EGYPTIAN ACADEMIC JOURNAL OF

BIOLOGICAL SCIENCES

TOXICOLOGY & PEST CONTROL

F

ISSN
2090-0791

WWW.EAJBS.EG.NET

Vol. 13 No. 2 (2021)

Citation: *Egypt. Acad. J. Biolog. Sci.* (F.Toxicology& Pest control) *Vol.13(2)pp71-81(2021)*
DOI: 10.21608/EAJBSF.2021.187542

Egypt. Acad. J. Biolog. Sci., 13(2):71-81(2021)

Egyptian Academic Journal of Biological Sciences
F. Toxicology & Pest Control
ISSN: 2090 - 0791
http://eajbsf.journals.ekb.eg/

Effect of Biological Insecticide, Chemical Insecticide and Phosphate Fertilizer on *Callosobruchus maculatus* (F) (Coleoptera: Chrysomelidae) and *Rhyzopertha dominica* (F.) (Coleoptera: Bostrychidae)

Rasha Essameldin Saber[1]; Mawaheb Mahmoud Zewar[1] and Mohamed Abdelsattar[2]
1-Plant Protection Research Institute, Stored Products Department, Agriculture Research Center.
2-Agricultural Engineering Research Institute, Department of Plant Molecular Biology Agriculture Research Center**.**
E-mail***** rashaessam35@gmail.com

ARTICLEINFO
Article History
Received: 28/5/2021
Accepted: 25/7/2021

***Keywords*:**
Vicia faba., *Triticum durum*.,*Callosobruchus maculatus*., *Rhyzopertha dominica*., chemical insecticide., biological insecticide and phosphate fertilizer

ABSTRACT

This study was carried out to evaluate toxicity and residual effect of chemical insecticide deltamethrin, biological insecticide *Beauveria bassianaa* and phosphate fertilizer. This Evaluation was conducted against *Callosobruchus maculatus* (F.) adults and *Rhyzopertha dominica* (F.). Results indicated that biological insecticide *was* the most effective material that still kills adults of *C. maculatus* and *R. dominica* till the end of the storage period (3months). The chemical insecticide caused a complete death for *C. maculatus* adults till the end of the storage period while it decreased with time for adults of *R. dominica*. All treatments did not affect seed germination and water absorption.

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INTRODUCTION

According to the United Nations Food and Agriculture Organization (FAO), agricultural production must increase by 50% by 2050 to meet global food demand. (Barian L. Beres *et al*., 2020). The quality of the seeds sown is critical to the successful production of any crop (Rasha *et al*., 2017). Storage pest has become an increasing threat to food safety; losses over 30% or more have been recorded in Africa (Lale & Ofuya, 2001).

Faba bean (*Vicia faba* L.) and Durum wheat (*Triticum durum* Desf.) are the most important food crops in the world. Faba beans have high protein contents they are a good source of minerals, vitamins, and numerous bioactive compounds. They have an important role in maintaining the sustainability of the agricultural system, as it is a very efficient crop in the symbiotic fixation of atmospheric nitrogen (Anestis *et al*., 2018).

Durum wheat is no longer just a staple crop for food security but it has become a major cash crop. The industry of pasta and couscous currently purchase durum grain at prices 10 to 20% higher than bread wheat (Sall A.T. *et al*., 2019).

Callosobruchus maculatus (Bean beetle) is one of the most important and devastating pests belonging to order Coleoptera and family Chrysomelidae. It is a major pest of economically

important leguminous seeds. This weevil is reported to cause up to 100% loss of stored cowpeas, causing severe qualitative and quantitative losses (Lale 1991).

Lesser grain borer *Rhyzopertha dominica* (F.) (Coleoptera: Bostrychidae) is a destructive pest of stored grain, (Potter, 1935; Edde, 2012). *R. dominica* generally infests stored wheat during the summer. They bore irregularly shaped holes and the larvae may develop inside the grain (Potter 1935). Larva and adults feeding in and on grain kernels may leave only dust and thin brown shells (A. S. A. Saad *et al*., 2018).

To avoid the disadvantages of chemical protectants, the use of natural products and some living organisms has been confirmed.Deltamethrin is a member of the chemical class of pyrethroids which are synthetic chemicals modeled after the pyrethrin components of pyrethrum and is one of the highly applicable insecticides (Marijana Pražić Golić *et al*., 2018). *Beauveria bassiana* is an entomo-pathogenic fungi which evaluated as good alternatives to chemical insecticides (Muhammad Akmal *et al.*, 2017). Fertilizers improve crop yield also influence crop suitability for insect development (Van Emden, 1966; Wooldbridge and Harrison, 1968; Kogan, 1994, Asiwe, J. A. N *et al*., 2009)

MATERIALS AND METHODS

Plant Materials:

Seeds of Vicia *faba* cultivar Noubaria 1 and grains *Triticum durum* of (BaniSweif 5) were used as tested plants.

Tested Insect:

A laboratory colony of *Callosobrucus maculatus* and *Rhizopertha dominica* were kept under laboratory conditions of 28± 1°c and 65 ±5% RH in Stored Grains Pest Research Department, Plant Protection Research Institute, Agriculture Research Center, Dokki, Giza, Egypt. Adults (1- 2 days old for *C. maculatus* / 1- 2 weeks old for *R. dominica*).

Chemical insecticide deltamethrin, biological insecticide beauveria bassiana and phosphate salt fertilizer (Table 1).

Table 1: Tested materials.

No	Trade name	Company	Active ingredient	Physical properties
1	Delta- tox 5% Ec	Arabian Chemical industries (ACI)	deltamethrin 5%	liquid
2	Biosect	Kafr El zayat pesticides and chemicals co.	beauveria bassiana 32 $x10^{12}$/kg	powder
3	Superelfalah	Egypt for fertilizers and chemicals (Afco)	phosphate salt 30% P2O5	granules

Toxicological Studies:

A sample of 10 g of disinfected faba bean or wheat seeds was placed in a glass tube (3 x7.5 cm) and separately mixed with each concentration of tested materials as shown in Table (2). The tubes were shaken vigorously to ensure a uniform coating of faba bean and wheat seeds with the tested materials. Three replicates for every treatment were infested by 25 adults (1- 2 days old for *C. maculatus* / 1- 2 weeks old for *R. dominica*) of the tested insect. The tubes were covered secured with elastic bands and kept in the incubator under constant conditions of 28± 1°c and 65 ±5% RH. Mortality counts of *C. maculates* and *Rhyzopertha dominica* adults were recorded in all experimental treatments after 1 day for C. maculates and 3 days for *Rhyzopertha dominica* from exposure. The percentage of mortality was taken and was calculated according to Abbott´s (1925). The slope values of established lines, LC25, LC50, LC75, LC90, LC95 and LC99 were estimated after 1 day for C. maculates and 3 days from insect exposure (Bliss, 1935).

Residual Activity of the Tested Materials on *C. maculatu*s and *R. dominica* at Storage Periods:

Each tested concentration of LC95 of detamethrin, *beauveria bassiana* and phosphate salt fertilizer was mixed to500 gm of *V. faba* seeds/ *T. durum* grains separately. They were kept in a glass jar, covered tightly under laboratory conditions of 28 ±1ºC. Treated seeds were stored for a duration of 3 months. Mortality of *C. maculatus* and *Rhyzopertha dominica* adults were carried out every two weeks till 3 months (Finney, 1971) by adding Twenty-five adults of *C. maculatus*/ *R. dominica* to 10 gm of *V.faba* seeds/ *T. durum* grains of each of the three replicates in the presence of untreated control. The mortality values were corrected by using Abbott´s (1925) formula.

Seeds/ Grains Germination:

100 g of faba bean or wheat seeds were taken and treated with LC95 of deltamethrin, *beauveria bassiana* and phosphate salt fertilizer. Twenty-five *V. faba* seeds/ wheat grains were placed in Petri dishes; lined with two layers of cotton and filter paper then soaked with water. Each treatment and control were replicated four times using untreated control. After three days, germinated grains were recorded (Anonymus, 1966). The percentage of germination was calculated and the percentage of reduction in germination was determined. The above steps were repeated at the end of the storage period to determine the percentage of germination**.**

Water Absorbance:

V. faba seeds/ *T. durum* grains (5gm) were treated with LC_{95} for each protectant and then placed in tubes measuring 3 × 7.5 cm immersed in water with an untreated sample which was considered as a control sample. Each sample was replicated 3 times. The increase in seeds weight was recorded after different times of treatment (1, 4, and 24 hours) according to (Schoonhoven, 1978). The aforementioned steps were repeated after 90 days to estimate the percentages of water absorption after the end of the storage period**.**

RESULTS AND DISCUSSION

The mortality percentage of *C. maculatus* after one day of exposure to *Vicia faba* treated with different concentrations of deltamethrin, *B. bassiana*, and phosphate salt increased with increasing concentration. the highest value 85.33% observed for seeds treated with 0.05 ml/kg of deltamethrin, 85.00% after treatment with $6.4X10^{10}$ cell/kg of *Buvaria bassiana* and 84.00 % with 1.8 gm/kg of phosphate salt (Table 2) and Figure (1).

Table 2: Mortality percentage of *C. maculatus* adult after treatments *Vicia faba* with different concentrations of deltamethrin, *Buvaria basssiana* and phosphate fertilizer.

Tested materials	Rate of treatment	Mortality % after 1 day
1- Chemical insecticide Deltamethrin (ml/kg)	$5X10^{-5}$ $5X10^{-4}$ $5X10^{-3}$ 0.05	16.00 52.00 64.00 85.33
2- Bio-insecticide *Buvaria bassiana* (cell/kg)	$9.6X10^{9}$ $1.6X10^{10}$ $3.2X10^{10}$ $6.4X10^{10}$ $9.6X10^{10}$	25.33 30.67 64.00 85.00 100.0
3- Fertilizer superelfalah (Phosphate salt gm/kg)	0.15 0.6 1.5 1.8	33.33 57.33 78.67 84.00

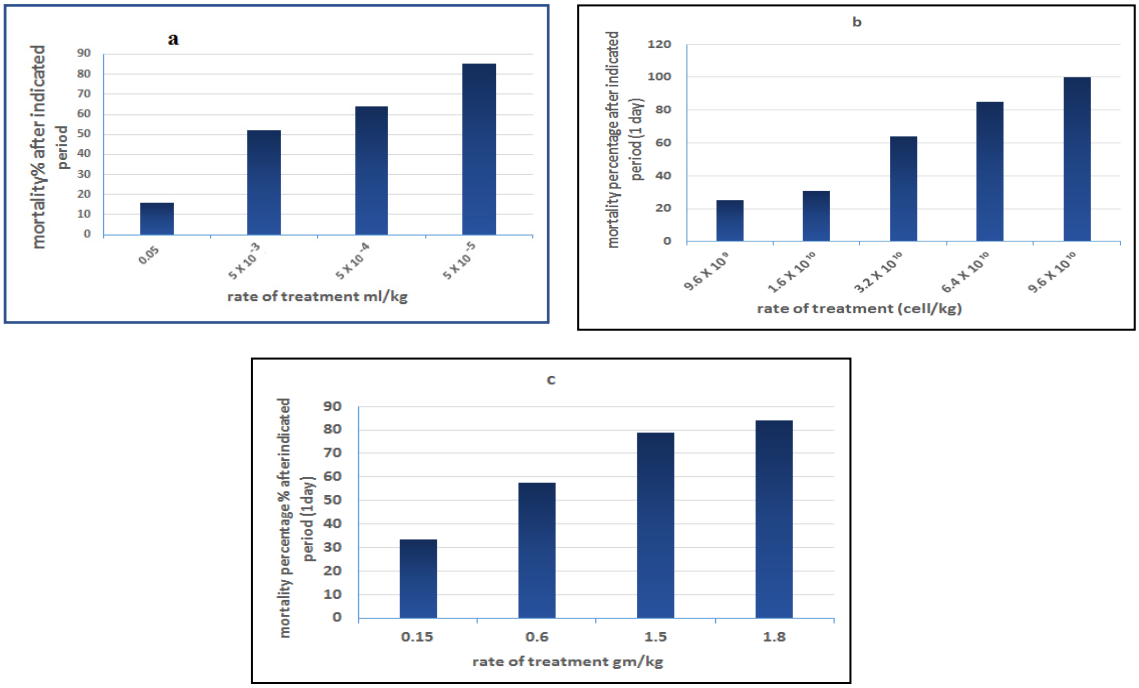

Fig.1: Mortality percentage of *C. maculatus* adult after different treatments with deltamethrin (a), *Buvaria bassiana* (b) and phosphate salt (C)**.**

For *R. dominica* the mortality percentage after exposure to *T. durum* seeds treated with tested materials for 3 days increased with increasing concentration. mortality recorded 81.33% when wheat grains were treated with $5x10^{-4}$ ml/kg of deltamethrin; 97.33% when treated with $1.28X10^{10}$ cell/kg of *Buvaria bassiana* and 84.00% when treated with 1.2 gm/kg of phosphate salt (Table 3) and Figure (2). This agrees with (Z. Mahdneshin *et al*., 2009, Yacoub Ahmad Batta., 2008).

Table 3: Mortality percentage of *R. dominica* adult after treatments *T. durum* with different concentrations of the deltamethrin, *Buvaria basssiana* and phosphate salt.

Tested materials	Rate of treatment	Mortality % after 3 days
1- Chemical insecticide Deltamethrin (ml/kg)	$5 X 10^{-7}$ $5 X 10^{-6}$ $5 X 10^{-5}$ $5 X 10^{-4}$	38.00 68.00 74.67 81.33
2- Bio-insecticide *Buvaria bassiana* (cell/kg)	$1.6X10^{10}$ $3.2X10^{10}$ $6.4X 10^{10}$ $9.6X10^{10}$ $1.28 X10^{10}$	36.00 57.33 76.00 86.67 97.33
2- Fertilizer, superelfalah (phosphate salt gm/kg)	0.15 0.3 0.6 0.9 1.2	21.33 28.00 42.67 72.00 84.00

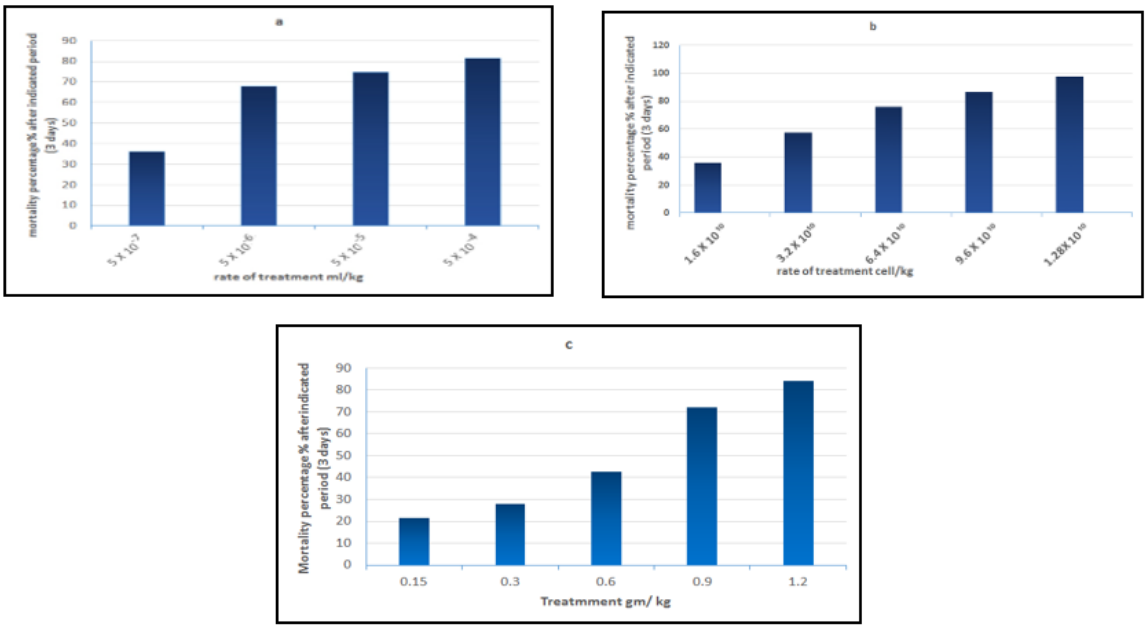

Fig. 2: Mortality percentage of *R. dominica* adult after different treatments with deltamethrin (a), *Buvaria bassiana* (b) and phosphate salt.

Data presented in (Table 4) demonstrated the LC_{25}, LC_{50}, LC_{75}, LC_{90}, LC_{95} and LC_{99} values after one day of exposure for *C. maculatus* and 3 days for *R. dominica* with the three test materials using Ldp line.

Table 4: Toxicological evaluation of deltamethrin, *Buvaria bassiana*, and phosphate salt (superelfalah) after one day of exposure against tested insect *C. maculatus*/ 3 days against *R. dominica*; (Concentrations = average of three replicate).

Conc	Deltamethrin (ml/kg)		*Buvaria bassiana* (cell/kg)		Superelfalah (gm/kg)	
	C. maculatus	*R.dominica.*	*C.maculatus*	*R. dominica.*	*C. maculatus*	*R. dominica.*
LC25	0.0001	—	11.42	1.22	0.106	0.228
LC50	0.001	0.0000012	21.74	2.54	0.355	o.505
LC75	0.0114	0.000068	41.39	5.32	1.192	1.116
LC90	0.1034	0.002577	73.88	10.32	3.545	2.279
LC95	0.3878	0.02269	104.49	15.35	6.804	3.493
LC99	4.624	1.34198	200.23	32.32	23.111	7.778

Results showed that there was a negative correlation between the mortality values and the time after application For *C. maculatus* the results showed 97.33% and 98.67% mortality at initial treatment then 100% up to three months treated with LC_{95} deltamethrin or LC_{95}*Buvaria bassiana* respectively. This agrees with (Ismail Oguz Ozdemir *et al*., 2020,) Data showed a sharp decline in the efficiency of the phosphate salt. It recorded 96% at initial treatment then, recorded 20% at the 4th week and no effect was recorded on after 8th week. This disagrees with (Antoin Sanon *et al*., 2010) who reported that deltamethrin failed to control *C.* maculatus after 3 months of storage (Table 5) and Figure (3)

Table 5: Mortality percentage of *C. maculatus* adult exposed to *V. faba* seeds treated with LC_{95} of tested materials at storage periods

Treatment	Mortality % at storage periods (weeks)						
	Initial	2nd week	4th week	6th week	8th week	10th week	12th week
LC_{95} Deltamethrin	97.33	100	100	100	100	100	100
LC_{95} *Buvaria bassiana*	98.67	100	100	100	100	100	100
LC95 Superelfalah	96	34.68	20	13.32	---	---	---

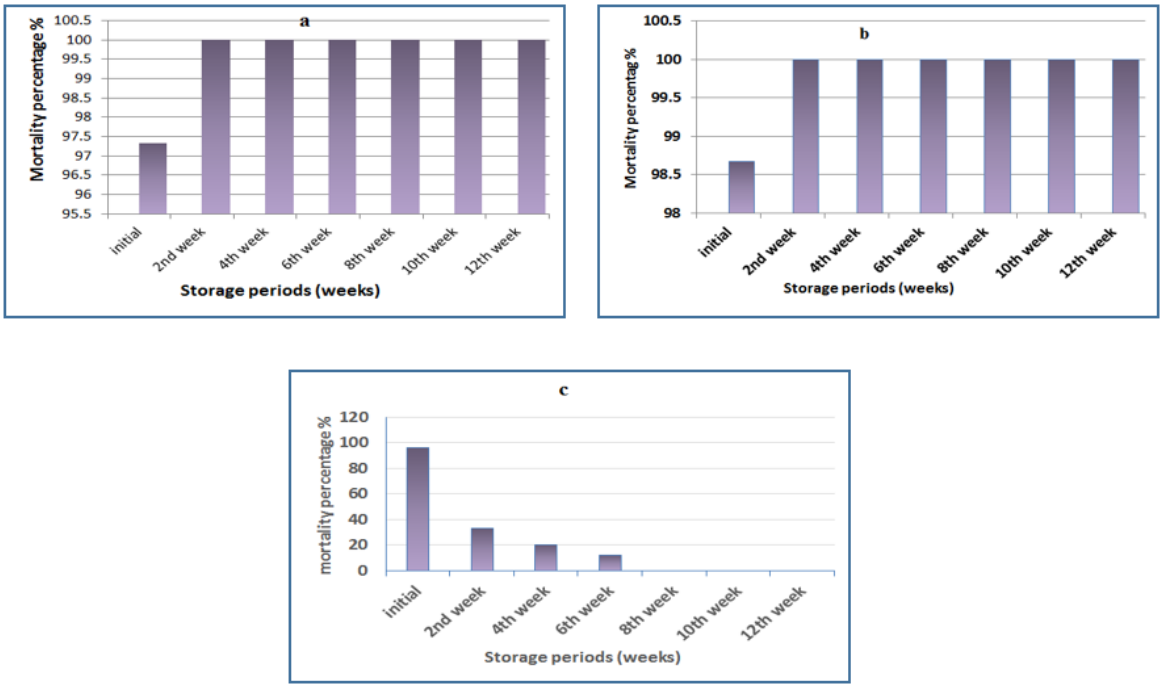

Fig.3: Mortality percentage of *C. maculatus* adult exposed to *Vicia faba* treated with LC_{95} of deltamethrin (a), *Buvaria bassiana* (b) and Superelfalah (c) at storage periods

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For *R. dominica* at initial treatment 98.67% mortality were recorded with adults affected by LC_{95} of *Buvaria bassiana*, then 100% at the second week up to the end of the storage period (3 months) indicating the high residual effect of this bio-insecticide. Mortality percentage recorded 89% by the end of the storage period when *R. dominica* was exposed to *T. durum* treated with LC_{95} deltamethrin which agrees with (Nasr, M. E. H. and S.M. Mahgoub., 2017). A sharp decline in the efficiency of phosphate salt occurred after six weeks which recorded 46.67% at the 6th week (Table 6 and Fig.4).

Table 6: Mortality percentage of *R. dominica* adult exposed to *T. durum* grains treated with LC_{95} of tested materials at storage periods.

Treatment	Mortality % storage periods (weeks)						
	Initial	2nd week	4th week	6th week	8th week	10th week	12th week
LC_{95} Deltamethrin	97.33	90.86	90	90	90	90	89.33
LC_{95} *Buvaria bassiana*	98.67	100	100	100	100	100	100
LC95 Superelfalah	98.67	84	54.76	46.67	-	-	-

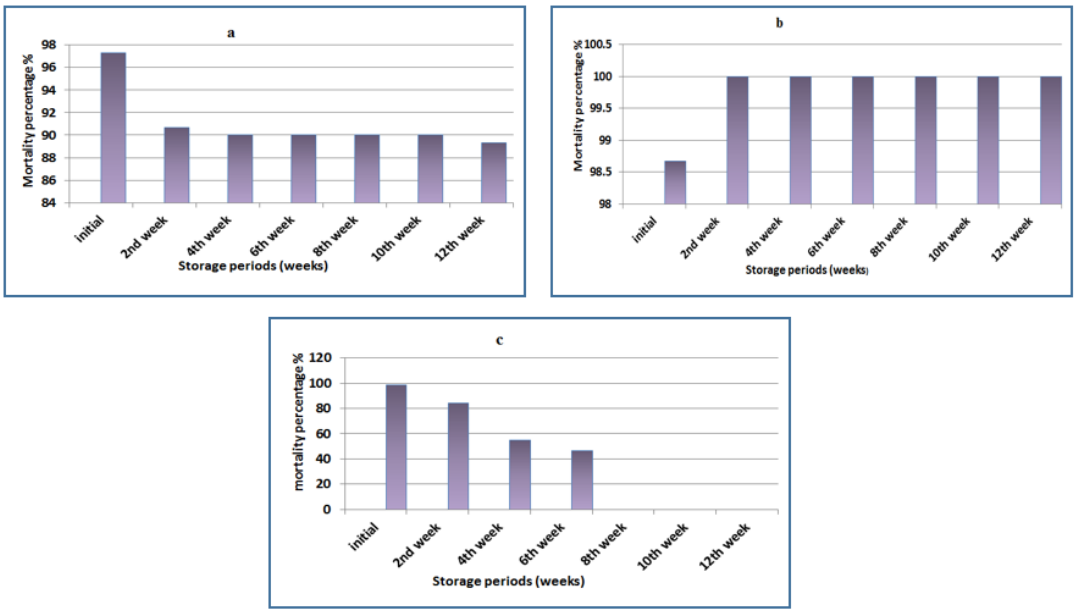

Fig.4: Mortality percentage of *R. dominica* adult exposed to *T. durum* grains treated with LC_{95} of deltamethrin (a), *Buvaria bassiana* (b), and superelfalah (C) storage periods.

Germination Test:

Data showed that germination of *V. faba* seeds treated with deltamethrin at the LC_{95} concentration remained almost equal to the control (100%) at the initial and after storage period (3 months) while *Buvaria bassiana* and phosphate salt indicated a 1.33% reduction in germination at the initial time only (Table 7 and Fig. 5). This agrees with (Nasr, M. E. H. and S.M. Mahgoub., 2017 El-Khayat., 2000) disagree with (Yacoub Ahmad Batta., 2008, J. M. Adesina *et al*., 2012).

Table 7: Germination percentage of *V. faba* seeds treated with deltamethrin, *Buvaria bassiana* and superelfalah at initial and after storage periods.

Treatment ml/Kg		**Control**	**LC_{95} deltamethrin**	**LC_{95} *Buvaria bassiana***	**LC_{95}** superelfalah
Germination %	**Initial**	100	100	98.67	98.67
	After storage	100	100	100	100

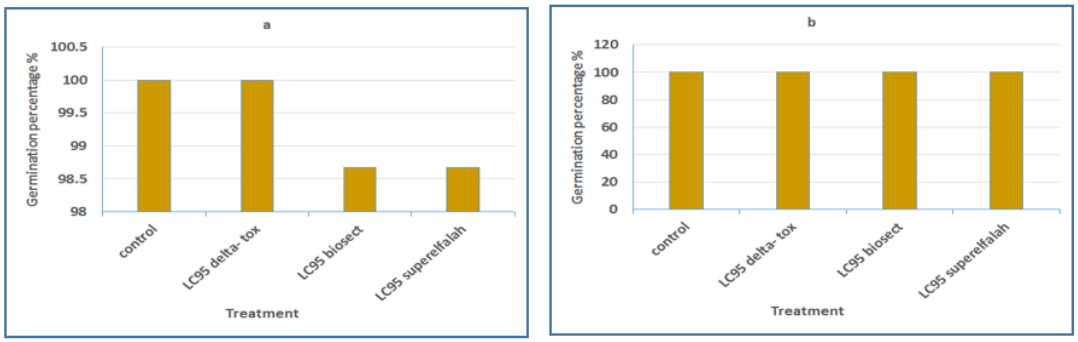

Fig.5: Germination percentage of *V. faba* seeds treated with deltamethrin, *Buvaria bassiana* and superelfalah at initial (a) treatment and after storage periods (b).

Data in Table (8) showed that germination of wheat grain treated with LC_{95} of deltamethrin remained almost equal to the control (100%) at the initial and after the storage period. *Buvaria bassiana* and phosphate salt indicated a reduction in germination at the initial time only (Fig. 6).

Table (8): Germination percentage of *T. durum* grains treated deltamethrin, *Buvaria bassiana* and superelfalah at initial treatment and after storage periods.

Treatment ml/Kg		Control	LC_{95} Deltamethrin	LC_{95} *Buvaria bassiana*	LC_{95} superelfalah
Germination %	Initial	100	100	98.67	97.33
	After storage	100	100	100	100

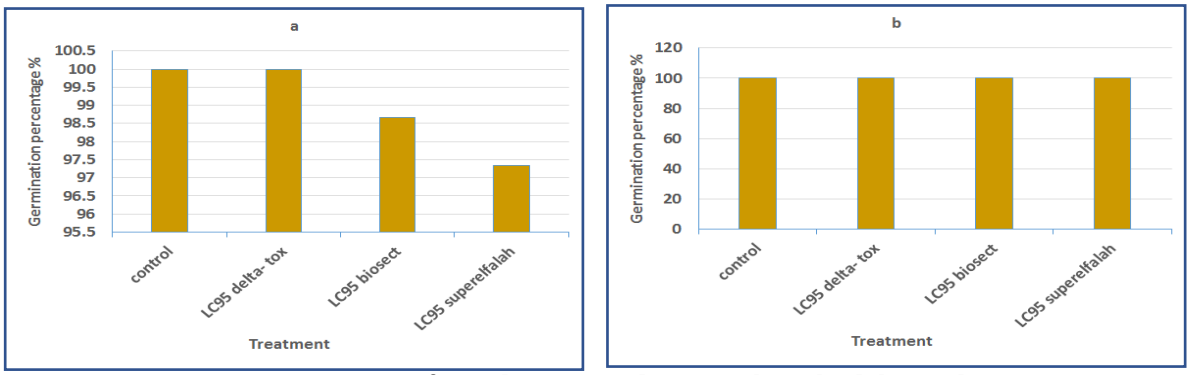

Fig.6: Germination percentage of *T. durum* seeds treated with deltamethrin, *Buvaria bassiana* and superelfalah at initial (a) treatment and after storage periods (b).

Water Absorption:

Water absorption of *V. faba* seeds and *T. durum* grains after the indicated times of dipping (1, 4, 24 hours). water absorption increased with the extension of the submerging period (Tables 9&10) and Figures (7& 8). There is no significant difference in the percentage of water absorption was recorded between the treated and untreated *V. faba* seeds/ *T. durum* grains.

Table 9: Water absorption percentage of *V. faba* treated with deltamethrin, *Buvaria bassiana* and phosphate salt at initial and after storage periods.

Tested material		Water absorption % for *V.faba*		
		1 hour	4 hour	24 hour
Initial	Control	13.55	37.50	107.17
	Deltamethrin	22.23	45.70	104.80
	Buvaria bassiana	18.30	44.08	115.04
	Superelfalah	22.00	33.07	102.79
Storage	Control	1.38	29.30	105.01
	Deltamethrin	7.39	46.85	106.89
	Buvaria bassiana	13.78	43.06	111.79
	Superelflah	4.59	32.90	102.80

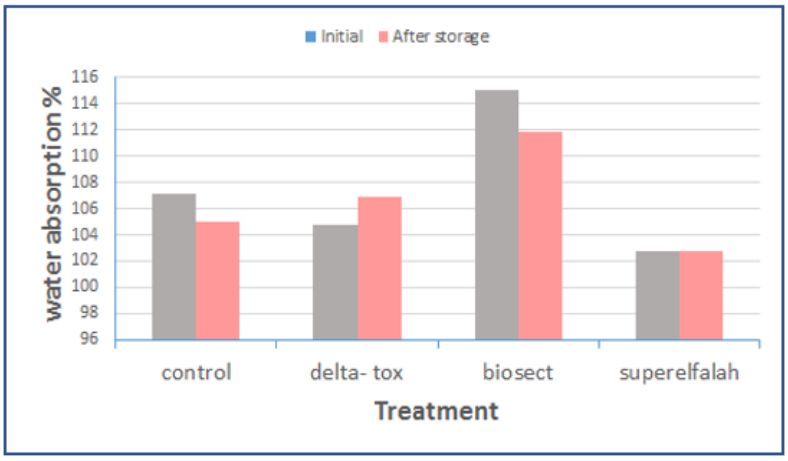

Fig.7. Water absorption percentage after 24 hours of *V. faba* seeds treated with deltamethrin, *Buvaria bassiana* and phosphate salt at initial and after storage periods.

Table 10: Water absorption percentage of *T. durum* treated with deltamethrin, *Buvaria bassiana* and superelflah at initial and after storage periods**.**

Tested material		Water absorption %for T. durum		
		1 hour	4 hour	24 hour
Initial	Control	13.96	23.21	47.02
	Deltamethrin	13.20	24.74	47.35
	Buvaria bassiana	14.10	24.13	49.33
	Phosphate salt	13.05	24.35	46.15
Storage	Control	11.29	21.97	43.36
	Deltamethrin	9.99	18.26	43.81
	Buvaria bassiana	14.39	18.28	43.96
	Superelfalah	10.09	21.48	40.36

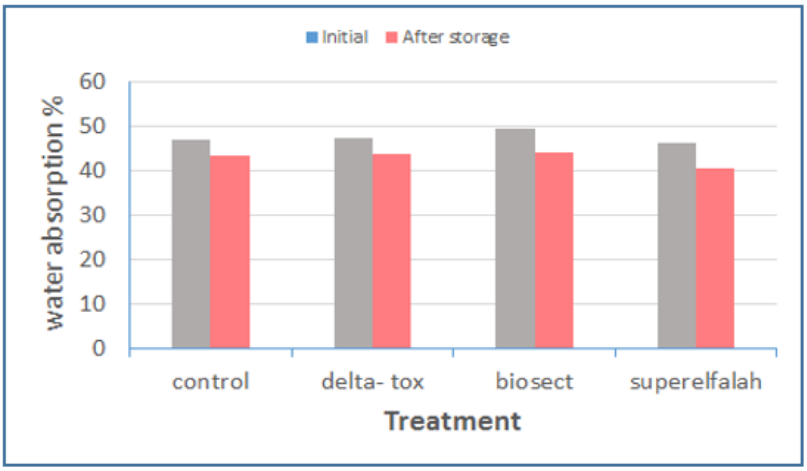

Fig.8. Water absorption percentage after 24 hours of *T. durum* grains treated with deltamethrin, *Buvaria bassiana* and phosphate salt at initial and after storage periods.

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ARABIC SUMMARY

تأثير المبيد الحيويوالمبيد الكيميائى والسماد الفوسفاتي على خنفساء اللوبيا وثاقبة الحبوب الصغرى

رشا عصام الدين صابر[1] مواهب محمود زيور[1] محمد عبد الستار[2]

1- قسم افات الحبوب والمواد المخزونة- معهد بحوث وقاية النباتات- مركز البحوث الزراعية
2- معهد بحوث الهندسة الوراثية الزراعية- مركز البحوث الزراعية

أجريت هذه الدراسة لتقييم تأثير كلا من المبيد الكيميائي (دلتاميثرين) والمبيد الحيوي (بيوفاريا باسيانا) و السماد الفوسفاتي على حشرتي خنفساء اللوبيا وثاقبة الحبوب الصغرى. واشتمل البحث دراسة التأثير السمي وقدرة كل مادة على الحفاظ على بذور الفول وحبوب القمح من الإصابة الحشرية طوال فترة التخزين (3 أشهر) بتركيزLC95 كما تمت دراسة تأثير هذه المبيدات على إنبات البذور/ الحبوب وامتصاصها للماء. أظهرت النتائج أن المبيد الحيوي كان الأكثر تأثيرا حيث أنه حتفظ بقدرته على قتل الطور البالغ لكلتا الحشرتين حتى إنتهاء فترة التخزين. بينما اختلف تأثير لمبيد الكيميائي حيث تسبب في قتل جميع حشرات الطور البالغ لخنفساء اللوبيا حتى إنتهاء فترة التخزين فيما قل تأثيره على الطور البالغ لثاقبة الحبوب الصغرى ليصل إلى 89,33% بنهاية فترة التخزين. كان تأثير السماد الفوسفاتي هو الأقل لكلتا الحشرتين. لم تأثر جميع المعاملات على معدل إنبات البذور أو إمتصاصها للماء.