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The Relative Potency of some Pesticides against Cotton Leafworm, *Spodoptera littoralis* (Boisd.) and their Side Effect on the Natural Enemy, *Chrysoperla carnea* (Stephens)

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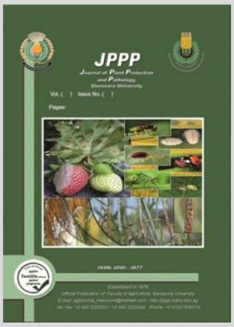
Abdullah, R. R. H.*

Plant Protection Research Institute, Agriculture Research Center, Dokki, Giza, Egypt.



ABSTRACT

The cotton leafworm, *Spodoptera littoralis* (Boisd.) is one of the dangerous economic insects on different crops in Egypt because it causes more damages in crops especially cotton and leafy vegetables. On the other hand, the natural enemy, *Chrysoperla carnea* (Stephens) is an important predator in all fields because it is able to control many insect pests and mites. In this study, six commercial pesticides (Chlorpyrifos, Alpha-cypermethrin, Methomyl, Abamectin, Biossiana, and Dipel) and two new compounds under the study (Crude extract of *Beauveria bassiana* metabolites and synthesis compound from Cyanoacetamide derivatives) were tested to determine their toxicity against the 2nd instar larvae of *S. littoralis* and their side effects against the 2nd instar larvae of *C. carnea.* All tested compounds had different degrees of toxicity against both tested insects. Based on the values of LC_{50}, toxicity index and relative potency, Alpha-cypermethrin was the highest toxic pesticide against *S. littoralis* and *C. carnea* followed by Methomyl and Chlorpyrifos but Biossiana was the least toxic one. In addition, the toxicity of tested chemical pesticides against *C. carnea* was more than their toxicity against *S. littoralis*. While, the toxicity of tested biopesticides against *C. carnea* was less than their toxicity against *S. littoralis*. In general, the sensitivity of the predator *C. carnea* to the tested biopesticides was less in comparison with chemical pesticides. Therefore, this study recommends using biopesticides besides the slight using of chemical pesticides to preserve the natural enemies and environment and achieve the best pest control.

Keywords: Toxicity index, Insecticidal activity, Biopesticides, Chemical pesticides

INTRODUCTION

The cotton leafworm, *Spodoptera littoralis* (Boisd.) (Lepidoptera: Noctuidae) is a destructive prolific and highly polyphagous insect in Egypt. It causes various ravages for cotton plants as well as other field crops and vegetables. Likewise, It is a major pest of great economic importance in many countries since it attacks a multitude of host plants. (Lobna *et al*., 2013 and Heidi *et al*., 2015 and Abdullah and El-Sharkawy , 2019). Many of pesticides were used to control *S. littoralis* as chemical pesticides (Chlorpyrifos, Alpha-cypermethrin, methomyl and act…) and biopesticides (*Bacillus thuringiensis* , Abamectin, Spinosad and act…) (EL-Khayat, *et al.* 2012).

The synthesized chemical pesticides have high efficiency to control *S. littoralis* and many of agricultural pests. Likewise, they have more dangerous to non-target organisms and the environment in general (Estay and Bruna 2002; Lietti, *et al*. 2005; Desneux, *et al*. 2007). Also, the biopesticides have enough value of efficacy to control several agricultural pests as well as keeping on the environment and human health. In addition, the use of pesticides results in the exposure of natural enemies of pests to these pesticides. Therefore, using safe pesticides that have low toxicity to natural enemies is necessary for the conservation of natural enemies populations (Tanaka, *et al*. 2000).

The common green lacewing, *Chrysoperla carnea* (Stephens) (Neuroptera: Chrysopidae) is one of the most common arthropod predators with a wide prey range including aphids, eggs and neonates of lepidopteran insects, scales insects, whiteflies, mites, and other soft bodied insects (McEwen, *et al*. 2001). *C. carnea* appears to be a good candidate for use in I.P.M. programs information (Aziza, *et al.* 2007).

The aim of this study is to measure the toxicity and the relative potency of some commercial pesticides and two new compounds under the study against cotton leafworm *S. littoralis*. Also, the side effect of all tested compounds was investigated against the natural enemy *C. carnea* under laboratory condition.

MATERIALS AND METHODS

The tested compounds:

a) Commercial pesticides:

Six commercial pesticides were kindly obtained from Plant Protection Research Institute, Agriculture Research Center, Dokki, Giza, Egypt. These pesticides belong to different pesticides groups as organophosphate, pyrethroid, carbamate, metabolites of microorganisms, and their spores. They are Chlorzan 48% EC (Chlorpyrifos), Alpha Z 10% EC (Alpha-cypermethrin), Newmel 90% SP (Methomyl), Mectyam 1.8% EC (Abamectin), Biossiana 2.5% WP (*Beauveria bassiana*) and Dipel 6.4% DF (*Bacillus thuringiensis*). These pesticides were used as the standard compounds to determine the relative potency and toxicity index of some new compounds. All compounds and their concentrations are listed in Table (1).

b) New compounds under the study:

Two new compounds were kindly obtained from the previous studies (Abdullah, 2019 & El-Sharkawy and Abdullah

*** Corresponding author.**
E-mail address: redakenany@yahoo.com
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2020). The first compound was the secondary metabolites of *Beauveria bassiana* which was extracted from a broth culture of the fungus by ethyl acetate solvent as described by Abdullah, (2019). The second compound was the synthesis compound from cyanoacetamide derivatives ((E)-2-cyano-N-(2-hydroxyphenyl)-3-(methylthio)-3-(phenylamino) acrylamide) as described by El-Sharkawy and Abdullah (2020). Both compounds and their concentrations are listed in Table (1).

The tested insects:

a) Cotton leafworm, *S. littoralis*:

The Laboratory strain of cotton leafworm *Spodoptera littoralis* Boisd. (Lepidoptera: Noctuidae) was obtained from Plant Protection Research Institute, Agriculture Research Center, Dokki, Giza, Egypt. Larvae were reared in laboratory by using clean castor leaves as described by El-Defrawi, *et. al.* (1964) to obtain the 2nd instar larvae for bioassay test.

b) The natural enemy, *C. carnea*:

The first instar larvae of the predator *Chrysoperla carnea* Stephens (Neuroptera: Chrysopidae) were kindly obtained from Dr. Ahmed Hassan Abd-Elwahab, Plant Protection Research Institute, Agriculture Research Center, Dokki, Giza, Egypt. The larvae were reared on the *Sitotroga cerealella* (Olivier) eggs but the adult stage was provided with water and a nutritious artificial diet as described by Vogt, *et al.* (2000). The rearing condition was 25 ± 2ºC, 60-80% (RH) and a photoperiod of 16:8 (L:D) h.

Table 1 . The tested compounds and their concentrations.

No	Trade name	Common name	Formulation	Pesticide group	The recommended field rate	The tested concentrations (ppm)
1	Alpha Z	Alpha cypermethrin	10% EC	Pyrethroid	60ml/100L	50 100 150
2	Chlorzan	Chlorpyrifos	48% EC	Organophosphate	1L/fed	500 1000 1500
3	Newmel	Methomyl	90% SP	Carbamate	300gm/fed	250 500 1000
4	Mectyam	Abamectin	1.8% EC	Avermectin fermentation of soil actinomycete *Streptomyces avermitilis*	40ml/100L	50 100 150
5	Biossiana	*Beauveria bassiana*	2.5% WP	Spores of *Beauveria bassiana*	250gm/100L	500 1000 1500
6	Dipel	*Bacillus thuringiensis*	6.4 % DF	Crystal toxins of *Bacillus thuringiensis*	300gm/fed	500 1000 1500
7	Crud extract	Secondary metabolites of *Beauveria bassiana*	100% Secondary metabolite	Fermentation products	new	1000 2000 4000
8	Synthesis compound	Cyanoacetamide derivatives	100% active ingredient	(E)-2-cyano-N-(2-hydroxyphenyl)-3-(methylthio)-3-(phenylamino) acrylamide	new	1000 2000 4000

Bioassay experiment:

a) Cotton leafworm, *S. littoralis*:

Leaf-dip bioassay method was conducted as described by Tabashnik, *et al*. (1991). Ten larvae of the 2nd instar were put in plastic jar (10 cm in diameter and 15 cm in height) and starved about 4 hours. Then disks (6 cm in diameter) of clean castor leaves were prepared and dipped 10 seconds in the prepared serial concentrations of pesticide solutions as mentioned in Table (1). The treated leaves were allowed to air dry then presented to larvae. Untreated castor leaves were presented to other larvae group as the control treatment. Each concentration of each compound in addition to the control treatment were repeated four times. The live larvae in each treatment were recorded every day until seven days after treatment. The mortality percentages were corrected by Abbot's formula (Abbot, 1925). Lethal concentration values, confidence limits and slope were calculated as (Finney, 1971), toxicity index and relative potency between all tested pesticides were estimated according to Sun's equation (Sun, 1950).

b) The natural enemy, *C. carnea*:

The bioassay test was conducted as the described method by Maia, *et al.* (2016) with some modification. Thirty larvae of the 2nd instar of the predator *C. carnea* were put in the plastic plates (6 cm in diameter and 3 cm in height) individually (to avoid cannibalism). The larvae were starved about four hours before the treatment. Serial concentrations from each compound were prepared as mentioned in Table (1). The castor leaves which contain different stages of the two-spotted red mite (*Tetranychus urticae*) were obtained from untreated castor tree by pesticides. Then the castor leaves were cut into fragments contains about 10 individuals of mites. Then the prepared fragments were dipped 10 seconds in the prepared concentrations of each compound and allow to air dry. Then the treated fragments presented to *C. carnea* larvae. Another untreated fragments were presented to other thirty larvae as control treatment. Also, small fragments of sponge were dipped in the prepared concentrations of each compound until saturation. One fragment of treated sponge was presented to each larva as a contaminated drink source. Another group of sponge fragments were dipped in tap water and presented to control group of larvae. New fragments of untreated castor leaves infested by mites were added every 24 hours as a food source to all *C. carnea* larvae. The dead larvae of *C. carnea* was recorded every day until seven days after treatment. Also, toxicity index and relative potency were estimated according to Sun's equation (Sun, 1950). The mortality percentages were corrected by Abbot's formula (Abbot, 1925).

Abbot's formula:

$$\text{Corrected \% mortality} = \frac{\text{Survival number in control} - \text{Survival number in treatment}}{\text{Survival number in control}} \text{X } 100$$

Sun's formula:

$$\text{Toxicity index} = \frac{LC_{50} \text{ of the most effective compound}}{LC_{50} \text{ of the other compound}} \text{ X } 100$$

$$\text{Relative Potency} = \frac{LC_{50} \text{ for least effective compound}}{LC_{50} \text{ of the other compound}}$$

RESULTS AND DISCUSSION

Results:

The toxicity of eight compounds; six commercial pesticides and two new compounds under the study against the 2nd instar larvae of cotton leafworm, *S. littoralis* under laboratory conditions are shown in Table (2). The susceptibility of *S. littoralis* to tested pesticides varied according to the insecticide group. In general, the larvae were more susceptible to pesticides which are belonged to synthetic chemical pesticides as Alpha Z 10% EC (Alpha-cypermethrin), Chlorzan 48% EC (Chlorpyrifos), Newmel 90% SP (Methomyl) than other pesticides which are belonged to biopesticides as Mectyam 1.8% EC (Abamectin), Biossiana 2.5% WP (*Beauveria bassiana*) and Dipel 6.4% DF (*Bacillus thuringiensis*). On the other hand, both new compounds have a middle effect between the synthetic chemical pesticides and biopesticides. The toxicity index indicated that Alpha-Z was the more toxic pesticide followed by Newmel but the lowest toxic pesticide was Biossiana followed by Dipel against the 2nd instar larvae of cotton leafworm, *S. littoralis* as shown in Figure (1).

Table 2. Toxicity of the tested compounds on the 2nd instar larvae of *S. littoralis* after three days post-treatment.

Treatments	LC_{50} ppm	Confidence limit ppm Lower	Confidence limit ppm Upper	Slope ± SE	Chi Square χ^2	r	Toxicity index %	Relative potency
Alpha Z	73	62	83	2.76±0.39	2.31	0.97	100.00	39.86
Chlorzan	199	51	322	2.07±0.53	0.14	0.99	36.68	14.62
Newmel	117	35	186	1.55±0.37	0.06	0.99	62.39	24.87
Mectyam	240	184	430	2.67±0.56	3.73	0.93	30.42	12.13
Biossiana	2910	2028	7505	2.17±0.52	0.05	0.99	2.51	1.00
Dipel	1791	1355	2072	2.76±0.46	2.66	0.96	4.08	1.62
Crud extract	1542	1328	2182	1.51±0.30	1.31	0.97	4.73	1.89
Synthesis compound	1469	1152	1763	1.9±0.31	1.68	0.98	4.97	1.98

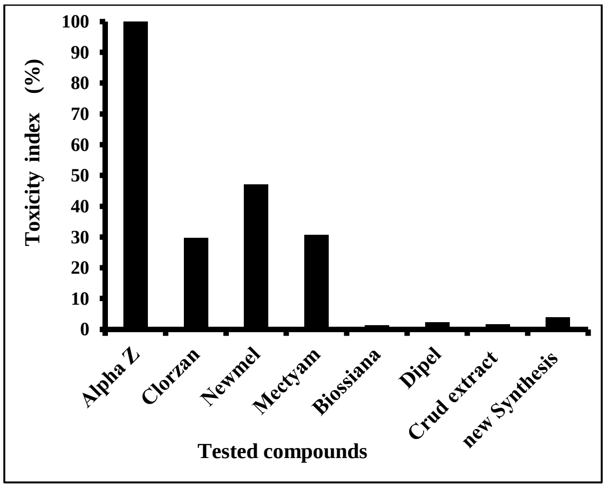

Figure 1. The efficiency of the tested compounds against the 2nd instar larvae of *S. littoralis* based on the toxicity index values.

The side effect of all tested compounds was assessed against the natural enemy, *C. carnea* under laboratory conditions. Table (3) shows the susceptibility of *C. carnea* to eight compounds; three compounds belonged to commercial chemical pesticides and three compounds belonged to commercial biopesticides in addition to two new compounds under the study. Based on the LC_{50} values, the commercial chemical pesticides (Alpha Z 10% EC (Alpha-cypermethrin) and Newmel 90% SP (Methomyl)) were more toxic compounds against the predator *C. carnea* followed by Mectyam 1.8% EC (Abamectin), Chlorzan 48% EC (Chlorpyrifos) and the new synthesis compound. On the other hand, the compounds Biossiana 2.5% WP (*Beauveria bassiana*), Dipel 6.4% DF (*Bacillus thuringiensis*), and the new crude extract compound have a low toxic effect. Likewise, based on the toxicity index and relative potency values, it is clear that Alpha Z 10% EC (Alpha-cypermethrin) was the most toxic compound against *C. carnea* followed by Newmel 90% SP (Methomyl) and Mectyam 1.8% EC (Abamectin) with toxicity index values of 100, 47.12 and 30.82, respectively. While, Biossiana 2.5% WP (*Beauveria bassiana*) and the new crude extract compound were the least toxic compounds with a toxicity index of 1.30 and 1.57, respectively as shown in Figure (2).

Table 3. Toxicity of the tested compounds against the 2nd instar larvae of *C. carnea* after three days post-treatment.

Treatments	LC_{50} ppm	Confidence limit ppm Lower	Confidence limit ppm Upper	Slope ± SE	Chi Square χ^2	r	Toxicity index %	Relative potency
Alpha Z	49	38	58	2.71 ±0.42	0.03	0.99	100.00	76.63
Chlorzan	165	24.7	287	2.36±0.68	1.89	0.91	29.70	22.76
Newmel	104	34	162	2.10±0.48	1.37	0.97	47.12	36.11
Mectyam	159	135	208	2.69±0.45	1.40	0.98	30.82	23.62
Biossiana	3755	2375	17712	2.44±0.70	0.94	0.98	1.30	1.00
Dipel	2118	1545	4827	1.64±0.41	0.01	0.99	2.31	1.77
Crud extract	3123	1850	5910	2.16±0.45	1.41	0.97	1.57	1.20
Synthesis compound	1260	681	2762	1.53±0.48	0.23	0.98	3.89	2.98

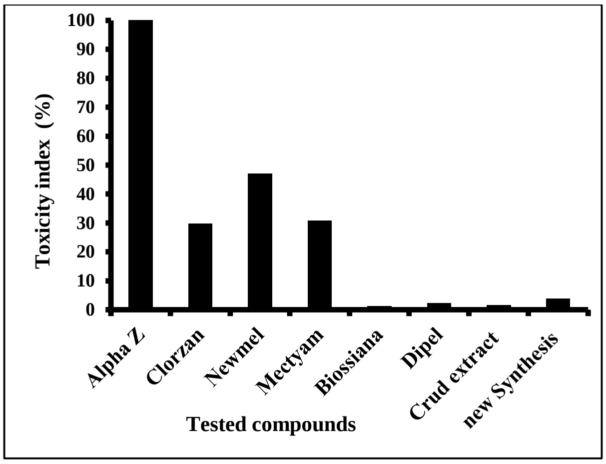

Figure 2. The efficiency of the tested compounds against the 2nd instar larvae of *C. carnea* based on the toxicity index values.

In general, as shown in Figure (3) the synthetic chemical compounds (Alpha-cypermethrin, Chlorpyrifos, Methomyl, and the new synthesis compound) in addition to Abamectin were more toxic effect against the natural enemy *C. carnea* than their toxicity against the pest *S. littoralis*. But the other compounds which belonged to the biopesticides group as Biossiana, Dipel, and the new crude extract compounds have a less toxic effect against the natural enemy, *C. carnea* with an acceptable toxic effect against cotton leafworm, *S. littoralis*.

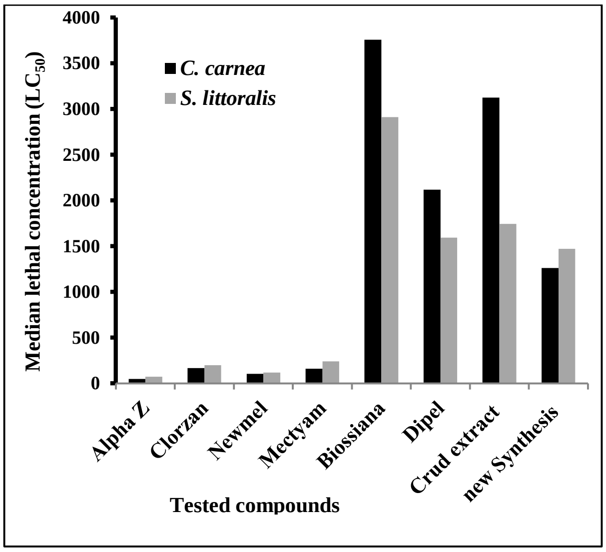

Figure 3. The efficiency of tested compounds against *S. littoralis* and *C. carnea* under laboratory conditions based on LC_{50} values.

Discussion

The selectivity of insecticides and their effects on natural enemies are important aspects of IPM programs (Metcalf, 1980; Hardin, *et al.* 1995; Desneux, *et al.* 2007). Chemical control is the most common method for pest control (Cooper and Dobson 2007; Song and Swinton 2009) and its use has increased in various cultures, especially in developing countries (Song and Swinton 2009; Meissle, *et al*. 2010; Lu, *et al.* 2012; Pedlowski, *et al.* 2012). The synthetic chemical insecticides are presently the main method for controlling the cotton leafworm, *S. littoralis* in Egypt and will likely continue to be used until more biologically based management studies could be developed (Alotaibi, 2013).

In the present study, the chemical pesticides, Alpha-cypermethrin, Chlorpyrifos, and Methomyl had high toxicity against cotton leafworm, *S. littoralis* in comparison with the other tested biopesticides such as Abamectin, *Bacillus thuringiensis*, *Beauveria bassiana*, and the crude extract of secondary metabolites of *B. bassiana*. These results are in agreement with Fetoh, *et al.* (2015) who studied the effectiveness of Emamectin benzoate and *Bacillus thuringiensis* as biopesticides and Chlorpyrifos as chemical pesticide against *S. littoralis* and determined the best compound for controlling this economic insect. They found that Chlorpyrifos has high toxic effect against the 2nd and 4th inster larvae compared with the tested biopesticides.

Also, the results in this work are in harmony with the obtained results by El-Khayat, *et al.* (2012) who evaluated the toxicity of some chemical and bio- pesticides against the cotton leafworm, *S. littoralis* under laboratory condition. They found that the tested chemical pesticides as Chlorpyrifos gave the highest significant toxic effect but the tested biopesticides as Dipel 2x (*Bacillus thuringiensis)* recorded the least significant toxic effect. The same results were obtained by Abd El-Latief, (2001) who tested various insecticides against eggs and larvae of *S. littoralis*. He mentioned that, Dipel 2x (*Bacillus thuringiensis*) had slight or low insecticidal activities against the 2nd and 4th instar larvae of *S. littoralis* compared with other tested compound. Likewise, the obtained data are in agreement with findings of Al-Shannaf, *et al.* (2006) when they used conventional insecticides (Indoxacarb and Methomyl) and biocides (Spinosad and Viroset) against *S. littoralis*.

On the other hand, results in this study indicated that the susceptibility of the predator *C. carnea* to the biopesticides is less than chemical pesticides. Where, based on LC_{50} values the toxicity effect of biopesticides was the lowest compared with the effect of chemical pesticides against the 2nd instar larvae of *C. carnea*. These results are in agreement with Croft, (1990) and Medina, *et al.* (2008). They mentioned that the insecticides Deltamethrin and Chlorpyrifos were not safe to the natural enemy *C. carnea* because they are broad-spectrum chemicals. Likewise, they found that organophosphate Chlorpyrifos was by far the most toxic product for L_3 *C. carnea*. The same strong lethal effect was reported by Cordeiro, *et al.* (2010) in other *Chrysoperla spp*. by Malathion pesticide to L_1 larvae. Also, other organophosphates exhibit a high toxic effect to *C. carnea* and *C. externa* larvae. In this study, Abamectin pesticide had median toxic effect between the tested chemical pesticides and biopesticides. In the other studies, different results were found. Hassan, *et al.* (1991) and Sattar, (2010) reported that Abamectin is a safe pesticide to *C. carnea*. However, Maia, *et al.* (2016) mentioned that Abamectin significantly reduce *C. carnea* population. Accordingly, the Abamectin was ranked as slightly harmful pesticide. Also, this results agreed with the obtained results of Giolo, *et al.* (2009) on the 1st instar larvae of *C. carnea* which was treated by Abamectin. Similarly, my results are harmony with the obtained results by Sabry and El-Sayed (2011), who tested some chemical and biochemical pesticides against 2nd instar larvae of *C. carnea*. They found that chlorpyrifos was more toxic than lambda-cyhalothrin, cypermethrin, spinosad and buprofezin. However, lambda-cyhalothrin was highly toxic to the adult of *C. carnea* compared to the other pesticides. In addition, Buprofezin and Spinosad were the least toxic to the 2nd instar larvae and adults of *C. carnea* respectively. Also, according to the percent of mortality, the pesticide toxicity was classified into harmful pesticide (chlorpyrifos), moderately harmful (lambda-cyhalothrin and cypermethrin), slightly harmful pesticides (Spinosad), and harmless pesticide (Buprofezin).

CONCLUSION

The results in this study confirmed that the tested biopesticides have a suitable toxic effect against the cotton leafworm, *S. littoralis* with a slightly toxic effect against the natural enemy, *C. carnea*. On the other hand, chemical pesticides have a highly toxic effect on both insects. Therefore, this study recommends using biopesticides besides the slight using of chemical pesticides to preserve the natural enemies and environment and achieve the best pest control.

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الفاعلية النسبية لبعض مبيدات الآفات ضد دودة ورق القطن وأثرها الجانبي على العدو الطبيعي أسد المن

رضا راضي حسن عبدالله*

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

تعتبر دودة ورق القطن من الحشرات الاقتصادية الخطيرة على المحاصيل المختلفة في مصر لأنها تسبب المزيد من الأضرار في المحاصيل وخاصة القطن والخضروات الورقية. من ناحية أخرى ، يعد العدو الطبيعي أسد المن من أهم المفترسات في جميع الحقول لأنه قادر على مكافحة العديد من الآفات الحشرية والعناكب. في هذه الدراسة ، تم اختبار ستة مبيدات حشرية تجارية (كلوروبيريفوس ، ألفاسيبرمثرين ، ميثوميل ، أبامكتين ، بيوسيانا و دايبل) ومركبين جديدين قيد الدراسة (المستخلص الخام من نواتج التمثيل الغذائي لفطر *Beauveria bassiana* ومركب اصطناعي من مشتقات السيانوأسيتاميد) لتحديد مدى سميتها ضد يرقات العمر الثاني لحشرة دودة ورق القطن وأثرها الجانبي على يرقات العمر الثاني للمفترس أسد المن. أوضحت النتائج أن جميع المركبات المختبرة لها درجات مختلفة من السمية ضد الحشرات المختبرة. وبناءً على قيم التركيز المميت النصفي ودليل السمية وكذلك الكفاءة النسبية كان المبيد ألفاسيبرمثرين أكثر المبيدات سمية ضد يرقات دودة ورق القطن ومفترس أسد المن يليه الميثوميل ثم الكلوربيريفوس ولكن أظهر مبيد البيوسيانا التأثير الأقل سمية بين المبيدات المختبرة. بالإضافة إلى ذلك ، كانت سمية المبيدات الكيماوية المختبرة ضد يرقات المفترس أسد المن أكثر من سميتها ضد يرقات دودة ورق القطن. بينما كانت سمية المبيدات الحيوية المختبرة ضد يرقات المفترس أسد المن أقل من سميتها ضد يرقات دودة ورق القطن. بشكل عام ، كانت حساسية المفترس أسد المن للمبيدات الحيوية المختبرة أقل مقارنة بالمبيدات الكيميائية. لذلك توصي هذه الدراسة باستخدام المبيدات الحيوية إلى جانب الاستخدام المحدود من المبيدات الكيماوية للحفاظ على الأعداء الطبيعية والبيئة مع تحقيق أفضل مكافحة للافات.