

EFFICACY OF SOME HONEY BEE PRODUCTS AGAINST THE SPIDER MITE, *Tetranychus urticae* KOCH

Abd–El Halim, Salwa M.

plant protection Dept., Fac. of Agriculture, Fayoum Univ, Fayoum, Egypt

ABSTRACT

The efficiency of propolis and dark honeybee comb were studied against egg and adult female stages of phytophagous mite, *T.urticae* Koch. at constant temperature (25 ± 1 C°) and relative humidity ($70 \pm 5\%$). Data showed that the propolis extract appears more effective against *T.urticae* eggs (LC50 0.52 g/ml) than dark honeybee comb (LC50 0.84 g/ml), while the dark honeybee comb extract was more effective than propolis extract (LC50 0.38 and 1.12 g/ml), respectively against adult females. The biological aspects of *T.urticae*, incubation period of eggs, life cycle, generation period, adult female longevity and female fecundity were affected by treating eggs and adult females with LC50 value of the propolis and dark honeybee comb extracts.

INTRODUCTION

The Phytophagous mite, *Tetranychus urticae* Koch is one of the most economically important mite species of wide spread on different hosts all over the world. Continuous application by using acaricides on agriculture crops causes many problems such as development of resistant strains and destroying natural enemies, beside residue contamination of humanities, mammalian toxicity and pollution of the environment.

Therefore, there is a vital interest in discovering natural substances and using it as pesticides without harming the environment.

Propolis is a natural product collected by honeybees and used in the construction and protection of the beehive (Ghisalberti 1979). It was effective against number of bacterial strains, viruses and fungus (Harman 1983, Lisa et al. 1989, Davey 1990, Dobrowolski et al. 1991, Amoros et al. 1992a, 1992b and Liska 1994), and it was effective to *Varroa destructor* (Anderson and Trueman) mites (Assegid et al 2002). In Egypt, Hussein et al. 1989, found repellency effect of propolis extract against cowpea seed beetle *Callosobruchus maculatus* Fab., also propolis extract showed a repellent effect of *Varroa jacobsoni* Oud. (Mohamed et al., 2001). On the other hand, propolis and dark honeybee comb extracts were effective to root-knot nematode *Melodogyne javanica* (Abou-zeid et al., 1994) and according to obtained results by Medhat 1998, adding dark honeybee comb to the soil, can decrease the harmful effect of *M. javanica* which infects tomato plants and increase the plant growth.

The present study was conducted to investigate the effect of propolis and dark honeybee comb extracts against eggs and adult females of *T. urticae*. Consequently, the changes occurring in the biological aspects after treatment with the previous extracts at LC50 levels should assume considerable importance.

MATERIALS AND METHODS

Rearing technique of mites:

A pure culture of spider mite, *T. urticae* was reared on castor bean *Ricinus comunis* (L.) leaves in an incubator at $25 \pm 1^\circ\text{C}$ and relative humidity ($70 \pm 5\%$)

Extraction procedure

Samples of propolis and dark honey bee combs (old comb) were obtained from the apiary of faculty of agriculture at Fayoum University.

I- Propolis extraction

Fifteen grams of propolis extracted were cut in small pieces and mixed with 150 ml of 96 % ethanol. The mixture was then kept in a brown dark bottle and shaken several times daily for five days. There after, the mixture was filtered through filter paper Watman No.1. The filtrate became a thick highly viscous which turned into a gummy matter when the ethanol evaporated (Boeru and Derevici, 1976).

II- Dark comb (old bees wax) extract

Dark comb used in the present study were over four years old. These combs were placed in a refrigerator for 48 hours, then, they were broken down into very small pieces using a sharp cutter. Extracts were obtained by mixing 500 gm of dark comb pieces with 1500ml. of 96% ethanol. The mixture was shaken several times a daily for five days, then filtered through a piece of Nylon cloth, afterwards, through filter paper Watman No. 1.

The extracts of propolis and dark comb were arbitrarily termed as standard (S), and dissolved in 10 ml distilled water each, and one drop of betweeen 80was added and mixed well.

Toxicity Tests

Ovicidal action:, Twenty adult females of *T. urticae* were allowed to lay eggs on the lower surface of castor bean leaf discs (2.5 cm in diameter). Four discs were placed in each Petri dish on moist cotton and considered as four replicates. The females were removed after 24 hours and the deposited eggs were counted. Castorbean leaf discs were treated by dipping technique for 20 sec. and left to dry. Four concentrations of each one were used to calculate the toxicity line. The slope of the Ldp.line, LC 50 and LC 90s were determined according to Finney (1952). The numbers of non-hatched eggs were counted and percentage of mortality was corrected by using abbott's formula (1925).

Toxicological effect on the adult female: Castorbean leaf discs (2.5 cm diameter) were treated by dipping technique for 20 sec. and left to dry. Twenty adult females in the same age were transferred on each leaf disc. Four concentrations of each extracts were used as indicated before in eggs treatment. Each treatment was replicated four times.

Effect of propolis and dark comb extracts on the biology of *T. urticae*

Changes in the biology of *T. urticae* were determined after treating eggs and adult females with propolis and dark comb extracts at the LC50 level.

Data concerning the changes occurring during the ontogeny of *T. urticae* was investigated incubation period, life cycle, generation period, longevity and fecundity were recorded.

Statistical analysis:

The obtained data were statistically analyzed for standard deviation (S.D.) and L.S.D. test was used to estimate the significant differences between the untreated and treated groups.

All experiments were incubated under controlling of 25 ± 1 °C and $70 \pm 5\%$ R.H.

RESULTS AND DISCUSSION

Toxicity of Propolis and Dark honeybee comb Extract on Eggs and Adult female stages of *T. urticae*.

1.1-The effect on hatchability

Data in Table1 indicated that propolis extract was more toxic than dark honey bee comb extract at LC50 and LC90 levels gave 0.52 and 1.88 g/ml, respectively, while dark honey comb extract was the least effective showing LC50 of 0.84 and LC90 of 3.92 g/ml. The relative potency levels at LC50 and LC90 show that propolis extracts was 1.62 and 2.08 times as toxic as dark honey bee comb extract, respectively.

1.2- The effect on mortality

As shown in Table 1 dark honey bee comb was more active than propolis extract against adult stage *T. urticae*. At the LC50 and LC90 values dark honey bee comb gave 0.38 and 2.18 g/ml whereas propolis extract was the lower toxic indicating LC50 of 1.12 and LC90 of 6.56 g/ml.

The relative potency levels at LC50 and LC90 exhibit that dark honey bee extract was 2.95 and 3.01 times as toxic as propolis extract, respectively as shown in Table 1.

Table 1: Toxicity of Propolis and Dark honey bee comb Extracts on Egg and Adult female stages of *T. urticae*

Extract	LC50 g/ml	LC90 g/ml	Slope	Toxicity Index(%)		Relative Potency	
				Lc50	lc90	Lc50	lc90
Eggs							
Propolis	0.52	1.88	1.04	(100)	(100)	1.62	2.08
Dark honey bee comb	0.84	3.92	0.57	61.90	47.95	(1.00)	(1.00)
Adult females							
Dark honey bee comb	0.38	2.18	0.71	(100)	(100)	2.95	3.01
Propolis	1.12	6.56	1.31	33.92	38.58	(1.00)	(1.00)

Effect of Propolis and Dark honeybee comb Extracts on the Biological Aspects of *T.urticae*.

After treating *T.urticae* eggs and adults with LC50 concentrations of propolis and dark honeybee comb extracts, changes in the biology were determined.

After egg treatment: The results in Table 2 showed that the propolis treatment significantly increased the incubation period of the eggs (5.00 days) more than dark honeybee comb (3.90days) compared with 3.7 days for untreated eggs. Treated eggs resulted in considerable prolongation of the

lifecycle as well as generation periods of *T.urticae*, reaching to 13.30 and 14.70 days for propolis, 10.70 and 12.40 days for dark honeybee comb as compared with 9.20 and 10.14 days for control, respectively.

As shown in Table 2, it was found that the propolis and honeybee comb extracts shortened the female adult longevity of *T.urticae* . significantly, the means being 4.80 and 6.60 days , respectively, compared with a mean of 17.30 days for the control.

Data also showed that the total number of deposited eggs/ female were highly affected when using the same extracts, the means being 15.50 and 23.20 eggs/ female as compared with control which was 87 eggs.

It is interesting to note that in case of egg treatment with propolis extract, six females from a total number seventeen didn't lay any egg, while longevity of females varied between (1-5 days).

Table 2: Effect of Treatment with Propolis and Dark honey bee comb Extracts on Biological Aspects of *T. urticae* Eggs

Treatment	Average period ± SD in Days				
	Incubation	Life cycle	Generation	Longevity	Fecundity
Propolis	5.00 ± 0.20	13.30 ± 0.90	14.70 ± 0.78	4.80 ± 0.87	15.50 ± 12.12
Dark honey bee comb	3.90 ± 0.30	10.70 ± 1.00	12.40 ± 1.11	6.60 ± 2.15	23.20 ± 17.64
Un-treated	3.70 ± 0.46	9.20 ± 0.41	10.14 ± 0.92	17.30 ± 2.69	87.00 ± 12.15
L.S.D.	0.27	0.74	0.89	2.32	13.05

After adult treatment: As shown in Table 3 the propolis and dark honey bee comb treatments reached to 5.10 and 5.40 days, respectively, significantly increased the incubation period of the eggs as compared with control(3.70 days). The life cycle of *T. urticae* was significantly prolonged with the treatment of the previous extracts, which averaged 12.20 and 13.80 days for propolis and dark honey bee comb, respectively, compared with 9.20 days from untreated females. Longevity of females was significantly shortened to 6.80 and 6.30 days for the same extracts, respectively compared with control check (17.30 days). Dark honey bee extract was significantly more effective on female fecundity (17.70 eggs/ female) than propolis extract (32.80 eggs/ female) compared with a mean of 87 eggs / female for the control.

Table 3: Effect of Treatment with Propolis and Dark honey bee comb Extracts on Biological Aspects of *T. urticae* Adults

Treatment	Average period ± SD in Days				
	Incubation	Life cycle	Generation	Longevity	Fecundity
Propolis	5.10 ± 0.30	12.20 ± 0.60	14.00 ± 0.63	6.80 ± 1.08	32.80 ± 7.56
Dark honey bee comb	5.40 ± 0.49	13.80 ± 0.87	15.50 ± 0.81	6.30 ± 1.01	17.70 ± 12.32
Un-treated	3.70 ± 0.46	9.20 ± 0.41	10.14 ± 0.92	17.30 ± 2.69	87.00 ± 12.15
L.S.D.	0.49	0.61	0.79	1.68	11.67

For available literature, Abouzeid *et al* 1994, revealed that propolis and old honey bee comb (dark comb) caused highest mortality rate to juveniles of *Meloidogyne incognita*, *M. javanica* and *Rotylenchulus reniformis*.

Assegid *et al* 2002, found that treatment of *varroa destructor* (Anderson and Trueman) mites with propolis has displayed both narcotic and lethal effects. The length of narcosis and rate of mortality showed dependence on the concentration of propolis used, the duration of contact time and the extraction procedure.

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**فعالية مستخلصات البروبوليز و اقراص شمع نحل العسل الداكنة ضد العنكبوت الاحمر تترانيكس يورتيكا
سلوى محمد عبد الحليم
قسم وقاية النبات - كلية الزراعة - جامعة الفيو**

اجريت دراسة فاعلية مستخلصات البروبوليز و اقراص الشمع القديمة على كل من طورى البيض و الاناث البالغة لكاروس العنكبوت الاحمر العادى عند درجة حرارة $25 \pm 10^{\circ}C$ و رطوبة نسبية $70 \pm 5\%$.
و اظهرت نتائج السمية ان مستخلص البروبوليز كان اكثر فاعلية على البيض بينما المستخلص الشمعى كان اكثر فاعلية على الاناث البالغة.
وكانت التركيزات التى تسبب موت 50% من الافراد هي 0.25 و 0.84 بالنسبة لطور البيض, 0.38 و 1.12 بالنسبة لطور الاناث البالغة و ذلك عند المعاملة بمستخلصى البروبوليز و الشمع على الترتيب, كما تائرت الطواهر البيولوجية عند معاملة البيض والاناث البالغة للعنكبوت الاحمر العادى بالتركيزات التى تسبب موت 50% فقد زادت فترة حضانة البيض و طالت دورة الحياة كما قصرت فترة حياة الانثى و قلت خصوبتها.

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