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**Inhibited reproductive capacity of Egyptian cotton leaf worm *Spodoptera littoralis* (Boisd.) (Lepidoptera: Noctuidae) by the chitin synthesis inhibitor Novaluron.**

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**ABSTRACT**

For evaluating the effects of Novaluron on various parameters of the reproductive capacity of *Spodoptera littoralis*, a concentration range of 1.00-0.0001ppm was applied on the newly moulted penultimate (5<sup>th</sup>) instar larvae and a concentration range of 0.10-0.0001ppm was applied on the newly moulted last instar larvae. A predominant inhibitory effect of Novaluron was exhibited on the oviposition efficiency since the oviposition rate was seriously regressed in no certain trend, regardless the time of larval treatment and concentration level. Treatment of penultimate or last instar larvae with Novaluron resulted in drastically reduced fecundity in a dose-dependent course. A reducing action of Novaluron was exerted also on fertility after treatment of larvae with different concentration levels, regardless the time of treatment. After treatment of penultimate or last instar larvae with Novaluron, the embryonic development had been generally subjected to a retarding effect since the incubation period was pronouncedly prolonged, regardless the concentration level.

**Keywords:** Embryonic development, fecundity, fertility, oviposition efficiency.

**INTRODUCTION**

Over the last few decades, the intensive use of broad-spectrum insecticides against the Egyptian cotton leaf worm, *Spodoptera littoralis* Boisduval (Lepidoptera: Noctuidae) has led the development of resistance to many registered pesticides making their control even more difficult (Smagghie *et al.*, 1999; Miles and Lysandrou, 2002; Aydin and Gurkan, 2006). Owing to its socioeconomic importance, the insect is subject to extensive research, much of which is envisioned to finding new ways to control it as a pest and to improve the effects of known pest control methods (Hussain, 2012). At present, using insect growth regulators (IGRs) is considered as the possible alternative way of conventional synthetic insecticides for controlling this pest (Raslan, 2002). They have novel modes of action which disrupt the physiology and development of the target pest. Such compounds tend to be selective and generally less toxic to non-target organisms than conventional insecticides (Gurr *et al.*, 1999). Depending on the mode of action, IGRs had been recently grouped in

chitin synthesis inhibitors (CSIs) and substances that interfere with the action of insect hormones (i.e. juvenile hormone analogues, and ecdysteroids) (Tunaz and Uygun, 2004).

CSIs interfere with chitin biosynthesis in insects (Hajjar and Casida, 1978; Gijswijt *et al.*, 1979) and thus prevent moulting, or produce an imperfect cuticle (Hammock and Quistad, 1981). These compounds are effective suppressors of development for the entire life cycle of insect pests (Verloop and Ferrell, 1977; Grosscurt, 1978). They also affect the hormonal balance in insects, thereby resulting in different physiological disturbances (Deul *et al.*, 1978; Deloach *et al.*, 1981; Soltani *et al.*, 1984). Novaluron is a relatively new benzoyl phenyl urea CSI with good activity against several insects, such as *Leptinotarsa decemlineata* (Cutler *et al.*, 2007; Alyokhin *et al.*, 2009), *Bemisia tabaci* and *Trialeurodes vaporariorum* as well as leafminer *Liriomyza huidobrensis* (Kim *et al.*, 2000) and *Spodoptera* and *Helicoverpa* species (Ishaaya *et al.*, 2001, 2002, 2003). The compound has no appreciable effect on parasitoids and has probably a mild effect on other natural enemies (Ishaaya *et al.*, 2001, 2002). Also, it has low mammalian toxicity (Barazani, 2001; Ishaaya and Horowitz, 2002). Its residues tend to dissipate with half-life of 2.08 days and the safe use of it on tomatoes, and possibly on other crops in Egypt was established (Malhata *et al.*, 2014).

The noctuid *S. littoralis* one of the most destructive pests in the tropical and subtropical areas of the world (Hill, 1987). It is damaging vegetables in North Africa, cotton in Egypt and the glasshouses plant and flower production in Southern Europe (El-Aswad *et al.*, 2003; Roques *et al.*, 2008). It attacks plants in 44 families containing at least 112 species of plants of varying economic importance (Sarto and Monteys, 1988; Lal and Naji, 1990). When large numbers of the pest are present, complete crop loss is possible (Khalil, 1988). The current study was conducted aiming to evaluate the effects of Novaluron on various reproductive parameters of *S. littoralis*.

## MATERIALS AND METHODS

### Experimental insect.

A sample of *S. Littoralis* pupae was kindly obtained from the culture of susceptible strain maintained for several generations in Plant Protection Research Institute, Agricultural Research Center, Doqqi, Giza, Egypt. In laboratory of Entomology, Faculty of Science, Al-Azhar University, Cairo, a culture was reared under laboratory controlled conditions (27±2°C, 65±5% R.H., photoperiod 14 h L and 10 h D). Rearing procedure was carried out according to Ghoneim (1985) and improved by Bakr *et al.* (2010). Larvae were provided daily with fresh castor bean leaves *Ricinus communis*. The emerged adults were provided with 10% honey solution on a cotton wick as a food source. Moths were allowed to lay eggs on branches of *Nerium oleander*. The egg patches were collected daily and transferred into Petri dishes for another generation.

### Larval treatments with Novaluron:

Novaluron (1-[3-chloro-4-(1,1,2-trifluoro-2-trifluoro-methoxy-ethoxy) phenyl]-3-(2,6-difluorobenzoyl)urea) (Rimon, Mosquiron®100EC, Pestanal®, Chemtura Corporation, Middlebury, CT) was purchased from Sigma-Aldrich Chemicals (<https://www.sigmaaldrich.com>). Its molecular formula is C<sub>17</sub>H<sub>9</sub>ClF<sub>8</sub>N<sub>2</sub>O<sub>4</sub>. A series of concentration levels using distilled water (1.0, 0.1, 0.01, 0.001 & 0.0001 ppm) was prepared. Newly moulted larvae of penultimate (5<sup>th</sup>) instar were treated with all

concentrations but the newly moulted larvae of last instar were treated with 0.1, 0.01, 0.001 & 0.0001 ppm. Fresh castor bean leaf discs were dipped in each concentration for 5 minutes and air dried before introduction to larvae for feeding. Control congeners were provided with water-treated leaf discs. Ten replicates of treated and control larvae (one larva/replicate) were kept separately in glass vials. The larvae were left to feed on treated leaf discs for 24 hrs and then were provided with untreated fresh leaf discs every day. The larvae (control and treated) were carefully handled until the adult emergence just after which all reproductive parameters were recorded.

### Reproductive parameters:

The emerged adult females from each treatment were kept separately in glass jars (1 L) and coupled with normal adult males (1:2), of the same age, obtained from the main culture. Each jar was provided with pieces of sterilized cotton soaked in 10% honey solution for feeding and clean fresh branches of *N. oleander* as an oviposition site. The egg patches were collected daily and carefully transferred to Petri dishes for counting the eggs.

Oviposition rate was calculated as follows:

$$\text{Number of laid eggs per } \text{♀} / \text{reproductive lifetime (in days)}.$$

The laid eggs were counted for calculating the number of eggs per female (Fecundity). The laid eggs were kept in Petri dishes under the same controlled laboratory conditions as previously mentioned. Just after the oviposition, eggs were observed until hatching elapsing an incubation period (in days). The hatchability (Fertility) was usually expressed in hatching percentage of laid eggs. Sterility index was calculated according to Topozada *et al.* (1966) as follows:

$$\text{Sterility Index} = 100 - [(a/b / A/B) \times 100]$$

where: a: mean number of eggs laid per female in the treatment. b: percentage of hatching in the treatment. A: mean number of eggs laid per female in the controls. B: percentage of hatching in the controls.

### Statistical analysis of data:

Data obtained were analyzed by the Student's *t*-distribution, and refined by Bessel correction (Moroney, 1956) for the test significance of difference between means.

## RESULTS

### Oviposition efficiency of *S. littoralis* as affected by Novaluron:

In a preliminary experiment, treatment of penultimate or last instar larvae of *S. littoralis* with 10.0 ppm of Novaluron resulted in no adults due to the larval and pupal mortalities. Therefore, the used dose range for assessing the effects of Novaluron on reproductive parameters was 1.0-0.0001 ppm. In insects, the oviposition rate can be used as an informative indicator of the oviposition efficiency. Data assorted in Table (1) clearly show a predominant inhibitory effect of Novaluron on the oviposition efficiency since the oviposition rate was seriously regressed, regardless the concentration level applied on penultimate instar larvae.

Although such effect could not be exhibited in certain trend, the severely slow down rates were determined at the higher two concentration levels (24.63±1.37 and 16.14±2.04, respectively, vs. 192.64±4.33 of control females) denoting the most potent prohibiting action of Novaluron on the oviposition efficiency at these two

concentration levels after treatment of penultimate instar larvae.

A similar suppressing action of Novaluron on the oviposition rate, or oviposition efficiency, of the adult females was exerted after treatment of last instar larvae, regardless the concentration level as obviously shown in Table (2). Depending on data of this table, the strongest inhibitory effect of Novaluron was exhibited at the highest concentration level since it enforced the adult females to lay eggs in the slowest rate ( $108.33 \pm 4.17$  vs.  $192.64 \pm 4.33$  of control adult females).

#### Effect of Novaluron on the reproductive capacity of *S. littoralis*:

In the light of data distributed in Table (1), treatment of penultimate instar larvae with Novaluron resulted in drastically reduced fecundity in a dose-dependent course. The female fecundity dropped at the higher two concentration levels ( $78.00 \pm 22.14$  and  $100.00 \pm 11.00$  eggs/treated ♀, respectively, vs.  $1733.40 \pm 57.23$  eggs/control ♀).

Table 1: Oviposition efficiency and reproductive potential of *S. Littoralis* as affected by the treatment of newly moulted penultimate instar larvae with Novaluron.

Conc. (ppm)	Oviposition rate	Fecundity (mean eggs±SD)	Fertility (%)	Sterility index (%)	Incubation period (mean days±SD)
1.00	$024.63 \pm 1.37$ d	$078.00 \pm 22.14$ d	$37.23 \pm 0.00$ d	98.33	$4.00 \pm 0.00$ d
0.10	$016.14 \pm 2.04$ d	$100.00 \pm 11.00$ d	$72.00 \pm 0.00$ d	95.83	$4.00 \pm 0.00$ d
0.01	$140.08 \pm 4.12$ d	$721.00 \pm 18.39$ d	$44.70 \pm 4.24$ d	81.00	$4.00 \pm 0.00$ d
0.001	$132.58 \pm 3.67$ d	$945.00 \pm 76.88$ d	$65.60 \pm 0.57$ d	63.45	$3.00 \pm 1.00$ d
0.0001	$168.57 \pm 3.25$ c	$1128.85 \pm 62.94$ d	$84.20 \pm 1.94$ d	43.91	$3.43 \pm 0.53$ d
<b>Control</b>	$192.64 \pm 4.33$	$1733.40 \pm 57.23$	$97.80 \pm 0.78$	---	$2.20 \pm 0.45$

Conc.: concentration level.. Mean  $\pm$  SD followed with the same letter (a): insignificantly different ( $P > 0.01$ ), (b): significantly different ( $P < 0.05$ ), (c): highly significantly different ( $P < 0.01$ ), (d): very highly significantly different ( $P < 0.001$ ).

A similar reducing effect of Novaluron on the female fecundity was remarkably exhibited after treatment of last instar larvae. As shown in Table (2), the reducing effect was found in a dose-dependent manner. Also, the most reducing effect was exhibited at the highest concentration level ( $787.50 \pm 109.60$  eggs/treated ♀ vs.  $1733.40 \pm 57.23$  eggs/control ♀).

Table 2: Oviposition efficiency and reproductive potential of *S. Littoralis* as affected by the treatment of newly moulted last instar larvae with Novaluron.

Conc. (ppm)	Oviposition rate	Fecundity (mean eggs $\pm$ SD)	Fertility (%)	Sterility index (%)	Incubation period (mean days $\pm$ SD)
0.10	$108.33 \pm 4.17$ d	$787.50 \pm 109.60$ d	$66.85 \pm 1.91$ d	86.90	$4.00 \pm 0.00$ d
0.01	$150.79 \pm 5.67$ c	$1251.67 \pm 76.54$ d	$75.43 \pm 17.67$ d	44.38	$3.67 \pm 0.58$ d
0.001	$173.39 \pm 4.51$ b	$1378.75 \pm 124.19$ d	$84.88 \pm 2.18$ d	30.90	$3.25 \pm 0.50$ d
0.0001	$152.81 \pm 3.97$ c	$1149.17 \pm 135.63$ d	$82.53 \pm 1.14$ d	44.13	$3.33 \pm 0.52$ d
<b>Control</b>	$192.64 \pm 4.33$	$1733.40 \pm 57.23$	$97.80 \pm 0.78$	---	$2.20 \pm 0.45$

Conc., a, b, c, d: See footnote of Table (1).

Another parameter of the reproductive capacity is fertility (egg viability). Data arranged in Table (1) unexceptionally show a deranging action of Novaluron after treatment of penultimate instar larvae with different concentration levels. Although the reduction of fertility could not be detected in certain trend, the most powerful reducing action of Novaluron was exerted at 1.00 and 0.01 ppm ( $37.23 \pm 0.00$  and  $44.70 \pm 4.24\%$ , respectively, compared to  $97.80 \pm 0.78\%$  hatching eggs of controls). Moreover, the sterility index was found in a dose-dependent manner.

A similar fertility reduction was, also, recorded after treatment of last instar larvae with Novaluron. Referring to data depicted in Table (2), the strongest reducing action

of Novaluron on fertility was exerted at the highest concentration level ( $66.85 \pm 1.91$  vs.  $97.80 \pm 0.78\%$  hatching eggs of controls). In addition, the highest sterility index was calculated in 86.90 at the highest concentration level while the lowest one was calculated in 30.90 at 0.001 ppm.

#### **Effect of Novaluron on the embryonic development of *S. littoralis*:**

In insects, the egg incubation period can be used as a good indicator of the embryonic developmental rate, i.e., longer period usually denotes slower developmental rate and *vice versa*. After treatment of penultimate instar larvae with different concentration levels of Novaluron, data of the egg incubation period were listed in Table (1). Generally, the embryonic development had been subjected to an extended inhibitory effect of the used compound since the incubation period was pronouncedly prolonged. At the higher three concentration levels, the most prolonged period was estimated in  $4.00 \pm 0.00$  days, compared to  $2.20 \pm 0.45$  days of control eggs.

A similar prohibiting effect of Novaluron on the embryonic development was exhibited after treatment of last instar larvae since the incubation period was significantly lengthened, regardless the concentration level. The most prolonged period ( $4.00 \pm 0.00$  days, vs.  $2.20 \pm 0.45$  days of control eggs) was recorded only at the highest concentration level (Table 2).

## **DISCUSSION**

Reproduction in insects is mainly controlled by corpus allatum hormone (juvenile hormone, JH), which is also responsible for protein metabolism, and is specifically needed for egg maturation. The insect growth regulators (IGRs) have been found to render treated insects either sterile or less fecund (Metwally *et al.*, 1972). The IGR-treated insects may develop as morphologically deformed adults who would be non-viable or at least their reproductive capacity is reduced (Williams and Amos, 1974). The effects of IGRs on reproduction can be grouped into the following categories: i) reproductive behaviour, ii) oviposition, iii) hatchability of eggs (ovicidal and embryocidal), and iv) sterilization of adults (Mondal and Parween, 2000). On the other hand, ecdysteroids have essential functions in controlling the processes involved in insect reproduction, i.e., vitellogenesis, ovulation of matured eggs and spermatocyte growth (Wigglesworth, 1984; Hagedorn, 1985).

#### **Inhibited oviposition efficiency of *S. littoralis* by Novaluron:**

In insects, the oviposition rate can be used as an informative indicator for the oviposition efficiency. In the present study, Novaluron exhibited a predominant inhibitory effect on the oviposition efficiency of *S. littoralis*, since the oviposition rate was drastically regressed, in no certain trend, irrespective of the time of larval treatment or concentration level. This result is in agreement with those reported results of decreased oviposition rate (or index) of the same lepidopteran insect by tebufenozide (Bakr *et al.*, 2005) or flufenoxuron (Bakr *et al.*, 2010). It also agrees, to a great extent, with the inhibited oviposition of *Schistocerca gregaria* by flufenoxuron and lufenuron (Soltani-Mazouni and Soltani, 1994) or tebufenozide (Al-Dali *et al.*, 2008), *Plodia interpunctella* by the ecdysteroid agonist RH-5849 (Smagghe and Degheele, 1994), *Musca domestica* by other IGRs (Abdel-Moty *et al.*, 1996) and *Callosobruchas maculata* by Cyromazine (Al-Mekhlafi *et al.*, 2011) but disagrees with the stimulated oviposition of *Gryllus bimaculatus* by some ecdysteroid agonists (Behrens and Hoffmann, 1983).

The prohibited oviposition, in the current work, may be explained as a result of inhibition of ovarian DNA synthesis or the interference of Novaluron with vitellogenesis *via* certain biochemical processes, as will be mentioned later. However, this CSI may exert a reverse action to those exerted by the ecdysteroid agonists which stimulate the neurosecretory cells to release a myotropic ovulation hormone (Soltani-Mazouni, 1994; Smaghe *et al.*, 1996; Parween *et al.*, 2001).

#### **Reduced fecundity and fertility of *S. littoralis* by Novaluron:**

The available literature contains a lot of results of reduced fecundity of *S. littoralis* after treatment of larvae with different IGRs, such as diflubenzuron (Rofail, 1981; Aref *et al.*, 2010), chlorfluazuron (Morsi, 1985), lufenuron (Shaaban, 1993; Abdel-Rahman *et al.*, 2007; Gaaboub *et al.*, 2012), flufenoxuron (Shaaban and Mourad, 1994), methoxyfenozide (Ishaaya *et al.*, 1995; Pineda *et al.*, 2009). Also, fecundity of other insect species was reduced by various IGRs, such as *Rhyzopertha dominica* by methoprene (Daglish and Pulvarenti, 1997); *Platynotai daeusalis*, *Helicoverpa zea* and *Cydia pomonella* (Carpenter and Chandler, 1994; Brown, 1996; Biddinger and Hull, 1999) and *Ephestia kuehniella* (Khebbab *et al.*, 2008) by the ecdysteroid agonist tebufenozide; *Argyrotaenia velutinana* and *Choristoneura rosaceana* (Sun *et al.*, 2000), *L. botrana* (Saenz-de-Cabezón *et al.*, 2005) and *Spodoptera litura* (Shahout *et al.*, 2011) by the ecdysteroid agonist methoxyfenozide; *L. decemlineata* and *Aubeonymus mariaefranciscae* (Farinos *et al.*, 1999), and *Tenebrio molitor* (Taibi *et al.*, 2003) by the ecdysteroid agonist halofenozide (RH-0345); etc. In addition, fecundity in some insects was reduced after treatment with different CSIs, such as *S. litura* after treatment with chlorfluazuron (Perveen and Miyata, 2000), *M. domestica* after treatment with lufenuron (Hamadah, 2003), *Dysdercus koenigi* after treatment with flufenoxuron (Khan and Qamar, 2011), *Anagasta kuehniella* after treatment with Diflubenzuron and hexaflumuron (Ashouri *et al.* 2014), etc. The present results are in accordance with these reported results because treatment of penultimate instar larvae of *S. littoralis* with Novaluron resulted in considerably reduced fecundity in a dose-dependent course. A similar reducing effect on fecundity was pronouncedly exhibited after treatment of last instar larvae with Novaluron. On the other hand, our results disagree with some results because Fenoxycarb failed to affect fecundity of *Apis mellifera* (Thompson *et al.*, 2005), methoxyfenozide failed to affect fecundity of *Spodoptera exigua* (Christian-Lius *et al.*, 2010) and Novaluron and diflubenzuron failed to affect fecundity of *Halyomorpha halys* (Kamminga *et al.*, 2012). Moreover, feeding of *S. littoralis* larvae on leaves treated with a sublethal dose of methoxyfenozide resulted in a significantly increase in fecundity (Ishaaya *et al.*, 1995). These differences of effect can be understood by the different modes of action of IGRs, different susceptibilities of the insect species, time of treatment and other factors.

The reduced fecundity in *S. littoralis* adult females, after treatment of larvae with Novaluron, may be attributed to its interference with one or some processes from ovarian follicle development to egg maturation. In other words, Novaluron may inhibit the development of some ovarioles in larvae (Davey, 1993) or adversely affect the morphogenesis of ovipositor of adult females, ovarian growth and/or synthesis and metabolism of proteinaceous constituents during the oogenesis (Smaghe *et al.*, 1996; Salem *et al.*, 1997). Novaluron may cause some disorders in the ovaries, including cell death in the germarium, resorption of oocytes in the pre-vitellarium and vitellarium, formation of vitellin envelopes and undue proliferation of follicle cells sometimes resulting in malformation of the whole ovary (Metwally *et al.*, 1972;

Lucantoni *et al.*, 2006; Khan *et al.*, 2007). On the basis of hormonal regulation of insect reproduction, Novaluron may disturb the production and/or function of the gonadotropic hormone (juvenile hormone, JH) responsible for the synthesis of vitellogenins (yolk precursors) and vitellogenesis (Di Ilio *et al.*, 1999). In addition, it may be acceptable to suggest that the reduced reproductive output of *S. littoralis*, in the current work, may be due to an inhibitory effect of CSI on synthesis of both DNA and RNA, suboptimal nutrition owing to reduced feeding, altered mating behaviour as a result of sublethal intoxication, or a combination of factors.

Another parameter of the reproductive capacity in insects is fertility (egg viability). In the present study, Novaluron exerted a deranging action on fertility of *S. littoralis* after treatment of penultimate or last instar larvae with different concentration levels. The reduction in fertility could not be detected in a certain trend. To a great extent, these results are in compliance with those reported results of reduced fertility in the same lepidopteran by several CSIs, such as diflubenzuron (El-Badawy, 1979; Rofail, 1981; Aref *et al.*, 2010), chlorfluazuron (Morsi, 1985; Sammour *et al.*, 2008), flufenoxuron (Shaaban and Mourad, 1994), triflumuron (El-Naggar, 2013), lufenuron (Shaaban, 1993; Abdel-Rahman *et al.*, 2007; Sammour *et al.*, 2008; Adel, 2012; Gaaboub *et al.*, 2012) as well as the ecdysteroid agonist methoxyfenozide (Pineda *et al.*, 2009). Also, our results agree, to some extent, with many reported results of reduced fertility of other insects by various IGRs and CSIs, such as *S. litura* (Perveen and Miyata, 2000), *M. domestica* by Diofenolan (Hamadah, 2003), *Tribolium castaneum* by Novaluron (Kostyukovsky and Trostanelsky, 2004), *D. koenigi* by flufenoxuron (Khan and Qamar, 2011), *C. maculata* by Cyromazine (Al-Mekhlafi *et al.*, 2011), *Pectinophora gossypiella* by lufenuron and chlorfluazuron (Kandil *et al.*, 2012), *A. kuehniella* by diflubenzuron and hexaflumuron (Ashouri *et al.*, 2014), etc. In addition, a reduction in fertility was reported in different insect species by some ecdysteroid agonists, such as *H. zea* (Carpenter and Chandler, 1994) and *E. kuehniella* (Khebbeb *et al.*, 2008) by tebufenozide; *T. Molitor* by Halofenozide (Taibi *et al.*, 2003); *A. velutinana* and *Ch. rosaceana* (Sun *et al.*, 2000), *L. botrana* (Saenz-de-Cabezonet *et al.*, 2005), *S. exigua* (Christian-Lius *et al.*, 2010), *S. litura* (Shahout *et al.*, 2011), *E. kuehniella* (Bouzera and Soltani-Mazouni, 2014) by methoxyfenozide and *P. gossypiella* by chromafenozide (Kandilet *et al.*, 2012).

For explicating the fertility reduction in *S. littoralis* by Novaluron, some suggestions can be provided herein, excluding those reported interpretations after treatment of adults (delayed lethal effects and transovarial movement of CSI) because only the larvae had been treated with CSI in the present study. It is well known that the maturation of insect eggs depends basically on the vitellogenins, precursor materials of vitellins including proteins, lipids and carbohydrates, all of which are necessarily required for the embryonic development (Kanost *et al.*, 1990; Soltani and Mazouni, 1992; Chapman, 1998). These materials are synthesized primarily by fat body during the immature stages (Telfer, 2009) or by the ovary *in situ* (Indrasith *et al.*, 1988). Wherever the site of synthesis of these materials, Novaluron may disturb their production and/or accumulation in adult females leading to reduction of fertility in *S. littoralis*, in the present study. This suggestion may be substantiated by the reduction in reproductive parameters previously reported in studies using ecdysteroid agonists against several insect species (Knight, 2000; Sun *et al.*, 2003; Taibi *et al.*, 2003; Pineda *et al.*, 2006; Osorio *et al.*, 2008). On the other hand, the tested CSI may indirectly affect the fertility *via* its disruptive effect on "potency" or opening of the intracellular spaces in follicular epithelium or generally inhibited the role of JH (gonadotropic hormone) responsible for the regulation of vitellogenin deposition into

oocytes (Davey and Gordon, 1996). Also, the reduction in fertility may be due to the penetration of residual amounts of CSI in mothers into their eggs and disturbance of embryonic cuticle synthesis. So, the fully mature embryos had weakened chitinous mouth parts that were insufficiently rigid to perforate the surrounding vitellin membrane and free from the eggs (Mass *et al.*, 1980; Marco and Vinuela, 1994; Sallam, 1999; Sammour *et al.*, 2008). The reduced fertility of *S. littoralis*, in the current study, may be due to serious effect of Novaluron on the survival of developing embryos at certain stages as recorded in decreasing hatching percentage. Because the molecular studies revealed the effects of some IGRs on insect reproduction owing to the perturbation of gene expression in the hierarchy cascade of vitellogenesis and/or choriogenesis (Sun *et al.*, 2003), Novaluron may interfere with the gene expression resulting in a reduction of the developed embryos in *S. littoralis*, in the present study.

#### **Retarded embryonic development of *S. littoralis* by Novaluron:**

In insects, the egg incubation period can be used as a good indicator for the embryonic developmental rate, i.e., longer period usually denotes slower rate and *vice versa*. In the present study, treatment of penultimate or last instar larvae of *S. littoralis* with Novaluron resulted generally in retarded embryonic development since the incubation period was remarkably prolonged, regardless the concentration level. Unfortunately, the literature, available to us, contains no reported results of affected incubation period in the same lepidopteran by IGRs. In other insects, increasing Cyromazine concentrations led to increasingly prolonged incubation period of *C. maculatus* eggs (Al-Mekhlafi *et al.*, 2011) and treatment with LC<sub>50</sub> of lufenuron, chlorfluazuron and chromafenozide resulted in significantly prolonged incubation period of *P. gossypiella* eggs (Kandilet *et al.*, 2012). Also, prolonged incubation period was reported for some insect species by extracts of certain plants, such as *Euprepocnemis plorans* by Margosan-O (a neem preparation) (Abdel Ghaffar, 1998), *Schistocerca gregaria* by *Fagonia bruguieri* extracts (Basiouny, 2008), the same locust by *Nigella sativa* seed extracts (Hamada, 2009) and the same locust by *Punica granatum* peel extracts (Ghoneim *et al.*, 2014). The delayed embryonic development in *S. littoralis* after treatment of larvae with Novaluron, in the present study, may be due to its effect on ecdysteroids responsible for the regulation of embryogenesis at certain stages, especially those originating from the ovary *in situ* (Chapman, 1998).

#### **CONCLUSION**

Since *S. littoralis* has developed resistance to the majority of conventional synthetic insecticides, the use of compounds with different mode of action, like Novaluron, might be considered a part of the integrated pest management since it disruptively affected the reproductive capacity leading to a reduction of the pest population.

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## ARABIC SUMMARY

تنشيط القدرة التكاثرية لدودة ورق القطن المصرية سيبودوبتيرا ليتوراليس (حرفشيات الأجنحة: الليلييات) بمشبط تخليق الكيتين نوفالورون

كارم السيد غنيم، محمد علي طناني، خالد شوقي حمادة، أحمد لطفي بسيوني، حسن عمر وهيب  
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من أجل تقويم تأثيرات مركب نوفالورون في مختلف معايير القدرة التكاثرية لدودة ورق القطن، تمت معالجة يرقات الدور قبل الأخير (الخامس) بالتركيزات 1.0-0.0001 ج ف م، ومعاملة يرقات الدور الأخير بالتركيزات 0.1-0.0001 ج ف م. أبدى المركب تأثيراً تثبيطياً سائداً في كفاءة وضع البيض، مادام معدله قد تراجع تراجعاً كبيراً- في غير اتجاه محدد- بصرف النظر عن التركيز المستعمل وتوقيت المعاملة. وأياً كان توقيت المعاملة، فإن المعاملة بمركب نوفالورون قد أدت إلى اختزال إنتاجية الإناث اختزالاً عنيفاً، توازى مع مستوى التركيز. كما أنه قد بذل فعلاً اختزالياً بكافة تركيزاته، بغض الطرف عن توقيت المعاملة، وسجلت الدراسة الحالية، أيضاً، تعطيل عام للإنباء الجنيني، مادامت فترة حضانة البيض قد طالت إطالة ملحوظة، بصرف النظر عن التركيز المستعمل.