INTERACTIONS AMONG THE LARVAL ENDOPARASITOID *Microplitis rufiventris* KOK. (HYMENOPTERA; BRACONIDAE), ITS HOST *Spodoptera liltoralis* (BOISD.) (LEPIDOPTERA; NOCTUIDAE) AND SPINTOR 24 SC. Mahmoud, Basma. A. and Fawzia. A. Hassanien

Department Of Biological Control, Plant Protection Research Institute, Dokki, Giza, Egypt

ABSTRACT

Spintor is a new insecticide from natural resource, the active ingredient is spinosyns A and D, which are secondary metabolites from the aerobic fermentation of *Saccharopolyspora spinosa* on nutrient media.

Effect of Spintor treatment on the larval parasitoid *Microplitis rufiventris* Kok. was evaluated. Pre- and post-feeding of *S. littoralis* 2nd instar larvae on castor leaves treated with different concentrations of Spintor had adverse effect on the development of the parasitoid immature stages. Prolongation of the development periods (egg-larval duration and pupal stage) and decreasing of adult longevity were recorded when concentrations of 0.188 and 0.125ml/L water were tested. At concentrations of 0.188, 0.125, 0.0625 and 0.0313ml/L percentages of immerged parasitoids were decreased.

The combined effect of Spintor and parasitism led to an obvious increase in host mortality compared to the treatment with Spintor alone: 4-days Lc50 values for parasitized and unparasitized larvae were 0.194 and 0.239 ml/L respectively. **Keywords:** *Microplitis rufiventris, Spodoptera littoralis,* Spintor.

INTRODUCTION

Recent years have witnessed great efforts to substitute, completely or partially, chemical control of agricultural pests by biological control or at least integrate them to minimize the agricultural costs, reduce the pollution of environment and deplete healthy hazards accompanying synthetic insecticides applications.

Biological control includes effective utilization of natural insect enemies such as parasitoids and predators as well as specific types of entomopathogens and natural insecticides. Spintor is a new chemical class of insecticides that are registered by EPA. The active ingredient are the two active naturally occurring metabolites spinosyns A and D produced by a soil bacterium called *Saccharopolyspora spinosa*. It is used to control insect pests, including fruit flies, caterpillars, leafminers, thrips, sawflies and leaf beetles. Spinosad is recommended for use of an IPM program since it will not harm beneficial insects or predatory mites (Schoonover and Larson, 1995; Thompson *et al.*, 2000).

The braconid parasitoid *Microplitis rufiventris* Kok. is reported as an important solitary endoparasitoid of some serious noctuid larvae, including *Heliothis armigera* Hbn., *Spodoptera littoralis (Boisd.), S. latebrosa* and *S. exigua Hbn.* (El-Minshawy, 1963; Hammad *et al.*, 1965; Hafez *et al.*, 1976 and Hegazi, 1973).

Mahmoud, Basma. A. and Fawzia. A. Hassanien

The aim of this work is to study, the effect of Spintor on some biological aspects of *M. rufiventris* reared in *S. littoralis* larvae fed on caster lean leaves treated with Spintor at different concentrations. Response of parasitized *S. littoralis* larvae to Spintor was also tested.

MATERIALS AND METHODS

Rearing technique:

The cotton leafwarm *S. littoralis* ,as a laboratory strain, has been reared on castor bean leaves for several generations in controlled rearing room under constant conditions of 22±1°C and 65±2% RH in the Biological Control Department, Plant Protec. Res. Inst. A.R.C., Giza, Egypt.

Laboratory rearing of the braconid parasitoid, *M. rofiventris* was accomplished on *S. littoralis* 2nd instar larvae in another rearing room under the same conditions following the method described by Tawfik *et al.* (1980).

Insecticide: Spintor 24 sc.

It is a mixture of spinosyn A and spinosyn D, which produced during fermentation of a soil actenomycete that are registered by EPA as spinosad to control variety of insects and has a high activity towards Lepidoptera (Tompson and Hutchins, 1999). It is potentially potent compound for control of *S. littoralis* (Pineda *et al.*, 2004).

Spintor is a product of Dow AgroSciences recommended to control *S. littoralis* larvae at a rate of 50 ml/feddan.

Laboratory tests:

The following experiments were conducted to evaluate the effect of Spintor treatments on the larval parasitoid *M. rufiventris*.

- 1. In case of feeding of *S. littoralis* 2nd instar larvae on Spintor treated leaves on the parasitoid before parasitization (pre-feeding).
- 2. In case of feeding of *S. littoralis* 2nd instar larvae on Spintor treated leaves after parasitization (post-feeding).
- 3. Response of parasitized S. littoralis 2nd instar larvae to Spintor.

I. Effect of prefeeding of *S. littoralis* 2^{nd} instar larvae on Spintor treated leaves on the parasitoid:

S. littoralis 2nd instar larvae were fed for 48 h on castor bean leaves that were dipped in Spintor solution at concentrations of 0.188, 0.125 (the recommended rate), 0.0625 and 0.0313 ml Spintor/L water. 100 *S. littoralis* larvae in 5 replicates (20 larvae/replicate) were tested for each concentration. Larvae of each replicate were exposed individually to the mated females of the parasitoid in plastic vials (7x2cm) for 24h. The parasitized host larvae of each replicate were transferred to plastic jars 20x10cm and fed on clean castor bean leaves. In the control treatment 100 host larvae (2nd instar) were fed on clean castor bean leaves that were dipped in water and exposed to the parasitoid as mentioned before. Percentages of host mortality, parasitoid duration, percentage of adult emergence and adults longevity were recorded.

II. Effect of feeding *S. littoralis* 2nd instar larvae on Spintor treated leaves after parasitization:

S. littoralis 2nd instar larvae were exposed to mated females of the parasitoid, as mentioned before, for 24 h. Immediately after removal of the parasitoid, larvae were transferred to jars containing castor leaves treated with Spintor at concentrations of 0.188, 0.125, 0.0625 and 0.0313 ml Spintor/L water for 48h , then fed on clean castor bean leaves. 100 host larvae in 5 replicates (20 larvae/replicate) were used for each concentration. Larvae of each replicate were confined in plastic jars 20x10 cm covered with a piece of moslin. Larvae of control treatments (100 larvae) were exposed to the parasitoid and fed on castor bean leaves dipped in water.

Percentages of host mortality, duration of the parasitoid (egg-larval stage and pupal stage). Percent of adult emergence and longevity of adults were recorded.

III. Response of the parasitized larvae to Spintor:

S. littoralis 2^{nd} instar larvae were exposed to mated females of the parasitoid as mentioned before, then fed on castor bean leaves treated with spintor at concentrations of 0.188, 0.125, 0.0625 and 0.0313 ml/L water. 100 larvae in 5 replicates were used for each concentration. 100 unparasitized *S. littoralis* 2^{nd} instar larvae in 5 replicates were fed on castor leaves treated with the formerly mentioned concentrations.

Percentages of larval mortality in each case were recorded. Lc50 and slope values for parasitized and unparasitized *S. littoralis* larvae were calculated.

RESULTS AND DISCUSSION

I. Effect of prefeeding of *S. littoralis* 2nd instar larvae on Spintor treated leaves on the parasitoid *M. rufiventris*:

Data presented in Table (1) indicated that, when *S. littoralis* larvae were fed on Spintor treated castor bean leaves at concentrations of 0.188, 0.125, 0.0625 and 0.0313 ml/L water for 48h before parasitization, percentages of emerged adults of the parasittoids were 8, 16, 28 and 38%, respectively compared to 80% in the untreated control. These results indicated the adverse effect of Spintor on the development of the parasitoid immature stages.

Data in Table (1) show also that the egg-larval duration of the parasitoid was not affected at concentrations of 0.0625 and 0.0312 ml Spintor /L water, however, the higher concentrations (0.125 and 0.188 ml/L) prolonged the duration in comparison with untreated control. Also, a reduction in longevity of the parasitoid adults was observed.

In case of prefeeding host larvae on Spintor treated leaves at concentrations of 0.188, 0.125 and 0.0625 ml/L, adults longevity reached 2.3, 2.9 and 4.2 day, respectively compared to 6.2 day in untreated control.

Mahmoud, Basma. A. and Fawzia. A. Hassanien

	Parasitism %	% of emerged	Duration in days		Longevity Of
Conc. ml/L	(No. of cocoons %)	•	Egg-larval	Pupal	Adults
		parasitolus	stage	stage	Addits
0.188	11	8	15.2	7.6	2.3
0.125	20	16	14.4	7.4	2.9
0.0625	32	28	12.4	6.7	4.2
0.0313	40	38	12.0	6.2	6.1
Untreated	83	80	12.0	6.0	6.2

Table (1): Effect of prefeeding of *S. littoralis* 2nd instar larvae on Spintor treated leaves on the parasitoid *M. rufiventris*.

II. Effect of feeding of *S. littoralis* 2nd instar larvae on Spintor treated leaves after parasitization:

From data shown in Table (2) it is clear that spintor treatments affected negatively the percentage of emerged adult parasitoids, it reached 15, 30, 39 and 45% when spintor was used at concentrations of 0.188, 0.125, 0.0625 and 0.0133 ml/L, respectively. While, in the control treatment it reached 78%. Data indicated also that treatment of parasitized host larvae with high concentrations of Spintor (0.188 and 0.125 ml/L) had various effects on the parasitoid, including prolongation of the development periods (egg-larval duration and pupal stage) and decreasing the adults longevity.

Duration of egg-larval stage reached 15.4 and 14.2 days when using concentrations of 0.188 and 0.125 ml/L were tested, respectively, at the same concentrations the duration of pupal stage reached 7.8 and 7.2 days.while longevity of adults decreased to 2.6 and 3.2 days respectively.

From data shown in Table (1) and (2), it is obvious that the females of *M. rufiventris* did not discriminate between healthy and spintor treated host larvae, they laid their eggs in both. These results are in agreement with those mentioned by Levin *et al.* (1983) who found that *Apanteles glomeratus* distinguished between parasitized and nonparasitized healthy *Pieris rapae* but did not discriminate between healthy and G.V. treated larvae, ovipositing even in G.V. killed or moribund larvae.

It can be concluded also that prefeeding and postfeeding of the host larvae on Spintor treated leaves was harmful for the parasitoid, since the percentages of immerged parasitoids were decreased in both treatments in comparison with control treatment.

	Parasitism %	% of emerged.	Duration	in days	Longevity Of
Conc. MI/L	(No. of cocoons %)	Ŭ	Egg-larval	Pupal	Adults
		parasitolus	stage	stage	Addits
0.188	18	15	15.4	7.8	2.6
0.125	34	30	14.8	7.2	3.2
0.0625	43	39	12.6	6.4	4.7
0.0313	48	45	12.2	6.2	6.2
Untreated	80	78	12.0	6.2	6.1

Table (2): Effect of post-feeding of *S. littoralis* 2nd instar larvae on Spintor treated leaves on the parasitoid *M. rufiventris*.

Spintor treatments prevent successful development and pupation of *M. rufiventris*, mainly because of the early mortality of the host. These are in agreement with the findings of El-Magharby *et al.* (1988) who stated that treatment of parasitized host larvae with microbial pesticides had determintal effects on the parasitoid, including prolongation of the developmental period abnormality of the cocoons within the host and reduced the percentage of adult parasitoid emergence. On the other hand, Blumberg *et al.* (1997) stated that prefeeding *Heliothis armigera* with lethal concentrations of Dipel 2 x (*B.t.* Kurstaki) did not prevent *Microplitis croceipes* from ovipositing in the treated host larvae. They added that development of parasitoid immatures in host larvae prefed for 24 or 48h with the dietary *B. thuringiensis* was not adversely affected.

III. Response of parasitized S. littoralis 2nd instar larvae to Spintor:

Data illustrated in Table (3) indicated that the combined effect of spintor and parasitism led to an obvious increase in mortality compared to treatment with spintor alone, when concentrations of 0.188, 0.125, 0.0625 and 0.0313 ml/L were tested, percentages of mortality reached 90, 84, 78 and 72%, as compared to 68, 45, 30 and 20% in unparasitized host larvae. These results are similar to the findings of Ahmed *et al.* (1978) who recorded higher mortality percentages in *Lymantrai dispar* (L.) larvae parasitized by *Apanteles melanoscelus* Ratz. and treated with *B. thuringiensis* than when host larvae were treated with the parasitoid or the bacterium alone. Also, similar results were reported by Salama *et al.* (1996); Atwood *et al.*(1997) and Amy and Kenneth (1998).

Salgado (1997) reported that spinosad activity is characterized by cessation of feeding and paralysis of exposed insects. He added that growers and scouts should wait a minimum of two to three days to evaluate the control. Therefore, 4-days Lc_{50} and slope values for parasitized and unparasitized *S. littoralis* larvae were statistically calculated. Probit analysis indicated that 4-days Lc_{50} values for parasitized and unparasitized larvae were 0.194 and 0.239 ml/L, respectively, while slope values were 1.532 and 1.646 (Table 3).

Mahmoud, Basma. A. and Fawzia. A. Hassanien

Concentration	Total mortality% of	Total mortality% of		
Concentration	unparasitized larvae	parasitized larvae		
0.188	68	90		
0.125	45	84		
0.0625	30	78		
0.0313	20	72		
4-days Lc ₅₀	0.239	0.194		
Slope	1.646	1.532		

 Table (3): percentage of mortality in parasitized and unparasitized

 S. littoralis 2nd instar larvae treated with Spintor.

REFERENCES

- Ahmed, S.; J.R. O'Neill; D.L. Mague and R.K. Nowalk (1978). Toxicity of *Bacillus thuringiensis* to gypsy moth larvae parasitized by *Apanteles melanoscalus*. Environ. Ent., 7: 73-76.
- Amy, B.C. and F.R. Kenneth (1998). Effects of parasitoid strain and host instar on the interaction of *Bacillus thuringiensis* subsp. *Kurstaki* with the gypsy moth (Lepidoptera: Lymantriidae) larval parasitoid *Cotesia melanoscela* (Hymenoptera: Braconidae). Environ. Ent., 27(1): 137-147.
- Atwood, D.W.; S.Y. Young III and T.J. Kring (1997). Development of *Cotesia marginiventris* (Hymenoptera: Braconidae) in tobacco budworm (Lepidoptera: Noctuidae) larvae treated with *Bacillus thuringiensis* and Thiodicarb. J. Econ. Ent., 90(3): 751-756.
- Blumberg, D.; A. Novon; S. Keren; S. Goldenberg and S.M. Ferkovich (1997). Interactions among *Helicoverpa armigera* (Lepidoptera: Noctuidae), its larval endoparasitoid *Microplitis croceipes* (Hymenoptera: Braconidae) and *Bacillus thuringiensis*. J. Econ. Ent., 90(5): 1181-1186.
- El-Maghrabi, M.M.A.; Hegab, A.; Yousif Khalil, S.I. (1988). Interactions between *Bacillus thurengiensis* Berl., *Beauvaria bassiana* (Bals.) Vull., and the host/parasitoid system *Spodoptera littoralis* (Boisd.)/*Microplitis rufiventris* Kok. J. Appl. Ent., 106: 417-421.
- El-Minshawy, A.M. (1963). Studies on the morphology and biology of Spodoptera exigua Hb. and Spodoptera lateborsa Led. in Alexandria. M.Sc. Thesis, Faculty of Agric. Univ. of Alex.
- Hfez,Mostafa;M.F.S.Tawfik;A.K.Azab and Amira A. Ibrahim (1976). Survey and economic importance of parasites of the cotton leafworm, *Spodoptera littoralis* (Biosd.) in Egypt. Bull. Soc. Ent., Egypt, 60: 179-189.

- Hammad, S.M.; A.M. El-Minshawy and A. Salama (1965). Studies on *Microplitis rufiventris* Kok. (Hym.; Braconidae) Bull. Soc. Ent., Egypt, 49: 215-219.
- Hegazi, E.M. (1973). Studies on certain natural enemies attacking the immature stage of certain Lepidopterous pests. Parasities of egg-larval and larval stage of the cotton leafworm *Spodoptera littoralis* (Boisd.) M.Sc. Thesis, Fac. of Agric. Univ. of Alexandria, Alex., Egypt.
- Levin, D.B.; J.E. Laing; R.P. Jaques and J.E. Corrigan (1983). Transmission of the granulosis virus of *Pieris rapae* (Lepidoptera: Pieredae) by the parasitoid *Apanteles glomeratus* (Hymenopetra: Braconidae). Environ. Entomol., 12: 166-170.
- Pineda, S.; F. Budia; M.I. Schneider; A. Gobbi; E. Vinuela; J. Valle and P. DelEstal (2004). Effects of two biorational insecticides, spinosad and Methoxyfenozide, on *Spodoptera littoralis* (Lepidoptera: Noctuidae) under laboratory conditions. J. Econ. Entomol., 97(6): 1906-1911.
- Salama, H.S.; F.N. Zaki and M.M. Sabbour (1996). Effect of *Bacillus thuringiensis* endotoxin on *Apanteles litae* Nixon and *Bracon instabilis* Marsh. (Hym: Braconidae), two parasitoids of the potato tuber moth *Phthorimia operculella* Zelle (Lep., Geleshiidae). J. Appl. Ent., 120: 565-568.
- Salgado, V.L. (1997). The mode of action of spinosad and other insect control products. Down to earth, 52(1), 35-44.
- Schoonover and Larson (1995). Laboratory activity of spinosad on nontarget beneficial arthropods. Arthorod Management Tests, 20: 357.
- Tawfik, M.F.S.; Mostafa Hafez and A.A. Ibrahim (1980). Immature stages of *Microplitis rufiventris* Kok. (Hym., Braconidae). Dtsch. Ent. Z., 27, 1-3: 39-50.
- Thompson, G.D.; R. Dutton and T.C. Sparks (2000). Spinosad- a case study: on example from a natural products discovery program. Pest Management Science, 56: 696-702.
- Thompson, G.; S. Hutchins (1999). Spinosad- a new class of fermentationderrived insect control agents. Pesticide Outlook, 10(2): 78-81.

سبنتور، مبيد حشري جديد ناتج عن مصدر طبيعي حيث أن المادة الفعالة به هي سيتوزين أ ، د وهما من النواتج الثانوية التي تتكون عند الإكثار الهوائي للميكروب Saccharopolyspora على البيئات المعذية.

درس تأثير معاملة دودة ورق القطن بهذا المبيد على الطفيل ميكروبليتس روفي فنترس (طفيل يرقة) وذلك في حالتي:

- اً. تغذيبة يرقبات العمر الثباني ليدودة ورق القطبن على أوراق خبروع معاملية بالمبييد بتركيزات مختلفة ثم تعريضها للطفيل.
- ب. تعريض يرقات العمر الثاني لدودة ورق القطن للطفيل ثم تغذيتها على أوراق خروع معاملة بالمبيد بالتركيزات المختلفة.
- فى كلا الحالتين وجد أن للمبيد تأثير سلبى على الأطوار غير الكاملة للطفيل فعند استخدام التركيزين ١٨٨، ١٢٥، مللتر/لتر ماء وجد أن فترة نمو الطفيل (من البيضة حتى العمر اليرقى الأخير) تكون أطول من مثيلاتها فى حالة اليرقات الغير معاملة بالمبيد، كما أن فترة طور العذراء تكون أطول بينما تقل فترة حياة الحشرة الكاملة.
- معاملة اليرقات بالتركيزات ٠,١٨٨ ، ٠,١٢٥ ، ٠,٠٦٢ ، ٠,٠٣١٣ ، مللتر/لتر أدت إلى نقص واضح في نسبة خروج الحشرات الكاملة للطفيل.
- اتضح من الدراسةً أن التأثير المشتَرك للطفيل والمبيد أدى إلى زيادة واضحة في النسبة المئوية لموت الآفة بالمقارنة إلى استخدام المبيد منفرد.
- تم حساب قيم التركيز القاتل لـ ٥٠% من يرقات الأفة فوجد أنها ٠,١٩٤ ، ٢٣٩. مللتر/لتر ماء في حالة اليرقات المتطفل عليها واليرقات الغير متطفل عليها على التوالي.