------	--------	--------
4	4.50 a	--------	--------	--------	--------
5	--------	--------	--------	--------	--------
L.S.D	0.406				

Means followed by the same letter in each column are not significantly different at P>0.05.

While the adult males and females longevity of S. *cerealella* was significantly decreased in all treatments of ozone at all exposure periods comparing with control.

F_1 generation:

Data in Table (1) show also, that the adult emerged from treated egg failed at mating at all exposure times with comparison control (100%) at F_1 generation. The results were obtained by the authors differed by the different species of insects, the concentration and exposure time between ozone and the insect. Leesch (2002) reported that high concentrations of ozone at 10.000 ppm for a 4 h. exposure period did not kill 100% of the eggs of *Plodia interpunctella*. Al-ahmdi, *et al.,* (2009) showed that egg of *Oryzaphilus surinamensis* was susceptible to ozone as compared to larvae and adults and 100% percent mortality for eggs was achieved using 7 ppm ozone for 1h. Niakousari *et al*. (2010) reported that a concentration of 4.000 ppm of ozone for 2 h. resulted in only 80% mortality of *P. interpunctella* eggs on date fruits. Abo-El-Saad *et al*., (2011) concluded that ozone exhibited less effect against eggs of *E. cautella,* and exposing eggs to 2.0 ppm ozone for 12h. resulted in 91% hatchability and 50% emergence respectively. *Keivanloo et al*., (2014) showed that not surprisingly, ozone had less effect on egg mortality of *P. interpunctella*. At the highest ozone concentration× exposure period treatment and the mortality of eggs were (56.66%). Our results indicate that increasing in exposure period or in gas concentration of ozone resulted in decreasing in hatchability percent of treated eggs, adult survival and developmental periods of tested insect comparing with control in the P_1 generation, the results showed also, the adult emerged from treated egg failed at mating and not resulted any progeny at all exposure periods of tested insect at F_1 generation. This could be most likely attributed to the toxic effect of ozone at high concentrations may be due to modification of the egg shell protein polymer by the oxidant, rendering it more resistant to hatching enzyme (Grotmol *et al*., 2003) and that possible that the outer layer on the eggs provide some additional barrier to the ozone. Furthermore, these factors may be responsible for some of the inconsistencies in overall trends in the mortality data for eggs. The metabolic status and integrity of the outer layer may vary, resulting in some individuals being more susceptible to ozone than others (McDonough *et al.,* 2011).

Effect of ozone treatment on larvae:

Parental generation:

The data in Table (3) clearly show that both exposure time to ozone and the gas concentrations have obvious influence on adult emergence of *S. cerealella*. The lowest percentages of adult emergence (22.52, 7.10 and 0.65%) were recorded at 5h. exposure time when concentration of ozone was 1, 3 and 5g/m^3, respectively, comparing with control (100%). While the adult males and females longevity of S. *cerealella* was significantly decreased in all treatments of ozone with exposure periods comparing with control.

F_1 generation:

Data in Table (4) show that ozone treatment on *S. cerealella* did not significantly decrease mating percent at 1 and 3 g/m^3, while at 5 g/m^3 percentage of mating was significantly decreased by increasing exposure times to ozone, reaching to 33.33% at 5h. exposure times compared with control (100%). While the fecundity of females mated with males emerged from treated larvae of *S. cerealella* was significantly decreased by increasing exposure times to ozone, reaching to the lowest values (34.16, 63.66and 16.66 egg/ female) at 5 h. exposure time at 1, 3 and 5 g/m^3, respectively, comparison with control (100.4 egg/ female) at F_1 generation.

Table 3: Effect of O_3 gas (g/m^3) on the biological aspects of *S. cerealella* treated as full grown larvae

Exposure time (h)	Adult emergence %	Male longevity (d)	Female longevity (d)
$1g/m^3$			
0	100.0 a	5.00 a	6.00 a
0.5	58.48 b	3.30 b	3.30 cd
1	54.67 b	3.00 b	3.00 d
2	42.86 c	3.10 b	3.20 cd
3	32.89 d	3.20 b	3.70 bc
4	26.32 de	3.40 b	4.00 b
5	22.52 e	3.20 b	3.30 cd
L.S.D	6.81	0.399	0.502
$3g/m^3$			
0	100.0 a	5.00 a	6.00 a
0.5	30.52 b	3.00 b	3.40 b
1	23.19 c	3.00 b	3.20 b
2	19.88 d	3.00 b	3.50 b
3	14.62 e	3.00 b	3.40 b
4	12.11 f	3.00 b	3.20 b
5	7.10 g	3.00 b	3.30 b
L.S.D	1.01	0.225	0.43
$5g/m^3$			
0	100.0 a	5.00 a	6.00 a
0.5	23.17 b	3.00 b	3.60 b
1	19.36 c	3.10 b	3.40 bc
2	12.25 d	3.00 b	3.30 bc
3	8.84 e	3.00 b	3.00 c
4	5.79 f	3.00 b	3.60 b
5	0.65 g	3.00 b	3.00 c
L.S.D	0.94	0.192	0.495

Means followed by the same letter in each column are not significantly different at P>0.05.

Table 4: Effect of O_3 gas (g/m^3) on the biological aspects of F_1 generation of *S. cerealella* treated as full grown larvae

Exposure time (h)	Mating %	Egg laid per female	hatchability %	Egg incubation period (d)
$1g/m^3$				
0	100.0 a	103.8 a	88.02 a	3.30 c
0.5	88.90 a	43.60 b	38.22 b	3.80 bc
1	77.77 a	43.66 b	32.36 bc	4.50 a
2	75.00 a	39.40 b	32.15 bc	4.50 a
3	75.00 a	50.76 b	26.96 c	4.00 ab
4	88.90 a	51.33 b	19.72 d	3.60 bc
5	66.66 a	34.16 b	15.76 d	4.00 ab
L.S.D	26.07	18.04	6.25	0.469
$3g/m^3$				
0	100.0 a	100.3 a	88.02 a	3.30 c
0.5	83.33 a	75.00 b	31.83 b	4.30 a
1	83.33 a	75.16 b	25.81 bc	4.40 a
2	83.33 a	63.16 b	21.63 cd	3.80 bc
3	83.33 a	59.20 b	17.60 d	4.00 ab
4	66.66 a	53.00 b	17.70 d	4.30 a
5	66.66 a	63.66 b	8.68 e	4.00 ab
L.S.D	37.65	23.94	6.09	0.366
$5g/m^3$				
0	100.0 a	100.3 a	88.02 a	3.30 c
0.5	75.00 ab	48.90 b	29.03 b	4.00 ab
1	58.35 bc	61.83 b	19.68 bc	3.70 bc
2	50.00 bc	57.50 b	16.32 cd	4.00 ab
3	41.66 c	43.18 b	12.26 cde	4.20 a
4	41.66 c	57.83 b	5.85 de	4.40 a
5	33.33 c	16.66 c	0.00 e	-------
L.S.D	29.16	20.53	11.76	0.395

Means followed by the same letter in each column are not significantly different at P>0.05.

Data also, clearly show that both exposure time to ozone and the gas concentrations have obvious influence on egg hatchability of *S. cerealella*. The lowest egg hatchabilities (15.76, 8.86 and 0.0%) were recorded at 5h. exposure time when concentration of ozone was 1, 3 and 5g/m^3, respectively.

While egg incubation period increased from 3.30 days at control to 4 and 4days at 5h.exposure time at 1 and 3g/m^3, while increased at 5 g/m^3 to 4.4 days at 4h. exposure time at F_1 generation.

Similar results were obtained by Kells *et al*. (2001) found that a higher mortality rate for the larval stage compared with other stages of *P. interpunctella* exposed to 50 ppm of ozone for 3 days. Osman (2009) studied the effect of ozone on *E. kuehniella* at 1 g /m^3 for different exposure periods of 0.5, 1, 2, 3, 4 and 5 h. and found that larvae required not less than 6 days after ozone exposure to reveal the full effect in the mortality rate. Niakousari *et al.,* (2010) have proved that exposing samples to higher than 2000 mg/L^1 O_3 for 120 min resulted in complete mortality of larvae of *P. interpunctella* in Kabkab dates. Hussain (2014) showed that corrected mortalities percentage increased gradually by increasing each of exposure time to ozone gas and period after treatment when *E. cautella* larvae treated by 80 ppm. and noticed that the mortality percent was 38.92 % at 1 h. exposure time, followed significantly by 66.55, 88.37, 98.55 and 100 % at 2, 3, 4 and 5 h. exposure times, respectively. Husain *et al*., (2015) showed that efficacy of ozone gas (22 ppm) was examined against larval mortality of *E. cautella*, and the immediate and delayed larval mortality was recorded after each exposure time. Ozone possessed the strongest fumigant toxicity causing 100% mortality with all varieties after 24 h. exposure and was more effective than any other gases. Obtained results from the present study showed that biological aspects of treated larva with ozone gas were affected with increasing in exposure period or in gas concentration resulted decreasing in adult emergence and developmental periods of tested insect comparing with control in the P_1 generation, the results showed also, that ozone gas affected the progeny resulted from emerged adults of treated larva of tested insect in F_1 generation.

Effect of ozone treatment on pupae:

Parental generation:

The data in Table (5) clearly show that both exposure time to ozone and the gas concentrations have obvious influence on adult emergence of *S. cerealella*. The lowest percentages of adult emergence (28.88, 7.33 and 2.66%) were recorded at 5h. exposure time when concentration of ozone was 1, 3 and 5g/m^3, respectively comparing with control (100%).While the adult males and females longevity of S. *cerealella* was significantly decreased in all treatments of ozone with exposure periods comparing with control.

F_1 generation:

Data in table (6) show that ozone treatment on *S. cerealella* did not affect significantly on the mating percent at 1 g/m^3, while at 3 g/m^3 percentage of mating was significantly decreased by increasing exposure times to ozone, reaching to 16.66% at 5 h. and reached to 0% at 3, 4 and 5 h. exposure times at 5 g/m^3 comparing with control (100%). While the fecundity of females mated with males emerged from treated pupae of *S. cerealella* was significantly decreased by increasing exposure times to ozone, reaching to the lowest values (41.33 and 26 egg/ female) at 5 h. exposure time at 1 and 3 g/m^3, respectively and reached to 18.66 egg/ female at 2h. exposure time at 5g/m^3 with comparison control (100.4 egg/ female) at F_1 generation.

Table 5: Effect of O_3 gas (g/m^3) on the biological aspects of *S. cerealella* treated as 5 old pupae.

Exposure time (h)	Adult emergence %	Male longevity (d)	Female longevity (d)
$1g/m^3$			
0	100.0 a	5.00 a	6.00 a
0.5	87.30 b	3.50 b	3.40 b
1	71.11 c	3.30 b	3.30 b
2	65.80 cd	3.40 b	3.50 b
3	62.59 d	3.40 b	3.50 b
4	58.88 d	3.30 b	3.60 b
5	28.88 e	3.10 b	3.60 b
L.S.D	7.24	0.396	0.426
$3g/m^3$			
0	100.0 a	5.00 a	6.00 a
0.5	36.75 b	3.20 b	3.40 b
1	30.04 c	3.00 b	3.50 b
2	21.71 d	3.10 b	3.20 b
3	13.33 e	3.10 b	3.20 b
4	10.67 e	3.00 b	3.30 b
5	7.33 f	3.00 b	3.20 b
L.S.D	2.92	0.208	0.386
$5g/m^3$			
0	100.0 a	5.00 a	6.00 a
0.5	21.02 b	3.00 b	3.30 bc
1	13.01 c	3.20 b	3.30 bc
2	7.356 d	3.00 b	3.40 bc
3	6.666 d	3.00 b	3.60 bc
4	6.346 d	2.70 b	3.80 b
5	2.66 e	3.00 b	3.20 c
L.S.D	1.11	0.217	0.48

Means followed by the same letter in each column are not significantly different at P>0.05.

Table 6: Effect of O_3 gas (g/m^3) on the biological aspects of F_1 generation of *S. cerealella* treated as 5 day old pupae

Exposure time (h)	Mating %	Egg laid per female	hatchability %	Egg incubation period (d)
$1g/m^3$				
0	100.0 a	103.8 a	88.02 a	3.30 c
0.5	83.33 a	62.20 bc	32.00 b	3.90 b
1	83.33 a	61.55 bc	30.30 b	4.00 ab
2	100.0 a	52.33 cd	17.58 c	3.70 bc
3	100.0 a	75.00 b	14.20 c	3.80 b
4	75.00 a	58.83 bcd	13.46 c	4.30 a
5	100.0 a	41.33 d	11.70 c	3.90 b
L.S.D	28.31	17.34	10.97	0.356
$3g/m^3$				
0	100.0 a	103.8 a	88.02 a	3.30 b
0.5	91.66 a	66.50 b	26.18 b	3.50 b
1	75.00 ab	64.48 b	22.88 b	4.30 a
2	66.67 abc	45.00 bcd	21.23 b	4.00 a
3	50.00 abc	41.00 cd	17.65 b	4.00 a
4	33.33 bc	57.33 bc	15.52 b	4.00 a
5	16.66 c	26.00 d	0.00 c	--------
L.S.D	46.81	20.99	13.71	0.435
$5g/m^3$				
0	100.0 a	103.8 a	88.02 a	3.30 b
0.5	75.00 a	66.00 b	23.57 b	4.00 a
1	66.66 a	63.66 b	19.49 b	3.90 a
2	66.66 a	18.66 c	16.33 b	4.00 a
3	0.00 b	------	------	--------
4	0.00 b	------	------	------
5	0.00 b	------	------	------
L.S.D	28.31	19.17	8.09	0.261

Means followed by the same letter in each column are not significantly different at P>0.05.

Data also, clearly show that both exposure time to ozone and the gas concentrations have obvious influence on egg hatchability of *S. cerealella*. The lowest egg hatchabilities (11.7 and 0.0 %) were recorded at 5h. exposure time when concentration of ozone was 1and 3g/m^3, and its reduced to 16.33% at 2h. exposure time at 5 g/m^3 respectively. While egg incubation period increased from 3.30 days at control to 3.9 and 4 days at 5 and 4h.exposure time at 1and 3 g/m^3, and at 5 g/m^3 reached to4 days at 2h. exposure time at F_1 generation. Similar results were obtained by Leesch (2002) reported that laboratory treatment of ozone alone resulted in high mortalities on the pupae of *P. interpunctella* at high concentration (300 ppm) at short exposure time (4h.).

Isikber *et al*., (2007) stated that toxicity of initial concentration of 300ppm gaseous ozone against pupae of *E. kuehniella* at short exposure time (2h.) was studied and mentioned that ozone gave a complete mortality for pupae. Bonjour *et al*., (2011) evaluating the efficacy of ozone fumigation against the major grain pests in stored wheat, reported that pupae of *P. interpunctella* were more susceptible than eggs and larvae. James (2011) reported that pupae of *G. mellonella* were more resistant to ozone than larvae. Abo El-saad (2011) exposed that pupae of *E. cautella* to 2.0 ppm of ozone at various exposure times 2, 4, 8, and 12 h. and showed that percentage of adult emergence was reached to 50% after 12 h. exposure time. The results from the present study indicate that increasing in exposure period or in gas concentration of ozone resulted decreasing in adult emergence from treated pupa and prolonged the developmental periods of tested insect comparing with control in the P_1 generation, the results showed also, that ozone gas affected on progeny resulted from emerged adults of treated pupa of tested insect in F_1 generation.

Effect of ozone treatment on adults:

Parental generation:

The data in Table (7) clearly show that percentage of adult mortality of *S. cerealella* has increased by increasing exposure times to ozone, reaching to 100% after 4 days from treatment at 0.5, 1, 2 and 3 h. after 3 days from treatment at 4h. and after 2days from treatment at 5 h. exposure time, respectively at 1g/m^3, while reached to 100% after 3days from treatment at 0.5 and 1 h. after 2 days from treatment at 2, 3 and 4 h. and after 1day from treatment at 5h. exposure time, respectively at 3 g/m^3 and at 5 g/m^3, reached to 100% after 3days from treatment at 0.5 h. after 2 days from treatment at 1, 2, and 3h.and after 1day from treatment at 4 and 5h.exposure time, respectively. While the adult males and females longevity of S. *cerealella* was significantly decreased in all treatments of ozone with exposure periods comparing with control.

F_1 generation:

Data in Table (8) showed that ozone treatment on *S. cerealella* did not affect significantly on the mating percentage except at 4 and 5h. exposure times whereas that percentage of mating was high significantly reduced to (0%) at 1 g/m^3, while at 3 g/m^3 percentage of mating was significantly decreased by increasing exposure times to ozone, reaching to 0.0 % at 3 and 4 h. and at 5 g/m^3, the treated adult failed at mating at all exposure times with compared with control (100%). While the fecundity of treated females mated with treated males of *S. cerealella* was significantly decreased by increasing exposure times to ozone, reaching to 42 and 15.75 egg/ female at 3 and 2 h. exposure time at 1and 3 g/m^3, respectively, with comparison control (100.4 egg / female) at F_1 generation.

Table 7: Effect of O_3 gas (g/m^3) on the biological aspects of *S. cerealella* treated as one day old adult

Exposure Period (h)	Mortality %				Male longevity (d)	Female longevity (d)
			$1g/m^3$			
	1d	2d	3d	4d		
0	0.00 e	0.00 d	0.00 c	0.00 b	5.00 a	6.00 a
0.5	7.40 d	36.11 c	58.88 b	100.0 a	3.80 b	3.80 b
1	10.00 d	37.03 c	62.22 b	100.0 a	3.60 b	3.70 b
2	19.49 c	41.48 c	71.11 b	100.0 a	3.40 bc	3.50 b
3	11.56 d	52.38 b	91.66 a	100.0 a	3.00 c	3.40 b
4	30.00 b	57.17 b	100.0 a	---------	3.50 bc	3.70 b
5	92.37 a	100.0 a	-------	-------	2.00 d	2.00 c
L.S.D	5.68	5.56	18.23	45.05	0.488	0.597
			$3g/m^3$			
0	0.00 g	0.00 d	0.00 b	-----	5.00 a	6.00 a
0.5	18.00 f	51.28 c	100.0 a	----	2.40 b	2.50 b
1	34.06 e	73.13 b	100.0 a	-----	2.50 b	2.50 b
2	66.70 d	100.0 a	-----	----	2.40 b	2.60 b
3	80.73 c	100.0 a	----	-----	2.00 c	2.80 b
4	90.90 b	100.0 a	-----	----	2.00 c	2.00 c
5	100.0 a	------	----	-----	----	-----
L.S.D	3.19	4.65	0.941	----	0.304	0.406
			$5g/m^3$			
0	0.00 f	0.00 c	0.00 b	-----	5.00 a	6.00 a
0.5	50.00 e	80.55 b	100.0 a	----	3.00 b	3.90 b
1	60.06 d	100.0 a	-----	-----	3.00 b	3.80 b
2	68.83 c	100.0 a	----	----	3.00 b	4.00 b
3	85.93 b	100.0 a	-----	-----	3.00 b	4.00 b
4	100.0 a	-----	----	----	----	----
5	100.0 a	----	-----	-----	----	----
L.S.D	6.22	7.96	0.925		3.57	0.212

Means followed by the same letter in each column are not significantly different at P>0.05

Table 8: Effect of O_3 gas (g/m^3) on the biological aspects of F_1 generation of *S. cerealella* treated as one day old adult

Exposure Time (h)	mating %	Egg laid per female	hatchability %	Egg Incubation period (d)
		$1g/m^3$		
0	100.0 a	100.4 a	88.02 a	3.30 b
0.5	90.00 a	83.00 a	47.72 b	4.30 a
1	100.0 a	53.80 b	25.86 c	4.00 a
2	80.00 a	47.20 b	25.06 c	4.00 a
3	100.0 a	42.00 b	25.48 c	4.00 a
4	0.00 b	------	----	------
5	0.00 b	------	----	--------
L.S.D	21.89	22.59	10.80	0.384
		$3g/m^3$		
0	100.0 a	100.4 a	88.02 a	3.30 b
0.5	80.00 a	37.20 b	30.98 b	3.50 ab
1	60.00 ab	22.50 c	30.00 b	3.90 a
2	40.00 bc	15.75 c	32.04 b	3.80 a
3	0.00 c	------	----	----
4	0.00 c	------	----	----
5	--------	------	----	----
L.S.D	40.22	8.34	10.23	0.402
		5g/m3		
0	100.0 a	---------	--------	-------
0.5	0.00 b	------	----	-----
1	0.00 b	------	----	-----
2	0.00 b	------	----	-----
3	0.00 b	------	----	-----
4	------	------	----	-----
5	------	------	----	-----
L.S.D	7.95			

Means followed by the same letter in each column are not significantly different at P>0.05.

Data also, clearly show the percentage of egg hatchability of *S. cerealella* was significantly decreased by increasing exposure times to ozone. The egg hatchability

decreased sharply from 88.02 % at control to 25.48 and 32.04% at 3 and 2h. exposure time at 1and 3g/m^3, respectively. While egg incubation period increased from 3.30 days at control to 4 and 3.8 days at 3 and 2h. exposure time at 1 and 3g/m^3 respectively, at F_1 generation. Similar results were obtained by Kells *et al.,* (2001) indicated that high mortality was achieved for adults of *Sitophilus zeamais* and *T. castaneum* exposed to 50 ppm ozone for 3 days. Zakladnoy *et al.,* (2003) found that ozone application in concentration of 1.35 g/m^3 caused 100% mortality after 1and 3 days post treatment for adult of *S. oryzae* and *S. ganarius,* respectively.

Sousa *et al.*, (2008) tested ozone on phosphine-resistant insects in the laboratory, and found them to be susceptible to ozone at a concentration of 0.321 g/m^3. Subramanyam *et al*., (2014) exposed adults of *R. dominica* (F.) to ozone concentration of 0.43 or 0.86 g/m^3 for 15-36 h. or 4-30 h. The authors found that the toxicity of ozone to *R. dominica* adults was delayed with greater mortalities occurring five days after exposure compared to one day after exposure, and found that Phosphine-resistant adults of *T. castaneum* and *R. dominica* were highly susceptible to ozone concentrations of 0.43 or 0.86 g/m^3 after a 24 h. exposure period. Our results from the present study demonstrate that higher sensitivity of moths to ozone, where the ozone gas increased the adult mortality percent reached to complete mortality and affected on progeny resulted from treated adults of tested insect except the treated adults with ozone concentration 5g/m3, where failed at mating and not resulted any progeny at all exposure periods in F_1 generation. This is higher mortality for adults may be indicated that during the degradation of ozone to diatomic oxygen, free radicals may be formed from reactive oxygen species. In addition, O_3 may cause the per oxidation of polyunsaturated fatty acids, resulting in the destruction of critical molecules, such as DNA and proteins. Thus, these effects, either alone or together, may result in cell damage and the death of the insects that are exposed to ozone gas thus reducing the instantaneous growth rate of insect pests (Holmstrup *et al.,* 2011).

REFERENCES

Abbott, W.S. (1925). Method of computing the effectiveness of insecticides. J. Econ. Entomol., 18: 265-267.

Abo-El-Saad M. M., Elshafie H.A., Al Ajlan A. M. and Bou-Khowh I.A. (2011). Non-chemical alternatives to methyl bromide against *Ephestia cautella* (Lepidoptera: Pyralidae): microwave and ozone. ABJNA, 2: 1222-1231.

Al-Ahmadi, S. S., Ibrahim, R. A. and Ouf, S. A. (2009). Possible control of fungal and insect infestation of date fruits using ozone. Biosci., Biotech. Res. Asia, 6(1): 17-28.

Bonjour E.L., Opit G.P., Hardin J., Jones C.L., Payton M.E. and Beeby R.L. (2011). Efficacy of ozone fumigation against the major grain pests in stored wheat. J. Econ. Entomol., 104(1):308–316.

Cogburn, R.R. and Bollich, C.N. (1980). Breeding for host plant resistance to stored rice insects (355-358). In "Biology and Breeding for Resistance to Arthropods an Pathogens in Agricultural Plants" Texas A&M University.

Duncan, D.B. (1955). Multiple range and multiple F tests. Biometrics11:1-24.

EPA, United States Environmental Protection Agency (1999). Alternative disinfectants and oxidants guidance manual. Publication: 815 R 99014.

FAO (Food and Agricultural Organization) (1988). Global survey of pesticide susceptibility of stored grain pests.Report of the 9th session of the FAO working party of experts on pest resistance to pesticides, June 18-22. Rome, Italy. 17p.

Garamoon A. A., Elakshar F. F. and Sawah M. (2009). Optimization of ozone generation at low resonance frequency.Eur.Phys. J. Appl. Phys., 48: 21002.

Grotmol, S., Dahl-Paulsen and Totland, G.K (2003). Hatchability of eggs from Atlantic cod, turbot and Atlantic halibut after disinfection with ozonated seawater. Aquaculture, 221: 245-254.

Holmstrup, M., Sørensen, J.G., Heckmann, L.H., Slotsbo, S., Hansen, P. and Hansen, L. S. (2011). Effects of ozone on gene expression and lipid peroxidation in adults and larvae of the red flour beetle (*Tribolium castaneum).* J. Stored Prod. Res., 47: 378-384.

Husain, M., Rasool, K. G., Tufail, M., Alhamdan, A. M., Mehmood, K. and Aldawood, A. S. (2015). Comparative efficacy of CO_2 and Ozone gases against *Ephestia cautella* (Lepidoptera: Pyralidae) larvae under different temperature regimes. J. Insect Sci., 15 (1): 1-5.

Hussain, H. B. (2014). Biological aspects and control of *Ephestia cautella* (Walk.) (Lepidoptera: Phycitidae) in stored dates. Bull. Entomol. Soc. Egypt, 91:187-198.

Isikber, A. A. and Athanassiou, C.G. (2015). The use of ozone gas for the control of insects and micro-organisms in stored products. J. Stored Prod. Res., 64:139-145.

Isikber, A.A., Oztekin, S., Ulusoy, R., Ozsoy, S. and Karcı, A. (2007). Effectiveness of gaseous ozone alone and in combination with low pressure or carbon dioxide against *Ephestiakuehniella* (Zell.) (Lepidoptera: Pyralidae) at short exposure time. OIBC/SROP Bulletin, 30: 205-213.

James R.R. (2011). Potential of ozone as a fumigant to control pests in honey bee (Hymenoptera: Apidae) Hives. J. Econ. Entomol., 104 (2): 353–359.

Keivanloo, E., Namaghi, H.S., Khodaparas, M.H. (2014). Effects of low ozone concentrations and short exposure times on the mortality of immature stages of the Indian meal moth, *Plodia interpunctella* (Lepidoptera: Pyralidae). J. Plant Prot. Res., 54(3): 267-271.

Kells, S. A., Mason, L. J., Maier, D. E. and Woloshuk, C. P. (2001). Efficacy and fumigation characteristics of ozone in stored maize.J.Stored Prod. Res., 37: 371-382.

Kim, J.G., Yousef, A.E. and Dave, S. (1999). Application of ozone to control insects, molds, and mycotoxins. In: Donahaye, E. J. Navarro, S., Varnava, A. (Eds.) Proceeding of the International Conference on Control Atmosphere and Fumigation of Stored Products Cyprus. April 21-26, 1996.

Law, S.E. and Kiss, E.G. (1991). Instrumentation for ozone-based insect control in agriculture. Automated agriculture for the 21st century: proceedings of the 1991 symposium. Chicago, IL. December, 16-17.

Leesch, J.G. (2002). The mortality of stored-product insects following exposure to gaseous ozone at high concentrations. p. 827–831. In: "Advances in Stored Product Protection" (P.F. Credland, D. M. Armitage, C.H. Bell, P.M. Cogan, E. Highley, eds.). Proc. 8th Int. Working Conference on Stored- Product Protection, York, UK, 22–26 July 2002, 965 pp.

Legeron, J.P. (1984). Ozone disinfection of drinking water. In: Rice, R. G., Netzer, A. (Eds), Handbook of Ozone Technology and Applications. Boston, Butterworth, Vol. II, pp. 99-121.

McDonough, M .X., Mason, L. J. and Woloshuk, C.P. (2011). Susceptibility of stored product insects to high concentrations of ozone at different exposure intervals. J. Stored Prod. Res., 47:306-310.

Mylona, K., Kogkaki, E., Sulyok, M. and Magan, N. (2014). Efficacy of gaseous ozone treatment on spore germination, growth and fumonisin production by *Fusarium verticillioides* in vitro and *in situ* in maize. J. Stored Prod. Res., 59:178-184.

Niakousari, M., Erjaee, Z. and Javadian, S. (2010). Fumigation characteristics of ozone in postharvest treatment of Kabkabdates (*Phoenix dactylifera* L.) against selected insect Infestation. J. Food Prot., 73 (4): 763–768.

Osman, M. A. M. (2009). Susceptibility of some stored product insects to gaseous ozone treatment under laboratory conditions. Bull. Entomol. Soc. Egypt, Econ. Ser., 35: 157- 170.

Ross, R. (1999). Current status of domestic and international controls for methyl bromide and the status alternatives. In: Moy, J.H., Wong, L. (Eds.), the Use of Irradiation as a Quarantine Treatment of Food and Agricultural Commodities. University of Hawaii, Manoa, pp. 4–10.

Savi, G. D., Piacentini, K.C. and Scussel, V. M. (2015). Reduction in residues of deltamethrin and fenitrothion on stored wheat grains by ozone gas. J. Stored Prod. Res., 61:65-69.

Sousa, A.H., Faroni, L.R., Guedes, R.N., Totola, M.R. and Urruchi, W. I. (2008). Ozone as a management alternative against phosphine-resistant insect pests of stored products.J. Stored Prod. Res., 44: 379-385.

Subramanyam, B. E., Savoldelli, S., Sehgal, B., Maier, D.E. and Ren, Y.L. (2014). Efficacy of ozone against stored grain insect species in wheat: laboratory and field observations. Proceedings of the 11th International Working Conference on Stored Product Protection, Chiang Mai, Thailand 24–28 November 2014, pp.489-498.

Suffet, I.H., Anselme, C. and Mallevialle, J. (1986). Removal of tastes and odors by ozonation. In: Proceedings of the American Water Wo11.7 and 0 % rks Association Annual Conference, Seminar on Ozonation. Denver, CO, AWWA.

Teotia, T.P.S. and Singh, Y. (1976). Studies on the growth of population of *Sitotroga cerealella*. Bull. Grain Technol., 14 (9-17).

Zakladnoy, G.A., Saeid, E. K. and Korteva, E. F. (2003). Biological effectiveness of ozone against grain pests, rice weevil and wheat weevil. Storage and handling agric.raw, 4:59-61

ARABIC SUMMERY

تاثير الاوزون على فراشة الحبوب ونسلها

محمد عبد الغفار محمود ، حمدى احمد محمد ، ابراهيم لبيب ابراهيم ، حسن عبد الرحيم جاد
قسم وقاية النبات – كلية الزراعة – جامعة الازهر بالقاهرة

تعتبر فراشة الحبوب واحدة من اهم الافات التى تصيب عدد من الحبوب المخزونة مثل الذرة والقمح وتم معاملة الاطوار المختلفة من هذة الافة لثلاث تركيزات من غاز الاوزون (١,٣,٥جم/م^{٣}) وستة مدد تعريض مختلفة تتراوح من نصف ساعة الى ٦ ساعات مقارنة بالمعاملة القياسية .اشارت النتائج الى ان زيادة كلا من التركيز او مدة التعريض للاوزون تؤدى الى زيادة معدل الموت فى كل الاطوار فى جيل الاباء .اوضحت النتائج ايضا ان غاز الاوزون اثر على النواحى الحيوية للنسل الناتج من الاطوار المعاملة (سواء كانت بيض او يرقات او عذارى او حشرة كاملة) فى الجيل الاول