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Comparative study on the Efficacy of *Agrotis segetum* Granulosis virus (*Agse*GV^{EG}) against *Agrotis ipsilon* under semi-field conditions

Magda Khattab
Plant Protection Research Institute, ARC, Dokki, Giza, Egypt
Department of Cutworms and Mole Cricket

ABSTRACT

The present work was carried out to evaluate the efficacy of *Agrotis segetum* granulosis virus (*Agse*GV^{EG}) against *Agrotis ipsilon* larvae in semi-field experiments using spray and wheat bran bait method. A Fluorescent brightener (Tinopal UNPA-GX) was added for enhancement of *Agrotis segetum* granulosis virus (*Agse*GV^{EG}). Results showed that, addition of F. brightener 1% to *Agse*GV^{EG} suspension using surface contamination technique of semi synthetic diet increased the susceptibility of 3rd and 4th instars of *A. ipsilon* larvae; the LC_{50} was 2.08×10^{8} OB / ml and 6.10×10^{8} OB/ml, respectively. The LT_{50} values of *Agse*GV^{EG} (9.07×10^{10} OB/ml) +F. brightener 1% was7.67 and 7.85 days for 3rd and 4th instars. In addition, semi-field conditions, F. brightener 1% as sun screen enhanced *Agse*GV^{EG} persistence on Cotton foliage after spray, the calculated lethal inactivation median time for 50 % reduction in virulence (lethal median inactivation time, LIT_{50}) for 2nd instar larvae was0.97 or 2.27 days for virus alone-treatment and *Agse*GV+ F. brightener additive, respectively. The calculated potency value for F. brightener was 2.335 Fold.

Mixture of *Agse*GV^{EG}+ F. brightener 1% in bran bait formulation increased mortality% of *A. ipsilon* in two concentrations of *Agse*GV^{EG} (9.07×10^{9} and 9.07×10^{8} OB/ ml) + F. brightener 1% and decreased the number of cut cotton plants infested with 3rd instar larvae of *A. ipsilon* compared to untreated control.

The damage reduction to Cotton seedlings with *A. ipsilon*, was 83.33% and 70% for 3rd and 4th instars larvae, respectively, in the *Agse*GV^{EG} (9.07×10^{9} OB/ml) + F. brightener 1% compared to control.

In conclusion in both spray and bran tested treatments larvae infected with *Agse*GV^{EG}+ F. brightener 1% died more rapidly than those infected with *Agse*GV^{EG} alone ; indicating that *Agse*GV^{EG} appears to have better potential as a control agent for *A. ipsilon* by adding enhancement additive.

Keywords: Cutworms, *Agrotis segetum*, Granulosis virus (*Agse*GV^{EG}), Occlusion Bodies (OB), brightener additive, *Agrotis ipsilon*, Spray, Bran bait formulation.

INTRODUCTION

The term cutworms is used to describe the caterpillars of a range of noctuid moths that spend a large portion of their time in the soil and have a tendency to feed wastefully on a wide range of crops by severing plants near the soil surface. They have been recorded as pests on the seedlings of many vegetable crops. In Egypt, most of the recorded damages are produced by the larvae of *Agrotis ipsilon* and the closely-related *A. segetum*. Cutworms' attacks about 50 plant species, e.g., maize, faba bean, wheat, cotton, berseem, soybean, tomatoes, potato, cantaloupe, cucumber and many other vegetable plant species and can be very wasteful feeders, destroying far more plant seedlings than they consume, (Bourner *et al.*, 1992).There are numerous

problems associated with treating cutworms; in particular they are hard to locate until the larvae are large and the damage has been done, since at this stage the larvae spend most of their time below ground where they feed on untreated parts of the crop (Bowden *et al*., 1983).

Control of cutworms depends exclusively on various kinds of insecticides that have been used with varying success for many decades at least. Baits based on wheat bran, finely spread, have met with some success.

Baculoviruses have high potential to be used as microbial control agent against lepidopteron pests due to their high pathogenicity, environmental safety, and low stability, i.e., restricted host range. Two baculovirus species have been isolated from *A. segetum* larvae, the granulovirus *Agse*GV and the nucleopolyedrovirus *Agse*MNPV (Allaway and Payne, 1983; Lipa and Ziemnicka, 1971; Lipa *et al*., 1971 and Khattab, 2012. Both types of viruses showed bio control potential in the field (Caballero *et al*., 1991; Bourner *et al.*, 1992).

A granulovirus of *A. segetum* (*Agse*GV) is effective in spray formulation against both *A. segetum* and *A. ipsilon* in field trial (e.g. Shah *et al.,* 1979; Zethner *et al*., 1987), and has also been used in a wheat bran bait trial in Spain (Caballero *et al*.,1991; Khattab, 2007).

One way to increase the activity of the pathogen is to incorporate it with small quantities of synergistic substances such as F.brighteners. The enhancers decrease, slightly, the time to mortality and decrease the amount of virus necessary to produce mortality (Shapiro, 1992&Shaprio and Dougherty, 1994).

Several additives have been used to increase efficacy of several baculoviruses by decreasing the amount of virus needed for a 50 % effective lethal concentration (LC_{50}) and lethal median time (LT_{50}) e.g., Neem extract (El-Salamouny *et al*., 1997; Khattab *et al*., 2004), Boric acid has been demonstrated to potentiate the activity of several baculoviruses (Shapiro and Bell, 1982; Morris *et al.,* 1995; Khattab, 2007). Fluorescent brighteners (= Optical brighteners) are commonly used in soaps, detergents, and fabric softeners. These compounds absorb ultraviolet energy and convert it to visible light (acts as UV protectants) (Shapiro, 1992), but they can also act as viral enhancers by decreasing the LC_{50} and the LT_{50}. The optimum concentration of brightener for enhancing viral activity was 1 %. Adding brightener reduced the time required for the virus to kill larvae by 30-72 % (Li and Otvos, 1999). F. brighteners enhanced several baculoviruses e.g., *Spodoptera frugiperda* MNPV (*Sf*MNPV) (Hamm and Shapiro, 1992), *Lymantria dispar Ld*NPV (*Lydi*NPV) and *Autographa californica* (*Auca*MNPV) (Dougherty *et al*., 1996), *Anagrapha falcifera* NPV (*Anfa*NPV) (Vail *et al*., 1996) and *A. segetum* NPV (*Agse*NPV) and *Agse*GV (Khattab *et al*., 2004).

The present work describes a semi - field trial to compare the efficacy of granulovirus *A. segetum* (*Agse*GV^{EG}) Egyptian isolate in both spray and wheat bran bait formulations with and without enhancement additive Florescent brightener to control 3rd and 4th instar *A. ipsilon* larvae on cotton seedlings.

MATERIALS AND METHODS

Granulosis virus:

Agrotis segetum granulovirus virus (*Agse*GV^{EG}) was obtained from Cutworms and Mole Cricket Department at Plant Protection Research Institute, ARC, Dokki, Giza, Egypt. It was prepared as water suspension in Tris buffer at pH 8 and virus concentration was $9.07x10^{10}$, 9.07×10^{9} and 9.07×10^{8} OB/ ml.

Tinopal:

Fluorescent brightener (Tinopal UNPA-GX) was obtained from (Sigma®). It was obtained as a powder to be used as a sun screen for increasing *Agse*GV^{EG} stability and persistence under field conditions. It was prepared as water suspension and tested at the concentration 0.1 %.

Tests insect:

Laboratory strain of *A. ipsilon* was provided by Cutworms Department at Plant Protection Research Institute, ARC. *A. ipsilon* larvae were mass reared on synthetic diet described by Shorey and Hale (1965), but formaldehyde was removed from diet component when it is used in bio-assay tests.

Laboratory Bioassay tests:

Bioassay tests were performed using surface contamination of synthetic diet with *Agse*GV^{EG} concentrations (Khattab, 2007). A bioassay plates (L1CEFA, Bad-Salzuflen Germany) were used. Triton X -100 at concentration 2.5% was mixed with virus suspension to reduce *Agse*GV^{EG} OB clumping. *Agse*GV^{EG} was tested at concentrations 9.07×10^{10}, 9.07×10^{9} and 9.07×10^{8} OB/ ml against 3 rd and 4 th instar larvae of *A. ipsilon*. For each concentration, five replicates were used (20 larvae each) after mixing virus suspension with brightener1%. In addition, five more replicates were used as control by adding distilled water only. Mortality % was recorded after 2 and 4 days post-feeding on virus contaminated diet. Abbot formula (1925) was used to calculate corrected mortality %. The LC_{50} and LT_{50} values were calculated.

Semi - field Treatments:

Seeds of the cotton variety (Giza 86) were cultivated in pots (12.5 cm in diameter × 12.5 cm in length) on May 2013. The efficacy of the Egyptian isolate granulovirus of *A. segetum* (*Agse*GV^{EG}) with or without F. brightener 1% additive was evaluated against of *A. epsilon* 2nd, 3rd and 4th instar larvae on cotton seedlings. The virus was applied by two application methods, spray and bait formulations.

The spray treatments:

Spray of (*Agse*GV^{EG}) was applied to the cotton seedlings using a hand sprayer at concentration of (**9.07×10^{9}** OB/ ml) in addition to 2.5 % (Triton X -100) as a wetting agent. The nozzle was directed towards the plants and spraying took place during sunset. After drying, sprayed leaf from each treatment was collected (as a zero time) then a daily collection was made through 1, 2 and 4 days. 2nd instar larvae were fed on the collected treated leaves for 24hrs. Mortality% of larvae or pupae due to viral infection was done at two days intervals (Elnagar *et al*. 2003).

Wheat bran bait treatments:

Infestation levels of 3rd and /or 4th instars larvae of *A. ipsilon* was calculated in 20 plants per concentration/ treatment on the soil into the pots near the stem of the cultivated cotton plants when the seedlings had 3-4 leaves.

Wheat bran bait was used as bait at a rate of 50 gm.+1ml *Agse*GV^{EG}/ concentration (two concentrations of *Agse*GV^{EG} 9.07×10^{9} and 9.07×10^{8} OB / ml were tested) alone or mixed with F. brightener 1% additive. The wheat bran bait formulations were applied manually in the infested pots on soil surface around the cotton seedling after 2hrs. Control pots were treated with wheat bran bait mixed water only. To compare level of virus infection, samples of *A. ipsilon* were examined from the treated pots at times, 24 hr., 48 hr. and 72 hr. and 7 days after treatment (Khattab, 2007).

Larval diagnosis:

After 7 days of treatment the soil surface pots, soil samples were inspected to determine the number of dead and alive larvae by shaking them onto a cloth and then

alive larvae were transferred individually to semi-synthetic diet described by Shorey and Hale (1965) until death or pupation. Diagnosis of viral deaths was performed visually since the symptoms of viral infection are easily observed.

Damage evaluation:

(*Agse*GV^{EG}) treated plants with or without F. brightener 1% additive applied to the cotton seedlings as well as control plants were examined for *A. ipsilon* damage in the feature of plants or leaves cut after the release of larvae.

Statistical analysis:

Bioassay data were subjected to probit analysis using the method described by Ehab Mostafa Bakr (1998). The relative potencies of the treatments were calculated according to the changes in % mortality. Percent reduction of population was calculated according to the following equation (Mohamed *et al.,* 2000).

$$\%\,\mathrm{Re}\,duction = \frac{C-T}{C}\ X\ 100$$

Where C= the estimated parameter in check
T= the same parameter in treatment

RESULTS AND DISCUSSION

Laboratory bioassay:

Preliminary bioassay was conducted to estimate the relative virulence based on LC_{50} values of *A. segetum Agse*GV^{EG} alone or mixed with F.brightener1%.

Data in Table (1) show that mortality percentage of *A. ipsilon* 3rd instar larvae was (14.00, 4.08 and 0.00 %) after treatment with different concentrations of *Agse*GV^{EG}, while it was 0% at the same concentrations for the 4th instar larvae, respectively.

Table 1: Mortality response of *Agrotis ipsilon* 3 rd and 4th instar larvae fed on diet contaminated with the virulence (*Agse*GV^{EG}) alone and with the additive F.brightener1%:

Virus Treatments	Virus Conc. *OB /ml	Mortality (%) due to virus ± F.brightener1%	
		3 rd instar	4th instar
Virus alone	9.07×10^{10}	14.00	0.00
	9.07×10^{9}	4.08	0.00
	9.07×10^{8}	0.00	0.00
Virus + F.brightener1%	9.07×10^{10}+F.1%	93.75	90.00
	9.07×10^{9}+F.1%	85.41	82.00
	9.07×10^{8}+F.1%	64.00	52.00
LC_{50}(OB,s/ml) Virus+F.brightener1%		$2.08x10^{8}$	$6.10x10^{8}$

* OB = Occlusion Bodies

Mixing *Agse*GV^{EG} with F. brightener at concentration 1% increased mortality percentage of 3rd and 4th instar larvae of *A. ipsilon*. Mortality percentage was 93.75, 85.41 and 64.00 for the 3rd instar larvae of *A. ipsilon* at *Agse*GV^{EG} concentrations (9.0×10^{10}, 9.07×10^{9} and 9.07×10^{8} OB/ ml) mixed with 1% of F. brightener, respectively. While mortality percentage was 90.00, 82.00 and 52.00% for the 4th instar larvae of *A. ipsilon* at the same concentrations of *Agse*GV^{EG} mixed with F. brightener, respectively.

Taking into account the LT_{50} values, it is clear that LT_{50} values for 3rd and 4th instar larvae of *A. ipsilon* decreased with increasing viral tested concentration mixed

with F. brightener 1%.

The LT_{50} values for the 3rd instar larvae were 7.67, 8.48 and10.03 days at 1%F-brightener mixed with virus concentrations at 9.07×10^{10}, 9.07×10^{9} and 9.07×10^{8} OB, s/ml, respectively. The LT_{50} values for the 4th instar larvae were 7.85, 7.94 and 10.33 days at the same concentration of virus, respectively (Table 2).

Table 2: Lethal median (LT_{50}) values of *Agse*GV^{EG} combined with Fluorescent brightener 1% against *Agrotis ipsilon* 3 rd and 4th instar larvae:

Virus Conc. OB /ml + F. brightener1 %	LT_{50} Value (days)	
	3 rd instar	4th instar
9.07×10^{10}	7.67	7.85
9.07×10^{9}	8.48	7.94
9.07×10^{8}	10.03	10.33

Otherwise, no mortality occurred to *A. ipsilon* 3rd and 4th instar larvae treated with F. brightener1% alone.

It is clearly obvious that, the addition of F. brightener-28 additive to the *Agse*GV^{EG} has increased their virulence against *A. ipsilon* larvae by reducing both the LC_{50} and LT_{50} values as reported previously by (Khattab *et al*., 2004).

Semi-field test:

Spray test:

Effect of solar radiation on *Agse*GV^{EG} efficacy against the 2*nd* instar larvae of *A. ipsilon* after feeding on treated Cotton plants are shown in (Table 3). Data in Table (3) show that increasing time exposure of treated cotton plants by virus separately or mixed with F. brightener reduced mortality percentage of *A. ipsilon* 2*nd* instar larvae. Mortality % after larval feeding on treated Cotton with virus was 73.46, 48.00, 34.00 and 16.00 % at exposure times 0, 1, 2 and 4 days, respectively. Mixing *Agse*GV^{EG} with F. brightener at 1% increased mortality percentage while mortality % was 81.63, 78.00, 52.00 and 32.00 % at different time exposure of treated Cotton to solar radiation, respectively (Table 3).

Table 3: Effect of the virulence of *Agrotis segetum* GV (*Agse*GV^{EG}) alone and with F. brightener 1% additive sprayed against *Agrotis ipsilon* 2nd instar larvae on Cotton foliage exposed to natural sunlight during May, 2013:

Exposure time (days)	Mortality(%) due to			
	AgseGV^{EG} alone		*Agse*GV^{EG} +F.brightener 1%	
	Mortality %	*Reduction %	Mortality %	Reduction %
0	73.46	-	81.63	-
1	48.00	33.33	78.00	2.5
2	34.00	52.77	52.00	35.0
4	16.00	77.77	32.00	60.0
LIT_{50} (day)	0.974	-	2.275	-
Potency(Fold)			2.335	

*Reduction= Reduction in virulence

The calculated lethal median inactivation time for 50 % reduction (LIT_{50}) was 0.97 &2.27days for virus alone-treatment and *Agse*GV^{EG}+ F. brightener additive, respectively. The calculated potency value was 2.335 Fold for F. brightener (Table 3).

The present results are in agreement with those previously reported on the effect of F. brightener-28 as an enhancing additive to NPV virulence (Hamm and Shapiro, 1992; Shapiro and Robertson 1992; Shapiro and Dougherty, 1994; Adams *et al*. 1994; Shapiro and Vaughan, 1995; Zou and Young, 1996; Vail *et al*., 1996; and El-Salamouny *et al*.(2001) who reported that susceptibility of *A. ipsilon* neonate larvae

was increased by addition of F. brightener-28 (Tinopal LPW) 0.1% to the tested viruses concentration of baculoviruses, *Agse* MNPV, *Auca* MNPV and *Mamestra brassicae Mabr* MNPV by the rate of 1806, 1040 and 336 fold, respectively. Also, Boughton *et al*., (2001) reported a potential effect of F. brightener on the newly isolated *Agrotis ipsilon* MNPV.

From the obtained results, it is worth to conclude that, F. brightener additive enhanced virus persistence on cotton foliage and showed the best UV protection effect to *Agse*GV^{EG} sprayed against *Agrotis ipsilon* under natural sunlight.

Wheat bran baits tests:

First observation of *A. ipsilon* larvae used in the present experiment demonstrated that the 3rd & 4th instars were often feed directly on the prepared bait .The role of F. brightener in enhancing *Agse*GV^{EG} persistence was also observed in semi-field tests using wheat bran bait. The reduction damage of *A. ipsilon* 3rd instar larvae after 7days of *Agse*GV^{EG}+ F. brightener 1%treatment was 83.33 % and 75.00 % (in two concentrations 9.07×10^9 and 9.07×10^8 OB/ ml), respectively. While % of cut plants by *A. ipsilon* larvae was 60 % after 72 hrs. and reached 100% after 7 days in control treatments (Table 4). On the other hand, the reduction of damage reached 70% in both above-mentioned tested concentrations of *Agse*GV^{EG} + F. brightener 1% in wheat bran bait applied against *A. ipsilon* 4th instar larvae. In control treatment 100% damage was obtained after 72 hrs. of treatment (Table 4). Results indicated that the mixture of *Agse*GV^{EG} + F. brightener 1% in wheat bran bait decreased the number of cut Cotton plants infested with 3rd instar larvae of *A. ipsilon* compared to untreated control (Table 4).

Table 4: Percentage damage cotton seedling treated with virus-wheat bran bait formulation + F. brightener 1% for controlling 3rd and 4th instar of *Agrotis ipsilon* on Semi-field test, during May, 2013:

Treatments	Insect instars	Virus Conc. (OB /ml)	*No. of cut plants at days				%of cut plants	% Reduction
			24hr.	48hr.	72hr.	7days		
Control	3 rd	-	0	0	12	0	60	-
	4th	-	0	0	20	0	100	-
Virus + F. brightener 1%	3 rd	9.07×10^9	0	0	0	2	16.67%	83.33
		9.07×10^8	0	0	0	3	25.00	75.00
	4th	9.07×10^9	0	0	0	6	30.00	70.00
		9.07×10^8	0	0	0	6	30.00	70.00

*No. of infested plants (20 plant).

Data presented in Table (5) show the mortality % among *A. ipsilon* larvae collected from Cotton plants 7 days after wheat bran bait application and fed on semi-synthetic diet. Mortality% in two concentrations of *Agse*GV^{EG} ($9.07x10^9$ and $9.07x10^8$ OB/ ml) + F. brightener 1% was(100 or 90%) and (60 or 50%) for3rd and 4th instar larvae, respectively.

The present results are in agreement with several authors who reported the successful use of *A. segetum* GV (*Agse*GV) for control *A. segetum* and the closely related *A. ipsilon* and reducing the cutworm damages in tobacco plots in Pakistan by72 and 100% when *Agse*GV was used against 2nd instar larvae of *A. ipsilon* (Shah *et al*., 1979), Denmark (Zethner, 1980) Also, *Agse*GV reduced the natural occurrence of cutworms (*A. ipsilon* and *A. segetum*) damage by 64.82% in tobacco, 85% in Okra, 77% in potato and 78% in sugar beet (Zethner *et al*., 1987) and Spain (Caballero *et al*., 1990, 1991) in tobacco, root crops and maize, respectively.

Boruner *et al*. (1992) found that, infection rates in *A. segetum* 2nd instar larvae after treatment with *Agse*NPV and *Agse*GV were 87.5 or 91% for NPV and 12.5 or

55% for GV in spray or bait treatment, respectively. Mixed inocula of *Agse*NPV and *Agse*GV gave intermediate results on maize and beet root (Bourner *et al*., 1994).

Table 5: Percentage viral mortality of *Agrotis ipsilon* larvae collected on7 days after wheat bran bait treatment (on semi- synthetic diet):

Treatments	Insect instars	Virus Conc. (OB /ml)	No. of collected larvae	No. of dead larvae		Mortality %
				Field	Lap.	
Control	3^{rd}	-	20	-	-	-
	4^{th}	-	20	-	-	-
Virus + F.brightener 1%	3^{rd}	9.07×10^{9}	6	14	6	100
		9.07×10^{8}	12	8	10	90.00
	4^{th}	9.07×10^{9}	20	-	12	60.00
		9.07×10^{8}	20	-	10	50.00

*No. of infested larva (20 larva)

Consequently, results obtained by the author, and those presented in the present work show that F. brightener additive 1%, may be used in viral formulations to increase their virulence and speed of kill and protects the virus from the adverse effects of sunlight (UV light) for several days.

Previous reports demonstrated that, baculoviruses were rapidly inactivated after exposure to natural sunlight under natural field conditions. Also, purified virus was more affected than the crude extract (Elnagar and Abul-Nasr, 1980; Tamez-Guerra *et al*., 2000; El-Salamouny *et al*., 2000 and Elnagar *et al*., 2003).

From all the previous results such strategies could be stated by early application in field trials using *Agse*GV^{EG} + F. brightener 1% during cutworm season, to enhance viral persistence and increase speed of kill as well as reduce larval feeding activity of *A. ipsilon*. Death of those larvae will in turn create a reservoir of virus in the soil and thatch that perpetuates control of *A. ipsilon* thought the growing season. Thus the soil acts as a reservoir of virus and provides inoculum to initiate new infections each year as reported by Fuxa and Richter (1994).

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REFERENCES

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

Adams, J.R.; Sheppard, C.A.; Shapiro, M. and Tompkins, G.T. (1994). Light and electron microscopic investigations on the histopathology of the midgut of gypsy moth larvae infected with *Ld*MNPV plus a fluorescent brightener. J. Invertebr. Pathol., 64: 156-159.

Allaway, G.P., Payne, C.C., 1983. A biochemical and biological Comparison of three European isolates of nuclear polyhedrosis viruses from *Agrotis segetum*. Arch. Virol. 75, 43–54.

Boughton, A.J. Lewis, L.C. and Bonning, B.C. (2001): Potential of *Agrotis ipsilon* nucleopolyhedrovirus for suppression of the black cutworm (Lepidoptera: Noctuidae) and effect of an optical brightener on virus efficacy. J. Econ. Entomol. 94: 1045-1052.

Bourner, T.C.;Vargas-Osuna, E.; Willims,T.; Alvrrez, C.S. and Cory, J.S.(1992): A comparison of the efficacy of nuclear polyhedrosis and granulosis viruses in spray and bait formulation for the control of *Agrotis segetum* (Lepidoptera: Noctuidae) in maize. Biocontrol Science and Technology, 2: 315-326.

Bourner, T.C.; Cory, J.S. and Popay, A.J. (1994). Nuclear polyhedrosis and granulosis viruses for the control of the common cutworm, *Agrotis segetum* (Lepidoptera:Noctuidae). Proc. Dorty-Seventh New Zealand Plant Protection Conf., Waitangi Hotel, New Zealand, 9-11 August, pp. 159-162.

Bowden, J.; Cochrane, J.; Emmett, B.J.; Minall, T.E. and Sherlock, P.L. (1983). A survey of cutworm attacks in England and Wales, and a descriptive population model for *Agrotis segetum* (Lepidoptera: Noctuidae). Ann. Appl. Biol., 102:29-47.

Caballero, P.; Vargas-Osuna, E. and Santiago-Alvarez, C. (1990). Field application of the granulosis virus of *Agrotis segetum* Schiff. (Lepidoptera: Noctuidae). Poletin de Sanidad Vegetal Plagas, 16 (1):333-337.

Caballero, P.; Vargas-Osuna, E. and Santiago-Alvarez, C. (1991). Efficacy of a Spanish strain of *Agrotis segetum* granulosis virus (Baculoviridae) against *Agrotis segetum* Schiff. (Lep., Noctuidae) on corn. J. App. Entomol., 112:59-64.

Dougherty, E.M.; Guthrie, K. and Shapiro, M. (1996). Optical brighteners provide baculovirus activity enhancement and UV radiation protection. Biological Control, 7 (1): 71-74.

Ehab Mostafa Bakr (1998) - http://www.ehabsoft.com

Elnagar, S. and Abul-Nasr, S. (1980). Effect of direct sunlight on the virulence of NPV (nuclear polyhedrosis virus) of the cotton leafworm, *Spodoptera littoralis* (Boisd.). Z. Angew. Entomol., 90:75-80.

Elnagar S.,El-Sheikh M.A.K.,El-Salamouny S.,Amin A.,and Khattab M.(2003). Screening of four lignin additives as UV protectants to Baculovirusses. Bull.ent.Soc.Egypte, Econ.Ser., 29,(165).

El-Salamouny, S.; Huber, J.; Elnagar, S. and El- Sheikh, M.A.K. (1997). Increasing the susceptibility to nuclear polyhedrosis viruses by synergistic additives. Proceeding of a Symposium, Unic. of Warwick, Coventry, UK, 16-18 April, pp. 289-292.

El-Salamouny, S.;El-Sheikh, M.A.K.; Elnagar, S. and Huber, J. (2000). Prolongation of the UV-persistence of nucleopolyhedroviruses by the lignin derivative product. 33rd Annual Meeting Society for Invertebr. Pathol., Guanajuats Mexico University of Guanajuato, 13-18 Aug., pp. 39.

El-Salamouny, S.; El-Sheikh, M.A.K.; Elnagar, S. and Huber, J. (2001). Enhancement effect of fluorescent brightener on infectivity of three nucleopolyhydroviruses against the black cutworm, *Agrotis ipsilon* (Hufn.). The First Cong. on Integrated Pest Management, Cairo Univ., 22-23 April, 36 pp.

Fuxa, J.R. and Richter, A.R. (1994). Distance and rate of spread of *Anticarsia gemmatalis*(Lepidoptera, Noctuidae) nuclear polyhedrosis virus released into soybean. Environ. Entomol., 23:1308-16.

Hamm, J.J. and Shapiro, M. (1992). Infectivity of fall armyworm (Lepidoptera: Noctuidae) nuclear polyhedrosis virus enhanced by a fluorescent brightener. J. Econ. Entomol., 85 (6): 2149-2152.

Khattab, M. Magda, EL- Salamouny, S., EL- Sheikh, M.A.K., Amin, A., and Elnagar, S.(2004): Positive effect of Fluorescent brightener 28 and Neemazal T/S on the activity of *Agrotis sgetum* granulovirus tested against the black cutworm, *Agrotis ipsilon* larvae (Lepidoptera: Noctuidae).1stArab Conference of applied

Biological Pest Control, Cairo, Egypt, 5-7 April 2004 pp57-58.

Khattab, Magda (2007): Enhanced Activity of *Agrotis segetum* (Schiff.) (Lepidoptera: Noctuidae) granulovirus by boric acid additive. J. Agricultural Science; Suez Canal University, 3:1-9.

Khattab, Magda (2012): First Isolation, In Egypt, of a granulovirus (*Agse*GV^{EG}) From *Agrotis segetum* Schiffermuller (Lepidoptera:Noctuidae).Egypt.Acad.J.Biol.Sci. (G Microbiology) Vol.4 (1) 93-99.

Li, S.Y. and Otvos, I.S. (1999). Optical brighteners enhance activity of a nuclear polyhedrosis virus against western spruce budworm (Lepidoptera: Tortricidae). J. Econ. Entomol., 92(2): 335-339.

Lipa, J., Ziemnicka, J., 1971. Studies on the granulosis virus of cutworms *Agrotis spp*. (Lepidoptera:Noctuidae). Acta Microbiologica Polonica. 3:155–162.

Lipa, J., Ziemnicka, J., Gudz-Gorban, A., 1971. Electron microscopy of nuclear polyhedrosis virus from *Agrotis segetum* SchiV. and *A. exclamationis* L. (Lepidoptera: Noctuidae). Acta Microbiologica Polonica. B3 (20) No.1: 55–61.

Mohamed, Sondos; A. Badr, N.A. and Abd El-Hafez A. (2000): Efficacy of two formulations of pathogenic bacteria *Bacillus thuringiensis* against the first instar larvae of *Spodoptera littoralis* and *Agrotis ipsilon.* J. Agric. Res., 78 (3): 1025-1039.

Morris, O.N., Converse, V. and Kanagaratnam, P. (1995). Chemical additive effect on the efficacy of Bacillus thuringiensis Berliner subsp. hurstaki against Mamestra configurata (Lepidoptera: Noctuidae) J. Econ. Entomol. 88: 815-824.

Shah, B.H.; Zethner, O.; Gul, H. and Chaudhry, M.I. (1979). Control experiments using *Agrotis segetum* granulosis virus against *Agrotis ipsilon* (Lep.: Noctuidae) on tobacco seedlings in northern Pakistan. Entomophaga, 24: 393-401.

Shapiro, M. (1992). Use of optical brighteners as radiation protectants for gypsy moth (Lepidoptera:Lymantriidae) nuclear polyhedrosis virus. J. Econ. Entomol., 85 (5) : 1682-1686.

Shaprio, M. and Bell. R.A. (1982). Enhanced effectiveness of *Lymantria disper* (Lepidoptera: Lymantriidae) nucleopolyhedrosis virus formulated with boric acid. Ann. Entomol. Soc. Am. 75: 346-349.

Shapiro, M. and Dougherty, E.M. (1994). Enhancement in activity of homologous and heterologous viruses against the gypsy moth (Lepidoptera: Lymantriidae) by an optical brightener. J. Econ. Entomol., 87 (2): 361- 365.

Shapiro, M. and Robertson, J.L. (1992). Enhancement of gypsy moth (Lepidoptera: Lymantriidae) baculovirus activity by optical brighteners. J. Econ. Entomol., 84 (4) : 1120-1124.

Shapiro, M. and Vaughn, J. L. (1995). Enhancement in activity of homologous and heterologous baculovirus infectious to cotton bollworm (Lepidoptera: Noctuidae) by an optical brightener. J. Econ. Entomol., 88 (2):265-269.

Shorey, H.H. and Hale, R.L.(1965). Mass rearing of the larvae of nine noctuid species on a simple artificial medium. J. Econ. Entomol., 58 : 522-524.

Tamez-Guerra, P.; McGuire, M.R. and Behle, R.W. (2000). Shelf life of *Anagrapha falcifera* nuclear polyhedrosis virus (*Af*MNPV) microcapsular lignin-based formulation. Society for Invertebrate Pathology, 33rd Annual Meeting Guanajuato, Mexico. Univ. of Guanajuato, 13-18 Aug., 2000.

Vail, P.V.; Hoffmann, D.F. and Tebbets, J.S. (1996). Effects of a fluorescent brightener on the activity of *Anagrapha falcifera* (Lepidoptera: Noctuidae) nuclear polyhedrosis virus to four noctuid pests. Biolo. Control, 7 (1): 121-125.

Zethner, O. (1980). Control of *Agrotis segetum* (Lepidoptera:Noctuidae) root crops

by granulosis virus. Entomophaga, 25:27-35.

Zethner, O.; Khan, B.M.; Chaudhry, M.L.; Bolet, B.; Khan, S.; Khan, H.; Gul, H.; Ogaard, L.; Zaman, M. and Nawaz, G. (1987). *Agrotis segetum* granulosis virus as a control agent against field populations of *Agrotis ipsilon* and *A. segetum* (Lepidoptera: Noctuidae) on tobacco, okra, potato and sugar beet in northern Pakistan. Entomophaga, 35 (5): 449-455.

Zou, Y.G. and Young, S.Y. (1996). Use of a fluorescent brightener to improve *Pseudoplusia includens* (Lepidoptera:Noctuidae) nuclear polyhedrosis virus activity in the laboratory and field. J. Econ. Entomol., 89 (1) : 92-96.

ARABIC SUMMARY

دراسات مقارنة لتأثيرالجرانيولوسيز فيروس (*Agse*GV^{EG}) لدودة اللفت القارضة *Agrotis segetum* على الدودة القارضة السوداء *Agrotis ipsilon* (حرشفية الأجنحة: نوكتويدى) تحت الظروف شبه الحقلية

ماجدة خطاب

معهد بحوث وقاية النباتات – مركز البحوث الزراعية- الدقى – الجيزة – مصر

إجريت دراسة لمقارنة تأثير معاملة الجرانيولوسيز فيروس لدودة اللفت القارضة *Agrotis segetum* المعزول في مصر، مع دراسة تأثير إضافة مادة العواكس الفلوروسنتية Fluorescent brightener (Tinopal UNPA-GX) بتركيز 1% كمادة منشطة للجرانيولوسيز فيروس بتطبيق الرش أو الطعم السام على الدودة القارضة السوداء *A. ipsilon* تحت الظروف شبه الحقلية.

وقد أظهرت نتائج الإختبارات الحيوية المعملية زيادة حساسية كلا من العمر اليرقى الثالث و الرابع للدودة القارضة السوداء للإصابة الفيروسية فى المعاملة المشتركة (*Agse*GV^{EG} +Tinopal %1) حيث كانت قيمة التركيز النصفى السام (LC_{50}) 2.08×10^{8} & 6.10×10^{8} OB/ml ، على التوالى. كما تراوحت قيمة الوقت النصفى السام (LT_{50}) 7.67و7.85يوم، على التوالى للمعاملة المشتركة (9.07×10^{10} OB/ml+ Tinopal %1).

أظهرت نتائج تجارب الدراسة شبه الحقلية ، على العمر اليرقى الثانى أن إضافة مادة العواكس الفلوروسنتية بتركيز 1% إلى محلول الرش أدى إلى زيادة مدة بقاء الفيروس على بادرات القطن ، حيث كانت قيمة (LIT_{50}) 0.97 و2.27 يوم لمعاملة الفيروس منفردا والمعاملة المشتركة، على التوالى.

كما أدت إضافة مادة العواكس الفلوروسنتية بتركيز 1% كمادة منشطة للفيروس إلى الطعم السام بالتركيز (9.07×10^{9} OB/ml و 9.07×10^{8} +Tinopal %1) إلى زيادة نسبة موت يرقات العمر اليرقى الثالث والرابع للدودة القارضة السوداء، و إنخفاض أعداد بادرات القطن المقروضة عند المعاملة المشتركة مقارنة بالغير معاملة (المقارنة) .

وقدرمعدل إنخفاض الضرر الناتج لبادرات القطن (المصابة) فى المعاملة المشتركة بالتركيز(OB/ml 9.07×10^{9} +Tinopal %1) ب 83.33 &70% لكل من العمراليرقى الثالث والرابع للدودة القارضة السوداء ، على التوالى مقارنة بالغير معاملة (المقارنة) .

توضح النتائج السابقة إمكانية إستخدام الجرانيولوسيز فيروس لدودة اللفت القارضة (*Agse*GV^{EG}) فى مكافحة الدودة القارضة السوداء *A. ipsilon* مع إضافة مادة منشطة حيث أظهرت المعاملة ب (*Agse*GV^{EG}) وإضافة Tinopal %1 وتطبيقها سواء بالرش أو كطعم سام يؤدي إلى موت يرقات الديدان القارضة السوداء أسرع من المعاملة بالفيروس منفردا" علي بادرات القطن.