#### EFFICIENCY OF KATEL-SOUS AND BLACK PEPPER SEEDS DUSTS UNDER **ATMOSPHERES** MODIFIED AGAINST SOME STORED PRODUCT INSECTS

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## **ABSTRACT**

These experiments were conducted inside gastight steel bins, each of about 0.5 m<sup>3</sup> volume and filled with approximately 450 kg wheat grains. The bins were situated on the ground of the agricultural experimental farm of the Faculty of Agric., Moshtohor, Tukh, Qalvubia, Grain temperature was 26±2 °C. Modified atmospheres (MAs) tested were 99% N2 and 30±5% CO2. Tested dusts used were katel-sous dust and powdered black pepper seeds. Tested insects were the adults of Sitophilus oryzae L., Rhizopertha dominica F., Tribolium castaneum Herbst., and active and diapausing larvae of Trogoderma granarium Everts. Obtained results indicated that katel-sous was more active than black pepper to all tested insects. Diapausing larvae of T. grananum were the least sensitive species to all the test dusts and MAs, followed by T. castaneum adults. The sensitivity of the tested insects to dusts or MAs varied according to insect species. The efficacy of the tested MA of CO2 against the tested insects was greater than the MA of N2. Percentage mortality of all tested insects was dependent on concentration and exposure period. The results also showed that the efficacy and combined action of tested dusts under MA of N2 or CO2 were pronounced additive, potentiation and antagonistic effects according to dusts concentration, exposure period and insect species.

## INTRODUCTION

Post-harvest losses are often more significant than crop losses which occur in the field. In Africa as much as 20-50% of the grain can be lost in maize and pulses because of insects' infestations (FAO, 1985). In addition to the poisonous gases that are used for stored-products' protection, the normal gases of the atmosphere can be altered to achieve control. The use and manipulation of natural components of the atmosphere, e.g. oxygen, nitrogen and carbon dioxide, to preserve food is referred to as modified atmospheres. Modified atmospheres techniques are widely used in the storage of perishable commodities such as grain, vegetables, cut flowers, etc. to control some insects in these products (Navarro and jay, 1987; Ofuya and Reichmuth, 1993; Hashem and Reichmuth, 1994; El-Lakwah et al., 1997; Mohamed, 1999 and El-Lakwah et al., 2004). The most extensive use of modified atmospheres for insect control is on grain and similar commodities. The atmospheres are modified by adding various levels of carbon dioxide and high levels of nitrogen. Frequent application of insecticides by methods that did not completely kill an insect infestation developed an insect population that became increasingly resistant to several insecticides and many effective insecticides have been banned for health and environmental reasons and only a few new insecticides have been developed to replace them. For these reasons there is a renewed interest in physical control methods even though most of these methods may be more expensive and not as effective in

eliminating or preventing an infestation as are chemical treatments. Non-silica dusts, such as rock phosphate, have been used in Egypt (Fam et al. 1974) and lime (calcium oxide) provides some control (Golob and Webley 1980). The main advantage of using inert dusts is their safety to humans and animals. Inert dusts in stored grains can provide continuous protection from insect infestations and do not affect the baking quality of wheat. Inert dusts kill insects by causing moisture to move out of the insect's body. The dust either scratches through the insect's waxy integument that normally prevents excessive loss of moisture to the dry grain and intergranular air. There is a common tradition among farmers at the subsistence level to use natural plant products such as black pepper dusts instead and these alternatives appear to have increased their importance.

The aim of the present investigation is to evaluate the effect of a modified atmosphere (CO2 or N2) associated with two tested materials (katel-sous dust and black pepper dust) against some stored product insects.

## **MATERIALS AND METHODS**

#### Tested insects:

Laboratory strains of the rice weevil (Sitophilus oryzae L.), the lesser grain borer (Rhizopertha dominica F.) and the red flour beetle (Tribolium castaneum Herbst.) were used as adlut stage in these experiments, in addition to active and diapausing larvae of Khapra beetle (Trogoderma granarium Everts).

## Insects rearing:

Tested insect species were reared in glass jars of approx. 250 ml volume, each jar contained about 200 g. of wheat kernels for *S. oryzae*, *R. dominica* and *T. granarium* or wheat flour in case of *T. castaneum* jars were covered with muslin cloth fixed with rubber band. Insect cultures were kept under controlled conditions of 26±2 °C and 55±5% RH in the rearing room of the laboratory. Active and diapausing larvae of 0.8±0.2 mg/l and 2.7±0.2 mg/l, respectively were used for the bioassay tests. The used active larvae were those of 3<sup>rd</sup> and 4<sup>th</sup> instars. Diapausing larvae were collected from a roll paper which had been placed on the top of the culture media. Diapausing larvae are known to be more tolerant to fumigants, inert gases and insecticides than active larvae taken from culture media (Bell *et al.*, 1984). Wheat grains were well treated by freezing at –18 °C for two weeks before application to eliminate any possible infestation by any other species. The moisture content of the grains was about 14%.

#### Gases used:

- -Carbon dioxide was provided as a pure gas in a pressure cylinder and monitored using a CO<sub>2</sub> Gas-Analyzer model 200-600 (Gow-Mac Instrument Co., USA).
- Nitrogen was provided as a pure gas of more than 99% gas in a pressure cylinder and monitored using an Oxygen Analyzer 572, Servomex, England.
   Tested materials:

-Katel-sous dust: Grain protectant contains 84% calcium triphosphate plus 16% sulpher, produced by Kafr El-Zayat Company for

Pesticides, Egypt, mixed with the grains at its recommended rate (1%) and half of it (0.5 w/w).

-Black pepper seeds(*Piper nigrum* L.): Obtained from herb shop, ground well in an electric mill, sieved using a 300 mesh sieve and kept in a clean glass jar. The pulverized seed powder was tested at 1 and 2 % w/w concentrations.

#### **Bioassay tests:**

The appropriate amount of the above mentioned dusts to give the required concentrations was mixed thoroughly with wheat grains or wheat flour. Batches of 30 adult insects (1-2 week-old) or 30 active and diapausing larvae of *T. granarium* were confined in the cloth bags (10x16 cm) each containing 50 g. treated or untreated wheat kernels for *S. oryzae*, *R. dominica* and *T. granarium* or wheat flour in case of *T. castaneum*. Each bag was closed well by a rubber band and placed into the steel bins (each of 0.5 m<sup>3</sup> volume). All tests were triplicated.

# Purging of the gases inside the bin:

After introducing the insect samples inside the bin, and to obtain 30% CO<sub>2</sub> concentration, the cylinder of CO<sub>2</sub> was connected with the upper valve of the bin through a polyethylene tube. The valve of the cylinder was opened for one and half minute, while the bottom valve of the bin was opened and closed after one minute from the initial purging of gas.

For achieving modified atmosphere of high nitrogen content (99%  $N_2$ ), the upper valve of the bin was opened for 5 minutes after the connection of  $N_2$ -cylinder with bin, while the bottom bin valve was opened for two minutes, after which it was closed tightly.

## Fumigation procedure:

Insect samples were fumigated with the different treatments for different exposure periods (2, 3, 5, 7 and 14 days) inside gastight steel bins during summer time at temperature ca. 26±2°C and 55±5% RH.

## Post-fumigation procedure:

Aeration was carried out after exposure periods, the mortality of the insect adults was determined after 48 hrs.

#### Calculation of Joint Action:

For the evaluation of the joint action of katel-sous dust and powdered black pepper seeds under modified atmospheres, the following equation was adopted as reported by Mansour *et al.* (1966):

# Co-toxicity Factor = (<u>%Observed Mortality</u> - <u>%Expected Mortality</u>) <sub>x</sub> 100 <u>%Expected Mortality</u>

This factor was used to classify the results into three categories. A positive factor of +20 or more meant synergistic effect, a negative factor of – 20 or more meant antagonism, and any intermediate value, i.e. between +20 and -20 was considered an additive effect.

# RESULTS AND DISCUSSION

Mortality percentages after treatments indicated that the efficacy of tested dusts and CO2 or N2 when each of them was used separately or in mixtures (the dusts under MA of CO2 or N2) was increased with extending the time of exposure with all the insect species.

A- Efficacy and Combined Action of Tested Dusts Under MA of 30±5% CO<sub>2</sub> to Various Insect Species at Grain Temperature of 26± 2 °C.

The results in table (1) present the efficacy and combined action of black pepper dust under MA of 30±5% CO2 to the adults of S. oryzae, R. dominica and T. castaneum as well as active and diapausing larvae of T. granarium at grain temperature of 26± 2 °C and 55± 5% RH. Mortality percentages resulting from mixtures of black pepper dust and CO2 were higher than those obtained from each treatment alone with the adults of S. oryzae, R. dominica and T. castaneum while mortality percentages resulting from the mentioned mixtures for the active and diapausing larvae of T. granarium were lower than mortalities resulted from CO<sub>2</sub> alone. Black pepper dust at 1% and 7days-exposure gave 65, 43 and 5% mortality with the adults of S. oryzae, R. dominica and T. castaneum, respectively. While, those were 28 and 38 as well as 6 and 8% after 14 days post-treatment at the low and high black pepper dust concentration for the active and diapausing larvae of T. granarium, respectively. However, mortalities resulted from CO<sub>2</sub> alone were 30, 6 and 3% at 2 days-exposure and these values increased to 97, 99 and 78% after 7 days-exposure for the adults of S. oryzae, R. dominica and T. castaneum, respectively. Meanwhile, these values were 25 and 16% at 5 days-exposure and increased to 77 and 66% after 14 days-exposure for the active and diapausing larvae of T. granarium, respectively. Treatment of black pepper dust at 2% under MA of 30±5% CO2 resulted in complete kill after one week exposure time for S. oryzae, R. dominica and T. castaneum adults, while this combined treatment resulted in 72 and 30% mortality after two weeks exposure period with the active and diapausing larvae of T. granarium, respectively. Co-toxicity values resulting from the mixtures of black pepper dust at 1 and 2% plus 30±5% CO2 indicated potentiation or additive effect at short exposure period while a negative effect was observed at longer period with S. oryzae adults. While in case of R. dominica. potentiation effect was observed at short exposure, but this effect turned to antagonistic effect at longer period. In case of T. castaneum, data indicated potentiation effect after 2 days post-treatment while after one week exposure time, this effect changed to be additive. Meanwhile, the active and diapausing larvae of T. granarium, using black pepper dust at 1 and 2% under MA of 30±5% CO2 resulted in antagonistic effect at the various tested exposure periods of 5 - 14 days.

Table (2) presents the efficacy and combined action of katel-sous dust at 0.5 and 1% concentration under MA of  $30\pm5\%$  CO<sub>2</sub> to the tested insects at grain temperature of  $26\pm2$  °C and  $55\pm5\%$  RH. The obtained mortality percentages showed that 1% katel-sous under MA of CO<sub>2</sub> caused 100% mortality of *S. oryzae* and *R. dominica* adults after two days exposure.

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while this combined treatment resulted in 70% mortality after one week exposure period with T. castaneum adult. Treatment of the active larvae of T. grananum with katel-sous dust at the lower concentration under MA of  $CO_2$  gave a higher mortality values (26-68%) than those of the higher concentration (20-30%). Co-toxicity values resulting from the mixtures of katel-sous dust at 0.5 and 1% plus  $30\pm5\%$   $CO_2$  indicated potentiation or additive effect with S. oryzae adults. While in case of R. dominica, potentiation or antagonistic effect was observed. Co-toxicity values of the mixtures showed a potentiation effect with the T. castaneum adults, but this effect turned to additive effect after one week exposure period. In case of active and diapausing larvae of T. grananum co-toxicity values resulting from addition of  $CO_2$  at  $30\pm5\%$  concentration to katel-sous dust at its two application rates induced antagonistic effect with all various periods of exposure.

Table (1): Efficacy and Combined Action of Black Pepper seeds Bp)

Dust under MA of 30±5% CO₂ to Various Insect Species at

Grain Temperature of 26± 2 °C and 55± 5% RH.

% Mortality at Various Treatments Type of Co-toxicity Exposure Bp% MA of Mixtures of Joint Period Factor (W/W) 30± 5% Action Bp+CO<sub>2</sub> (days) CO2 1 (a) 2 (b) (b) (b) (a) (b) (a) (a) S. oryzae-adults 1 30 98 223 216 0 97 5 7 90 98 99 2 d 1 8 d 44 52 94 100 100 -28 -32 a a 65 75 97 100 100 -38 -42 а a R. dominica-aduits 2 3 5 7 481 465 10 11 6 93 96 3 3 21 22 50 35 36 96 98 5 S 41 98 -23 -28 32 100 100 a a 43 58 -30 -36 99 100 100 а а T. castaneum-aduits 425 0 925 0 3 21 41 s 5 1 1 17 32 55 78 206 d s 2 3 34 43 70 19 89 đ 5 7 78 91 100 10 17 d d T. granarium active larvae 22 7 25 26 -44 -45 18 a a 11 24 31 32 42 -24 -24 a a 10 21 31 54 47 62 -37 -27 a a 28 38 77 62 72 -41 -37 а а granarium diapause larvae 2 1 16 4 8 -77 -56 a a 1 3 26 15 21 -44 -28 а a 10 5 5 46 16 28 -69 -45 a а 6 8 66 18 30 -75 -60 а a

a = antagonistic effect / d = additive effect / s = potentiation effect

Table (2): Efficacy and Combined Action of Katel-Sous (KS) Dust under MA of 30±5% CO₂ to Various Insect Species at Grain

Temperature of 26± 2 °C and 55± 5% RH.

	% N		at Variou		ents			Tyr	e of
Exposure Period (days)	KS% (W/W)		MA of 30± 5%	Mixtures of Ks+CO <sub>2</sub>		Co-toxicity Factor		Type of Joint Action	
(44,5)	0.5 (a)	1 (b)	CO2	(a)	(b)	(a)	(b)	(a)	(b)
			S. c	ryzae-ad	luits				
2	1	2	30	97	100	213	213	5	5
2 3 5 7	2	4	90	98	100	7	6	d	d
5	17	24	94	100	100	-10	-12	d	d
7	33	45	97	100	100	-23	-30	a	a
	•		R. do	minica-	adults	•		•	
2	61	71	6	95	100	42	30	8	5
2 3 5	83	90	50	97	100	-27	-29	а	a
5	93	98	98	100	100	-48	-49	а	а
7	95	99	99	100	100	-49	-50	а	а
	•		T. cas	taneum-	aduits	•			
2	1 0	1	3	10	25	233	800	5	5
3	1 1	1	17	53	41	194	278	5	5
2 3 5 7	8	18	34	96	55	129	92	8	5
7	15	33	78	100	70	8	-10	d	d
	•		T. grana	rium acti	ve iarvae			•	
5	18	22	25	26	20	-40	-47	a	а
7	32	41	31	28	27	-56	-43	а	а
10	37	43	54	48	28	-55	-51	а	а
14	46	64	77	68	30	-55	-54	а	а
	•	1	r. granari	um diapa	use larva	10		,	
5	6	21	16	15	52	-32	-46	а	a
7	15	32	26	20	84	-51	-54	а	а
10	23	42	46	22	96	-68	-68	а	а
14	28	53	66	24	98	-75	-75	а	а

a = antagonistic effect / d = additive effect / s = potentiation effect

# B- Efficacy and Combined Action of Tested Dusts Under MA of 99% N<sub>2</sub> to Various Insect Species at Grain Temperature of 26± 2 °C.

Data of the efficacy and combined action of black pepper dust at 1 and 2% under MA of 99%  $\rm N_2$  to the adults of *S. oryzae*, *R. dominica* and *T. castaneum* as well as active and diapausing larvae of *T. granarium* at grain temperature of 26± 2 °C and 55± 5% RH. are given in table (3). The results shows that, using MA of N2 alone or combined with the black pepper dust followed a time-dependent phenomenon. Generally, the results revealed that the combined treatment of black pepper dust under MA of 99%  $\rm N_2$  enhanced the resulted mortalities of all the tested insects at all exposure periods except with *T. granarium* active larvae after two weeks exposure period. Co-toxicity values resulting from the mixtures indicated mostly potentiation effects or sometimes additive actions with all the insects at various periods of exposure.

Table (3): Efficacy and Combined Action of Black Pepper (Bp) Seeds
Dust under MA of 99% N₂ to Various Insect Species at Grain

Temperature of 26± 2 °C and 55± 5% RH.

	% I		at Variou	s Treatr	nents			Typ	e of
Exposure Period (days)	(W	Bp% (W/W)		Mixtures of Bp+N <sub>2</sub>		Co-toxicity Factor		Joint Action	
	1 (a)	2 (b)	99% N <sub>2</sub>	(a)	(b)	(a)	(b)	(a)	(b)
			S. (	oryzae-a	duits				
2	0	1	25	96	98	284	277	8	8
3	1	7	58	98	100	66	2	8	5
5	44	52	94	100	100	-28	-32	a	а
7 .	65	75	100	100	100	0	0	d	d
	•		R. d	ominica-	adults				
2	10	11	8	38	48	111	153	8	8
3	21	22	11	91	93	184	182	5	5
5	32	41	15	93	94	98	68	5	5
7	43	58	48	96	100	6	-6	d	d
	•		T. ca	staneum	-aduits				
2	0	0	2	28	40	1300	1900	5	5
3	] 1	1	3	36	46	800	1050	5	5
3 5	2 5	3	6	75	81	838	800	5	5
7	5	7	24	85	88	193	193	5	5
			T. grana	irium act	ive larvae				
5	7	22	3	43	50	330	100	5	5
7	11	24	8	48	51	153	59	5	8
10	21	31	45	58	65	-12	-15	d	d
14	28	38	94	81	83	-34	-37	a	а
	•		T. granari	um diap	ause larv	ae			
5	1	2	5	38	46	533	557	5	5
7	1 1	3 5	6	45	46	543	411	5	5
10	5	5	15	51	60	155	200	5	5
14	6	8	66	65	73	-10	-1	d	d

a = antagonistic effect / d = additive effect / s = potentiation effect

Table (4) shows the results of efficacy and combined action of katel-sous dust at 0.5 and 1% under MA of  $99\%\ N_2$  to the tested insects at grain temperature of  $26\pm\ 2$  °C and  $55\pm\ 5\%$  RH. Mortality percentages apparently indicated that the efficacy of katel-sous dust was concentration-dependent, treatment of the all tested insects with katel-sous dust at the two applied concentrations under the above mentioned MA of  $N_2$  gave considerably higher mortality values than those of each component alone at various exposure periods. Co-toxicity values of the mixtures showed mostly potentiation or additive effects with the four insect species at the different periods of exposure.

Katel-sous dust at 1% under MA of 99%  $N_2$  resulted in complete kill after two days exposure period with the adults of S. oryzae, while these combined treatment resulted in complete kill after 5 and 14 days exposure period with T. castaneum adults and active larvae of T. granarium, respectively. 1% katel-sous under MA of  $CO_2$  caused 100% mortality of R. dominica adults after two days exposure period, while the same combined treatment caused 98% mortality of diapausing larvae of T. granarium after 14 days exposure period. Generally, the results revealed that katel-sous dust at 1% was more active than the black pepper dust to all tested insect species.

Table (4): Efficacy and Combind Action of Katel-Sous (KS)) Dust under MA of 99% N₂ to Various Insect Species at Grain Temperature of 26± 2 °C and 55± 5% RH.

F	% Mortality at Various Treatments							Type of	
Exposure Period (days)	(W	KS% (W/W)		Mixtures of Ks+N₂		Co-toxicity Factor		Joint Action	
	0.5 (a)	; 1 (b)	99% N <sub>2</sub>	(a)	(b)	(a)	(b)	(a)	(b)
			<b>S</b> . c	pryzae-ac	luits				
2 3	1	2	25	98	100	277	270	S	5
3	2	4	58	100	100	67	61	S	\$
5	17	24	94	100	100	-10	-15	d	d
7	33	45	100	100	100	0	0	d	d
	'		R. de	ominica∹	adults			•	
2	61	71	. 8	60	63	-13	-9	d	d
2 3	83	90	11	95	98	1	-3	d	d
5	93	98	15	98	100	-9	-12	d	d
7	95	99	48	100	100	-30	-32	а	а
1	•		T. cas	taneum-	adults			•	
2	1 0	1		95	97	4650	3133	s	s
3	1 1	1	2	97	98	2325	2350	s	s
3 5	8	18	6	98	100	600	317	S	s
7	15	33	24	100	100	156	75	s	5
ļ	•		T. grana	rium acti	ve larvae				
5	18	22	3	53	53	152	112	s	s
5 7	32	41	8	55	60	41	50	s	s
10	37	43	45	68	78	-17	-11	d	d
14	46	64	94	82	100	-41	-37	а	a
	,	•	T. granari		use larva				-
5	6	21	5	50	51	355	96	s	s
5 7	15	32	6	51	56	143	47	S	s
10	23	42	15	55	71	45	25	5	5
14	28	53	66	77	85	-18	-29	d	ď

a = antagonistic effect / d = additive effect / s = potentiation effect

Similar results were observed by Darwish, (1997); Mohamed, (1999) and El-Lakwah *et al.* (1998, 2000a and 2000b) in their experiments on the combined action of plant extracts in presence of various atmospheres of either CO<sub>2</sub> or N<sub>2</sub>. Also El-Lakwah *et al.* (2002) reported that the combined action of Dill and Cumin seed extracts under modified atmospheres of various carbon dioxide concentrations or modified atmospheres of very high nitrogen content resulted in either synergistic or additive effects with active and diapausing larvae of *T. granarium* at different periods of exposure.

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فاعلية مسحوق قاتل سوس ومسحوق بذور الفلفل الأسود تحت الأجواء المعدلية ضد بعض حشرات الحبوب المخزونة

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\* \* كلية الزراعة بدمياط - جامعة المنصورة.

أُجَريت التجارب داخل صُوامُع محكمة الغلق حَجَم كَلاَ مَنها ٢/١م٣ وكانتُ هذه الصوامع ممتلئــة بحوالي ٤٥٠ كيلو جرام من حبوب القمح وتم وضع الصوامع داخل مزرعة كلية الزراعة بمشتهر.

و قد أوضحت النتائج أن مسحوق قاتل سوس كان أعلى تأثيرا من مسحوق الفلف الأسود على جميع الحشرات تحت الدراسة، كذلك تباينت حساسية العشرات المختبرة للمسحوقين وكذلك للأجواء المعدلة على حسب نوع الحشرة.

كما أوضحت النتائج أن اليرقات الساكنة لخنفساء الصعيد كانب الأقبل حساسية لتأثير كلا المسحوقين وكذلك الأجواء المعدلة سواء من ثاني أكسيد الكربون أو النيتروجين عند استخدام كلا منهما منفر دا ثم تلاها في الأقل حساسية الحشرات الكاملة لخنفساء الدقيق الصدنية.

وقد أشارت النتائج كذلك إلى أن فعالية غاز ثاني أكسيد الكربون كانت أعلى من فعالية غاز النيتروجين عنسد استخدام كلا منهما منفردا، كذلك أظهرت النتائج أن نسبة الموت للحشرات تحت الدراسة توقفت على تُركيز المسحوق المستخدم، كما زاد معدل الموت بزيادة فترة التعريض.

كذلك أوضحت النتائج أن التاثير المشترك للمساحيق المختبرة تحت الأجواء المعدلة من كل من غازي ثاني لأكسيد الكربون والنيتروجين إما تأثير تقوية أو إضافة أو تضاد، وكان هذا التأثير متوقفا على نوع الحشرة، تركيز المسحوق وكذلك فترة التعريض.