Pest Technology ©2009 Global Science Books

Insecticidal Activity of Gibberellic Acid against *Spodoptera littoralis* (Lepidoptera, Noctuidae) and *Locusta migratoria migratoria* (Orthoptera, Acrididae)

Khemais Abdellaoui • Monia Ben Halima-Kamel* • Mohamed Habib Ben Hamouda

Department of Biological Sciences and Plant Protection, Higher Agronomic Institute of Chott-Mariem, 4042 Sousse, Tunisia

Corresponding author: * **Kamonia_tn@yahoo.fr**

ABSTRACT

Oral toxicity of gibberellic acid (GA_3), a plant growth regulator, was evaluated on *S. littoralis* (Lepidoptera, Noctuidae) and *L. migratoria migratoria* (Orthoptera, Acrididae) larvae. These insects were exposed to various concentrations of GA_3 incorporated into the diet. GA_3 significantly reduced food consumption of both insect species leading to larval weight loss. GA_3 toxicity was also demonstrated by some larval mortality caused by exuviation difficulties. Different types of malformations observed were due to difficulties in rejecting the nymphal integuments. Additionally, digestive tract softness, particularly for *L. migratoria migratoria*, was observed. A subsequent histological study of the foregut and gastric caeca revealed the cytotoxic effect of GA_3. In fact, we noted the destruction of epithelial cells and a total disorganization of the cellular structure of these organs. Consequently, this experiment led us to conclude that ingested GA_3 caused perturbation in development and death of both insect species, which may be caused by antifeedant properties and by cytotoxic effect via alteration of the digestive system.

Keywords: antifeedant, histology, phytophagous insects, phytoregulator, toxicity

INTRODUCTION

Among various biotic stresses that plants face, insect attack has been a major challenge leading to severe losses in crop yields. The use of chemical pesticides has been the main insect controlling measure in recent decades. Due to the threat of the use of synthetic insecticides, such as environmental pollution, development of insecticide resistance, insecticide-induced resurgence of insect pests and adverse effects on non-target organisms, alternative pesticides are becoming increasingly important (Pascual-Villalobos and Robledo 1998; Pavela *et al.* 2004).

New approaches to the development of insect control agents have been revealed through the description of natural compounds capable of interfering with the development and reproduction of target insects (Hoffman and Lorenz 1998). Much attention has been devoted to the use of plant constituents that have an insecticidal effect as "biocides" (Nasseh *et al.* 1993). The use of plant products to control pest populations is a new approach which has captured world wide attention (Isman 1993; Deborah *et al.* 2000). Plants have always been a rich source of natural compounds that can be utilized in the development of environmentally safe methods for insect control. The deleterious effects of certain purified phytochemicals or crude plant extracts on insects are manifested in several ways, including toxicity (Hiremath *et al.* 1997), growth retardation (Breuer and Schmidt 1995), feeding inhibition (Klepzig and Schlyter 1999; Wheeler and Isman 2001), oviposition deterrence (Dimock and Renwick 1991; Hermawan *et al.* 1994; Zhao *et al.* 1998), suppression of calling behaviour (Khan and Saxena 1986) and reduction of fecundity and fertility (Muthukrishnan and Pushpalatha 2001). Such a wide variety of effects provides potential alternatives for the use of synthetic chemical insecticides.

Certain plant families, particularly Meliaceae, Asteraceae, Rutaceae, Labiaceae, Annonaceae and Canellaceae, are viewed as exceptionally promising sources of plant-based insecticides (Schmutterer 1990; Ould El Hadj *et al.* 2006). Several other families which have entomotoxic properties have also been frequently reported (Hermawan *et al.* 1994; Sadek 1997; Rodriguez-Saona and Trumble 1999; Barbouche *et al.* 2001, 2002; Sadek 2003; Ammar 2007; Chaieb *et al.* 2007a; Idrissi Hassani and Hermas 2008).

Among the natural compounds produced by plants, growth regulatory compounds appear to influence directly or indirectly the patterns of growth and reproduction of associated phytophagous insects. As an example, gibberellic acid (GA_3) perturbs food consumption, the development or reproductive potential in Diptera *Ceratitis capitata* (Diptera, Tephritidae*)* (Barbouche and Ben Hamouda 1986) and *Bactrocera cucurbitae* (Diptera, Tephritidae) (Kaur and Rup 2003). Besides, some researchers have even recommended the use of plant growth regulators like GA_3, coumarin and indole-3-acetic acid as successful chemosterilants against some insect pests (Pandey *et al.* 1980; Kaur and Rup 2002). The present investigation is an attempt to explore the effects of various concentrations of GA_3 on *S. littoralis* and *L. migratoria migratoria* larvae.

MATERIALS AND METHODS

Insects

***L. migratoria migratoria*:** Insects used for testing came from a gregarious stock, which had been reared in breeding cages measuring 50 cm^3 and containing a few hundred specimens. The temperature was kept at 30 ± 1°C and a light/dark cycle of 12/12h was used. *L. migratoria migratoria* were fed once a day, in the morning, with fresh *Sorghum vulgare* leaves. Larvae in the 5^{th} instar (0-1 day old), used for biological tests were transferred individually in boxes of two litres placed in the same conditions than gregarious stock.

***S. littoralis*:** *S. littoralis* adults were captured using a Pennsylvanian luminous trap placed in agriculture area in Chott Meriem (Tunisia). These latter were placed in Plexiglass boxes and were

Received: 12 September, 2009. Accepted: 13 October, 2009.

Original Research Paper

Table 1 Effect of different concentrations of GA_3 on food intake (g/larva) in *S. littoralis* and *L. migratoria migratoria*.

Insect species	Concentrations	2 days	4 days	6 days	8 days
S. littoralis	0 ppm	0.83 a	1.87 a	2.72 a	2.80 a
	125 ppm	0.58 b	1.19 b	1.69 b	1.58 b
	625 ppm	0.53 b	0.87 c	1.14 c	0.98 c
	3125 ppm	0.39 c	0.65 c	0.75 d	1.02 c
L. migratoria migratoria	0 ppm	0.94 a	1.22 a	1.35 a	1.63 a
	125 ppm	0.91 b	0.96 ab	1.20 b	1.42 b
	625 ppm	0.65 c	0.84 bc	0.88 c	1.10 c
	3125 ppm	0.61 c	0.63 c	0.75 c	0.86 c

Means within a column and insect species followed by different letters are significantly different ($P < 0.05$).

Table 2 Fresh body weight (g) of *S. littoralis* and *L. migratoria migratoria* larvae treated by different concentrations of GA_3.

Insect species	Concentrations	0 days	2 days	4 days	6 days	8 days
S. littoralis	0 ppm	0.02 a	0.27 a	0.49 a	0.77 a	0.70 a
	125 ppm	0.02 a	0.12 b	0.29 b	0.53 b	0.56 b
	625 ppm	0.02 a	0.06 b	0.17 b	0.40 c	0.45 c
	3125 ppm	0.02 a	0.05 b	0.10 c	0.27 c	0.31 c
L. migratoria migratoria	0 ppm	0.53 a	0.89 a	1.09 a	1.22 a	1.45 a
	125 ppm	0.53 a	0.71 b	0.74 b	1.08 ab	1.12 ab
	625 ppm	0.44 a	0.67 b	0.66 b	0.97 ab	1.06 ab
	3125 ppm	0.32 a	0.51 c	0.68 b	0.81 b	0.92 b

Means within a column and insect species followed by different letters are significantly different ($P < 0.05$).

fed a 30% honey solution containing 1% *Acacia cyanophylla* pollen. Eggs were laid directly on the walls of the box. The adults were then removed in order to ensure the hatching and breeding of the young caterpillars. Larvae of the 3rd stage were reared individually in Petri dishes on a simplified artificial diet of Poitout and Bues (1974). The caterpillars were maintained in culture rooms at 25°C, 70% relative humidity and a 16L: 8D photoperiod with 4000 lux light intensity.

Biological tests

The effects of GA_3 (Sigma-Aldrich Chemie, GmbH) on the development and food consumption of the Asiatic migratory locust *L. migratoria migratoria* were investigated by exposing freshly emerged (0-1 day old) 5th instar larvae to fresh *S. vulgare* leaves treated by three different concentrations (125, 625 and 3125 ppm). Pure GA_3 used in this assay was dissolved in distilled water. In the control experiment, the larvae received the same quantity of distilled water. The larvae (n = 10 for each concentration) were weighed and kept separate in small plastic 2-L containers to standardize their state of hunger prior to the assay. Every morning, definite quantities of fresh *S. vulgare* leaves were provided to the insects, and aliquots of the same food were kept in the same conditions to calibrate the water lost from the food provided. Uneaten food was separated from the faeces and weighed. The insects were weighed every 2 days.

The effect of GA_3 on *S. littoralis* was performed with 3rd instar larvae (10-12 mg). The GA_3 solutions were incorporated in the same concentrations already mentioned in the artificial diet at the time of its preparation. Control insects were reared on a GA_3-free diet. Larvae were maintained in the same conditions as the stock culture. All larvae (n = 30 for each concentration) were individually weighed, every 2 days, and average weights were determined. The same methodology as *L. migratoria migratoria* was followed for the estimation of food intake.

Histology

The effect of GA_3 on the digestive system structure was investigated on the 5th larval instar of *L. migratoria migratoria*. Insects were sampled after 8 days of treatment. Histological procedures were conducted according to Martoja and Martoja (1967). Foregut and gastric caeca were dissected in physiological liquid (Ringer's solution) and fixed in Bouin's solution for 3 days. Transverse sections (7 µm) were stained with Mallory liquid.

Statistics

Results are expressed as means ± standard deviation. The significance between control and treated series was estimated using *S.N.K.* (Student-Newman-Keuls) test at the 5% level. All data were statistically analyzed by SPSS (Version 13.0.).

RESULTS

Effect on food consumption

GA_3 strongly deterred feeding of both insect species. Indeed, the results of feeding assay showed significant ($P<0.05$) variation in consumed food between the different tested concentrations. However, the severity of their effects was not of the same intensity compared with the control group. The influence of GA_3 on food consumption increased as its concentration increased. Indeed, the maximum reduction in both insect species was observed with the high concentration used (3125 ppm) compared with the other treatments (**Table 1**). After 8 days, GA_3 concentration significantly ($P<0.05$) decreased the *S. littoralis* larvae food intake by 44, 65 and 64%, respectively for 125, 625 and 3125 ppm compared to the control treatment. In contrast, for the *L. migratoria migratoria* larvae food intake decreased 13, 33, and 47% for the same treatments compared to the 0 ppm treatment (**Table 1**). The results reveal that GA_3 had a substantial toxic effect as was shown by the relatively low food intake of larvae fed on treated diets.

Effect on larval weight

Table 2 shows a significant difference ($P<0.05$) in the size of the control insects compared to those treated with different concentrations of GA_3. These results underline the possibility that the reduction in size can only be the consequence of insufficient feeding caused by anti-feedancy, which appears from the results of measuring the consumed diet. In both species, control larvae showed a rapid increase in weight until day 8. At this point, we noted that the weight of *S. littoralis* control larvae began to drop as the larvae ceased to feed prior to pupation. As the concentration of GA_3 increased, the mean weight reached by the insect decreased. On day 8 and at GA_3 = 3125 ppm, the mean fresh weights decreased by 56 and 37%, respectively for *S. littoralis* and *L. migratoria migratoria* compared to the 0 ppm treatment. The greatest mean fresh weights were observed with the untreated control insects. Indeed, when 8 days old, the mean weights noted were 0.7 ± 0.07 and 1.45 ± 0.28 g, respectively for both insect species. The reductions noted with all the concentrations tested compared to untreated animals persisted during the experimental period.

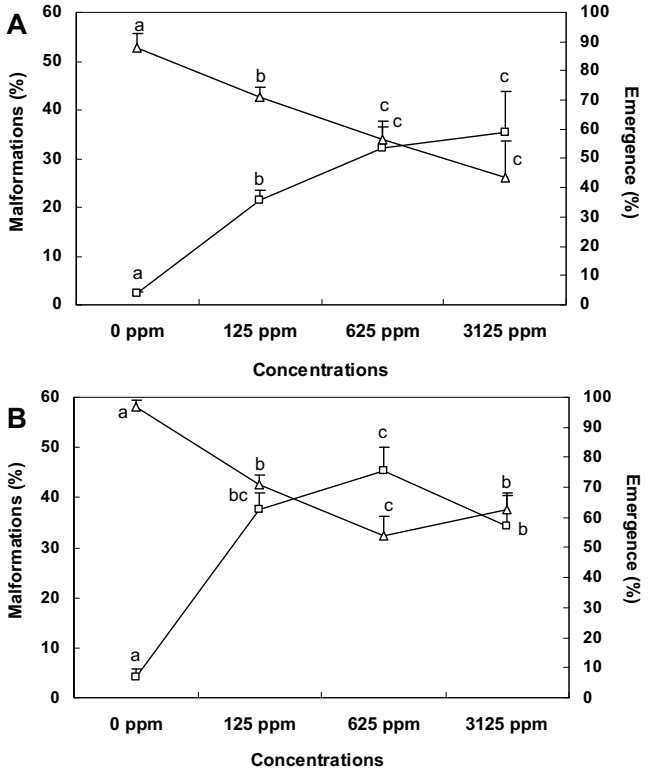

Fig. 1 Percentage of emergence and nymphal malformations of *S. littoralis* (**A**) and *L. migratoria migratoria* (**B**) treated by different concentrations of GA_3. Triangles: emergence, squares: malformations. Within concentrations, means followed by the same letters are not significantly different ($P<0.05$). (Bar = standard deviation).

Effect on larval development

Fig. 1A and **1B** show nymphal mortality, in particular with the highest concentration of GA_3 tested (3125 ppm). These mortalities are related to malformations affecting the chrysalis and the 5th larval instar of *L. migratoria migratoria*. Actually, the percentage of malformation is proportional to the concentration of GA_3 added in the diet and inversely proportional to the rate of emergence (**Fig. 1A, 1B**). Some malformations were observed in *S. littoralis* larvae that appeared at the beginning of nymphosis. Larval exuvia became more difficult to eliminate during the nymphal moult that generates a separation of the nymphal teguments and appearance of the nude zone. Moreover, these zones of the cuticle probably constitute an entered door for pathogenic microorganisms (**Fig. 2B**). The second type of malformation observed in *S. littoralis* was difficulties in larval exuviations (**Fig. 2A**). These difficulties were due to impossibility to reject the old cuticle causing larval mortality. The phenomenon was observed in particular at the anterior part of the body (cephalic and thoracic parts).

The same observations were also noted with *L. migratoria migratoria*. Indeed, the persistence of the larval cuticle and impossibility to reject the old integuments, causing mortality at the beginning of the imagos stage were noted (**Fig. 2C, 2D**). The imagos moulting from treated larvae were considerably smaller in their body dimensions than the untreated control locusts. In addition to these results we observed that GA_3 was responsible for other developmental perturbations on *L. migratoria migratoria* 5th instar and *S. littoralis* larvae. However, the mobility of the larvae decreased considerably and their movement became incoherent.

Cytotoxic effect

Our experiments showed that insects, particularly *L. migratoria migratoria*, having received GA_3 in their feeding demonstrated necroses at the level of the abdomen, probably due to intestinal intoxication (**Fig. 3**). For this reason we tried in this work to study the cytotoxic effect of GA_3 on the digestive tract.

Histological studies showed that various symptoms are due to structural modifications observed as well at the level of the gastric caeca and foregut. These modifications are presumably due to the cytotoxicity caused by GA_3.

***Gastric caeca*:** histological observations show that for treated insects, the gastric caeca presented notable histological modifications compared to the control. These cellular changes were revealed through epithelial burstings and a disruption of the muscular layer surrounding the caeca. In addition, the epithelium showed a grainy appearance, an altered edge of the microvillosities and some typical signs of cell necrosis. Indeed, we noticed that the apical brush border was packed less densely due to cell swelling and vacuolization and it was missing in some regions. The epithelial cell nuclei become hypertrophied. The chromatins were disorganized and condense in visible granules in the nucleoplasm. These symptoms are signs of cellular degeneration. Regenerative crypts were evident but appeared to be in the process of disintegration (**Fig. 4B**). On the contrary, in the control group the epithelial cell layer was well organized having a regular arrangement of epithelial cells. The brush border of these cells was well defined and the regenerative crypts were frequently localized at the base of epi-

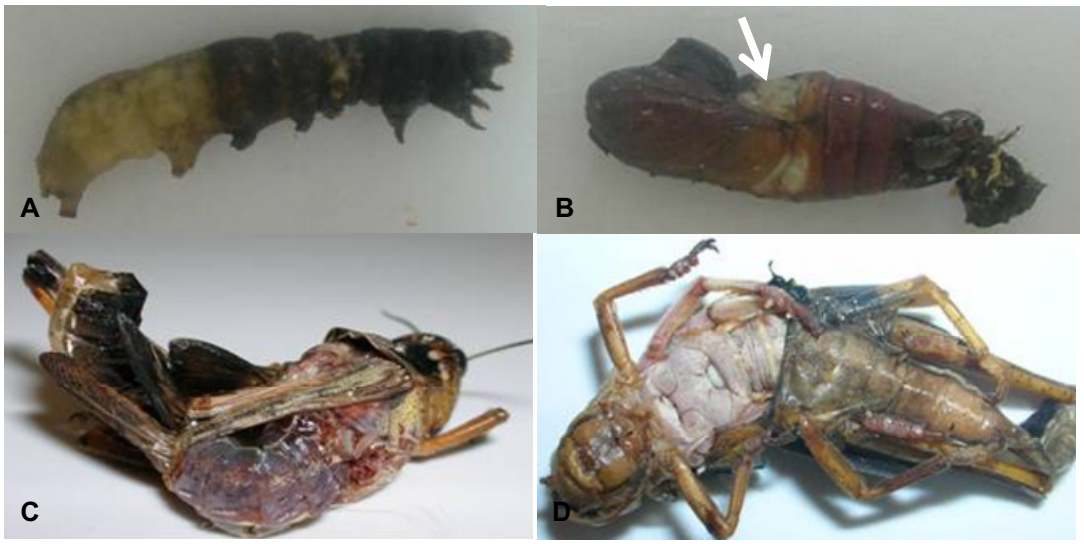

Fig. 2 Exuviation difficulties for *S. littoralis* (**A**) and *L. migratoria migratoria* larvae (**C** and **D**) after ingestion of GA_3. Impossibility to reject the old integuments causing the mortality of insects. Malformation at the nymphosis stage of *S. littoralis* and appears of nude zone causing the death of the nymph or the obtaining of the adults with severe malformations (**B**).

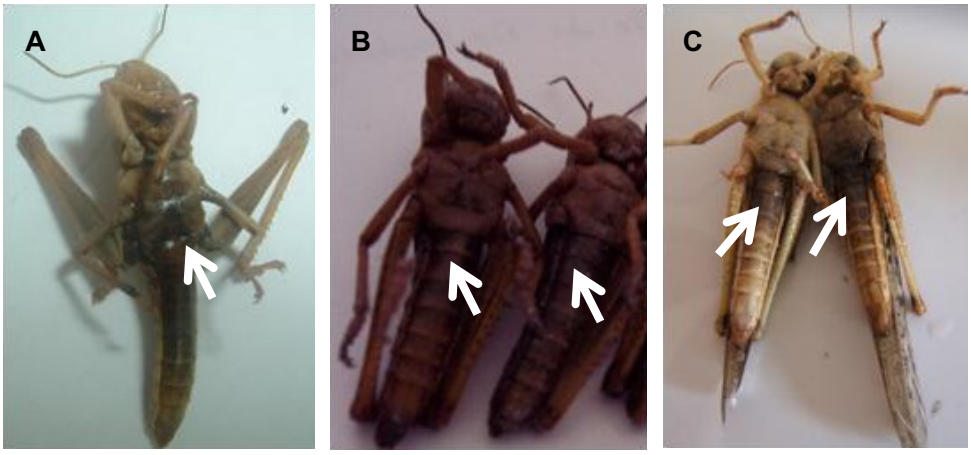

Fig. 3 Appearance of necroses at the level of the abdomen of larvae (**A, B**) and adults (**C**) of *L. migratoria migratoria* further to the ingestion of GA_3.

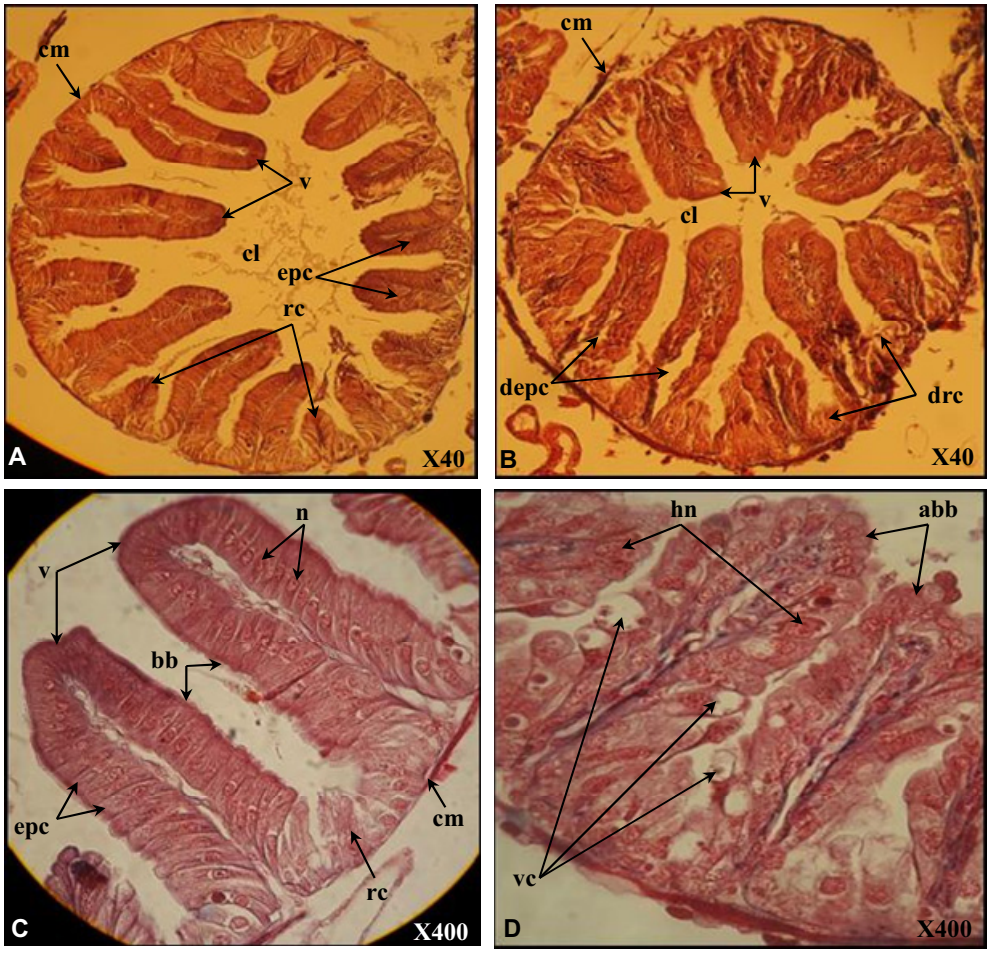

Fig. 4 Light microscope photographs of *L. migratoria migratoria* larval gastric caeca structure after 8 days of GA_3 treatment. abb: altered brush border, bb: brush border, cl: caeca light, ci: cuticle intima, cm: circular muscles, depc: destroyed epithelial cells, drc: degraded regenerative crypts, epc: epithelial cells, hn: hypertrophied nuclei, n: nuclei, rc: regenerative crypts, v: villosity, vc: vacuolarized cells. Gastric caeca for (**A, C**) untreated control and (**B, D**) for treated insects with 3125 ppm GA_3.

thelial folds (**Fig. 4A**).

Foregut: the same phenomena were observed on the level of the foregut with a reduction of the external circular muscle resulting in a relaxation of the intestine. The results showed the destruction of cellular structure manifested by separation of the intestinal epithelium and muscular layers. At the highest concentration (3125 ppm), the epithelial tissues were totally destroyed and we noted total disorganization of the cellular structure (**Fig. 5A, 5B**).

DISCUSSION

The GA_3 proved to have conspicuous antifeedant and toxic effects on the larvae of *S. littoralis* and *L. migratoria migratoria*. The experiments showed that GA_3 significantly reduced in a dose-dependant manner the food consumption of both insect species leading to a loss of weight among larvae. The phytohormone exhibited strong antifeedant and toxic activity against the larvae when applied either on fresh leaves or incorporated into artificial diet. Antifeedant substances are customarily classified into repellents (which repel an insect without making contact with the material), suppressants (which suppress biting activity after contact) or deterrents (which deter an insect from further feeding after ingestion of the material) (Chapman 1974; Schoonhoven 1982). Based on these definitions, the GA_3 had both suppressant and deterrent properties.

The toxicity of GA_3 was manifested by intestinal intoxi-

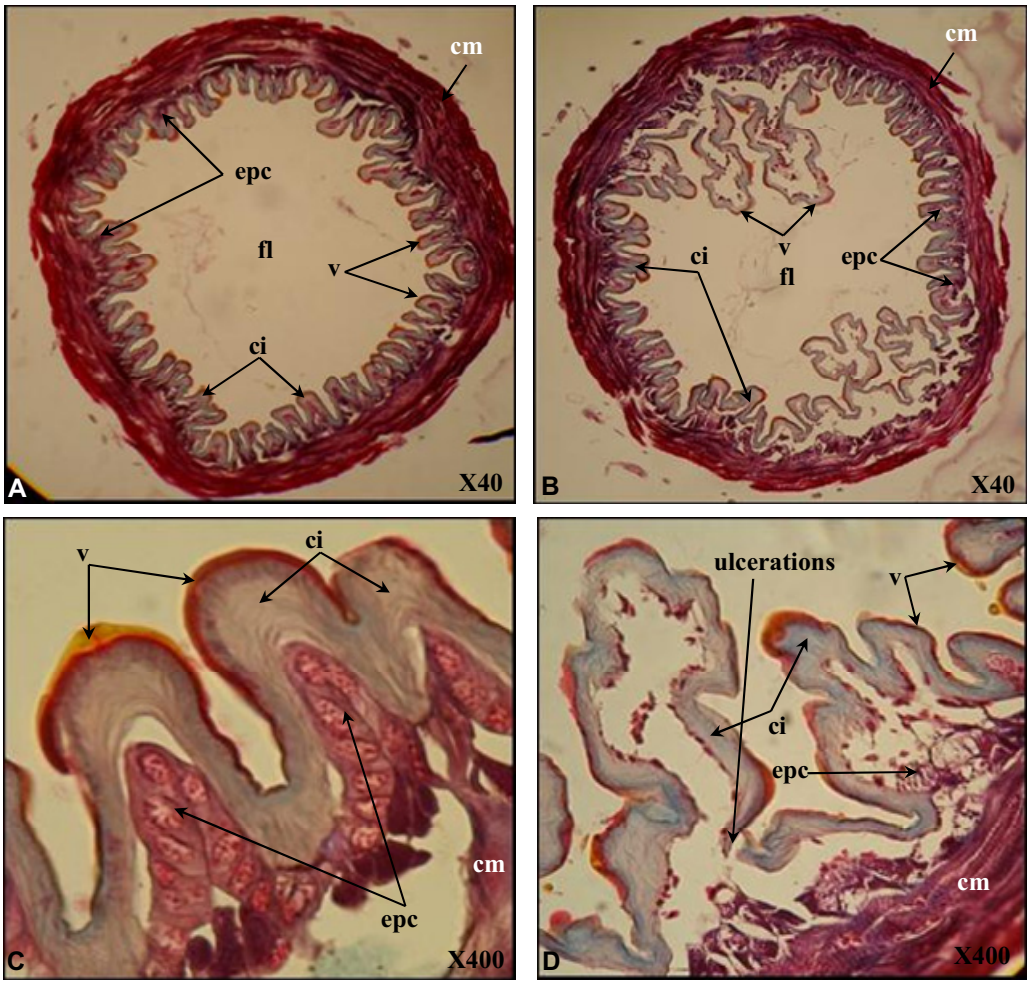

Fig. 5 Light microscope photographs of *L. migratoria migratoria* larval foregut structure after 8 days of GA_3 treatment. ci: cuticle intima, cm: circular muscles, epc: epithelial cells, fl: foregut light, v: villosity. Foregut for (**A, C**) untreated control and (**B, D**) for treated insects with 3125 ppm GA_3.

cation. The histological study revealed a cytotoxic effect on the digestive system of *L. migratoria migratoria.* Indeed, we noted a cell destruction of the foregut and the gastric caeca of treated animals compared to the control. We also remarked a total disorganization of the cells structure that provoked insect's death. The treatments have also decreased considerably the mobility and the dimensions of insects and caused exuviations difficulties.

Similarly, Barbouche (1986) and Barbouche and Ben Hamouda (1986), showed that gibberellic acid inhibits the development and the reproductive potential of *C. capitata*. Indeed, the larvae having gibberellins in their alimentation showed a low rate of insects in pupal stage even with low concentrations (20 and 30 mg/100 g of food).

Besides, Kaur and Rup (2002) reported that the topical treatment with gibberellic acid given to freshly emerged (0-1-day-old) male and female of the melon fruit fly *B. cucurbitae* showed a significant adverse influence on the development and the longevity of this fly. These authors noted in (2003) that some plant growth regulators like kinetin, coumarin, indole-3-acetic acid (IAA) and especially GA_3 exerted growth and development inhibitory effects on the fly. Treatment with the plant growth regulators also prolonged the fly's developmental period, reduced percentage emergence and increased percentage of abnormal flies emerging. At the highest concentration tested GA_3 caused 100 % mortality in first instar (Kaur and Rup 2003).

In the same way, Isman and Rodriguez (1983) showed that natural phytoregulator extracted from the *Parthenium* plant is capable of reducing to 88% the population of *Heliothis* reared on food containing the rate of 3 mM/kg of food. Paulson *et al.* (2005) showed that prohexadione-calcium, a plant growth regulator that inhibits gibberellin metabolism, significantly reduced *Cacopsylla pyricoloa* and *Aphis spiracola* populations in pear and apple trees respectively.

The effects of GA_3 on the digestive tract of *L. migratoria migratoria* larvae are comparable to those induced by the ingestion of the crude saponic extract of *Cestrum parquii* on *Schistocerca gregaria* digestive system (Chaieb *et al.* 2007b). The authors noted a dilation of the gastric caeca showing a quite visible light accompanied by a fall height of the epithelial folds after 6 hrs of treatment. Moreover, cellular perturbation and epithelial bursting were observed after 24 hrs. The histological study, conducted at the level of the foregut, showed a cytotoxicity manifested by a progressive destruction that began with a separation of the intestinal epithelium and muscular layers 6 hours after treatment, leading eventually to a total disorganization of the cells after 24 hours. Ammar and N'cir (2008) also showed that the light microscopy observations revealed that the foregut structure of the 5^{th} stage of *S. gregaria* fed with *C. parquii* leaves is modified at cuticular intima level where no exuvial space could be seen up to 7 days of treatment. By the 9^{th} day of treatment, this space started to appear but no new cuticle intimae were observed as in the case of the control. The height of the epithelial cells and the thickness of muscular layers were reduced significantly.

The way GA_3 acts on inhibiting food intake and development of insects is still unknown. The first results come from simple observations that indicated a reduction of the population of arthropods on the plants treated with GA_3 (Turner *et al.* 1970 *in* Barbouche 1986; Henneberry *et al.* 1982). Most of authors explain this phenomenon by the physiological changes of the treated plant that could provide a chemical defence against insect herbivores.

The histological study demonstrated that the antifeeding property of GA_3 was probably related to postingestive toxicity. Indeed, Månsson (2005) described antifeedants as

compounds that inhibit feeding by sensory perception i.e. giving plant material an unpalatable taste but may also reduce feeding by toxic, postingestive effects. However, some complementary studies are therefore necessary to verify the effect of this phytoregulator on the sensory perception of the larvae in order to better understand their antifeeding property.

Barbouche and Ben Hamouda (1986) supposed that the GA_3 acts either while reinforcing the action of ecdysteroids or while partially inhibiting the synthesis of the juvenile hormone (JH) because the GA_3 and JH have a common precursor which is the mevalonic acid. Kaur and Rup (2002) noted that the chemical configuration of GA_3, a terpenoid compound, is also similar to a JH and it was hypothesized that GA_3 interfered in the endocrinal metabolic processes involved in development and reproduction.

The increasing number of investigations on plant–insect chemical interactions in the last few decades has unveiled the potential of utilizing secondary plant metabolites, or allelochemicals, as pest control agents. This growing interest in botanical insecticides resulted from the need to provide an alternative in IPM programs to the synthetic insecticides, whose adverse effects on agroecological systems are proven. This study could also contribute to assess the possibility of using plants growth regulators (PGRs) as potential insecticides. Indeed, an insecticide does not have to cause high mortality on target organisms in order to be acceptable. Antifeedant and growth inhibiting activity reduce pest damage to products without even killing the pest. This antifeedant and growth-inhibiting activity can therefore be incorporated into other insect control techniques in the strategy of integrated pest management (IPM).

ACKNOWLEDGEMENTS

The authors are grateful to Pr. Hakim Feidi (Institut Supérieur Agronomique, Chott-Mariem) for his comments and critical reading of the manuscript.

REFERENCES

Ammar M (2007) Caractérisation physiologique de la transformation phasaire chez le criquet pèlerin *Schistocerca gregaria* Forskål (Orthoptera, Acrididae) sa contribution a la connaissance de la dynamique des essaims et influences de quelques composants végétaux sur le développement. Thèse de doctorat d'état en sciences agronomiques, Institut National Agronomique de Tunis, Tunisie 230 pp

Ammar M, N'cir S (2008) Incorporation of *Cestrum parquii* (Solanaceae) leaves in an artificial diet affected larval longevity and gut structure of the desert locust *Schistocerca gregaria*. *Tunisian Journal of Plant Protection* **3**, 27-34

Barbouche N (1986) Action des gibbérellines, hormones de croissance végétales, sur le développement et la physiologie de la reproduction de *Ceratitis capitata* (Dipt. Tephritidae). Doctorat de Spécialité, Faculté des Sciences de Tunis, Tunisie 126 pp

Barbouche N, Ben Hamouda MH (1986) Action des gibbérellines, hormones de croissance végétales, sur la physiologie de la reproduction de *Ceratitis capitata* (Dipt. Tephritidae) In: Proceedings of II International Symposium of Fruit Flies/crete Greece, pp 103-113

Barbouche N, Ammar M, Hajjem B, Lognay G (2001) Contribution à l'étude de l'activité biologique d'extraits de feuilles de *Cestrum parquii* l'Herit (Solanacée) sur le criquet pèlerin *Schistocerca gregaria* (Forsk). *Biotechnology, Agronomy, Society and Environment* **5**, 85-90

Barbouche N, Ben Salah H, Moumene K, Hajjem B, Lognay G (2002) Action des saponines présentes dans les feuilles de *Cestrum parquii* l'Herit sur la digestion du criquet pèlerin *Schistocerca gregaria* Forsk, 1775 (Ortho. Acrididae). *Revue de l'INAT* **17**, 185-197

Breuer M, Schmidt GH (1995) Influence of a short period treatment with *Melia azedarach* extract on food intake and growth of the larvae of *Spodoptera frugiperda* (Lepi. Noctuidae). *Journal of Plant Diseases and Protection* **102**, 633-654

Chaieb I, Ben Halima-Kamel M, Ben Hamouda MH (2007a) Toxicity experiments of the saponic extract of *Cestrum parqui* (Solanaceae) on some insect spices. *Journal of Entomology* **4**, 113-120

Chaieb I, Trabelsi M, Ben Halima-Kamel M, Ben Hamouda MH (2007b) Histological effects of *Cestrum parquii* saponins on *Schistocerca gregaria* and *Spodoptera littoralis*. *Journal of Biological Sciences* **7**, 95-101

Chapman RF (1974) The chemical inhibition of feeding by phytophagous insects. *Bulletin of Entomological Research* **64**, 339-363

Deborah AW, Murray BI, Pablo ESV, Arnason JT (2000) Screening of Costa Rican *Trichilia* species for biological activity against the larvae of *Spodoptera litura* (Lepi. Noctuidae). *Biochemical Systematics and Ecology* **29**, 347-358

Dimock MB, Renwick JAA (1991) Oviposition by field populations of *Pieris rapae* (Lepi. Pieridae) deterred by an extract of a wild crucifer. *Environmental Entomology* **20**, 802-806

Henneberry TJ, Bariola LA, Kittock DL (1982) Selective removal of miniature cotton Bolls in late season to reduce populations of diapausing pinte boll-worm. *Protection Ecology* **4**, 159-165

Hermawan W, Kajiyama S, Tsukuda R, Fujisaki K, Koboyashi A, Nakasuji F (1994) Antifeedant and antioviposition activities of the fraction of extract from a tropical plant, *Andrographis paniculata* (Acanthaceae), against the diamondback moth, *Plutella xylostella* (Lepi. Yponomeutidae). *Applied Entomology and Zoology* **29**, 533-538

Hiremath IG, Ahn YJ, Kim SI (1997) Insecticidal activity of Indian plant extracts against *Nilaparvata lugens* (Hom. Delphacidae). *Applied Entomology and Zoology* **32**, 159-166

Hoffman KH, Lorenz MW (1998) Recent advances in hormones in insect pest control. *Phytoparasitica* **26**, 1-8

Idrissi Hassani LM, Hermas J (2008) Effets de l'alimentation en *Peganum harmala* L. (Zygophyllaceae) sur le tube digestif du criquet pèlerin *Schistocerca gregaria* Forsk. (Orthoptera, Acrididae). *Zoologica Baetica* **19**, 71-84

Isman MB (1993) Growth inhibitory and antifeedant effects of azadirachtin on six noctuids of regional economic importance. *Pesticide Science* **38**, 57-63

Isman MB, Rodriguez E (1983) Feeding and growth of noctuid larvae on foliar material and extracts of guayule related species of *Parthenium* and F1 hybrids. *Environmental Entomology* **13**, 539-542

Kaur R, Rup PJ (2002) Evaluation of regulatory influence of four plant growth regulators on the reproductive potential and longevity of melon fruit fly *Bactrocera cucurbitae*. *Phytoparasitica* **30**, 1-7

Kaur R, Rup PJ (2003) Influence of four plant growth regulators on development of melon fruit fly *Bactrocera cucurbitae* (Coquillett). *Insect Science and its Application* **23**, 121-125

Khan ZR, Saxena RC (1986) Effect of steam distillate extracts of resistant and susceptible rice cultivars on behavior of *Sogatella furcifera* (Hom. Delphacidae). *Journal of Economic Entomology* **79**, 928-935

Klepzig KD, Schlyter F (1999) Laboratory evaluation of plant derived antifeedants against the pine weevil *Hylobius abietis* (Col. Curculionidae). *Journal of Economic Entomology* **92**, 644-650

Månsson PE (2005) Host selection and antifeedants in *Hylobius abietis* Pine Weevils. Thèse de doctorat, Swedish University of Agricultural Sciences Alnarp, 33 pp

Martoja R, Martoja M (1967) Techniques d'histologie animale. In: Masson Cie (Ed), Paris VI, 345 pp

Muthukrishnan J, Pushpalatha E (2001) Effects of plant extracts on fecundity and fertility of mosquitoes. *Journal of Applied Entomology* **125**, 31-35

Nasseh O, Wilps H, Rembold H, Krall S (1993) Biologically active compounds in *Melia volkensii*. Larval growth inhibitor and phase modulator against the desert locust *Schistocerca gregaria* (Forskål) (Orthoptera, Cyrtacanthacridinae). *Journal of Applied Entomology* **116**, 1-11

Ould El Hadj MD, Tankari Dan-Badjo A, Halouane F, Doumandji S (2006) Toxicité comparée des extraits de trois plantes acridifuges sur les larves du cinquième stade et sur les adultes de *Schistocerca gregaria* Forskål, 1775 (Orthoptera, Cyrtacanthacridinae). *Sécheresse* **17**, 407-414

Pandey GP, Teotia TPS (1980) Laboratory studies on chemosterilization of Angoumois grain moth, *Sitotroga cerealella* (Oliv.): screening of 33 compounds by pupal treatments. *Indian Journal of Entomology* **42**, 1-15

Pascual-Villalobos MJ, Robledo A (1998) Screening for anti-insect activity in Mediterranean plants. *Industrial Crops and Products* **8**, 83-194

Pavela R, Barnet M, Kocaurek F (2004) Effect of Azadirachtin applied systemically through roots of plants on the mortality, development and fecundity of the cabbage aphid (*Brevicoryne brassicae*). *Phytoparasitica* **32**, 286-294

Poitout S, Bues R (1974) Rearing of 28 spices of Noctuidae and two spices of Arctiidae on simplified artificial diet, rearing particularities according to spices. *Annales de Zoologie Ecologie Animale* **6**, 431-441

Rodriguez-Saona CR, Trumble JT (1999) Effect of avocadofurans on larval survival, growth, and food preference of the generalist herbivore, *Spodoptera exigua*. *Entomologia Experimentalis et Applicata* **90**, 131-140

Sadek MM (1997) Antifeedant and larvicidal effects of *Eichornia crassipes* leaves on the cotton leafworm *Spodoptera littoralis* (Boisd.). *Journal of the Egyptian German Society of Zoology* **24**, 209-232

Sadek MM (2003) Antifeedant and toxic activity of *Adhatoda vasica* leaf extract against *Spodoptera littoralis* (Lepi. Noctuidae). *Journal of Applied Entomology* **127**, 396-404

Schmutterer H (1990) Properties and potential of natural pesticides from the neem tree. *Annual Review of Entomology* **35**, 271-297

Schoonhoven LM (1982) Biological aspects of antifeedants. *Entomologia Experimentalis et Applicata* **31**, 57-69

Wheeler DA, Isman MB (2001) Antifeedant and toxic activity of *Trichilia americana* extract against the larvae of *Spodoptera litura*. *Entomologia Experimentalis et Applicata* **98**, 9-16

Zhao B, Grant GG, Langevin D, MacDonald L (1998) Deterring and inhibiting effects of quinolizidine alkaloids on spruce budworm (Lepi, Tortricidae) oviposition. *Environmental Entomology* **27**, 984-992