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INSECTICIDE RESISTANCE OF TOMATO BORER *Tuta absoluta* (MEYRICK) UNDER EGYPTIAN CONDTONS

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ABSTRACT

The efficacy of five insecticides [organophosphate (methamidophos), carbamate (methomyl), pyrethroid (Deltamethrin), spinosad (spintor) and imidaclopride (Confidor)] were determined against four strains of *Tuta absoluta* collected from four governorates in Egypt [Damytta (DAM), Marsa Matrouh (MAR), Behera (BEH) and Kafer El-Sheikh (KAF)]. Collected strains were bioassayed and compared with a reference susceptible strain. Insecticides were dissolved in acetone and topically applied to the mid-dorsal abdominal region of two-day old 4th instar larvae. LD50 values were estimated and the Resistance Factor (RF) for each insecticide was calculated (RF = LD50 value of each field strain / LD50 value of the susceptible strain). DAM and MAR strains recorded 29.72 and 10.62-fold resistance, respectively to methamidophos. DAM and MAR strains recorded 32.53 and 10.76-fold resistance, respectively to methomyl. DAM and MAR strains demonstrating 70.81 and 28.09-fold resistance, respectively to deltamethrin. DAM and MAR strains recorded 38.04 and 17.08-fold resistance, respectively to spinosad, while KAF and BEH strains demonstrated 14.92 and 12.14-fold resistance, respectively to confidor. These results are discussed in relation to the possible mechanisms of resistance present in the studied *T. absoluta* strains and underpin the resistance management strategy for the tomato borer in Egypt.

INTRODUCTION

The tomato borer, *Tuta absoluta* Meyrick, (Lepidoptera : Gelechiidae) is a serious pest of both outdoor and greenhouse tomatoes. Originated from South America, *T. absoluta* was reported since the early 1980s from Argentina, Brazil and Bolivia (Estay, 2000); the insect rapidly invaded many European and Mediterranean countries. It was first recorded from eastern Spain in late 2006 (Urbaneja *et al*., 2007), then Morocco, Algeria, France, Greece, Malta, Egypt and other countries (Mohammed, 2010; Roditakis *et al*., 2010). After hatching, young larvae produce large galleries in leaves, burrow into stalks, apical buds, and green and ripe fruits, causing considerable damage and yield losses. If no control measures are taken, then the pest can cause up to 80–100% yield losses in tomato crops in recently invaded areas. Because of the short generation time and the frequent applications of insecticide to manage *T. absoluta* resistance to several insecticides has developed. In 1999 significant resistance of *T. absoluta* to acephate and deltamethrin was reported (Castelo Branco *et al.* 2001). In the same year resistance to deltamethrin, lamba=cyhalothrin, mevinphos, metamidophos and esphenvalerate was reported in Chile (Salazar and Araya 2001). In 2000 resistance to Cartap was reported in Brazil (Siqueira *et al.* 2000,). In 2001 resistance to abamectin was additionally reported in Brazil (Siqueira *et al.* 2001). In 2005 an Argentine study confirmed *T. absoluta* resistance in that country to deltamethrin and abamectin as well as methamidophos (Liettii *et al.* 2005). It has been hypothesized that excessive insecticide applications commonly applied to the tomato crop during a single cultivation period, up to 36 sprays (sometimes tow sprays per day), could have led to the evolution of resistant populations, besides eliminating their natural enemies, and leading to additional occupational hazards (Gonçalves *et al*. 1994; Picanço *et al*. 1995). Thus, in order to avoid selection of resistant biotypes, a careful management with frequent changes of active ingredients is desirable.

The objective of the present study was to detect the existence of Egyptian populations of *T. absoluta* resistant to the main insecticides groups used against it and to quantify that resistance and its relationship with insecticide use in Egypt.

MATERIALS AND METHODS

Insect strains

The laboratory standard strain (the susceptible reference strain; SUS) insects were obtained from Plant Protection Research Institute, Dokki, Giza, Egypt, where it had been maintained in the absence of insecticides since April 2010. Four field strains were collected during 2012, MAR from Marsa Matrouh governorate, DAM from Damytta governorate, KAF from Kafer El-Sheikh governorate and BEH from Behera governorate. The individuals of populations were reared on tomato plants (*Lycopersicon esculentum* Mill. Salonaceae), without insecticide exposure, enclosed in cages are made of wood and covered by mesh muslin and maintained under laboratory conditions at a temperature and day length varying from 25 to 28C and from 12 to 14 h respectively, during the study period. The colonies were maintained for two generations in the laboratory before starting the bioassays.

Insecticides

Formulated of the five tested insecticides used for bioassays were; organophosphate Methamidophos (Tamaron 40%), carbamate Methomyl (Lannate 90% SP), pyrethroid Decis (Deltamethrin, 25 g/l-EC), Spinosad (Spintor 24% SC) and Imidacloprid (Confidor, 200 g/l-SL). Acetone was used as solvent.

Bioassay method

The experimental work was done on two-day old 4th larval stage. Insecticides were topically applied to the mid-dorsal abdominal region of the larvae using a micro syringe provided with a dispenser. Each insect received 0.2 µl of a solution of insecticide in acetone. Control groups were topically treated with acetone alone. 10 larvae for each dose were used to estimate the lethal dose 50% (LD50) values. Each bioassay used three replicates of five concentrations. After treatment, the larvae were individually placed in 3 cm^3 plastic vials (13 x 35 mm) and incubated at 28 ± 2°C. Mortality was recorded 24h after treatment under stereoscopic microscope. Insects were counted as dead if they were unable to walk.

Analysis of bioassays

Dose-response bioassay against standard strain and field strains were conducted using three batches of 10 insects at a minimum of five insecticide concentrations per bioassay. Each assay was repeated at least three times and results pooled for analysis, probit analysis of the concentration dependent mortality data were carried out using the software program POLO-PC (Anon., 1987). Resistance factors (RFs) were calculated by dividing the LD50 of the resistant strain by the LD50 of the susceptible strain.

RESULTS

Bioassay, methamidophos

Data in table (1) showed that DAM strain was relatively, the highest resistant to methamidophos (29.72 fold) while MAR, BEH and KAF strains were the least resistance to methamidophos (10.62, 11.45 and 13.55 fold, respictively).

Table(1): Comparative responses of *Tuta absoluta* strains tested against methamidophos.

strains	N.	LD50 (µg a.i./larvae)	95% CLs	slope	RFs
SUS	150	2.9	2.21-3.68	1.8	-
MAR	150	30.8	19.39-57.25	0.7	10.62
BEH	150	33.2	26.70-43.17	1.0	11.45
KAF	150	39.3	30.89-49.96	2.2	13.55
DAM	150	86.2	34.68-792.1	0.8	29.72

Bioassay, methomyl

There was significant variation in methomyl susceptibility among the insect strains studied. Data in table (2) indicate that DAM strain was relatively, the highest resistant to methomyl (32.53 fold) while BEH and KAF strains displayed moderate resistance to methomyl (28.86 and 24.15 fold, respictively) but MAR strain was the least resistance to methomyl (10.76 fold).

Table(2): Comparative responses of *Tuta absoluta* strains tested against methomyl.

strains	N.	LD50 (µg a.i./larvae)	95% CLs	slope	RFs
SUS	150	4.58	3.27-6.37	0.9	-
MAR	150	49.3	35.55-70.75	1.8	10.76
BEH	150	132.2	83.5-314.1	1.2	28.86
KAF	150	110.6	64.25-258.9	0.8	24.15
DAM	150	149	109.7-219.4	1.1	32.53

Bioassay, deltamethrin

BEH and KAF strains exhibited similarly resistance to deltamethrin (39.18 and 38.54 fold resistance, respectively). As for DAM strain, resistance factor was generally greater for deltamethrin (70.81 fold resistance). While MAR strain was the least resistance to deltamethrin (28.09 fold). (table 3).

Table(3): Comparative responses of *Tuta absoluta* strains tested against deltamethrin.

strains	N.	LD50 (µg a.i./larvae)	95% CLs	slope	RFs
SUS	150	1.1	0.61-1.54	1.5	-
MAR	150	30.9	24.60-40.18	1.5	28.09
BEH	150	43.1	33.0-60.23	1.5	39.18
KAF	150	42.4	18.1-91.8	0.8	38.54
DAM	150	77.9	53.9-121.8	0.9	70.81

Bioassay, spinosad

Data in table (4) indicate that resistance to spinosad was observed in all of the strains studied in comparison with the susceptible standard strain. MAR and KAF strains exhibited 17.08 to 18.33 fold resistance to spinosad, but BEH and DAM strains were more resistance to spinosad (31.96 to 38.04 fold).

Table(4): Comparative responses of *Tuta absoluta* strains tested against spinosad.

strains	N.	LD50 (µg a.i./larvae)	95% CLs	slope	RFs
SUS	150	0.24	0.179-0.321	1.6	-
MAR	150	4.1	2.76-6.14	1.3	17.08
BEH	150	7.67	6.252-8.946	1.4	31.96
KAF	150	4.4	3.28-5.70	1.9	18.33
DAM	150	9.13	7.076-11.97	0.9	38.04

Bioassay, confidor

Data in table (5) showed that all insect populations were slight resistance to confidor comparison with the susceptible standard strain. MAR, BEH, KAF and DAM strains exhibited 13.57, 12.14, 14.92 to 13.57 fold resistance, respectively to confidor.

Table(5): Comparative responses of *Tuta absoluta* strains tested against confidor.

strains	N.	LD50 (µg a.i./larvae)	95% CLs	slope	RFs
SUS	150	0.14	0.095-0.209	1.4	-
MAR	150	1.9	1.43-2.53	2.0	13.57
BEH	150	1.7	1.29-2.19	2.4	12.14
KAF	150	2.09	1.514-2.894	1.2	14.92
DAM	150	1.9	1.36-2.68	1.9	13.57

DISCUSSION

The results of current study revealed that all strains showed varied degrees of resistance to the five insecticides studied. The highest resistance was recorded at 70.81 fold resistance in DAM strain for deltamethrin, in contrast, the lowest resistant strain was MAR strain for methomyl (10.76 fold).

The variability of response to these insecticides among populations of the tomato borer, which showed different levels of resistance, is probably due to differences in the pattern of insecticides use at the different sites where the populations were collected.

Pesticide bioassays are useful for detecting the trends in resistance to insecticides. The different insecticide resistance levels suggest different selection pressure among populations, genetic diversity in the resistance mechanisms among strains, or both (Kerns and Gaylor, 1992). Among the known insecticide-resistance mechanisms, the biochemical ones (i.e. enhanced activity of detoxification enzymes and target site insensitivity) are frequently reported to be the most important (Brattsten *et al*., 1986; Mullin and Scott, 1992). Insect detoxification enzymes are important resistant mechanisms and insecticide synergists are very helpful in providing preliminary evidence of their involvement as resistance mechanisms (Brindley and Selim, 1984; Scott, 1990; Bernard and Philogea, 1993; Ishaaya, 1993). The persistence of an insecticide on a plant leads to the continuous selection of resistant individuals, which may contribute to a faster resistance evolution (Roush, 1989). Carbamates, organophosphates and pyrethroids are widely used in the last tow years to control *T. absoluta* in Egypt, tomato growers found that, in order to combat *T. absoluta*, one to two insecticide applications per day had to be applied so it was expected that some resistance would be present. Ten-fold greater than the susceptible strain, it has been suggested that insects should not be considered resistant until a resistance ratio of 10 is exhibited (Ahmad *et al*., 2008).

Resistance to spinosad and indoxacarb has been shown to be esterases mediated in *P. xylostella* and *Helicoverpa armigera* (Sayyed and Wright, 2006; Wang *et al*., 2009). Spinosad, a mixture of spinosyns A and D, is derived from the naturally occurring actionomycete, *Saccharopolyspora spinosa* (Sparks *et al.,* 1998). Because of its unique mode of action, involving the postsynaptic nicotinic acetylcholine and Gamma-aminobutyric (GABA) receptors, spinosad has strong insecticidal activity against insects (Salgado, 1998) especially Lepidoptera, *Spodoptera frugiperda* (Méndez *et al*., 2002), (e.g. *Helicoverpa armigera* (Wang *et a*l., 2009), Diptera (King and Hennesey 1996; Collier and Vanstynwyk, 2003; Bond *et al*., 2004), some Coleoptera (Elliott *et al.,* 2007) as well as stored grains (Hertlein *et al*., 2011). To date, spinosad is considered a good alternative control of Lepidopteran pests due to its high activity at low rates and its use in integrated pest management programs. The product possesses advantages in term of safety for farm workers and consumers due to its low mammalian toxicity and rapid breakdown in the environment (Sparks *et al.,* 1998).

Strains were slightly resistant to imidacloprid, presently, approximately 80% of imidacloprid applications in Egypt are foliar. It is possible that this is the application method most likely to decrease selection pressure for resistance in insect pests. This is because soil applied or seed treatments tend to persist to the extent that they may leave the population exposed to sub lethal doses over long periods. Imidacloprid is not yet strongly resisted in combination with the remaining efficacy that appears to exist for some carbamates, organophosphates and pyrethroids, it ought to be possible to institute simple alternation strategies that would go some way to solving the tomato borer problem whilst conserving insecticide susceptibility.

Resistance management should be a component of integrated pest management, which seeks to minimize pesticide usage through the application of alternative tactics such as cultural control and conservation of natural control through selective insecticides. Monitoring the susceptibility of different populations exposed to distinct active ingredients is essential.

REFERENCES

Ahmad, M., A.H. Sayyed and M.A. Saleem (2008): Evidence for field evolved resistance to newer insecticides in *Spodoptera litura* (Lepidoptera : Noctuidae) from Pakistan. Crop Prot., 27: 1367-1372.

Anon., (1987): POLO-PC-a user's guide to Probit or Logit analysis. 22pp. LeOra Software, California.

Bernard, C. B. and Philogea N.B.J.R. (1993): Insecticide synergists: role, importance, and perspectives. *Journal of Toxicology and Environmental Health,* **38**, 199 ± 223.

Bond J.G., Marina C.F., and Williams T. (2004): The naturally derived insecticide Spinosad is highly toxic to *Aedes* and *Anopheles* mosquito larvae. Medical and Veterinary Entomology 18: 50-56.

Brattsten, L. B.; Holyoke, L. W. Jr; Leeper, J. R. and K. F. Raffia. (1986): Insecticide resistance: challenge to pest management and basic research. *Science.* 231: 1255–1260.

Brindley Y. W. A. and Selim A. A. (1984): Synergism and antagonism in the analysis of insecticide resistance. *Environmental Entomology*, **13**, 348 ± 353.

Castelo Branco M., F.H. França, M.A. Medeiros and J.G.T. Leal (2001): Uso de inseticidas para o controle da traça-do-tomateiro e traça-das-crucíferas: um estudo de caso. Horticultura Brasileira 9: 60-63.

Collier T.R. and Vanstynwyk R. (2003): Olive fruitfly in California: prospects for integrated control. California Agriculture, 57: 28-32.

Elliott R.H., Benjamin M.C. and Gillott G. (2007): Laboratory studies of the toxicity of spinosad and deltamethrin to *Phyllotreta cruciferae* (Coleoptera : Chrysomelidae). Can. Entomol. 139: 534-544.

Estay P. (2000): Polilla del tomate *Tuta absoluta* (Meyrick). http://alerce.inia.cl/docs/informativos/informativo09. pdf. Accessed, 12 May 2010.

Gonçalves D.M.H.R., Picanço M.C., Ribeiro L.J. and Campos L.O. (1994): Seletividade de quatro inseticidas a *Polybia* sp.2 (Hymenoptera: Vespidae) predador de *Scrobipalpuloides absoluta* (Lepidoptera: Gelechiidae). *Horticultura Brasileira*, 21, 81.

Hertlein M.B, Thompson G.D., Subramanyam B. and Athanassiou C.G. (2011): Spinosad : A new natural product for stored grain protection. Journal of stored products research. Doi: 10.1016/j.jspr.2011.01.004 : 1-15.

Ishaaya I. (1993): Insect detoxifying enzymes. Their importance in pesticide synergism and resistance. *Arch. Insect Biochem. Physiol.*, 22: 263–276.

Kerns D.L. and Gaylor M.J. (1992): Insecticide resistance in field populations of the cotton aphid (Homoptera: aphididae). *Journal of Economic Entomology*, 85: 1-8.

King J.R. and Hennesey M.K. (1996): Spinosad bait for the Caribbinean fruitfly (Diptera: Tephritidae). Florida Entomologist. 79(4):526- 531.

Lietti M.M., Botto E. and Alzogaray R.A. (2005): Insecticide Resistance in Argentine Populations of *Tuta absoluta* (Lepidoptera: Gelechiidae). Neotropical Entomology. 34(1): 113-119.

Méndez W.A., Valle J., Ibarra J.E., Cisneros J., Penagos D.I. and Williams T. (2002): Spinosad and nucleopolyhedrovirus mixtures for control of *Spodoptera frugiperda* (Lepidoptera: Noctuidae) in maize. Biological control 25:195-206.

Mohammed A.S. (2010): New record for leafminer, *Tuta absoluta* (Lepidoptera: Gelechiidae) infested tomato plantations in Kafer El-Sheikh region. J. Agric. Res. Kafer El-Sheikh. Uni. 36(2): 238-239.

Mullin C. A. and Scott J. G. (1992): Biomolecular bases for insecticide resistance: classification and comparisons. In: *Molecular Mechanisms of Insecticide Resistance.* Ed. by Mullin, C. A.; Scott, J. G. New York: ACS, 1–13.

Picance O. M. C., Guedes R. N. C., Leite G. L. D., Fontes P. C. R. and SILVA E. A. (1995): IncideÃncia de *Scrobipalpuloides absoluta* (Meyrick) (Lepidoptera: Gelechiidae) em tomateiro sob diferentes sistemas de tutoramento e controle quõÂmico de pragas. *Horticultura Brasileira,* **13**, 180 ± 183.

Roditakis E., Papachristos D. and Roditakis N.E. (2010): Current status of the tomato leafminer *Tuta absoluta* in Greece. OEPP/EPPO Bulletin. 40: 163-166.

Roush R.T. (1989): Designing resistance management programs: how can you choose? *Pesticide Science*, 26:423 441.

Salazar E.R. and J.E. Araya (2001): Respuesta de la polilla del tomate, *Tuta absoluta* (Meyrick), a insecticidas en Arica. Agric. Téc. 61: 429-435.

Salgado V.L. (1998): Studies on the mode of action of Spinosad: insect symptoms and physiological correlates. Pesticides Biochemistry and Physiology. 60: 91-102.

Sayyed A.H. and D.J. Wright (2006): Genetics and evidence for an esterase-associated mechanism of resistance to indoxacarb in a field population of diamondback moth (Lepidoptera : Plutellidae) Pest Manag. Sci., 62: 1045-1051.

Scott J. G. (1990): Investigating mechanisms of insecticide resistance: methods, strategies, and pitfalls. In*: Pesticide Resistance in Arthropods*. Ed. by Roush, R. T.; Tabashnik, B. E. New York and London: Chapman & Hall, 39–57.

Siqueira H.A., Guedes R.N., Fragoso D.B. and Magalhães L.C. (2001): Abamectin resistance and synergism in brazilian populations of *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae). Int. J. Pest Manag. 47: 247- 251.

Siqueira H.A., Guedes R.N. and Picanço M.C. (2000): Insecticide resistance in populations of *Tuta absoluta* (Lepidoptera:Gelechiidae). Agric. Forest Entomol. 2: 147-153**.**

Sparks T.C., Thompson G.D., Kirst H.A., Hertlein M.B., Larson L.L. Worden T.N. and Thibault M.B. (1998): Biological activity of Spinosyns, new fermentain derived insect control agents on tobacco budworm (Lepidoptera: Noctuidae) larvae. J. Econ. Ent. 91: 1277-1283.

Urbaneja A., Vercher R., Navarro V., Garcia-Mari F. and Porcuna J.L. (2007): La polilla del tomate *Tuta absoluta*. Phytoma España. 194:16-23.

Wang D., Cong P.Y., Li M., Qui X.H. and Wang K.Y. (2009): Sublethal effects of Spinosad on survival, growth and reproduction of *Helicoverpa armigera* (Lepidoptera : Noctuidae). Pest. Man. Sci. 65: 223-227.

للمبيدات الحشرية *Tuta absoluta* (Meyrick) مقاومة نافقة الطماطم تحت الظروف المصرية

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أجريت هذه الدراسة بغرض تقييم حساسية بعض سلالات حشرة نافقة الطماطم التي تم جمعها من محافظات مختلفة لمجموعة من المبيدات تمثل أهم المجموعات الكيميائية المعروفة بغرض المساهمة في تطوير برامج المكافحة المتكاملة لحشرة نافقة الطماطم في مصر.

وقد أجريت تجارب التقدير الحيوي علي السلالات الآتية:-

سلالة SUS وهي سلالة حساسة تمت تربيتها في معامل معهد بحوث وقاية النباتات بمركز البحوث الزراعية بدون استخدام أي نوع من المبيدات عليها منذ عام 2010. سلالة DAM وهي سلالة جمعت من محافظة دمياط من علي نبات الطماطم في عام 2012. سلالة MAR وهي سلالة جمعت من محافظة مرسي مطروح من علي نبات الطماطم في عام 2012. سلالة BEH وهي سلالة جمعت من محافظة البحيرة من علي نبات الطماطم في عام 2012. سلالة KAF وهي سلالة جمعت من محافظة كفر الشيخ من علي نبات الطماطم في عام 2012.

وقد تم إجراء تجارب التقدير الحيوي علي سلالات نافقة الطماطم باستخدام المبيدات التالية:-

1- من مجموعة الفسفور العضوية مبيد التمارون Methamidophos .
2- من مجموعة البروثرويدات مبيد ديسيس Deltamethrin .
3- من مجموعة الكرباميت مبيد اللانيت Methomyl .
4- من مجموعة النيكوتينويد مبيد Imidaclopride.

5- المستخلص الميكروبي Spinosad.

أوضحت النتائج أن استجابة سلالات نافقة الطماطم لمبيد التمارون كانت متباينة حيث كان معامل المقاومة (RFs) 29.72 لسلالة DAM وكان 10.62 لسلالة MAR بينما كان معامل المقاومة لمبيد ديسيس 32.53 لسلالة DAM وكان 10.76 لسلالة MAR. كما أظهرت النتائج أن معامل المقاومة لمبيد اللانيت 70.81 لسلالة DAM وكان 28.09 لسلالة MAR. كما أوضحت النتائج أن معامل المقاومة لمبيد كونفيدور كان 14.92 لسلالة KAF وكان 12.14 لسلالة BEH بينما كان معامل المقاومة للمستخلص الميكروبي سبينوساد 38.04 لسلالة DAM وكان 17.08 لسلالة MAR.

من النتائج السابقة يتضح أن السلالة التي جمعت من محافظة دمياط كانت الأعلى مقاومة لجميع المبيدات التي استخدمت في الدراسة باستثناء مبيد كونفيدور بينما السلالة التي جمعت من محافظة مرسي مطروح كانت الأقل مقاومة لجميع المبيدات المستخدمة في الدراسة باستثناء مبيد كونفيدور أيضاً. كما بينت النتائج إن أكثر المبيدات سمية وتأثير علي جميع سلالات نافقة الطماطم المستخدمة في الدراسة كان مبيد كونفيدور. وبصفة عامة فان مقاومة حشرة نافقة الطماطم للعديد من المبيدات بهذه الدرجة الكبيرة يجب أن تؤخذ في الاعتبار عند إعداد برامج المكافحة المتكاملة لحشرة نافقة الطماطم في مصر.

قام بتحكيم البحث

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