Zagazig J. Agric. Res., Vol. 35 No. (5) 2008 1201 - 1212

EFFECT OF CARBON DIOXIDE GAS ON THE INDIAN MEAL MOTH, *PLODIA INTERPUNCTELLA* (HUB.) (LEPIDOPTERA: PHYCITIDAE)

Gharib, M.S.A.[1], A.A. El-Fishawi[2], M.M.I. Aamir[2], A.A. Shalaby[2], and D.T.M. El-Bana[2]

1. Plant Protection Research Institute, Dokki, Giza, Egypt.

2. Plant Protection Dept., Fac. Agric., Zagazig Univ., Egypt.

Accepted 27 /10 /2008

ABSTRACT: Different developmental stages of the Indian meal moth, *Plodia interpunctella* were exposed to a modified atmosphere of carbon dioxide in air-tight containers for different exposure periods. Also, some biological aspects of stages obtained from eggs and larvae that survived from CO2 treatments were studied. Cumulative mortality percentages of the different stages were increased as the exposure period to the tested gas (CO_2) increased. Complete mortality was recorded after 8, 8, 8 and 2 days for eggs, larvae, pupae and adults, respectively. Adult stage was the most sensitive to the gas than the other stages. High levels of larval mortality occurred among those descended from the gas- survived eggs especially of three days old. Percentage of larval mortality of obtained from gas-treated eggs increased as the exposure period to gas increased. Percentage of larval pupation clearly decreased for larvae obtained from gas- exposed eggs for the longest period. First larvae were more sensitive to the gas than the older ones especially in the shortest period of exposure. However, survived young larvae recorded the highest percentage of pupation than the third and fifth instars. Emergence of adults descended from eggs and larvae that survived from CO2 treatments clearly obtained as the exposure period to the gas increased.

Key words: Carbon dioxide gas, *Plodia Interpunctella*, stages, atmosphere, survived, mortality.

INTRODUCTION

Stored-grain pests cause severe quantitative and qualitative losses, which have serious implications for the availability of food, especially in those areas of the world where grain storage is poorly managed due to the lack of proper knowledge and technology (Sauer *et al.*, 1992 and Harein and Davis, 1992). In developing countries, the indiscriminate and improper use of insecticides to control the proliferation of stored grain pests has resulted in the development of insecticide-resistant strains which require an increase in pesticide dosage as well as frequency of applications to keep them under control (Champ and Dyte, 1979).

It is therefore imperative to develop alternative methods that are economically feasible and ecologically accepted to control stored grain pests. Carbon dioxide is now being used in several countries for the treatment of stored products, particularly grain bulk to control insects (Cossentine *et al.*, 2004 and Conyers and Bell, 2007). The action of CO_2 as a modified atmosphere treatment lies in its availability, relative convenience, and safety of application beside the fact that it does not leave toxic residues (Conyers *et al.*, 2001). Modified atmosphere treatments depend for their effectiveness in combating insects by removal of life supporting oxygen or addition of toxic levels of carbon dioxide which producing hypercarbonic or combination of the two in practical control treatments (Bell, 1984).

In practice, the effectiveness of any chemical control measure or non-chemical is based on insect mortality thus ignoring other effects that could be greater than those shown by mortality data alone. Alteration in the biological aspects of insects surviving from treatment with sub-lethal CO_2 concentrations in atmosphere was indicated by some investigators (Hashem and Reichmuth, 1994; Ismail *et al.*, 1995; Hashem, 2000; Aamir *et al.*, 2006 and 2008). The present work was undertaken to investigate toxicity of carbon dioxide gas on the different stages of *Plodia interpunctella*.

MATERIALS AND METHODS

Test Insect

A large batch of *P. interpunctella* (Hubner) was supplied by Stored Products Pests Dept., Plant Protection Institute, Dokki, Giza, Egypt as adults and reared under laboratory conditions.

Rearing medium used for rearing this insect was composed of dry date fruits with medium diet composed of crushed maize kernels (1000 gm) + sugar cane powder (100 gm) + glycerin (25 ml) + yeast powder (5 gm)+ tap water (75 ml). The adults were introduced to 1 kg glass jars half filled with commercial dry date fruits and the jars were kept in an incubator adjusted at 30± 1°C. and 60± 5% R.H. according to Subramanyam and Cutkomp (1987).

The following insect stages were exposed. Three ages of eggs viz., 1, 2, 3 days old were exposed to the gas. Five replicates of 20 eggs each were used. Eggs of each replicate were gently put in the plastic base of the wire gauze cage over small quantity (0.5 gm.) of medium diet. Three larval instars (first, third and fifth larval instars) were exposed to the gas. Five replicates of 10 larvae each of each instar were used for testing. Larvae of each replicate were confined in a wire gauze vial with small amount of medium diet and small pieces of dry date fruits. Pupae of different ages (1– 3 days old pupae) were used. Five replicates of 5 pupae each were gently caged in a wire gauze vial without any medium diet to be ready for gas exposure. Newly emerged adults (0– 1 day old) were used for exposure to CO_2 gas. Four replicates of 2 pairs of moths each were used and introduced in wire gauze cage supplied with small amount of medium diet to be ready for testing.

Gas Exposure Technique

Wire gauze cage was used as exposure unit. Copper wire gauze of 60 mesh/inch was used for making the cages of insect exposure to gases. These cages were cylindrical in shape (4 cm diameter x 7 cm height) supported at the bottom with a plastic base and at the other end with a plastic ring covered with a punctured covers or muslin cloth. The known urine collection plastic bags were used as gas exposure chambers. These bags are rectangular in shape (18 x 23 cm) and of 2000 ml capacity (Aamir *et al.*, 2008).

The cages of the different developmental stages of *P. interpunctella* which reared and prepared as mentioned before were put in the exposure chamber through a lateral cut and the bags were tightly sealed using a hand pressing sealer. The air inside the bag was allowed to get out by gently pressing on the bag and then the bag was connected to gas cylinder (CO_2) through a rubber hose and the long tube of the bag. The gas was allowed to purge very slowly inside the bag for about 20–30 seconds to ensure complete exchanging the air inside the bag with the tested gas without any pressure inside the bag. Valves of the treated bags were tightly closed and the bags were kept in an incubator adjusted at 30 ± 1°C for different periods of gas exposure (1, 2, 4, 6 and 8 days). At the end of each exposure periods, the bags were aerated and the wire gauze cages were taken out from the bags and kept at 30°C to record the mortality. Egg stage mortality was recorded after hatching where the hatched larvae were counted. Mortality of larvae, pupae and adults was directly recorded by observation and using of a fine brush. The mortality (%) data were corrected by Abbott (1925) and statistically analyzed according to Finney (1971) using the computer (Probit analysis program).

RESULTS AND DISCUSSION

Susceptibility of the Different Stages to CO_2

Data presented in Table 1 showed that mortality of insect increased as the exposure period to the gas increased. Adult stage was the most sensitive stage recording a complete kill after 2 days of exposure. Susceptibility of eggs differed from one age to another and from an exposure period to another for the same age. Eggs of two days old were less susceptible than the other two ages, while oldest eggs were the most susceptible. However, all eggs of different ages completely died after 8 days of exposure (Table 1). Larvae were less susceptible to the gas than eggs during the first three periods of exposure; older larvae were more tolerant than the young ones. A complete kill was occurred for all ages of larvae after 8 days of exposure. Pupal mortality took approximately the same trend recorded for larvae of the first instar. Similarly, a complete mortality of pupae was achieved after 8 days of exposure. The LT_{50} and LT_{90} values of both stages (eggs and pupae) were relatively close and less than that of larvae indicating that these two inactive stages were more susceptible to the gas than the larval stage. In other words, larval stage of this insect was the most tolerant stage to CO_2 at any period of exposure but completely died also after 8 days of exposure (Table 1).

Some Biological Aspects of Stages Obtained from Eggs that Survived Gas Treatment

Results in Table 2 indicate generally that the studied biological aspects of individuals obtained from gas-treated eggs were clearly influenced specially that developed from the oldest eggs. Larvae obtained from gas treated eggs recorded different levels of morality according to the age of treated eggs. High levels of larval mortality occurred among the larvae descended from the gas survived eggs specially those of three days old eggs. Percentages of larval mortality increased as the period of eggs exposure to gas increased. The highest mortality (79.6%) was recorded for the larvae obtained from three days old eggs exposed to gas for 6 days. It was noticed, however that pupation percentage of larvae obtained from gas- treated eggs was clearly affected by the gas exposure period. Percentage of larval pupation clearly decreased for larvae obtained from eggs exposed to the gas for the longest period compared with that descended from eggs exposed to the gas for short periods. There was no obvious effect for the age of gas-treated eggs on larval pupation. However, pupation was slightly reduced for larvae obtained from the old eggs especially that exposed to the gas for 6 days. Mortality of pupae obtained from the larvae hatched from gas-treated eggs was clearly affected by gas treatment, age of treated eggs and gas exposure period. High percentages of pupal mortality were recorded for the pupae obtained from the eggs of different ages exposed for 6 days to the tested gas. Emergence percentages of adults descended from gas treated eggs were pronouncedly influenced by age of tested eggs and gas exposure period. Emerged adults obtained from gas exposed eggs for longer periods was highly reduced than that exposed for shorter periods. Emergence of adults descended from young eggs exceed that descended from old eggs. The least percentage of adult emergence (20.91%) was recorded for the gas-treated eggs of three days old for 6 days.

Some Biological Aspects of Stages Obtained from Larvae that Survived Gas Treatment

Some biological aspects of the larvae at 1^{st}, 2^{nd}, 3^{rd} and 5^{th} instars that survived from CO_2 treatment were investigated. The percentage of mortality of pupae and adult emergence produced from survived larvae are presented in Table 3.

Table 1. Responses of the different stages of *Plodia interpunctella* exposed to carbon dioxide after different periods at 30 ± 1°C

	Treated stages	Mortality (%) at exposure periods(in days) 1	2	4	6	8	LT_{50} (in days)	LT_{90} (in days)	Slope
Eggs	One day old	-	53.75	71.93	89.25	100	2.04	5.71	2.71
	Two days old	-	41.93	60.54	84.36	100	2.62	6.71	3.15
	Three days old	-	62.57	84.12	95.66	100	1.65	4.29	3.10
Larvae	First instar	-	45.00	60.00	80.00	100	2.55	7.30	2.81
	Third instar	-	35.00	57.50	77.50	100	2.96	7.38	3.23
	Fifth instar	-	17.50	42.50	73.00	100	3.82	7.69	4.21
	Pupae	-	45.45	63.63	81.80	100	2.46	6.87	2.88
	Adults	87.50	100.00	-	-	-	-	-	-

Table 2. Some biological aspects of *Plodia interpunctella* stages obtained from eggs of different ages exposed to carbon dioxide gas for different periods

Treated eggs	Studied aspects		Exposure periods (in days) 2	4	6	8
One day old	% mortality of hatched larvae	Treated	18.91	38.47	45.46	-
		Untreated	6.42	7.80	6.67	12.2
	% pupation	Treated	81.08	61.53	54.54	-
		Untreated	93.58	92.20	93.33	87.8
	% mortality of pupae	Treated	20.00	43.75	66.67	-
		Untreated	2.55	5.17	8.09	11.69
	% emergence	Treated	80.00	56.25	33.33	-
		Untreated	97.45	94.83	91.91	88.31
Two days old	% mortality of larvae	Treated	13.73	36.80	48.70	-
		Untreated	3.80	5.20	7.90	10.81
	% pupation	Treated	86.27	63.20	51.30	-
		Untreated	96.20	94.80	92.10	89.19
	% mortality of pupae	Treated	22.07	49.68	70.55	-
		Untreated	1.32	4.10	5.72	7.58
	% emergence	Treated	77.93	50.32	29.45	-
		Untreated	98.68	95.90	94.28	92.42
Three days old	% mortality of larvae	Treated	32.91	52.37	79.60	-
		Untreated	3.85	5.13	7.80	10.53
	% pupation	Treated	74.65	63.60	47.50	-
		Untreated	96.15	94.87	92.20	89.47
	% mortality of pupae	Treated	16.80	60.60	79.09	-
		Untreated	1.34	5.41	8.46	7.58
	% emergence	Treated	83.2	39.40	20.91	-
		Untreated	98.66	94.59	91.54	92.42

Percentages of gas survived larvae that succeeded to pupate were clearly reduced as the exposure period was prolonged for the three tested larval instars. However, survived young larvae recorded the highest percent of pupation while those of the third and fifth instar recorded the lowest levels of pupation especially with the two former periods of exposure. High numbers of pupae obtained from the gas survived larvae were died and didn't complete to adults, especially of that exposed to the gas for longer period (6 days) and also that descended from the oldest larvae Table 3. It seems from the results that percentages of adult emergence that descended from gas survived larvae were highly reduced as the exposure period to gas increased and also as the mortality of pupae increased. There was no obvious effect of larval instar on the percentage of adult emergence (Table 3).

Table 3. Some biological aspects of *Plodia interpunctella* stages descended from larvae of different ages exposed to carbon dioxide gas for different periods

Larval instars	Studied aspects		Exposure periods (in days)			
			2	4	6	8
First instar	% pupation	Treated	81.81	81.25	50.00	-
		Untreated	94.87	92.30	89.74	84.20
	% mortality of pupae	Treated	16.67	38.46	75.00	-
		Untreated	2.78	5.60	8.34	11.20
	% emergence	Treated	83.33	61.53	25.00	-
		Untreated	97.22	94.40	91.66	88.80
Third instar	% pupation	Treated	73.07	58.82	44.44	-
		Untreated	97.43	94.87	92.10	89.47
	% mortality of pupae	Treated	26.32	50.00	75.00	-
		Untreated	2.71	6.90	8.34	8.60
	% emergence	Treated	73.68	50.00	25.00	-
		Untreated	97.29	92.10	91.66	91.40
Fifth instar	% pupation	Treated	69.69	65.21	50.00	-
		Untreated	95.00	94.87	94.90	92.89
	% mortality of pupae	Treated	26.09	46.67	60.00	-
		Untreated	0.00	2.71	8.90	5.90
	% emergence	Treated	73.91	53.33	40.00	-
		Untreated	100.00	97.29	91.90	94.10

These findings agree with those obtained by some authors such as Harein and Press (1968) who exposed larvae of *P. interpunctella* to mixtures of nitrogen, oxygen and carbon dioxide for 7 and 14 days. It was found that mortalities increased by extending the exposure period from 7 to 14 days. Marzke and Pearman (1970) who exposed larvae of the Indian meal moth, *P. interpunctella* to CO_2 for different periods found that larval percentage of mortality increased as the period of exposure increased. They also added that mortality varied with age and species of insects. Tunc (1983) studied the effect of low oxygen and high carbon dioxide atmospheres on the eggs and larvae of *P. interpunctella*. Results indicated that 0– 24 h. old eggs were killed by 100% in low oxygen atmospheres (1.8 and 4.2%) after 3 and 4 days of exposure respectively. Only 7% of the eggs could hatch in 5.7% CO_2 after 4 days of exposure. High carbon dioxide atmospheres (19.7–44.3%) caused 100% or little less morality of the eggs over 3 days exposure. It was also found that 13– 16 days old larvae were more tolerant than the eggs in both atmospheres. Suss and Locatelli (1991) mentioned that carbon dioxide concentration of 70% in silos containing 1000 tones of wheat infested with *Sitophilus granarius, S. oryzae, Rhyzopertha dominica, Tribolium castaneum* and *Plodia interpunctella* was more effective against the test insect at any time of sampling after treatment. Sauer and Shelton (2002) exposed the pupae of *P. interpunctella* to atmospheres of 60 or 80 or 98% CO_2 in N_2 or 60 or 80% CO_2 in air at 26.7 or 32.2 °C. and 60% R.H. At both temperatures, high CO_2 concentration treatments combined with nitrogen killed pupae faster than high CO_2 concentration treatments combined with air. Navarro *et al.* (2003) determined the effectiveness of vacuum or CO_2 in combination with increased temperature. For controlling *Tribolium castaneum, Ephestia cautella* and *P. interpunctella.* For *E. cautella* and *P. interpunctela* adults were the most susceptible, with LT_{90} of only 6 and 5h., respectively. Awadalla (2006) studied the effect of pure carbon dioxide against *Sitotroga cerealella* (Olivier), He reported that cumulative mortality of the different stages increased as the exposure period increased to reach 100% in the 4th day for adults, in the 7th day for eggs and in the 9th day for larvae and pupae, respectively.

REFERENCES

Aamir, M.M.I., A.A. EL-Fishawi, A.A. Shalaby and D.T.I. El-Bana. 2008. Control of *Sitophilus oryzae* (L.) by Malathion and two of the inert gases in a comparative study. Zagazig J. Agric. Res., 35 (3): 621- 635.

Aamir, M.M.I., K.A. Gouhar, M.M. El-Zohery and M.S. Awadalla. 2006. Efficacy of pure carbon dioxide gas against some insect species of stored products at different temperatures. Zagazig J. Agric. Res., 33 (2): 283 – 296.

Awadalla, M.S.M. 2006. Studies on certain stored grain pests and its control. Ph.D. Thesis, Fac,Agric., Zagazig University, 345. pp.

Abbott, W.S. 1925. A method of computing the effectiveness of insecticides. J. Econ., Entomol., 18 (3): 265-267.

Bell, C.H. 1984. Effect of oxygen on the toxicity of carbon dioxide to storage insects. In "Controlled Atmosphere and Fumigation in Grain Storage's" Ripp, B.E. (Ed.), Elsevier, Amsterdam, pp. 67-74.

Champ, B.R. and C.E. Dyte. 1979. Report of the FAO global survey of pesticide susceptibility of stored grain pests. FAO Plant Products / Protection Service No. 5, FAO, Rome.

Conyers, S.T. C.H. Bell, B. Harral and B.E. Liewellin, 2001. Project report no., 256. Practical and modeling studies on the use of modified atmospheres for insect and mite control in grain stores. HGCA-Project-Report. (256): 38 - 43.

Conyers, S.T. and C.H. Bell. 2007. A novel use of modified atmospheres: Storage insect population control. J. Stored Prod. Res. 43 (4): 367-374.

Cossentine, J.E., P.L. Sholberg, L.B.J. Jensen, K.E. Bedford and T.C. Shephard. 2004. Fumigation of empty fruit bins with carbon dioxide to control diapausing codling moth larvae and *Penicillium expansum* Link. Ex. Thom spores. Hort. Science, 39(2): 429-432.

Finney, D.J. 1971. Probit analysis, 3rd ed., Cambridge University Press: 333 pp.

Harein, P.K. and R. Davis. 1992. Control of stored grain insects. In : "Storage of cereals and their products", Saver, D.B. (Ed.), 4 Ed., American Association of Cereal Chemists, St Paul, MN, PP. 491-534.

Harein, P.K. and A.F. Press. 1968. Mortality of stored peanut insects exposed to mixtures of atmospheric gases at various temperatures. J. Stored Prod, Res., 4 : 77-82.

Hashem, M.Y. 2000. Role of bruchid beetle *Callosobruchus maculatus* (F.) in faba bean yield losses during storage and its control using inert gases. Integrated Protection of Stored Products, IOBC Bull. 23:275-283.

Hashem, M.Y. and C.H. Reichmuth. 1994. Interaction effects of high carbon dioxide or low oxygen atmospheres and temperature on hatchability of eggs of three stored product moths. J. Plant Diseases and Protection, 101 (2): 178-182.

Ismail, I.I., E.E. Omar and M.Y. Hashem. 1995. Effect of sub-lethal concentrations of CO_2 – air mixture on some biological aspects of *Sitophilus oryzae*. Egypt. J. Agric. Res., 73 (1): 167-173.

Marzke, F.O. and G.C. Pearman. 1970. Mortality of red flour beetle and Indian meal moth larvae in simulated peanut storages purged for short periods with CO_2 or N_2. J. Econ. Entomol., 63: 817-819.

Navarro, S., S. Finkelman, G. Sabio, A. Isikber, R. Dias, M. Rinder and A. Azrieli. 2003. Enhanced effectiveness of vacuum or CO_2 in combination with increased temperatures for control of storage insects. Advances in Stored Product Protection Proceedings of the 8th International Working Conference on Stored Product Protection, York, UK, 818-822.

Sauer, J.A. and M.D. Shelton. 2002. High temperature controlled atmosphere for post-harvest control of Indian meal moth (Lepidoptera: Pyralidae) on preserved flowers. J. Econ. Entomol., 95(5) : 1074-1078.

Sauer, D.B., R.A. Meronuck and C.M. Christensen. 1992. Micoflora in: "storage of cereal grains and their products", Sauer, D.B. (Ed.), 4th ed. American Association of Cereal Chemists, St Paul, MN, pp. 313 – 340.

Subramanyam, B. and L.K. Suss, L. and D.P. Locatelli. 1991. The use of carbon dioxide in eggs and larvae of *Plodia interpunctella* (Hubner). Zeitschrift Fur Angewandte Entomologie., 95: 53 – 57.

Tunc, I. 1983. The effect of low oxygen and high carbon dioxide atmospheres on the cereal pest control. Tecnica Molitoria, 42 (4): 333-338

Cutkomp. 1987. Total lipids and free fatty acid composition in male and female larvae of Indian meal moth and almond moth (Lepidoptera: Pyralidae). Great Lakes Entomologist, 20: 99-102.

تأثير غاز ثاني أكسيد الكربون علي فراشة جريش الذرة الهندية
Plodia interpunctella (Hub.)
(LEPIDOPTERA:PHYCITIDAE)

محروس سليمان غريب[١]-عادل عبد الحميد الفيشاوي[٢]-محمد محمد إبراهيم عامر[٢]
عطا عطا شلبي[٢]- دعاء ثابت البنا[٢]

١- معهد بحوث وقاية النباتات- الدقي- جيزة – مصر.

٢- قسم وقاية النبات – كلية الزراعة- جامعة الزقازيق.

عرضت الأطوار المختلفة لفراشة جريش الذرة الهندية *Plodia interpunctella* لغاز ثاني أكسيد الكربون في أوعية محكمة الغلق ولفترات تعريض مختلفة، كذلك دراسة بعض التغيرات البيولوجية (نسبة التعذر ونسبة خروج الحشرات الكاملة) في حياة الأفراد الباقية حية و الناتجة من بيض ويرقات (أعمار مختلفة) تم معاملتها بالغاز. يمكن تلخيص النتائج المتحصل عليها كالأتي: تم الوصول إلى نسبة موت كاملة (١٠٠٪) لجميع الأطوار بعد ٨، ٨، ٨ و ٢ يوم للبيض، اليرقات، العذارى والحشرات الكاملة على الترتيب. كانت الحشرات الكاملة أكثر الأطوار حساسية للغاز حيث سجلت موت كامل بعد أقصر فترة تعريض للغاز. كانت نسبة الموت عالية في اليرقات الناتجة من بيض معامل بالغاز وبخاصة الناتجة من البيض الأطول عمرا وظل حيا بعد المعاملة. نسبة موت اليرقات المنحدرة من بيض ظل حيا بعد المعاملة بالغازات كانت عالية مع زيادة فترة التعريض للغاز. نسبة تعذر اليرقات المنحدرة من بيض معامل بالغاز انخفضت بوضوح عندما زادت فترة التعريض للغاز. اليرقات الحديثة العمر الباقية حية بعد معاملة الغاز سجلت أعلي نسبة تعذر عن اليرقات الأخرى الأطول عمراً. انخفضت نسبة خروج الحشرات الكاملة المنحدرة من بيض أو يرقات معاملة بالغاز بزيادة فترة التعريض للغاز.