EFFECT OF THE SUBLETHAL CONCENTRATIONS OF TWO PLANT EXTRACTS, MALATHION AND PIRIMIPHOS-METHYL ON SOME BIOLOGICAL ASPECTS OF *TRIBOLIUM CASTANEUM* (HERBST)

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Abstract

The effect of the sublethal concentrations of acetone extract of *Piper cubeba* fruits ,acetone and petroleum ether extracts of *Thymus vulgaris* flowering buds ,malathion and pirimiphos-methyl on some biological aspects of *T. castaneum* survivors was studied in the laboratory at 30 ± 2°C and 70 ± 5% R. H.

The results showed that all treatments decreased significantly the number of eggs laid daily per femal during two weeks by 64.6, 51.2, 32.5, 39.0 and 27.8 % for the a forementioned treatments, respectively. On the other hand, no significant differences were achieved between all treatments and control for eggs hatchability, duration of pupal stage and sex ratio. The preoviposition period was prolonged significantly with *P. cubeba* acetone extract., while, it decreased obviously with *Thymus vulgaris* acetone extract. The incubation period of eggs was increased significantly with *P. cubeba*, but it reduced significantly with *Thymus vulgaris* petroleum ether extract. Duration of larval stage and total developmental period of immature stages were significantly decreased by using the sublethal concentrations of *T. vulgaris* acetone extract, malathion and pirimiphos-methyl.

INTRODUCTION

The post harvest losses caused by insect damage, transportation, microbial deterioration, method of storage, and other factors are estimated to about 10-25% world-wide. Synthetic chemicals are currently the method of choice to protect grains from insect damage, however their continuous usage has been led to the development of pest resistantce and the problems of toxic residues (Ahmed *et. al.*1981, Zettler and Cuperus 1990).

The bioactivity of several plant extracts, dusts and plant oils as pest control agents against stored product pests was studied by many investigators (Yung and Buvkholder, 1981, Yadav, 1984 Abo El-Ghar and El-Sheikh,1987, El-Lakwah *et. al.* 1992, 1993 a&b and 1995 and Abd El-Aziz 2002).

The effect of air containing 13% volatile substances from crushed seeds of *Azadirachta indica* (neem) on the reproductive biology of newly emerged adults of *Tribolium castaneum* on rice grains was tested .The volatile substances reduced the number of eggs laid by females by 27.5%, prolonged the development period from egg to larvae by 2.5 days and reduced the number of larvae emerging from eggs by 22.8%. Newly hatched to 12 days old larvae exposed to the volatile substances failed to develop further, whereas treated larvae more than 13 days old successfully completed their development into adults (Maheswaran and Ganesalingam 1988).

The effectiveness of organophosphorus insecticides against stored product insects was studied by various investigators (Abo El-Ghar and Badawy, 1961, Barbara and Linda, 1983, Patourel and Tayeb, 1988 and El-Lakwah *et. al.* 1993a).

The present work was carried out to study the effect of certain biological aspects of *Tribolium castaneum* adults treated with two plant extracts (namely acetone extract of *Piper cubeba* fruits, acetone and petroleum ether extracts of *Thymus vulgaris* flowering buds), malathion and pirimiphos – methyl.

MATERIALS AND METHODS

The effect of sublethal concentrations causing around 50-60% *T. castaneum* adult mortalities ,of acetone extract of *Piper cubeba* fruits, acetone and petroleum ether extracts of *Thymus vulgaris* flowering buds, malathion and pirimiphos-methyl on some biological aspects of *T. castaneum* was studied in the laboratory at 30 ± 2°C and 70 ± 5% RH.

The adults of *T. castaneum* (0-2 days-old) were exposed to each plant extract at 4% w/w concentration for 5days, and to 4ppm malathion, 3ppm pirimiphos- methyl (Actellic) for 3days only.These concentrations caused around 5o- 6o% insect mortality at the aforementioned of treatment. Then certain biological aspects of survivors were estimated.

Following parameters were recorded :average number of eggs laid daily per female, average total number of eggs laid per female during 2 weeks, pre-oviposition period, incubation period of eggs, hatchability of eggs, average duration of larval stage, average duration of pupal stage, total developmental periods of immature stages and sex ratio.

1- Number of eggs:

Tests were carried out to assess number of eggs laid by females during an observation period of 14 days. Unmated males and females after treatments were

paired in a glass tube (3 × 5 cm) containing a small amount of wheat flour and covered with muslin. Every day the number of eggs laid in the flour was counted and the flour was replaced. The total number of eggs laid by female during 14 days was recorded. Hatching of eggs was also recorded for determining its viability. Each treatment was replicated four times.

2-The incubation period and the developmental span:

For assessment the incubation period of eggs and the developmental periods for the various stages, one day-old eggs were taken and only one egg was put on an amount of wheat flour in glass tube (3×5cm) and then covered with muslin. Ten replicates were used for each treatment. The incubation period for every egg was recorded. The developmental stages were observed and their developmental periods were also estimated. Furthermore, larval mortality rate was calculated.

Statical analysis

The obtained data were statistically analysed at 5% probability level accordto Sendeing cor and Cochran (1967).

RESULTS AND DISCUSSION

The effect of the sublethal concentrations (causing around 50-60% adult mortality) of acetone extract of cubeb fruits (*Piper cubeba)*, acetone and petroleum ether extracts of *Thymus vulgaris* flowering buds, malathion and pirimiphos-methyl on some biological aspects of *T. castaneum* were studied in the laboratory at 30 ± 2°C and 70 ± 5 R. H. The obtained results are given in Tables (1 & 2 and 3).

Table 1. Number of eggs laid daily per female of *T. castaneum* adults exposed to the sublethal concentrations of two plant extracts, malathion and pirimiphos-methyl at 30 ± 2°C and 70 ± 5% R. H.

Treatment / Day	P. cubeba Acetone extract	T. vulgaris		Malathion	Pirimiphos-methyl	Control	L.S.D.
		Acetone extract	Petroleum ether extract				
	Mean number of eggs laid daily per female ±S.E.						
1	00	0.0	0.0	7.25±2.7	1.25±1.0	3.75±2.1	-
2	00	0.0	0.0	2.5±1.5	1.5±1.0	6.5±3	-
3	0.0	0.0	0.0	4.25±2.3	8.5±1.5	5.25±2	-
4	0.0	0.0	0.75±0.5	8.5±3.3	6.5±0.9	16±3	-
5	1.5±0.7	0.0	4.75±2.2	4.25±2.2	4.25±1.8	9.75±2.7	-
6	3.5±1.0	1.5±0.7	6.0±1.9	4.25±2.7	4.5±1.9	7±2.4	-
7	6.25±1.8	2.75±0.8	8.75±0.7	7.25±2.6	9.25±2	8.5±2.4	-
8	7.00±7.1	6.5±1.3	12.25±2.0	7.25±3.6	8.5±2.5	8.25±2.7	-
9	8.25±1.6	8.75±1.9	12.5±1.5	5.25±1.9	12.5±1.5	14±0.7	-
10	7.00±0.9	11.25±1.5	10.5 ± 1.0	12.75±2.4	10.75±1.0	11±0.4	-
11	5.25±0.7	9.75±1.0	11.25± 2.8	8.75±1.5	11.25±1.1	10.75±0.8	-
12	4.25±1.1	10.25±1.1	10.5± 4.3	7.75±2.3	11.25±2.4	11.0±2.1	-
13	5.8 ±1.3	12.25±1.3	9.25 ±3.8	5.75±2.2	9.75±0.8	21.25±1.7	-
14	5.00±1.9	11.25±1.3	16.00± 3.6	6.75±3.7	9.75±0.9	18.75±3.2	-
Mean No. of eggs laid female\day	3.82*± 0.6	5.29*±0.2	7.31*±1.0	6.6*±1.8	7.81*±0.7	10.84±0.8	2.9
Total No. of eggs laid per female during two weeks	53.75* ±1.5	74.25*±2.6	102.5*±2.7	92.5*±1.3	109.5*±1.8	151.75±2.6	4.8
Percent decrease in egg numbers	64.6%	51.2%	32.5%	39.0%	27.8%	-	-
Hatchability rate of eggs (%)	72.00± 2.2	73.00±3.4	66.00±4.8	63.0±4.1	74.0±2.9	79.0±4.8	NS

- Significant differences at 5% level.
  NS = Non significant.
  L. S. D. = Lethal significant differences.

Table 2. Number and duration of various larval instars of *T. Castaneum* for the different treatments at 30 ± 2°C and 70 ± 5% R. H.

Treatment / Larval stage	*P. cubeba* acetone extract	*T. vulgaris*		Malathion	Pirimiphos-methyl	Control	L.S.D
		Acetone extract	Petroleum ether extract				
	Mean duration of larval instars (days) ± S.E.						
First	3.25±0.7	4.50±0.7	5.00±0.7	5.00±0.6	4.25±0.8	4.00±0.4	NS
Second	5.00*±0.4	2.50*± 0.3	4.00±0.4	2.00*±0.0	2.75*±0.3	4.00±0.4	0.9
Third	3.50±0.7	2.25*±0.5	2.75*±0.5	2.25*±0.3	3.50±0.3	4.25±0.7	1.3
Fourth	6.00*±0.7	3.25±0.5	2.00*±0.0	2.25±0.3	2.50±0.5	3.50±0.5	1.4
Fifth	5.75±1.2	3.50±0.5	5.25±1.1	5.25±1.2	3.75±0.5	4.25±0.5	NS
Sixth	3.25±2.0	-	2.00±1.2	1.50±0.9	-	2.75±1.0	NS
Seventh	-	-	-	-	-	1.75+1.0	1.2
Larval stage period	26.75±2.1	16.00*±0.4	21.00±0.4	18.25*±0.7	16.75*±0.0	24.50±2.2	3.8

* = Significant differences at 5% level.
NS =Non significant.

Table 3. Effect of sublethal concentrations of two plant extracts, malathion and pirimiphos-methyl on certain biological aspects of *T. castaneum* at 30 ± 2°C and 70 ± 5% R. H.

Treatment / Biological parameters	*P. cubeba* acetone extract Mean±S.E	*T. vulgaris*		Malathion Mean±S.E.	Pirimiphos-methy Mean±S.E.	Control Mean± S.E.	L.S.D.
		Acetone extract Mean±S.E.	Petroleum ether extract Mean±S.E.				
Pre-oviposition period (day)	9.75*±1.5	2.50*±0.7	8.75*±0.9	5.00±0.4	7.75±0.9	6.25±1.0	2.7
Incubation period of eggs (day)	5.00*±0.0	3.54±0.3	5.75*±0.5	4.00±0.0	3.50±0.3	3.25±0.3	0.8
Duration of larval stage (day)	26.75±2.1	16.00*±0.4	21.00±0.4	18.25*±0.7	16.75*±0.0	24.50±2.2	3.8
Duration of pupal stage (day)	4.50±0.3	4.50±0.5	6.25±0.6	5.25±0.5	5.00±0.5	4.75±0.5	NS
Mortality of larval stage(%)	33.30	16.70	16.70	0.0	0.0	0.0	-
Total developmental period of immature stage (day)	36.25±2.1	24.00*±0.8	33.00±1.6	27.50*±0.3	25.25*±0.8	32.50±2.6	4.7
Sex ratio	1:1	1:1	1:1	1:1	1:1	1:1	-

* = Significant differences at 5% level.
NS = Non significant

1-*Piper cubeba* fruits acetone extract:

The results revealed that treatment of *T. castaneum* adults with the sublethal concentrations of Cubeb fruits acetone extract reduced significantly number of eggs laid daily per female during an observation period of two weeks by 64.6%. Also, pre-oviposition period and incubation period of eggs, the durations of second and fourth larval instars were obviously prolonged in comparison to untreated insects. Meanwhile, the larval mortality in the treatment reached 33.3%. On the other hand, no significant differences were found between the treatment and the control for egg hatchability, duration of the larval stage, pupal stage and total developmental period of immature stages and sex ratio.

2- *Thymus vulgaris* flowering buds extracts:

Treatment of *T. castaneum* adults with the sublethal concentrations of *T. vulgaris* flowering buds acetone extract resulted in lower egg- production than control during an observation period of two weeks by 51.2%. Also, pre-oviposition period, the durations of second and third larval instars, larval stage period and total developmental period of immature stages were significantly decreased in comparison to the control. Meanwhile, no significant differences between the treatment and control for incubation period of eggs, egg hatchability, pupal stage period and sex ratio were observed. Similarly, no significant differences were found between the treatment and the control for egg hatchability, pre-oviposition period, larval stage period, pupal stage period total developmental period of immature stages and sex ratio, when *T. castaneum* adults were treated with the sublethal concentration of petroleum ether extract of this plant. On the other hand, egg-production, and third and fourth larval instars durations were significantly decreased for petroleum ether extract of *Thymus vulgaris* compared with the control.

3- Malathion:

The results indicated that treatment of *T. castaneum* adults with sublethal concentration of malathion decreased significantly number of eggs laid per female, durations of second and third larval instars, period of larval stage and total developmental period of immature stages.

Contrarily, no significant differences were achieved between the treatment and the control for pre-oviposition period, incubation period of eggs, pupal stage period, eggs hatchbility, sex ratio and larval mortality.

4- Pirimiphos-methyl (Actellic):

Treatment of *T. castaneum* adults with the sublethal concentration of pirimiphos-methyl showed significantly lower egg- production than control. At the same time, significant decrease was obtained between the treatment and the control

for the duration of second larval instars, period of larval stage and total developmental period of immature stages. On the other hand, no significant differences were found between the treatment and the control for pre- oviposition period, incubation period of eggs, egg hatchability, pupal stage period, sex ratio and larval mortality.

In this respect, Maheswaran and Ganesaling (1988) studied the effect of air containing 13% volatile substances from crushed seeds of *Azadirachta indica* (neem) on the reproductive biology of newly emerged adults of *Tribolium castaneum* on rice grains. The volatile substances reduced the number of eggs laid by females by 27.5%, prolonged the development period from egg to larva by 2.5 days and reduced the number of larvae emerging from eggs by 22.8%. Newly hatched to 12-day-old larvae exposed to the volatile substances failed to develop further, whereas treated larvae more than 13 days old successfully completed their development into adults.

White and Sinha (1990) mentioned that the exposure of adults of the cucujid *Cryptolestes ferrugineus*, the silvanid *Oryzaephilus surinamensis* and the tenebriond *Tribolium castaneum* to filter papers treated with the organophosphorus insecticides malathion or chlropyrifos-methyl often affected offspring production in survivors. When groups of 5 male and 5 female survivors were placed on ground wheat at 30°C, *C. ferrugineus* and *O. surinamensis* adults produced fewer offspring with prolonged exposure to chlorpyrifos-methyl but malathion had no effect.

Tribolium castaneum adults produced significantly more offspring with increasing exposure to both malathion and chlorpyrifos-methyl, indicating stimulation of oviposition or a selection for increased fitness in survivors.

Also, the obtained results are in harmony with the findings of Spratt,(1984) and Ho *el, al.* (1995).

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تأثير التركيزات تحت المميته لمستخلصين نباتيين وكذلك الملاثيونوبيريميفوس-ميثيل على بعض الصفات البيولوجية لحشرة خنفساء الدقيق الكستنائي

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عند معاملة الحشرات الكاملة لخنفساء الدقيق الكستنائيه بالتركيزات تحت المميته (التى تعطى نسب موت تتراوح ما بين ٥٠-٦٠%) من المستخلص الأسيتونى للكباب الصينى والمستخلص الاسيتونى والاثير البترولى للبراعم الزهرية للزعتر وكذلك مبيدى الملاثيون والأكتلك فى المعمل على درجة حرارة ٣٠ ± ٢٥م، ٧٠% رطوبة نسبية، أظهرت النتائج انخفاضا معنويا فى عدد البيض الذى تضعه الا نثى يوميا خلال فترة أسبوعين مقارنة بالكنترول، وكانت نسبة الانخفاض فى أعداد البيض هى ٦٤,٦، ٥١,٢، ٣٢,٥، ٣٩,٠، ٢٧,٨% لكل من المستخلص الاسيتونى لثمار الكباب الصينى والمستخلص الاسيتونى للزعتر ،ومستخلص الاثير البترولى للزعتر، ومبيد الملاثيون، ومبيد البيريميفوس-ميثيل على التوالى.

بينما لم تظهر أى فروق معنوية بين المعاملات المختلفة والكنترول فى نسبة الفقس للبيض، وفترة طور العذراء ، والنسبة الجنسية، وكانت نسب الموت فى الأعمار اليرقية ٣٣,٣،١٦,٧، ١٦,٧، صفر، صفر% لكل المعاملات المختلفة على التوالى.

ونتج عن معاملة الحشرات الكاملة لخنفساء الدقيق الكستنائية بالتركيز تحت المميت من المستخلص الأسيتونى لثمار الكباب الصينى زيادة معنوية فى فترة ما قبل وضع البيض، وفترة حضانة البيض، وفترة العمر اليرقى الثانى والرابع مقارنة بالكنترول، ولكن لم تظهر أى فروق معنوية بين هذة المعاملة والكنترول فى فترة الطور اليرقى وكذلك فترة النمو الكلية للأطوار الغير كاملة.

وقد أعطى التركيز تحت المميت لمستخلص الأسيتون للبراعم الزهرية للزعتر انخفاضا معنويا فى فترة ما قبل وضع البيض، وفترة الأعمار اليرقية الثانى والثالث وفترة الطور اليرقى وكذلك مدة التطور الكلية للأطوار الغير كاملة لخنفساء الدقيق الكستنائية مقارنة بالكنترول، بينما لم تظهر أى فروق معنوية بين المعاملة والكنترول فى فترة حضانة البيض.

كما تبين وجود انخفاض معنوي فى فترة حضانة البيض، وفترة الأعمار اليرقية الثالث والرابع باستخدام التركيز تحت المميت لمستخلص الاثير البترولى للبراعم الزهرية للزعتر، بينما لم تظهر أى فروق معنوية مع فترة ما قبل وضع البيض، وفترة الطور اليرقى، والفترة الكلية لتطور الأطوار الغير كاملة.

كما أظهرت النتائج أيضا أنه بمعاملة الحشرات الكاملة لخنفساء الدقيق الكستنائية بالتركيز تحت المميت للملاثيون والأكتلك قد أدت الى انخفاض معنوي فى فترة الطور اليرقى، والفترة الكلية لتطور الأطوار الغير كاملة، بينما لم تظهر أى فروق معنوية بين المعاملات المختلفة والمقارنة فى فترة ما قبل وضع البيض، وفترة حضانة البيض لحشرة خنفساء الدقيق الكستنائية.