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IMPACT OF CERTAIN PLANT EXTRACTS ON ENZYMES ACTIVITY OF TOMATO LEAFMINER *Tuta absoluta*.

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ABSTRACT

The experiments were conducted in the laboratory Plant Protection Research Institute Dokki- Giza, to study the effect of *Anethum groveolenus* Dill, *Ambrosia maritime* Damaseia extract, *Cumminum cyminum* Cumin; *Syzyguim aromaticu*m Clove and *Allium maritime* Garlic on activity of some enzymes in(*Tuta absoluta Meyrick*) on tomato plants were investigated. After the tested compounds application, the results revealed that these compounds had various effects on the activity alkaline phosphatases, α and β esterases and carbohydrates hydrolyzing enzymes (trehalase, invertase and amylase) . The enzyme activity reduced or increased significantly. Activity of alkaline phosphatases, α and β esterases were higher in Damasiea on *T. absoluta*, in comparison to control. The percentage of increase of alkaline enzymes activity were +22.28 and +17.38 and +13.34 % in *T. absoluta* respectively. Also, in cumin and garlic extracts treatments. Generally, in case of *T. absoluta* treatments, the results of damasiea extract indicated high increase (+40.1% in comparison to control) in activity of beta- esterases and invertase enzymes but high decrease (+45.9%), in alpha- esterases activity only.

The results indicated that the damasiea extract had direct effects on the metabolism in the body *Tuta absoluta.*

INTRODUCTION

The tomato leafminer *Tuta absoluta* (Meyrick,) is a microlepidopteron has 12 generations per years (Siqueira *et al*. 2000), it was originally described in 1917 by Meyrick as *Phthorimaea absoluta*, based on individuals collected from Huancayo (Peru). Later, the pest was reported as *Gnorimoschema absoluta* (Clarke, 1962), *Scrobipalpula absoluta* (Povolny, 1964) or *Scrobipalpuloides absoluta* (Povolny, 1987), but was finally described under the genus *Tuta* as *T. absoluta* by Povolny in 1994. *Tuta absoluta* larvae feeds on the leaves of tomato *Lycopersicon sp.* by making tunnels in mesophyll of tomato leaf, also attack the stem and fruit of tomato plant. the activity of α and β - esterases. Cloves, cumin, garlic, dill extracts revealed significant decrease in the both enzyme activity in *T. absoluta*.

Tuta absoluta attacks several species of Solanacesous plants. There are host records on many *Solanum* species, including Tomato ,aubergine (*S. melongena),* pepino *(S. muricatum*) and black nightshade (*S. nigrum*). It hosts in other genera in the Solanaceae include peppers (*Capsicum* spp.), devil's apple (*Datura stramonium*) and tobacco tree (*Nicotina glauca*) (Korycinska and Moran, 2009). On potato, *T. absoluta* only attacks aerial

parts, thereby not directly impeding tuber development. Nevertheless, leaf feeding may indirectly lower potato yield and under appropriate climatic conditions, *T. absoluta* could become a pest for the potato crop (Derbalah *et al*., 2012 and Maiche, 2009).

MATERIALS AND METHODS

The experiments were conducted in the laboratory Plant Protection Research Institute Dokki- Giza al plants selected for this study are listed in Table (1), which includes four different families, five species, English name, Arabic name and the active parts which was used.

Table (1): Plants investigated for biological activity against *T. absoluta*.

Scientific Name	***Anethum groveolenus***	***Cumminum cyminum***	***Syzyguim aromaticu***	***Allium sativumi***	***Ambrosia maritime***
English name	Dill	Cumin	Clove	Garlic	Damaseia
Family	Umbelliferae	Umbelliferae	Myrtaceae	*Liliaceae*	Compostiae
Tested Part	Seeds	Seeds	Flowers	Globes	Leaves

Preparing of the plant extracts as described by Su (1985) for Dill and Cumin seeds, Clove flower and Damaseia leaves. Concentrations of 5, 10, 15 and 20 ml were prepared from the stock solution for the extracts of Dill, Cumin, Clove and Damaseia leaves for conducting the experiments but in the case of garlic, the globes of *Allium sativum* were cut into small pieces, then soaked in water (1g:1m1). The mixture was mixed in a household grinder. The extract was kept in glass stoppered bottles in refrigerator and its concentration was considered a 100% w/v. Different concentration was prepared by adding different quantities of the water to a constant volume of the initial extract. Following concentrations; 5, 10, 15 and 20 ml were prepared by diluting the liquid formulation in distilled water. The water extracts were used for the tests.

* Alkaline phosphatase (AlKP) was determined according to the method described by Powell and Smith (1954).
* Alpha and Beta- esterases (α-E, β-E) were determined according to the method of Van Asperen (1962) using α - naphthyl acetate and β - naphthyl acetate as substrates respectively.
* The methods to determine the digestion of trehalose, starch and sucrose by trehalase, amylase and invertase enzymes respectively were similar to those described by Ishaaya and Swiriski (1976).

Statistical analysis:

The mortality was corrected using Abbott's formula (1925).

The statistical analysis (ANOVA and Simple correlation) of the obtained data were performed by using SAS program (SAS Institute, 1988) which run under Windows. Also the difference between means was conducted by using Duncan's multiple range tests by this program.

RESULTS AND DISCUSSION

Data in Table (2), indicated that after tested plant extracts treatment, the activity of alkaline enzymes in the supernatant of the homogenate *T. absoluta* increased or decreased as affected by the tested plant extracts compared with control. In dill, damaseia and clove extracts treatment, alkaline enzymes activities increased significantly. The percentage of increase of alkaline enzymes activity were +22.28 and +17.38 and +13.34 % in *T. absoluta* In dill, damaseia and clove extracts treatment respectively. Also, in cumin and garlic extracts treatments, alkaline enzymes activity decreased significantly in *T. absoluta*, the percentages of enzyme decrease were -45.4 and -45.7 respectively.

Although, in damasiea extract treatment, alpha- esterase activity increased significantly, beta- esterase increased significantly in *T. absoluta*, the percentages of enzyme increase were +45.9 and +40.1 % respectively.

Data represented in Table (3), showed the changes in the activity of carbohydrate hydrolyzing enzymes (trehalase, invertase and amylase) of *T. absoluta* after treated with tested plant extracts. Trehalase activity increase significantly after application of clove, cumin, garlic, damasiea and dill extracts in *T. absoluta*, the percentages of enzyme increase were +41.7, +2.2 ,+ 2.6 ,+ 58.5 and +8.5 % in *T. absoluta* respectively.

Invertase activity had a different affect (a significant decrease or increase) in clove and cumin extracts treatments, the invertase activity decreased significantly in *T. absoluta* by -21.9 and -11.1 % respectively and it increased significantly in *T. absoluta* by +36.5, +14.7 and +1.6 % respectively. Invertase activity decreased significantly after application of clove and cumin extracts, Also invertase activity decrease significantly only in *T. absoluta* after treated with clove and cumin extracts.

Amylase activity was increased significantly in *T. absoluta* after the application of clove, cumin, garlic, damasiea and dill extracts the percentages of enzyme increase were +36.6, +40.2, + 63.1, + 32.3 and +21.2% in *T. absoluta* respectively.

The results also indicated that amylase and trehalase appeared as the most affected enzyme activity with high level of significant increased more than invertase enzymes.

The obtained in Table (4), shows the changes in the activity of α and β - esterases. Cloves, cumin, garlic, dill extracts revealed significant decrease in the both enzyme activity in *T. absoluta*. In cloves and cumin extracts treatment, percentages of α- esterases reduction were -87.8 and -27.4 % in *T. absoluta* respectively, while the reduction percentages of β- esterases activity were -14.2 and -23.3 % in *T. absoluta* respectively. In dill and garlic extracts treatment, percentages of α - esterase reduction were -6.7 and -7.4% in *T. absoluta* and the reduction of β- esterases were -2.4 and 6.7%. Generally, in case of *T. absoluta* treatments, the results of damasiea extract indicated high increase (+40.1% in comparison to control) in activity of beta-esterases and invertase enzymes but high decrease (+45.9%), in alpha-esterases activity only.

The results indicated that the damasiea extract had direct effects on the metabolism in the body *Tuta absoluta.*

These results are in harmony with (El-Ghar *et al*., 1995) who observed pronounced decrease in the carbohydrate hydrolyzing enzymes especially amylase and invertase after treated 5th larval instars of *Spodoptera littoralis* with sublethal concentrations of thuringeinsin (beta-exotoxin of *B. thuringiensis*). Also, Eid (2002) found Consult and Mimic decreased the invertase activity after 5 days of treatment, whereas Consult, Atabron and Cascade exhibited reduction in trehalase and invertase activities.

The activities of trehalase, invertase and amylase enzymes in larvae treated with spinosad and triflumuron were generally decreased than untreated larvae during different tested times (Mead *et al*., 2008). On the other hand, Khedr *et al*., (2005) reported that, when 4th larval instars were treated with Consult, Atabron, Match, Mimic and Cascade noticed increase in the carbohydrate hydrolyzing enzymes was recorded. Furthermore, the irregular effects of IGRs which ranged between decrease or increase during the tested time intervals was observed by (Mohamady, 2000). This contradiction in results may be due to difference in treatments, larval instar, concentrations used and tested times. The activities of trehalase and amylase were increased at the initial time intervals (after 24 hr.) than the last one (after 72 hr.). The reverse was true in the case of invertase enzyme. Abdel-Fattah *et al*., (1986) showed that the activities of the three enzymes were much higher at the initial time interval (Zero-time) than at the last one (96 hr.) at the three concentrations used of diflubenzuron and triflumuron (LC_{15}, LC_{30} and LC_{50}).

Great reduction was also showed in amylase activity of the 4th instars larvae of *S. littoralis* after fenvalerate treatment. As for invertase and trehalase enzymes activity was decreased after 48 hrs from treatment but after 72 hrs from treatment, the enzymes activities were increased (Mohamady, 2000).

Trehalase, amylase and invertase activities increased in *S. littoralis* to highest activities after 7 days of *Bacillus thuringiensis* treatment. Then these enzymes activities begin to decrease (Heba, 2005).

Table (2): Alkalin phosphatase activity of 4th larval instar of *Tuta absoluta* after treatment and control.

Treatment	Alkaline phosphatase	
	Enzyme activity (mean ± S. E)	%
Clove	1011±4[a]	+13.34
Cumin	585±20[b]	-45.4
Garlic	761.7±29[a]	-45.7
Damasiea	1047±6[c]	+17.38
Dill	1090.7±10[b]	+22.28
Control	892±5	----

Table (3): Carbohydrates hydrolyzing enzymes activities of *Tuta absoluta* treated and control.

Treatment	Trehalase		Invertase		Amylase	
	Enzyme activity (mean ± S. E)	%	Enzyme activity (mean ±S. E)	%	Enzyme activity (mean ±S. E)	%
Clove	321.7±8.7^{c}	+41.7	279.3±6.4^{b}	-21.9	101.5±2.1^{a}	+36.6
Cumin	232.0±5.6^{a}	+2.2	317.7±5.9	-11.1	104.2±1.5^{e}	+40.2
Garlic	287.0±1.7^{c}	+2.6	488.3±9.6^{a}	+36.5	121.2±1.5^{bc}	+63.1
Damasiea	360±12.1^{b}	+58.5	505.3±9.2^{a}	+14.7	98.3±1.5^{c}	+32.3
Dill	246.3±2.6^{e}	+8.5	363.7±6.1^{d}	+1.6	90.1±2.1^{f}	+21.2
Control	227.0±3.6	-	357.7±5.1	-	74.3±2.1	-

Table (4): Non-specific esterases activities of *Tuta absoluta* treated and control.

Treatment	Esterase's hydrolyzing			
	α- esterase		β- esterase	
	Enzyme activity (mean ± S. E)	%	Enzyme activity (mean ±S.E)	%
Clove	149.7±0.6^{d}	- 87.8	677.3±1.25^{b}	-14.2
Cumin	895.3±0.64^{d}	-27.4	606.0±0.5^{d}	-23.3
Garlic	1150.3±0.83^{a}	-6.7	771.0±0.53^{c}	-2.4
Damasiea	1801.3±0.64^{d}	+45.9	1107.7±0.46^{e}	+40.1
Dill	1142.3±1.33^{a}	-7.4	737.3±0.53^{c}	-6.7
Control	1234.0±0.5	-	790.3±0.32	-

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تـأثير بعـض المستخلصـات النباتيـة علـى نشـاط الإنزيمـات لحشـرة صـانعات انفـاق أوراق الطماطم توتا ابسليوتا

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تمـت هـذه الدراسـة فـي معهـد بحـوث وقايـة النباتـات الـدقي .جيـزة وذلـك لدراسـة تـأثير بعـض المستخلصات النباتية (الشبت, الكمون, القرنفل ,الثوم و الدمسيسة) على نشاط الإنزيمات في جسم العمر اليرقي الرابع لحشرة *Tuta absoluta*. وأظهرت النتائج أنه بعد تطبيق هذه المعاملات, تأثيرات متعددة على نشاط إنزيمـات الفوسـفاتيز والقاعـدى و الألفـا وبيتـا إسـتيريز بالأضـافة إلـي نشـاط إنزيمـات هضـم الكربوهيـدرات (التيريهاليز ,الأنفرتيز والأميليز) حيث وجد تأثيراً معنوياً إما بـالنقص أو الزيـادة فـي نشـاط هذه الإنزيمـات فـي بعض المعاملات. كما بينت النتائج الأتي:ازدياد نشاط إنزيم الفوسفات القلوي في حالة اليرقات المعاملة بكل من الشبت, الدمسيسـه, القرنفـل إلـى +٢٢.٢٨, + ١٧.٣٨ و+ ١٣.٣٤ مجـم % علـى التـوالي وانخفـض فـي حالـة اليرقات المعاملة بكل من الكمون و الثوم الى- ٤٥.٤ و-٤٥.٧ مجم % على التوالي . انخفض نشاط الإنفرتيز في اليرقات المعاملة بمستخلص القرنفل والكمون إلى- ٢١.٩ و-١١.١مجم% , وازداد في اليرقات المعاملة بكل من الثوم, الدمسيسه والشبت إلى+ ٣٦.٥ ,+ ١٤.٧ و+١.٦ مجم % على التوالي .- ازداد نشاط الأميليز في اليرقات المعاملة بمستخلص القرنفل ،الكمون ،الثوم ,الدمسيسه والشبت إلى+ ٣٦.٦ ,+٤٠.٢ ,+٦٣.١, +٣٢.٣ و+٢١.٢ مجم % على التوالي .

عموما يتضح أن نشاط إنزيمات الفوسفاتيز القاعدى, الألفا وبيتا إستيريز وكذلك نشاط إنزيمات و التيريهاليز ,الأنفرتيز والأميليز كان أعلى فى المعاملة بمستخلص الدمسيسة بالمقارنة بالكنترول .ولذلك يمكن استخدام مستخلص الدمسيسه في مكافحة حشرة *Tuta absoluta*. مع الأخذ في الاعتبار أن المستخلصات الأخرى أعطت نسبة موت عالية عن طريق تأثيرها على الإنزيمات ايضاً ولكن مستخلص الدمسيسه هو الأكثر تأثيراً.